



# Inhibitory Effect of Aqueous Extracts of Tree-legumes on Germination and Seedling Growth of Food Legume, Green Gram (*Vigna radiata* L.)

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## ABSTRACT

**Background:** Green gram (*Vigna radiata*), the third most important pulse crop, is grown in tank-fed, command areas after the harvest of paddy. In such areas, legume trees like *Delonix regia* and *Peltophorum pterocarpum* are commonly found and tree parts often fell in watercourses and were flown to crop fields along with irrigation water. Hence, it is necessary to study whether any allelopathic effect is present in these parts of trees which may affect the growth and development of green gram grown in farmland crops.

**Methods:** Collected different tree parts viz., bark, leaf and fresh flower, a matured pod of *Delonix regia* and *Peltophorum pterocarpum*, cleaned and soaked distilled water at weight/volume ratio of 1:10 for 24 hrs. Aqueous extracts were filtered and tested for inhibition of green gram. Pot culture experiments were conducted with 10 treatments and four replications. Germination (%), shoot length (cm), root length (cm), fresh weight (mg), dry weight (mg), seedling vigour index-I and vigour index-II in green gram were recorded and the magnitude of inhibition versus simulation in bioassay was compared through response index.

**Result:** Among the plant parts, *Delonix regia* flower and *Peltophorum pterocarpum* matured pods suppressed germination (45.00 and 60.00%), shoot length (13.87 and 13.39 cm), root length (4.27 and 10.49 cm), seedling vigour index-I (624.15 and 803.40) and seedling vigour index-II (960.75 and 948.00) respectively in green gram. This might due to presence of inhibitory chemicals in flowers of *Delonix regia* and matured pods of *Peltophorum pterocarpum* compared to other plant parts of trees.

**Key words:** Allelopathic, *Delonix regia*, Green gram, *Peltophorum pterocarpum*, Response index.

## INTRODUCTION

Green gram [*Vigna radiata* (L.) Wilczek], also known as mungbean, is the third most important pulse crop of India, cultivated in 33.70 lakh hectares with a production of 16.43 lakh tonnes and average productivity of 488 kg ha<sup>-1</sup> (Sajjan *et al.*, 2021) and is grown in tank-fed, command areas after harvesting of paddy. In such areas, legume trees like *Delonix regia* and *Peltophorum pterocarpum* are commonly found and tree parts such as flowers, leaves, pods, dead branches, and bark peels often fell in watercourses and were flown to crop fields along with irrigation water and sometimes, incorporated as green leaf manure. Hence, it is necessary to study whether any allelopathic effect is present in these parts of trees which may affect growth and development of green gram grown in farmland crops.

Secondary metabolites, known as allelochemicals produced by higher plants cause a phenomenon called allelopathy which may inhibit the growth of neighboring and other plants (Albuquerque *et al.*, 2011) and allelopathic potential of many plant species have been documented (Cheng and Cheng, 2015) and these allelopathic plant extracts are used as an alternative to synthetic herbicides for managing weeds (Iqbal *et al.*, 2009; Khaliq *et al.*, 2012; Mushtaq *et al.*, 2013). Two commonly found multipurpose tree species in farmlands and village lands viz., *Delonix regia* (Boj.) Raf. (flamboyant) and *Peltophorum pterocarpum* (DC.)

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K.Heyne (Copperpod, Golden or yellow flamboyant; Syn. *Peltophorum inermis* and *Peltophorum ferrugineum*) seasonally shed their flowers, leaves, dead branches and bark peels on farmlands. *Delonix regia* (Boj.) Raf. (flamboyant) is a 10-15 m tall tree native to Madagascar and spread the world over in tropical and subtropical regions, belongs to Fabaceae family (Fig 1) used as shade, shelter, timber, fuel and in apiculture too (Orwa *et al.*, 2009 a). Previously, it has been reported that *Delonix regia* substantially inhibited weeds under its canopy. Aqueous extracts of *Delonix regia* inhibited growth of *Centella asiatica*

and *Isachne nipponensis* upto 70%. The inhibitory effect of allelochemicals in plant extracts of a tree has been well documented (Mushtaq *et al.*, 2010).

*Peltophorum pterocarpum* (DC.) K. Heyne (Copperpod, Golden or yellow flamboyant; Syn. *Peltophorum inermis* and *Peltophorum ferrugineum*) also belongs to Fabaceae (Fig 2) native to tropical South-Eastern Asia and a popular ornamental tree grown around the world. It is a deciduous tree growing to 15-25 m tall, with a trunk diameter of up to 1 m, and found to contain aliphatic alcohol, fatty acids, amino acids, terpenoids, phenolics, flavonoids, alkaloids, steroids are isolated as phytochemicals in very minimal quantity from this plant (Huxley, 1992).

In line with illustrations that proof of allelopathy lies on demonstrating interference using suitable controls, describing symptoms, and quantifying growth reduction (Narwal, 2004), the present study was conducted to demonstrate allelopathic effects of different parts *viz.*, bark, leaf, fresh flower, fresh and matured pod of two tree species *Delonix regia* and *Peltophorum pterocarpum* on the green gram. Aqueous extracts of various parts mentioned above were prepared and tested for their inhibitory effect on germination, shoot and root length (cm) as well as fresh and dry weight through poly pot culture study.

## MATERIALS AND METHODS

Bark, leaf, fresh flower, fresh and matured pods of both tree species, *Delonix regia* and *Peltophorum pterocarpum* were collected from 20-year old stands at Agricultural College Campus and the study was carried out at the Department of

Agronomy, Agricultural College and Research Institute, Tamil Nadu Agricultural University, Madurai, India located at 9°54'N latitude and 78°54'E longitude. Thus collected tree parts were initially cleaned off from dirt and foreign bodies and then soaked separately in distilled water at a weight/volume ratio of 1:10 for 24 hrs. This ratio produces low osmolality of 10 per cent. After 24 hours, aqueous extracts were filtered through Whatman No.1 filter paper. Aqueous extracts of each tree part were filtered and tested for inhibition of germination and seedling growth of green gram [*Vigna radiata* (L.) R. Wilczek] cv. VBN 3. Treatments include aqueous extracts of Bark, leaf, flower, pod and seed of *Delonix regia*; Bark, leaf, flower, matured pod and fresh pod of *Peltophorum pterocarpum*, thus making to 10 treatments, each replicated four times. Distilled water served as control. Twenty-five seeds were sown at 27.08.2020 in poly pots of 25 × 25 cm size, filled with a nursery mixture of Red soil : Sand : Compost at a ratio of 2:1:1 in the field lab. Polypots were added with aqueous extracts frequently to avoid drying up. Distilled water served as control. Germination (%), shoot length (cm), root length (cm), fresh weight (mg), dry weight (mg), Vigour index-I {Standard germination (%) × seedling length (cm)} and Vigour index-II {Standard germination (%) × seedling dry weight (mg)} by Abdul-Baki and Anderson, (1973) were evaluated after two weeks. The magnitude of inhibition versus simulation in bioassay was compared through Response Index (RI) (Richardson and Williamson, 1988) is determined as follows,

$$\begin{aligned} \text{if } T > C, RI &= 1 - (C/T) \\ \text{if } T &= C, \text{ then } RI = 0 \\ \text{if } T < C, \text{ then } RI &= (T/C) - 1 \end{aligned}$$

Where

T is treatment mean and C controls mean. A negative RI reflects the proportional disparity in output [germination (%), shoot length (cm), root length (cm), fresh weight (mg) and dry weight (mg)] of test crop in treatment relative to output in control. Results were subjected to an analysis of variance (Gomez and Gomez, 1984) and mean RI values were tested for standard error.

## RESULTS AND DISCUSSION

Germination and seedling growth of green gram was inhibited by various aqueous extracts used and the effect showed mixed results. No uniformity in inhibition was noticed due to various aqueous extracts of the same species and it differs with tree species/parts. Flower aqueous extracts of *Delonix* had a more inhibitory effect on green gram while matured pod aqueous extracts of *Peltophorum* showed higher inhibition. Inhibition is thought to be due to phytotoxins present in extracts, instead of osmotic inhibition because the use of 10% extract ensures low osmolality (Orwa *et al.*, 2009b).

Easily visible effects including retarded germination rate (Williamson *et al.*, 1992), seeds darkening and swelling reduced radicle and shoot extension (Turk and Tawaha, 2003; Bhatt and Todaria, 1990), swelling/necrosis of root



Fig 1: *Delonix regia* at Agricultural College campus of Madurai.



Fig 2: *Peltophorum pterocarpum* at Agricultural College campus of Madurai.

tips, discoloration, (Fig 3), lack root hairs, reduced dry weight (Ayeni *et al.*, 1997) were observed and such visible effects were proved and reflected in response index.

Inhibition was measured by RI with a range of 1 to 5%. The highest suppression was registered in germination (45.00 and 60.00%), shoot length (13.87 and 13.39 cm) and root length (4.27 and 10.49 cm) by the flower of *Delonix regia* and a matured pod of *Peltophorum pterocarpum* aqueous extracts respectively. Though flower aqueous extracts of *Delonix* had high inhibition on germination, seedling growth, as against, it produced maximum fresh weight (354.65 mg) and dry weight (21.35 mg) of seedlings. But, matured pod aqueous extracts of *Peltophorum* which had the highest supersession on germination and seedling growth of green gram continue to maintain the inhibitory effect and produced the lowest fresh weight (227.50 mg) and dry weight (15.80 mg) of seedlings (Table 1). Seed vigour index-I and II data revealed that *Delonix regia* flower (624.15 and 960.75) and *Peltophorum pterocarpum* matured pods (803.40 and 948.00) produced very weak seedlings with the lowest vigour index values respectively when compared to aqueous extracts.

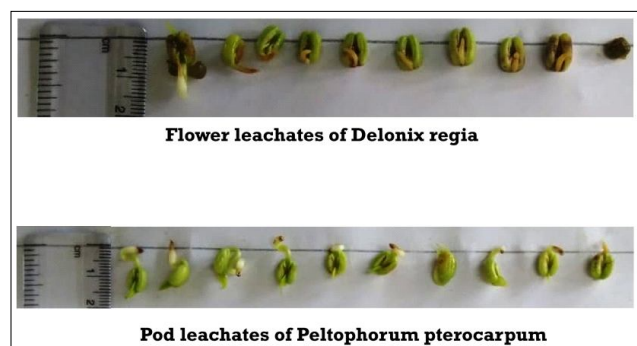
The RI indicated the highest negative values of germination (-0.430 and -0.241), shoot length (-0.187 and -0.215) and root length (-0.753 and -0.394) for flower

aqueous extracts of *Delonix regia* and matured pod aqueous extracts of *Peltophorum pterocarpum* respectively. Bark aqueous extracts *Delonix regia* had negative RI (-0.053) for fresh weight and all other tree parts had positive RI of fresh and dry weight. Bark (-0.079) and fresh pod (-0.102) aqueous extracts of *Peltophorum pterocarpum* registered negative RI and all other tree parts registered positive RI for the fresh and dry weight (Table 2). Considering cumulative effects, all aqueous extracts had affected germination and seedling growth and it was in the order of root length>germination>shoot length for *Delonix regia* aqueous extracts and root length>shoot length>germination for *Peltophorum* matured pods.

Among aqueous extracts of different tree parts of both species flower aqueous extracts of *Delonix regia* and matured pod aqueous extracts of *Peltophorum pterocarpum* had registered the lowest values of germination, shoot length, root length and highest negative values of RI (Fig 4).

*Delonix regia* flower aqueous extracts had a more inhibitory effect which may be occupied by a variety of allelochemicals (chlorogenic acid, 4-hydroxybenzoic, 3,4-dihydroxybenzoic, 3,5-dinitrobenzoic, L-azetidine-2-carboxylic 3,4-dihydroxybenzaldehyde, 3,4-dihydroxycinnamic and gallic acid) as reported by a team of scientists (Chou and Leu, 1992; Perveen *et al.*, 2018). Similarly, *Peltophorum pterocarpum* matured pod may contain more aliphatic alcohol, fatty acids, amino acids, terpenoids, phenolics, flavonoids, alkaloids, steroids chemicals as inferred by a group of scientists (Shyamal *et al.*, 2014; Taiwo *et al.*, 2013).

Studying the qualitative and quantitative distribution of carotenoids in flowers of *Delonix regia* indicated that flower petals contained 29 carotenoids viz. phytoene, phytofluene,  $\beta$ -carotene,  $\gamma$ -carotene, lycopene, rubixanthin, zeaxanthin, lutein while flower sepals contained 18 carotenoids (phytoene, phytofluene,  $\beta$ -carotene,  $\gamma$ -carotene, lycopene, etc), whereas filaments contain 20 (phytoene,  $\beta$ -carotene,  $\gamma$ -carotene, lutein, zeaxanthin, antheraxanthin, flavoxanthin, and other epoxy carotenoids) and anthers had the highest concentration of carotenoids with zeaxanthin accounting for



**Fig 3:** Allelopathic effect evident of seed discoloration on green gram.

**Table 1:** Inhibitory effect of *Delonix regia* and *Peltophorum pterocarpum* tree parts on green gram.

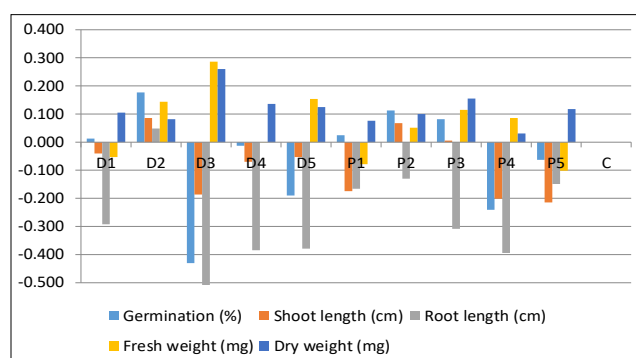
Treatment	Germination (%)	Shoot length (cm)	Root length (cm)	Fresh weight (mg)	Dry weight (mg)	Seedling vigour index-I	Seedling vigour index-II
D1- Bark	80.00±8.22	16.36±1.28	12.25±1.05	239.80±14.11	17.65±1.32	1308.80±10.52	1412.00±10.85
D2- Leaf	96.00±1.00	18.65±0.99	18.19±1.25	295.80±9.01	17.20±1.08	1790.40±0.99	1651.20±1.08
D3- Flower	45.00±17.80	13.87±1.36	4.27±0.37	354.65±16.25	21.35±2.15	624.15±24.21	960.75±38.27
D4- Pod	78.00±4.36	15.86±1.30	10.66±0.84	253.33±16.19	18.28±1.91	1237.08±5.67	1425.84±8.33
D5- Seed	64.00±8.12	16.14±1.14	10.75±1.28	299.22±19.19	18.06±1.34	1032.96±9.26	1155.84±10.88
P1- Bark	81.00±5.10	14.07±2.02	14.44±1.44	233.35±18.73	17.10±1.78	1139.67±10.30	1385.10±9.08
P2- Leaf	89.00±2.92	18.29±1.62	15.07±1.01	266.90±16.69	17.55±1.06	1627.81±4.73	1561.95±3.10
P3- Flower	86.00±5.79	17.15±1.21	11.98±0.79	286.05±15.46	18.70±1.22	1474.90±7.01	1608.20±7.06
P4- Matured pod	60.00±11.73	13.39±1.59	10.49±0.94	227.50±20.78	15.80±1.76	803.40±18.65	948.00±20.64
P5- Fresh pod	74.00±6.60	13.62±1.66	14.74±1.50	277.05±15.89	17.90±1.46	1007.88±10.96	1324.60±9.64
Distilled water	79.00±9.41	17.05±1.17	17.31±0.82	253.25±16.80	16.30±1.27	1346.95±11.01	1287.70±11.95

D - *Delonix regia*, P - *Peltophorum pterocarpum*. Data are mean values of four replicates with  $\pm$  standard error.

**Table 2:** Response index values for allelopathic effect of *Delonix regia* and *Peltophorum pterocarpum* tree parts on green gram.

Treatment	Germination	Shoot length	Root length	Fresh weight	Dry weight
D1- Bark	+0.013	-0.041	-0.292	-0.053	+0.105
D2- Leaf	+0.177	+0.086	+0.048	+0.144	+0.081
D3- Flower	-0.430	-0.187	-0.753	+0.286	+0.260
D4- Pod	-0.013	-0.070	-0.384	+0.000	+0.136
D5- Seed	-0.190	-0.053	-0.379	+0.154	+0.125
P1- Bark	+0.025	-0.175	-0.166	-0.079	+0.076
P2- Leaf	+0.112	+0.068	-0.130	+0.051	+0.100
P3- Flower	+0.081	+0.006	-0.308	+0.115	+0.155
P4- Matured pod	-0.241	-0.215	-0.394	+0.086	+0.031
P5- Fresh pod	-0.063	-0.201	-0.149	-0.102	+0.117
Distilled water	0.000	0.000	0.000	0.000	0.000

D - *Delonix regia*, P - *Peltophorum pterocarpum*.

**Fig 4:** Response index values of *Delonix regia* and *Peltophorum pterocarpum* tree parts on green gram.

90 per cent (Jungalwala and Chama, 1962). Further, alkaloids, flavonoids, proteins, tannins, carbohydrates, phenols, triterpenes, and steroids were also found in flowers of *Delonix regia* (Khursheed *et al.*, 2012; Rahman *et al.*, 2011; Shanmukha *et al.*, 2011; Shiramane *et al.*, 2011). The LC-MS studies characterized and confirmed the molecular structure of three major anthocyanins in water extract of *Delonix regia* flowers. Cyanidin 3-O-rutinoside and pelargonidin 3-O-rutinoside were identified in a concentration of 10.7 and 0.9 mg/l respectively (Adje *et al.*, 2008) and GC-MS analysis of flower extract revealed the presence of benzenetriol, butyl-8-methylnonyl ester, lupeol, and vitamin E as major compounds (Rani *et al.*, 2011).

About eighty-three phytochemicals have been reported in *Peltophorum pterocarpum* pods that include aliphatic alcohol, fatty acids, amino acids, terpenoids, phenolics, flavonoids, alkaloids, steroids (Nathan *et al.*, 2012) and matured pods had Quercetin, Rhamnetin, Rhamnetin, Meratin and Propelargonidin compounds (Polasek *et al.*, 2013; Menon *et al.*, 1982).

Parafiniuk and Czerwińska (2019) reported that bark aqueous extracts of eight tree species inhibited germination of maize, pigeon pea and sesame, with most inhibition in germination and radicle growth and plumule elongation in *Ailanthus excelsa*, followed by *Acacia nilotica*, *Dalbergia sissoo* while Swaminathan (1996) stated aqueous extracts

of bark generally inhibiting the growth of sesame most and pigeon peas least. On the contrary same author (Swaminathan *et al.*, 1990) in his earlier studies observed that aqueous extracts *Parthenium hysterphorus* weed has affected germination of four multi-purpose tree species viz., *Acacia leucophloea*, *Casuarina equisetifolia*, *Eucalyptus tereticornis* and *Leucaena leucocephala* and inhibition of germination and seedling growth is attributed to parthenin, an unsaturated lactone found in plant parts of weed species.

## CONCLUSION

The present study indicated allelochemicals causing inhibitory effects may be present more in flower parts of *Delonix regia* and matured pods of *Peltophorum pterocarpum* as they showed more inhibition to germination and seedling growth of green gram.

## Conflict of interest

All authors declared that there is no conflict of interest.

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