



## Phyto-allelopathic effect of different trees leaves' aqueous extracts on seed germination and seedling growth of *Echinochloa crus-galli* (L.) Beauv

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### ABSTRACT

*Echinochloa crus-galli* (L.) Beauv is the most frequently reported troublesome weed in rice fields as it is aggressive, difficult to control and reduces rice yields significantly. An experiment was conducted during 2021 with an objective to assess the allelopathic effect of ten trees' leaves aqueous extracts on the seed germination and seedling growth of *E. crus-galli*. Leaves of ten tree species were separately dried and the aqueous extracts of each of them were diluted to obtain the three concentrations *i.e.* 5, 10 and 15% of each. Three concentrates of each of the tree leaves extract were used as treatments. The 15% concentration of the leaves extracts of all tree species exhibited highest efficacy in reducing germination and growth of *E. crus-galli*. Amongst all tree species studied, *Aegle marmelos* (L.) Corrêa tree leaves aqueous extract, at all concentrations caused greater allelopathic effect and maximum seedling root and shoot inhibition with lowest vigour index and seedling weight of *E. crus-galli*.

*Echinochloa crus-galli* (L.) Beauv is known to be one of the worst weeds occurring in rice fields (Rao 2021) as it causes severe rice yield losses by depleting 60–80% of soil nitrogen. It is a plant with C<sub>4</sub> photosynthetic pathway which makes it physiologically advantageous when it is grown as a weed in C<sub>3</sub> crops like rice. *Echinochloa* species seedlings look very similar to rice plants which make it difficult to manage by manual weeding as farmers sometimes unknowingly transplant these weeds onto rice field (Rao and Moody 1988) and causing huge rice yield losses (Rao and Moody 1987). The use of plants with strong allelopathic properties for weed control has shown promising results (Duke *et al.* 2000). Leaf extracts of tree species are a potent source of metabolites and toxic effects of these are species specific (Krumsri *et al.* 2020). The phytochemicals have the ability to reduce and delay germination, induce mortality of seedling leading to reduction in growth and yield. Thus, incorporating allelopathy in agricultural weed management programs may reduce the usage of herbicides (Kaur *et al.* 2017). Hence, the present study was conducted to assess the allelopathic potential of various tree species' leaves aqueous extracts against *E. crus-galli*.

The study was conducted during 2021 in Department of Botany, Punjab Agricultural University

(PAU), Ludhiana, Punjab. The seeds of *E. crus-galli* were procured from the Department of Agronomy, PAU, Ludhiana and stored under optimum storage conditions till use. The ten trees selected for the study include: *Aegle marmelos* (L.) Corrêa, *Albizia lebbek* (L.) Benth., *Azadirachta indica* A. Juss., *Eucalyptus tereticornis* Sm., *Leucaena leucocephala* (Lam.) de Wit, *Murraya koenigii* (Linn.) Spreng, *Populus deltoides* W. Bartram ex Marshall, *Salix alba* L., *Syzygium cumini* (L.) Skeels. and *Toona ciliata* M. Roem. Healthy and fully expanded leaves of selected tree species were collected from the trees growing in the Research Farm of Department of Forestry and Natural Resources, PAU during the months of March to August. The collected leaves were dried in hot-air oven at 60°C for one week and then grinded in electric grinder so as to obtain fine powder and sieved through 40 mesh sieve. The extracts were obtained by adding dry powdered tissues in distilled water at 1:1 w/v proportion for 48 hours. Then the extract was filtered through double layered muslin cloth; centrifuged at 4000 g for 30 min and the supernatant was filtered through Whatman No. 1 filter paper. The obtained extracts served as the crude extract (100 % concentration) and it was used as a stock solution for the study (Hussain *et al.* 2012). Three diluted concentrations (5, 10 and 15%) were prepared from stock solution through dilution of 100% concentrate.

*E. crus-galli* seeds were surface sterilized with 5% sodium hypochlorite solution for 5 minutes and then rinsed twice with running tap water for 3-5 minutes prior to the germination test to avoid fungal contamination. Twenty-five *E. crus-galli* seeds were placed in a 9-cm diameter Petri dish lined with two pieces of Whatman no. 1 filter paper. The Petri dishes were sealed with parafilm and placed at 30°C in an environmental chamber. Different concentrations of leaf extracts were applied on the inner side of the cover of Petri dish. The number of germinated seeds was counted at 15 days after sowing (DAS) or until there was no further germination. A similar set up with distilled water served as control.

The *E. crus-galli* seed germination percentage was calculated based on the number of normal seedlings on 15<sup>th</sup> day of germination.

$$\text{Germination percentage} = \frac{\text{Number of seeds germinated}}{\text{Total no. of seeds placed in petri dish}} \times 100$$

The percentage inhibition of germination, per cent root inhibition and per cent shoot inhibition were calculated using the equation:

$$I = 100 - (E2 \times 100 / E1)$$

Where, I represents percentage inhibition, E1 represents response of control and E2 represents response of treatment. Ten *E. crus-galli* seedlings were selected at random, gently blotted dry and then fresh weight was recorded and expressed in milligrams. For dry weight determination, *E. crus-galli* seedlings which were used for recording fresh weight were dried in oven at 60°C for 3 days and their dry weight was recorded. The *E. crus-galli* seedling vigour index I and II were calculated as per Abdulbaki and Anderson (1973).

Seed germination was calculated following formula stated by Association of Official Seed

Analysts (1983).  $= \sum n1/d1 + n2/d2 + n3/d3 + \dots$ , where 'n' is the number of germinated seeds; 'd' is the number of days.

Primary root length and shoot length were measured at the end of 15<sup>th</sup> day. Ten normal *E. crus-galli* seedlings from each replication were taken at random. The root length and shoot length of *E. crus-galli* seedlings were measured from point of attachment to cotyledon till the tip of root and shoot, respectively. Mean root length and shoot length was computed and expressed in centimetres. Total seedling length was measured as length from shoot tip to the root tip from seedlings selected at random. The mean of ten seedlings was computed and expressed in centimetres.

Total phenolic content was assayed following the procedures given by Bray and Thorpe (1954). The method of Balabaa *et al.* (1974) was used for total flavonoid content. Total alkaloid content was estimated following the procedures given by Shamsa *et al.* (2008) and total tannins content was determined following the procedures given by Sadasivam and Manickam (1992). Total terpenoid content was determined using standard protocol of Ghorai *et al.* (2017). Total soluble sugars were assayed using standard methodology of Dubois *et al.* (1956). They are expressed in units mg/g dry weight (DW). The experiments were carried out using completely randomised design (CRD). The statistical analysis of data was performed using duncan multiple (DMRT) range test through SPSS statistical software. All the differences were considered statistically significant at the probability levels of ( $p < 0.05$ ).

### Phytochemical content of leaves

The maximum phenol content was recorded in the extract of *E. tereticornis* (32.91 mg/g DW) and *S. cumini* (30.90 mg/g DW), followed by *A. marmelos* extracts with 24.00 mg/g DW of total phenols (Table

**Table 1. Secondary metabolites composition in leaves of selected tree species**

Tree species	Total soluble sugars (mg/g DW)	Total phenols (mg/g DW)	Total flavonoids (mg/g DW)	Total tannins (mg/g DW)	Total alkaloids (mg/g DW)	Total terpenoids (mg/g DW)
<i>Salix alba</i>	6.39 <sup>bcd</sup>	14.57 <sup>b</sup>	5.94 <sup>bcd</sup>	4.2 <sup>bc</sup>	4.77 <sup>bcd</sup>	1.93 <sup>b</sup>
<i>Populus deltoides</i>	6.67 <sup>bcd</sup>	21.35 <sup>ab</sup>	5.4 <sup>bcd</sup>	6 <sup>abc</sup>	12.43 <sup>a</sup>	15.04 <sup>a</sup>
<i>Eucalyptus tereticornis</i>	6.33 <sup>bcd</sup>	32.91 <sup>a</sup>	9.23 <sup>a</sup>	1.75 <sup>c</sup>	9.91 <sup>ab</sup>	3.67 <sup>b</sup>
<i>Syzygium cumini</i>	8.01 <sup>abc</sup>	30.9 <sup>a</sup>	6.67 <sup>abcd</sup>	2.36 <sup>bc</sup>	4.54 <sup>abcd</sup>	5.38 <sup>b</sup>
<i>Aegle marmelos</i>	11.04 <sup>a</sup>	24 <sup>ab</sup>	8.33 <sup>ab</sup>	10.38 <sup>a</sup>	8.96 <sup>abc</sup>	5.96 <sup>b</sup>
<i>Murraya koenigii</i>	4.15 <sup>cd</sup>	11.35 <sup>b</sup>	5.08 <sup>cd</sup>	7.01 <sup>ab</sup>	6.13 <sup>bcd</sup>	4.71 <sup>b</sup>
<i>Azadirachta indica</i>	3.61 <sup>d</sup>	11.23 <sup>b</sup>	4.47 <sup>cd</sup>	4.2 <sup>bc</sup>	7.15 <sup>abcd</sup>	7.09 <sup>b</sup>
<i>Toona ciliata</i>	9.06 <sup>ab</sup>	12.94 <sup>b</sup>	3.79 <sup>d</sup>	2.6 <sup>bc</sup>	6.86 <sup>abcd</sup>	3.92 <sup>b</sup>
<i>Luecaena leucocephala</i>	6.79 <sup>bcd</sup>	18.99 <sup>ab</sup>	7.11 <sup>abc</sup>	4.58 <sup>bc</sup>	1.86 <sup>d</sup>	2.67 <sup>b</sup>
<i>Albizia lebbeck</i>	7.3 <sup>abcd</sup>	19.81 <sup>ab</sup>	6.26 <sup>abcd</sup>	4.18 <sup>bc</sup>	2.7 <sup>cd</sup>	3.97 <sup>b</sup>

Values depicted by same letter are not significantly different as per DMRT ( $p < 0.05$ )

1), while, minimum levels of total phenols were recorded in the extracts of *A. indica* (11.23 mg/g DW) which was statistically at par with the phenol levels in the extracts of *M. koenigii* (11.35 mg/g DW), *T. ciliata* (12.94 mg/g DW) and *S. alba* (14.57 mg/g DW). The recorded total flavonoids were significantly higher in *E. tereticornis* extracts (9.23 mg/g DW) followed by *A. marmelos* (8 mg/g DW) while, the lowest flavonoid levels were recorded in *T. ciliata* (3.79 mg/g DW) and *A. indica* (4.47 mg/g DW). Polyphenols and flavanoids were reported to cause strong inhibitory effects on seed germination and early seedling growth of *E. crus-galli* (Poonpaiboonpipat and Jumpathong 2019). Higher total soluble sugars were recorded in *A. marmelos* (11.04 mg/g DW), followed by *T. ciliata* (9.06 mg/ml) and *S. cumini* (8.01 mg/g DW) with lowest sugar content in *A. indica* (3.61 mg/g DW) closely followed by *M. koenigii* extracts (4.15 mg/g DW).

Significantly higher tannins were recorded in *A. marmelos* (10.38 mg/g DW) and lowest in *E.*

*tereticornis* (1.75 mg/g DW), among all tree species leaf aqueous extracts. Alkaloids are the metabolites chiefly responsible for the medicinal and allelopathic properties among plant species. Significantly maximum alkaloid content was recorded in the leaves aqueous extracts of *P. deltoids* at 12.43 mg/g DW and lowest in those of *L. leucocephala* at 1.86 mg/g DW, among all tree species extracts. Total terpenoids were found to be significantly highest in the leaves extracts of *P. deltoids* at 15.04 mg/g DW, while all other leaf extracts terpenoid levels were statistically at par amongst each other. Terpenoids are essential allelochemicals as they are highly potent leading to electrolyte leakage, lipid peroxidation, loss of cell water, disruption of respiration which adversely affected seed germination (Araniti *et al.* 2013). The trees which leaves extracts were screened for phytochemical constituents seemed to have the potential to act as a source of allelopathic chemicals that may be used to improve the current weed management practices.

**Table 2. Effect of aqueous leaf extracts of selected tree species on *E. crus-galli* seed germination related parameters**

Tree species	Concentration	Germination (%)	Germination inhibition (%)	Germination speed
	Water (control)	89.67 <sup>a</sup>	0 <sup>f</sup>	18.46 <sup>a</sup>
<i>Salix alba</i>	5%	81.67 <sup>abcd</sup>	8.92 <sup>cdef</sup>	15.16 <sup>ab</sup>
	10%	72.05 <sup>abcdef</sup>	20.76 <sup>abcdef</sup>	12 <sup>bcdef</sup>
	15%	64.88 <sup>cdef</sup>	27.66 <sup>abcd</sup>	10.08 <sup>bcdefg</sup>
<i>Populus deltoides</i>	5%	81.96 <sup>abcd</sup>	8.58 <sup>cdef</sup>	15.25 <sup>ab</sup>
	10%	70.51 <sup>abcdef</sup>	21.36 <sup>abcdef</sup>	13 <sup>bcd</sup>
	15%	64.97 <sup>cdef</sup>	27.57 <sup>abcd</sup>	10.67 <sup>bcdef</sup>
<i>Eucalyptus tereticornis</i>	5%	78.67 <sup>abcd</sup>	12.33 <sup>cdef</sup>	11.33 <sup>bcdef</sup>
	10%	72.67 <sup>abcdef</sup>	18.93 <sup>abcdef</sup>	9 <sup>cdefg</sup>
	15%	54.33 <sup>f</sup>	39.42 <sup>a</sup>	7.37 <sup>efg</sup>
<i>Syzygium cumini</i>	5%	82.27 <sup>abcd</sup>	8.22 <sup>cdef</sup>	13 <sup>bcd</sup>
	10%	73.71 <sup>abcde</sup>	17.79 <sup>bcdef</sup>	12 <sup>bcdef</sup>
	15%	66.77 <sup>bcdef</sup>	25.48 <sup>abcde</sup>	10.03 <sup>bcdefg</sup>
<i>Aegle marmelos</i>	5%	65.76 <sup>bcdef</sup>	26.65 <sup>abcde</sup>	9 <sup>cdefg</sup>
	10%	58.2 <sup>ef</sup>	35.09 <sup>ab</sup>	7.33 <sup>efg</sup>
	15%	53.95 <sup>f</sup>	39.85 <sup>a</sup>	4.99 <sup>g</sup>
<i>Murraya Koenigi</i>	5%	84.82 <sup>ab</sup>	5.37 <sup>ef</sup>	12.67 <sup>bcde</sup>
	10%	82.78 <sup>abc</sup>	7.64 <sup>def</sup>	11 <sup>bcdef</sup>
	15%	72.73 <sup>abcdef</sup>	18.84 <sup>abcdef</sup>	9 <sup>cdefg</sup>
<i>Azadirachta indica</i>	5%	84.28 <sup>abc</sup>	5.99 <sup>def</sup>	9.33 <sup>cdefg</sup>
	10%	76 <sup>abcde</sup>	15.24 <sup>bcdef</sup>	7.85 <sup>defg</sup>
	15%	62.71 <sup>def</sup>	30.05 <sup>abc</sup>	6.82 <sup>fg</sup>
<i>Toona ciliata</i>	5%	86.57 <sup>a</sup>	3.43 <sup>f</sup>	13.33 <sup>abcd</sup>
	10%	83.4 <sup>abc</sup>	6.98 <sup>def</sup>	11.67 <sup>bcdef</sup>
	15%	74.04 <sup>abcde</sup>	17.43 <sup>bcdef</sup>	9.09 <sup>cdefg</sup>
<i>Lucaena leucocephala</i>	5%	83.18 <sup>abc</sup>	7.24 <sup>def</sup>	13.33 <sup>abcd</sup>
	10%	75.37 <sup>abcde</sup>	15.9 <sup>bcdef</sup>	12.42 <sup>bcde</sup>
	15%	66.93 <sup>bcdef</sup>	25.33 <sup>abcde</sup>	11.58 <sup>bcdef</sup>
<i>Albizia lebbbeck</i>	5%	88.55 <sup>a</sup>	1.23 <sup>f</sup>	13.61 <sup>abc</sup>
	10%	85.03 <sup>ab</sup>	5.17 <sup>ef</sup>	11 <sup>bcdef</sup>
	15%	80.06 <sup>abcd</sup>	10.7 <sup>cdef</sup>	10.33 <sup>bcdefg</sup>

Values depicted by same letter are not significantly different as per DMRT ( $p < 0.05$ )

### Effect on germination

The tested leaf extracts were very effective in decreasing seed germination of *E. crus-galli* (Table 2). The inhibition of *E. crus-galli* seeds germination showed a concentration dependent trend with the degree of inhibitory proportional to the leaves aqueous extract concentration (Table 2). The highest per cent seed germination inhibition was with 15% formulation followed by 10 and 5%. Among ten tree species, *A. marmelos* leaves aqueous extract caused the maximum germination inhibition (Table 2). The inhibitory effect could be due to interference of leaf extracts on seed physiological processes like cell division and enlargement (Chowhan *et al.* 2011) which confirm reports of Nadeem *et al.* (2021) and Mondal *et al.* (2020). Lower rate of seed germination could be attributed to the presence of phytotoxic metabolites in the leaf aqueous extracts of trees which reduced *E. crus-galli* seeds germination index. These findings support the results of Khan *et al.* (2016) who reported that the germination kinetics of weed seeds were significantly reduced due to

extracts of different species. The phytotoxicity of plant extracts affected weed seed germination and seedling growth. This study revealed that the magnitude of inhibition on seed germination traits, seedling growth and biomass increased with incremental extract intensity and showed linear dose dependent variation as reported by Phuwawat *et al.* (2012) and Akacha *et al.* (2013), while examining the effect of aqueous leaf extracts of *Melia azedarach* on *E. crus-galli*.

### Effect on seedlings growth parameters

The minimum *E. crus-galli* seedling length was observed when treated with *A. marmelos* extracts followed closely by *S. cumini* and *E. tereticornis* (Table 3). Minimum *E. crus-galli* seedling root length (0.65 cm) and minimum shoot length (1.27 cm) was recorded with 15% extracts of *E. tereticornis* and *Aegle marmelos*, respectively. These observations indicated that allelopathic aqueous extracts generally have rather significantly more pronounced effect on

**Table 3. Effect of aqueous leaf extracts of selected tree species on seedling growth related parameters and percentage root and shoot inhibition of *E. crus-galli***

Tree species	Concentration	Root length (cm)	Shoot length (cm)	Total seedling length (cm)	Root inhibition (%)	Shoot inhibition (%)
	Water (control)	2.28 <sup>a</sup>	6.79 <sup>a</sup>	9.08 <sup>a</sup>	0 <sup>f</sup>	0 <sup>h</sup>
<i>Salix alba</i>	5%	1.83 <sup>ab</sup>	5.32 <sup>abc</sup>	7.16 <sup>abc</sup>	19.61 <sup>ef</sup>	21.72 <sup>fgh</sup>
	10%	1.63 <sup>abcd</sup>	5.14 <sup>abcd</sup>	6.77 <sup>abcd</sup>	28.65 <sup>cdef</sup>	24.45 <sup>efgh</sup>
	15%	1.13 <sup>bcdef</sup>	4 <sup>bcdefg</sup>	5.13 <sup>bcdefgh</sup>	50.72 <sup>abcde</sup>	40.76 <sup>bcdefg</sup>
<i>Populus deltoides</i>	5%	1.26 <sup>bcdef</sup>	4.76 <sup>abcde</sup>	6.02 <sup>bcdefg</sup>	44.48 <sup>abcde</sup>	29.97 <sup>defgh</sup>
	10%	1.09 <sup>bcdef</sup>	3.9 <sup>bcdefg</sup>	4.99 <sup>bcdefghi</sup>	52.15 <sup>abcd</sup>	42.68 <sup>bcdefg</sup>
	15%	0.96 <sup>def</sup>	3.4 <sup>bcdefgh</sup>	4.36 <sup>cdefghi</sup>	57.72 <sup>abc</sup>	49.81 <sup>abcdefg</sup>
<i>Eucalyptus tereticornis</i>	5%	1.08 <sup>cdef</sup>	3.43 <sup>bcdefgh</sup>	4.51 <sup>bcdefghi</sup>	52.67 <sup>abcd</sup>	49.28 <sup>abcdefg</sup>
	10%	0.92 <sup>def</sup>	2.97 <sup>cdefgh</sup>	3.89 <sup>defghi</sup>	59.75 <sup>abc</sup>	56.11 <sup>abcdef</sup>
	15%	0.65 <sup>f</sup>	2.38 <sup>efgh</sup>	3.03 <sup>ghi</sup>	71.62 <sup>a</sup>	64.93 <sup>abcd</sup>
<i>Syzygium cumini</i>	5%	1.21 <sup>bcdef</sup>	3.67 <sup>bcdefg</sup>	4.88 <sup>bcdefghi</sup>	46.91 <sup>abcde</sup>	46.06 <sup>bcdefg</sup>
	10%	1 <sup>cdef</sup>	2.86 <sup>defgh</sup>	3.86 <sup>defghi</sup>	56 <sup>abcd</sup>	57.76 <sup>abcde</sup>
	15%	0.81 <sup>ef</sup>	2.06 <sup>fgh</sup>	2.86 <sup>hi</sup>	64.56 <sup>ab</sup>	69.37 <sup>abc</sup>
<i>Aegle marmelos</i>	5%	0.93 <sup>def</sup>	2.54 <sup>efgh</sup>	3.47 <sup>efghi</sup>	59.11 <sup>abc</sup>	62.76 <sup>abcd</sup>
	10%	0.82 <sup>ef</sup>	1.79 <sup>gh</sup>	2.61 <sup>hi</sup>	63.76 <sup>ab</sup>	73.72 <sup>ab</sup>
	15%	0.69 <sup>f</sup>	1.27 <sup>h</sup>	1.96 <sup>i</sup>	69.6 <sup>a</sup>	81.29 <sup>a</sup>
<i>Murraya Koenigi</i>	5%	1.07 <sup>cdef</sup>	4.13 <sup>bcdefg</sup>	5.2 <sup>bcdefgh</sup>	53.17 <sup>abcd</sup>	39.27 <sup>bcdefg</sup>
	10%	0.89 <sup>def</sup>	3.63 <sup>bcdefgh</sup>	4.52 <sup>bcdefghi</sup>	60.98 <sup>abc</sup>	46.11 <sup>bcdefg</sup>
	15%	0.77 <sup>f</sup>	2.66 <sup>efgh</sup>	3.42 <sup>fghi</sup>	66.42 <sup>a</sup>	60.71 <sup>abcd</sup>
<i>Azadirachta indica</i>	5%	1.55 <sup>bcde</sup>	4.43 <sup>bcdef</sup>	5.98 <sup>bcdefg</sup>	32.53 <sup>bcdef</sup>	34.55 <sup>cdefgh</sup>
	10%	1.26 <sup>bcdef</sup>	3.67 <sup>bcdefg</sup>	4.93 <sup>bcdefghi</sup>	45.56 <sup>abcde</sup>	45.97 <sup>bcdefg</sup>
	15%	1.1 <sup>bcdef</sup>	2.63 <sup>efgh</sup>	3.73 <sup>efghi</sup>	52.68 <sup>abcd</sup>	60.79 <sup>abcd</sup>
<i>Toona ciliata</i>	5%	1.33 <sup>bcdef</sup>	5.13 <sup>abcd</sup>	6.46 <sup>abcde</sup>	42.6 <sup>abcde</sup>	24.64 <sup>efgh</sup>
	10%	0.87 <sup>ef</sup>	4.16 <sup>bcdefg</sup>	5.02 <sup>bcdefgh</sup>	62.18 <sup>ab</sup>	38.66 <sup>bcdefg</sup>
	15%	0.64 <sup>f</sup>	3.47 <sup>bcdefgh</sup>	4.11 <sup>defghi</sup>	71.3 <sup>a</sup>	48.72 <sup>abcdefg</sup>
<i>Lucaena leucocephala</i>	5%	1.74 <sup>abc</sup>	5.67 <sup>ab</sup>	7.41 <sup>ab</sup>	23.99 <sup>def</sup>	16.65 <sup>gh</sup>
	10%	0.68 <sup>f</sup>	4.58 <sup>abcde</sup>	5.26 <sup>bcdefgh</sup>	69.11 <sup>a</sup>	32.37 <sup>defgh</sup>
	15%	0.81 <sup>ef</sup>	3.33 <sup>bcdefgh</sup>	4.14 <sup>cdefghi</sup>	64.27 <sup>ab</sup>	50.73 <sup>abcdefg</sup>
<i>Albizia lebbeck</i>	5%	1.24 <sup>bcdef</sup>	5.11 <sup>abcd</sup>	6.35 <sup>abcdef</sup>	46.08 <sup>abcde</sup>	24.63 <sup>efgh</sup>
	10%	1.54 <sup>bcde</sup>	3.99 <sup>bcdefg</sup>	5.53 <sup>bcdefgh</sup>	32.96 <sup>bcde</sup>	40.94 <sup>bcdefg</sup>
	15%	1.16 <sup>bcdef</sup>	3.46 <sup>bcdefgh</sup>	4.62 <sup>bcdefghi</sup>	49.31 <sup>abcde</sup>	48.96 <sup>bcdefg</sup>

Values depicted by same letter are not significantly different as per DMRT (p <0.05)

inhibition of seedlings root growth than the shoot growth (Randhawa *et al.* 2002, Singh *et al.* 2009, Aslani *et al.* 2014, Scavo *et al.* 2019, Saad *et al.* 2019). Such an outcome is expected because plant root is often the first tissue to be in contact with allelochemicals present in them (Singh *et al.* 2009). All the leaf extracts were found to have an inhibitory effect on the root and shoot growth (Table 3). Roots were most sensitive to these extracts and exhibited highest degree of inhibition with extracts of *E. tereticornis* (71.62%), followed closely by *T. ciliata* (71.3%) and *A. marmelos* (69.6%) (Table 4). Highest root inhibition was recorded with *A. marmelos* and *E. tereticornis* extracts and lowest with *S. alba* extracts at all concentrations. Among various tree species extracts, *A. marmelos* recorded highest and *Salix alba* recorded lowest degree of shoot inhibition at all concentration levels. The chemicals present in these extracts inhibit shoot and seedling growth by inhibiting cell division and elongation and interferes with enzymes involved in mobilization of nutrients necessary for seedling emergence (Kong *et al.* 2019).

**Effect on seedlings vigour and biomass**

The highest vigour index I (616.26) and II was recorded with *L. leucocephala* at 5% concentration (Table 4). Among treatments, *E. crus-galli* seeds treated with *S. alba* and *A. lebbeck* extracts were most vigorous while, *E. crus-galli* seeds treated with *A. marmelos* and *E. tereticornis* extracts were least vigorous as they have the highest and lowest values of seed vigour index I and II, respectively. Similar trends were recorded for the *E. crus-galli* seedling fresh and dry weight (Table 5). Among treatments, *E. crus-galli* seeds treated with *S. alba* recorded highest fresh weight and dry weight of *E. crus-galli* seedlings followed by *A. lebbeck* while, *E. crus-galli* seeds treated with *A. marmelos* extracts recorded lowest fresh and dry weight of *E. crus-galli* seedlings at all concentration levels followed by *E. tereticornis* extracts. Minimum *E. crus-galli* seedling dry weight observed with the leaf aqueous extract application may be attributed to phytotoxic compounds released in higher concentration from their leaves which imparted growth inhibitory action (Ding *et al.* 2007).

**Table 4. Effect of aqueous leaf extracts of selected tree species on vigour and biomass of *E. crus-galli***

Tree species	Concentration	Vigour index I	Vigour index II	Fresh weight (mg)	Dry weight (mg)
	Water (control)	814.12 <sup>a</sup>	523.89 <sup>a</sup>	9.04 <sup>a</sup>	5.84 <sup>a</sup>
<i>Salix alba</i>	5%	585.11 <sup>abc</sup>	339.17 <sup>b</sup>	7.42 <sup>abc</sup>	4.16 <sup>b</sup>
	10%	481.28 <sup>bcdef</sup>	210.84 <sup>bcdefgh</sup>	6.65 <sup>abcd</sup>	2.96 <sup>bcde</sup>
	15%	332.74 <sup>bcdefgh</sup>	163.38 <sup>bcdefgh</sup>	5.03 <sup>cdefgh</sup>	2.53 <sup>bcde</sup>
<i>Populus deltoides</i>	5%	493.49 <sup>bcdef</sup>	324.76 <sup>bc</sup>	7.99 <sup>ab</sup>	3.96 <sup>bc</sup>
	10%	352.2 <sup>bcdefgh</sup>	272.27 <sup>bcd</sup>	6.48 <sup>abcde</sup>	3.86 <sup>bc</sup>
	15%	283.92 <sup>cdefgh</sup>	146.24 <sup>cdefgh</sup>	4.81 <sup>cdefgh</sup>	2.24 <sup>cde</sup>
<i>Eucalyptus tereticornis</i>	5%	355.05 <sup>bcdefgh</sup>	214.92 <sup>bcdefgh</sup>	5.43 <sup>bcdef</sup>	2.74 <sup>bcde</sup>
	10%	282.17 <sup>cdefgh</sup>	124.05 <sup>defgh</sup>	3.58 <sup>fgh</sup>	1.7 <sup>de</sup>
	15%	165.57 <sup>gh</sup>	66.04 <sup>h</sup>	2.54 <sup>gh</sup>	1.22 <sup>e</sup>
<i>Syzygium cumini</i>	5%	401.38 <sup>bcdefgh</sup>	250.67 <sup>bcdefg</sup>	6.02 <sup>bcdef</sup>	3.05 <sup>bcde</sup>
	10%	284.5 <sup>cdefgh</sup>	184.7 <sup>bcdefgh</sup>	4.65 <sup>cdefgh</sup>	2.51 <sup>bcde</sup>
	15%	190.79 <sup>fgh</sup>	114.56 <sup>defgh</sup>	3.83 <sup>defgh</sup>	1.72 <sup>de</sup>
<i>Aegle marmelos</i>	5%	228.46 <sup>efgh</sup>	170.82 <sup>bcdefgh</sup>	4.56 <sup>cdefgh</sup>	2.6 <sup>bcde</sup>
	10%	151.92 <sup>gh</sup>	85.35 <sup>efgh</sup>	3.61 <sup>efgh</sup>	1.47 <sup>de</sup>
	15%	105.46 <sup>h</sup>	71.04 <sup>gh</sup>	2.36 <sup>h</sup>	1.32 <sup>e</sup>
<i>Murraya Koenigi</i>	5%	439.85 <sup>bcdefg</sup>	232.19 <sup>bcdefgh</sup>	5.59 <sup>bcdef</sup>	2.74 <sup>bcde</sup>
	10%	374.96 <sup>bcdefgh</sup>	234.09 <sup>bcdefgh</sup>	5.2 <sup>bcdefgh</sup>	2.83 <sup>bcde</sup>
	15%	249.41 <sup>efgh</sup>	190.33 <sup>bcdefgh</sup>	4.43 <sup>defgh</sup>	2.61 <sup>bcde</sup>
<i>Azadirachta indica</i>	5%	503.89 <sup>bcde</sup>	206.55 <sup>bcdefgh</sup>	4.67 <sup>cdefgh</sup>	2.46 <sup>bcde</sup>
	10%	376.18 <sup>bcdefgh</sup>	161.77 <sup>bcdefgh</sup>	4.07 <sup>defgh</sup>	2.12 <sup>cde</sup>
	15%	236.38 <sup>efgh</sup>	82.07 <sup>fgh</sup>	3.49 <sup>fgh</sup>	1.31 <sup>e</sup>
<i>Toona ciliata</i>	5%	559.22 <sup>abcd</sup>	255.7 <sup>bcdef</sup>	5.81 <sup>bcdef</sup>	2.96 <sup>bcde</sup>
	10%	418.95 <sup>bcdefg</sup>	227.04 <sup>bcdefgh</sup>	5.53 <sup>bcdef</sup>	2.72 <sup>bcde</sup>
	15%	304.95 <sup>cdefgh</sup>	169.28 <sup>bcdefgh</sup>	4.41 <sup>defgh</sup>	2.3 <sup>bcde</sup>
<i>Lucaena leucocephala</i>	5%	616.26 <sup>ab</sup>	265.85 <sup>bcde</sup>	6.23 <sup>bcdef</sup>	3.19 <sup>bcde</sup>
	10%	397.12 <sup>bcdefgh</sup>	174.35 <sup>bcdefgh</sup>	4.94 <sup>bcdefgh</sup>	2.3 <sup>bcde</sup>
	15%	277.27 <sup>defgh</sup>	178.04 <sup>bcdefgh</sup>	4.69 <sup>cdefgh</sup>	2.66 <sup>bcde</sup>
<i>Albizia lebbeck</i>	5%	561.87 <sup>abcd</sup>	326.02 <sup>bc</sup>	6.25 <sup>bcdef</sup>	3.68 <sup>bc</sup>
	10%	471.54 <sup>bcdef</sup>	213.91 <sup>bcdefgh</sup>	5.25 <sup>bcdefg</sup>	2.52 <sup>bcde</sup>
	15%	370.13 <sup>bcdefgh</sup>	191.89 <sup>bcdefgh</sup>	4.5 <sup>defgh</sup>	2.4 <sup>bcde</sup>

Values depicted by same letter are not significantly different as per DMRT (p <0.05)

On the basis of this study, it was concluded that *Aegle marmelos* leaves aqueous extract at all concentrations tested, has greater phyto allelopathic effect on *E. crus-galli*

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