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Allelopathic potential of weed extracts on seed germination and seedling growth of *Vigna radiata* L. Wilzek and *Vigna mungo* L.

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Abstract: **The present investigation was carried out to evaluate and compare the effects two weed on the germination indices and seedling growth of *Vigna radiata* (Green gram) and *Vigna mungo* (Black gram). Fresh weight and dry weight of tested plant samples were also determined. Differential inhibitory effect was observed for two weed plants on two tested crops. Germination energy, speed of emergence, germination index, coefficient ratio of germination and seed vigour index of all the extracts treated sets decreased as compared to control sets. But mean germination time and phytotoxicity were increased in all the treated sets.**

Keywords: Allelochemicals, **Germination, Seedling growth.**

I. INTRODUCTION

Allelopathy has its importance in influencing plant growth both in natural and agricultural ecosystem (Chatterjee et al. 2012). Allelopathy is considered as direct or indirect chemical effects exerted by one plant germination, growth, or development of the other neighboring plant (Mishra, 2015). Weeds are plants those have potential to enter into disturbed or cultivated habitats occupied by man and inhibit or replace the native plant populations or cultivated plants (Navas, 1991). According to Batish et al (2007) plant extracts and plant residues of soil most commonly contain allelopathic substances. The allelochemicals are found in different plant parts such as leaves, flowers, roots, fruits, stem, rhizomes and seeds. In natural and agricultural systems they are released into the soil through root exudation, leaching, volatilization and decomposition of plant residues (Sisodia and Siddiqui, 2010). The allelochemicals released by weeds reported to decrease the seed germination and growth of many crops (Batish et al. 2005). Thus weed has impotent implication on agriculture and may somehow adversely affect productivity of crop plants. Germination behaviour and other physio-biochemical responses of test species are considered as some common indices for assessment of allelopathic action of plants or plant parts. (Bhattacharjee et. al. 2003). The present study was carried out to evalute the effect of aqueous extracts of *Lantana camara* L. and *Clerodendrum viscosum* on *Vigna mungo* and *Vigna radiata*. *Lantana camara* belong to family Verbenaceae and considered as noxious and invasive weed. It is one of the worst weeds of the world, and also widely distributed in India (Mishra, 2015). *Lantana camara* reported to inhibit or suppress germination rate, growth and development of crops by releasing allelochemicals to the rhizosphere of adjacent crop plants (Qasem, 2006). *Clerodendrum viscosum* was recorded as weed species from the tea plantations of Meghalaya (Sen et al. 2016). *Clerodendrum viscosum* of Verbenaceae family is distributed throughout India as undergrowth is usually known to very common crop field weed (Ghosh, 2012). It was observed that leaf extract of *Clerodendrum viscosum* inhibited seed germination and the effect increased with concentration (Datta and Chakraborti, 1975).

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Vigna mungo (L.) Hepper is commonly known as black gram or urdbean of family Fabaceae .It is short duration crop and one of the important crops cultivated extensively in India. *V. mungo* is usually sown in the late rainy season (August/September). Thus the crop may be seriously damaged due to heavy infestation of weeds (Prasada Babu et al. 2014). Mungbean (*Vigna radiata* (L.) Wilczek), also known as green gram belongs to Fabaceae family is mainly grown either as a subsistence monocrop or intercrop during kharif season.in India. It is typically warm season crop. Mungbean is a nutritious grain legume and constitutes an important source of cereal based diets throughout India (Gupta and Pratap 2016).

II. MATERIALS AND METHODS

Certified seeds of *Vigna radiata* (L.) Wilzek var PDM-139 and *Vigna mungo* (L.) var PU-31 were procured from State Agriculture Research Station, Tripura (west). Aqueous extracts of whole plant of *Lantana camara* L. and *Clerodendrum viscosum* vent were evaluated for their effects on seed germination and seedling growth of two tested crop plants.

A. Plant sampling and preparation of extracts:

Lantana camara L. and *Clerodendrum viscosum* vent plants were collected from MBB College campus (N 23˚82′77″, E 091˚29′88″). Plant parts were washed several times with water. Collected plant parts were dried, grounded and sieved. 1% aqueous extracts of the powered parts were prepared and stored in bottles. The extracts filtered through muslin cloth and stored in dark bottles and properly labeled (Joshi and Joshi, 2016). Extracts were marked as EL-Extracts of *Lantana camara* and ECl-Extracts of *Clerodendrum viscosum*. For all the experimental sets the seeds were presoaked in water for 6 hours. In case of treated experimental sets seeds were primed in weed extract for 20 minutes after presoaking in water.

B. Pot experiment:

Germination trays were filled with soil mixture (clay: sand in ratio of 3:1). Pre-soaked seeds were sown in the prepared soil at a depth of 0.5 to 1.0 cm in each tray for two tested crops and control sets. Experimental sets were irrigated with prepared aqueous extracts on every alternate day. The control sets were irrigated with water. Two replicates were prepared for each experimental set with complete randomized block design. Germination percentage was determined on 3^{rd} day, 5^{th} day, 7^{th} day & 10^{th}day using following formula (Javed and Panwar, 2013). Germination percentage and the lengths of epicotyl, hypocotyl and seedlings were estimated on the basis of data collected. Germination was daily monitored and emergence of radical from seeds was considered as criteria for germination (International Seed Testing Association, 1999). Seeds germinated in each day were marked. At 10th day, fresh weight and dry weight of seedlings (mg) were also determined. Experimental sets were marked as follows:-

EC1 - *V. mungo* control set; EC2 – *V. radiata* control set; EL1 – *V. radiata* treated with *L. camara* extract; EL2 – *V. mungo* treated with *L. camara* extract; ECl1 – *V. radiata* treated with *C. viscosum* extract; ECl2 – *V. mungo* treated with *C. viscosum* extract.

The potential allelopathic effects of extracts of donor species on germination of seeds of target plants were evaluated considering the following parameters:

Germination energy (GE) was calculated as

$$GE = \left(\frac{\text{Percentage of germinated seeds at the starting day of germination}}{\text{Total number of seeds set for bioassay}}\right) \text{x } 100$$

Modified from Ruan et al. (2002).

Speed of emergence (SE) $SE = \left(\frac{\text{Number of germinated seeds at the starting day of germiantion}}{\text{Number of germinated seeds at the final days of measurement}}\right)$ x 100

Islam et al. (2009).

Relative germination ratio (RGR) was calculated by the following formula

$$RGR = \left(\frac{G}{Gr}\right) \text{x } 100$$

Where, G the germination ratio of tested plant, and Gr is the germination ratio of control.

Rho and Kil (1986).

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Germination index (GI) was calculated as follows:

$(GI) = \sum \frac{GT}{Tt}$ or

$$GI = \frac{\text{Number of germinated seeds}}{\text{Day of first count}} + \frac{\text{Number of germinated seeds}}{\text{Day of second count}} + \cdots + \cdots + \frac{\text{Number of germinated seeds}}{\text{Days of final or last count}}$$

AOSA (1983)

Following formula was used for calculation of Coefficient of the rate of germination (CRG)

$$CRG = \left[\frac{N1+N2+N3+N4+\cdots+Nn}{(N1xT1)+(N2xT2)+(N3xT3)+(N4xT4)+\cdots+(NnxTn)}\right] \text{ x } 100$$

where N_1 =number of germinated seeds on time T_1 , N_2 =number of germinated seeds on time T_2 , Nn =number of germinated seeds on time Tn

Bewley and Black (1985) and Chiapusio et al. (1997).

The formula used for Mean germination time (MGT) was:-

$$MGT = \frac{\sum TiNi}{S}$$

Where, Ti=Number of days after beginning of the experiment, Ni=Number of seeds germinated on the day, S=Total number of seeds germinated

Ellis and Roberts (1981)

Seedling vigour index (SVI) $SVI = \left(\frac{\text{Seedling length (mm)x Germination percent}}{100}\right)$ Islam et al. (2009).

The elongation ratio were calculated by the following equations

$$Rs = \left(\frac{Ms}{Mc}\right) \text{x } 100$$

Where, Rs is the relative elongation ratio of shoot, Ms the mean shoot length of tested plant, Mc the mean length of control.

Rho and Kil (1986).

$$Rr = \left(\frac{M}{Mc}\right) \text{x } 100$$

Where, Rr is the relative elongation ratio of root and M the mean root length of tested plant, Mc the mean length of control.

Rho and Kil (1986).

Tolerance index (TI) was calculated as

$$TI = \left(\frac{\text{Longest root in treatment}}{\text{Longest root in control}}\right) \text{ x } 100$$

(Turner and Marshal, 1972).

The percentage of the toxicity of the seeds was calculated by the following formula

$$\text{Phytotoxicity } \% = \left(\frac{\text{Radicle length of the control} - \text{Radicle length of the treated sample}}{\text{Radicle length of the control}}\right) \text{x } 100$$

(Chiou and Muller 1972).

Percentage of inhibition or stimulation was calculated as

$$\text{Percentage of inhibition or stimulation } = \left(\frac{Lt - Lc}{Lc}\right) x100$$

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Where Lt is the length of hypocotyl, epicotyl or seedling of treated target plant, Lc is the corresponding length of control. The negative values indicate to inhibition (or phytotoxicity), while positive values indicate to stimulation percentage.

C. Statistical analysis

The values were calculated using Origin version 7.0. One way analysis of variance (ANOVA) was conducted to test the significance among the Germination indices and seedling growth parameters of *V. radiata* and *V. mungo* in different experimental sets.

III. RESULT AND DISCUSSION

During the period of germination, the seeds were observed daily to detect the onset of germination. Emergence of radical from seeds was considered as indication for seed germination. The study revealed that the whole plant extracts decreased the Germination Percentage (GP) of seeds of receptor crop in comparison with respective control sets. However, between control sets of receptor crops, crop 2 showed better germination ability as compared to crop 1. Among extract treated sets decrease in GP was most pronounced in test crop 1 treated with *L. camara* extract. At the same time, *L. camara* extract induced more inhibitory effect in GP of receptor crop than *C. viscosum* extract. Except EL1 and ECl1 in all other experimental sets GP was increased from 3rd day to 10th day of germination (Table 1).

TABLE I: GERMINATION PERCENTAGE (GP) OF *V. RADIATA* AND *V. MUNGO* IN DIFFERENT EXPERIMENTAL SETS

Days	Experimental sets					
	EC1	EC2	EL1	EL2	ECl1	ECl2
3rd	68	75	34	52	55	62
5th	70	81	45	55	60	66
7th	75	90	48	60	62	66
10th	82	93	48	62	62	68

Moreover early seedling growth was considered to be the most responsive parameter to test the phytotoxicity (Gong et al. 2001). The root and shoot lengths were decreased in extract treated sets as compared to control sets of receptor crops (Fig 1). The weed extracts induced inhibitory effect on root and shoots length of both the receptor crops. Maximum decrease in root and shoot length was observed in EL1 set. *L. camara* extract exert more pronounced inhibition in root and shoot length of target plants than *C. viscosum* extract (Fig 1). Thus whole plant extracts of *C. viscosum* and *L. camara* produced suppressive effects on the growth of root and shoots length of the two tested crops. However, more pronounced effect was observed in case of *L. camara* treated sets.

Apart from germination percentage, a number of indices have been considered by researchers to study the inhibitory effect of phytotoxic substances on germination process (Anjum and Bajwa 2005). Germination index (GI) is directly correlated with germination percentage. Thus greater the value of GI, the greater will be germination percentage. SVI is an important parameter to determine the potentiality of plant germination and growth. Germination energy, speed of emergence, germination index, coefficient ratio of germination and seed vigour index of all the extracts treated sets decreased as compared with control sets. The speed of the germination was expressed by the mean germination time; the lower the value of the MGT the earlier the germination. But mean germination time and phytotoxicity were increased in all the treated sets. Moreover, relative germination ratio, relative seedling length as well as relative elongation ratio of root and shoot in different extracts treated sets exhibited distinctive values. Maximum tolerance index were observed in control (100.00%). But Tolerance index was highest in ECl2 set considering only extract treated sets. Inhibitory effect was more in EL1 set for root, shoot as well as seedling growth. Among extract treated sets, GE, SE, GI, SVI and Relative seedling length were maximum in ECl2 set, whereas lowest values were obtained for EL1 set. But RGR was highest in ECl1 set (Table 2).

To find out the significant effect of germination indices and seedling growth parameters (Table 2) on *V. radiata* and *V. mungo* in different experimental sets condition for the analysis of variance (ANOVA One Way) was performed. Here, most of the most of the experimental sets showed significant effect at $p<0.05$ level. Regarding biomass production, the

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result of the present study indicates that in extract treated sets of both the recipient crops fresh and dry weight decreased as compared with the control sets (Fig 1).

According to the previous reports the leaf extract of *Lantana camara* delayed the germination significantly in the receptor crops in comparison to the control treatment (Ahmed et al. 2007) ; high concentration of *Lantana camara* root leachate induced inhibition of germination in mungbean (Shaukat and Siddiqui, 2002). Present experimental results correlated with the above findings. In a previous report aqueous extract of leaf, stem and root of sorghum exhibited allelopathic effect on seedling growth of mung bean. Here the effect of different concentrations was not significant for germination percentage. However germination rate and mean germination time decreased significantly (Moosavi et al. 2011). The above findings correlated with present experimental results. The leaf aqueous extract of *L. camara* reported to exert significant inhibitory effect on germination index, tolerance index. In *Vigna mungo* phytotoxicity increased in treated set whereas germination index and tolerance index decreased (Nadirsha and Yogamoorthi, 2016). Leaf-litter dust of *Lantana camara* also exerted significant inhibitory effect on vegetative growth and yield of green gram (Gantayat et al. 2014). These can be correlated with present inferences related to inhibitory activity on different growth parameters in *L. camara* treated experimental set. In an earlier report the seedling growth of the two crops *Cicer arietinum* and *Lathyrus sativus* was inhibited by the extracts of *Clerodendrum inerme* and *C. viscosum*. Germination rate index and coefficient of velocity of germination were decreased and mean germination time increased in extract treated sets in respect to controls. The extent of germination delay also differes in two recipient crops. Similar results observed in present experiment where the allelopathic effect was exerted by the *Clerodendrum viscosum* on recipient crops as seedling growth, GRI and CRG were decreased and at the same time MGT was increased in respect to control sets (Roy and Roy, 2017). *Clerodendrum viscosum* reported to induce inhibitory effect on seed germination and seedling growth of different agricultural crops in laboratory condition (Debnath et al. 2017). It was reported that leaves of *C. viscosum* consists of a natural chemical retardant clerodin which might be induced the allelopathic effect on seed germination and root growth of recipient weeds (Datta and Chakrabarty (1982). These may also be related to the allelopathic effects of *C. viscosum observed on two target plants in present experiment.* The *Parthenium hysterophorus* showed inhibitory effects on germination, shoot length, root length, vigour index, tolerance index, root length, shoot length, fresh and dry weight of *Phaseolus vulgaris* seedlings. (Tahseen et al. 2015). Present study documented the similar suppressive effects on target plants. Inhibitory effect of 5.0% leachate concentration of weed *Parthenium* on seed germination, growth and dry matter of *Vigna mungo* was observed by Prasada Babu et al. 2014. Moreover *Calotropis procera* leachates reported to inhibit the germination and growth of soybeans. Statistical analysis showed significant differences in the coefficient of velocity (COV), plumule length, fresh weights of roots and shoots compared to the control experiments (Ayeni and Akinyed, 2014). This finding can be corroborates with the present result. The present experimental results can be supported by findings of earlier work where tea extracts reduced SVI, seed germination percentage of wheat and maize in all treatments than control. Moreover, tea extracts also had a significant effect on MGT of the wheat (Waris et al. 2016) which substantiate with the present result.

Inhibitory effects of two weed plant tested in the present study were different on receiving plant species. As stated in earlier report that the different weeds have different allelopathic effect on target plants. Not only that the stages of target plants also respond differentially to the same donor plant. The variation might be due to the alteration in type, total amount and properties of allelochemicals produced by different donor weeds. The suppression of seed germination of target plants in the present study by extracts of weeds might be also due to the inhibitory effect of different allelochemicals like water soluble phenolics, alkaloid or terpenoids as stated by Ramadan et al. (2018).

TABLE II: GERMINATION INDICES AND SEEDLING GROWTH PARAMETERS OF *V. RADIATA* AND *V. MUNGO* IN DIFFERENT EXPERIMENTAL SETS

Experimental sets	Parameters														
	Germination Energy (GE)	Speed of Emergence (SE)	Relative Germination Ratio (RGR)	Germination Index (GI)	Coefficient Ratio of Germination (GRG)	Mean Germination Time (MGT)	Seed Vigour Index (SVI)	Relative Seedling Length	Relative Elongation Ratio of Shoot (Rs)	Relative elongation Ratio of Root (Rr)	Tolerance index (TI)(root)	Phytotoxicity (%)	Inhibitory Effect (Root)	Inhibitory Effect (Shoot)	Inhibitory Effect (Seedling)
EC1	16	19.51	0	143.85	16.05	3	17.425	0	0	0	100	0	0	0	0
EC2	21	22.58	0	169.3	16.2	2.69	22.785	0	0	0	100	0	0	0	0
EL1	6	12.5	58.53	55.8	14.64	4.77	5.064	49.64	47.87	53.14	52.5	46.85	-46.85	-52.12	-50.35
EL2	11	17.74	66.66	89.25	15.33	3.83	8.53	56.16	51.26	65.81	69.13	34.18	-34.68	-48.73	-43.83
EC11	8	12.9	75.6	85.18	15.05	3.54	8.84	67.15	64.89	71.6	72.5	28.39	-28.39	-35.1	-32.84
EC12	13	19.11	73.11	90.13	15.01	4.41	11.51	69.1	64.61	76.72	82.95	23.27	-23.27	-35.38	-31.3

Different alphabets differ significantly at $p<0.05$ along with each sampling sites.

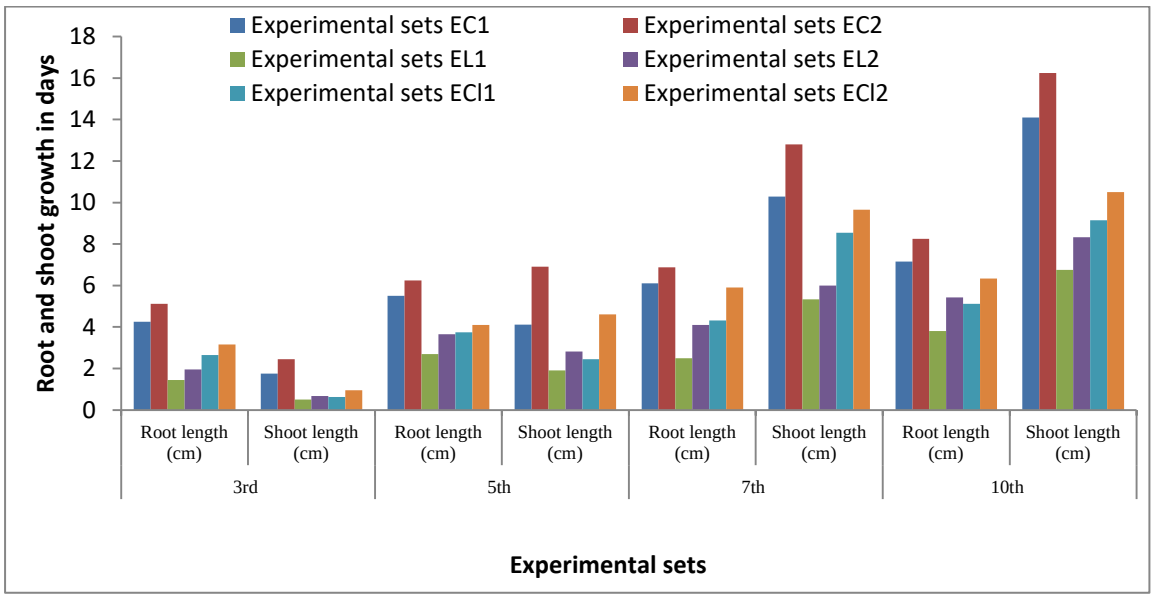

FIGURE I: GROWTH PARAMETERS OF *V. RADIATA* AND *V. MUNGO* IN DIFFERENT EXPERIMENTAL SETS

IV. CONCLUSION

Inhibitory effects were induced by weed extracts on seed germination and seedling growth parameters of two tested crop. Experimental result showed differential effect of weed extract on tested crop. Thus, suppressive effects of extracts were crop dependent. However, the diversity of allelochemicals present in the extracts was not assessed. Their remains a scope for assessing the plant defense mechanism of crop plant against such allelochemicals released by weeds. However extensive field trial along with the phytochemical analysis of weed extracts may be required for comprehensive evaluation of allelopathic potential of these weeds or donor plants on receptor plant.

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