

## **Inhibitory and stimulatory effects of *Melia dubia* cav. drupe pulp on *Capsicum annuum* L. and *Vigna radiata* L. in laboratory and pot culture bioassays**

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### **ABSTRACT**

We determined the *Melia dubia* Cav. drupe pulp phytochemicals and the effects of drupe pulp aqueous extracts (0, 25, 50, 75 and 100 %) on germination in laboratory bioassays. Besides, in pot culture the effects of pulp powder (25, 50, 75 and 100 g pot<sup>-1</sup>) were determined on germination, seedling growth, biomass and quantitative and qualitative attributes of *Capsicum annuum* L. and *Vigna radiata* (L.) R. Wilczek till crop maturity. Through GCMS analysis, 24 phytochemical compounds were detected in pulp. In laboratory bioassays, aqueous extracts significantly inhibited the germination of bell pepper (*C. annuum*) but not of green gram (*V. radiata*). In pot culture studies, pulp powder applied at 25 and 50 g had synergetic effects on the growth, development and biomass of both the test crops (2 months after drupe pulp treatment). The pulp powder at 50 g lead to maximum growth and biomass in *C. annuum*. Pulp powder at 50 g pot<sup>-1</sup> only enhanced the plant height of *C. annuum* by 5.83 %, over control, while, higher doses were inhibitory. On the contrary all the pulp powder doses stimulated the growth and seed yield of *V. radiata*, but the 100 g pulp was less stimulatory. This indicated that the *Melia dubia* drupe pulp is not harmful to the test plants; instead act as organic material to enhance the plant growth. Thus test crops could be grown in *M. dubia* based agroforestry system.

**Key Words:** Agroforestry, bell pepper, *Capsicum annuum*, GC-MS, germination, green gram, inhibition, *Melia dubia*, phytochemical, pot culture, seedling growth, stimulation, *Vigna radiata*, yield.

### **INTRODUCTION**

In agroforestry land use system, complex allelopathic interactions occur which could be positive or negative. The overall effects of trees on understorey vegetation/crops depends on the positive and negative effects in response to spatial and temporal interactions in both shoot and root growth. Agroforestry system components may have some phytochemicals with positive or negative effects on germination, growth and yield performance of companion crops. The component parts (leaf litter, exfoliating bark, flowers, fruits/pods, etc.) of donor tree may have stimulatory or inhibitory effects on the performance of associated crops. These interactions involve the production and release of chemical substances from the plants, which can promote or inhibit the growth and development. Compared to forestry trees, agroforestry tree species have been little investigated for the allelopathic influences (13,37) as they co-exist with the agricultural crops and their allelopathic compatibility may be crucial to determine the success of an agroforestry system.

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Nowadays, fast growing tree species are being promoted in agroforestry to sustain food, livelihood and environmental security *vis-a-vis* developing productive land use systems to reduce the impact of climate change (20). In agroforestry systems, compatibility of all components specially trees and crops are decisive for its success. Allelopathy is one of the important aspects that determines the tree-crop compatibility. Therefore, it is imperative that tree species with minimum or no negative effects on understory crops should be selected in agroforestry. One of such species is *Melia dubia* Cav. (family Meliaceae), commonly known as *Malabar neem* (Fig. 1). It is a large fast-growing deciduous tree found in India from 100-1500 m altitude. Its timber is mainly used in plywood and pulp industries, making furniture, agricultural implements, construction etc. and can be harvested after 4-5 years (35). Hence, it is being promoted in agroforestry across India (10,26). It produces huge number of fruits which may have alleged allelochemicals. Therefore, *M. dubia* drupe pulp may contain phytochemicals with inhibitory or stimulatory effects on associated crops.



Figure 1. Young (left) and middle aged (right) trees of donor specie *Melia dubia*

India is one of the largest producer of capsicum (2). It is used as conventional nutritional food and also as non-food (spiritual, ethnobotanical) uses (25). Mungbean (*Vigna radiata* L.) is an important nutritive crop in human diet and also improves the soil fertility by fixing the atmospheric nitrogen (28,31). Hence, this study aimed to (i). Find the *M. dubia* Cav. drupe pulp phytochemicals, (ii). Influence of drupe pulp aqueous extracts on germination of test crops in laboratory bioassays and (iii). Effects of pulp powder on germination, seedling growth, biomass and yield of test crops : *Capsicum annum* and *Vigna radiata* till crop maturity in pot culture.

## MATERIALS AND METHODS

The study was done in our laboratory and green house, at ASPEE College of Horticulture and Forestry, Navsari Agricultural University, Navsari (20.95° N latitude, 75.90° E longitude, altitude: 10 m above msl), Gujarat, India from December, 2019 to June, 2020. Mean annual rainfall: 1096 mm, mean maximum and minimum temperatures: 40 °C and 17 °C, respectively.

### **Drupe pulp chemical analysis**

*M. dubia* Cav. drupe pulp chemical compounds were determined by Gas Chromatography-Mass Spectrometry (GC-MS) as per Kumar *et al.* (21).

### **Preparation of aqueous extracts**

Fresh and mature *M. dubia* drupes were collected during January from the forests in Northern Western Ghats parts of Gujarat and de-pulped. Pulp was dried in partial shade and grounded. Two hundred g grounded pulp was added to 1.0 L distilled water. The solution was stirred and kept at room temperature (20-25 °C) for 24 h. The filtrate was centrifuged and supernatant was decanted (19) and resultant stock solution was of 20 % concentrations. From the stock solution dilutions of 25, 50, 75, 100 % concentrations were prepared with distilled water for laboratory bioassay.

### **Laboratory bioassay**

Fifty seeds of bell pepper (*Capsicum annuum* L. var. Indra) and green gram (*Vigna radiata* (L.) R. Wilczek var. GM-6) were sown equidistant in 90 mm dia petri-dishes lined with Whatman No. 1 filter paper. Each treatment was replicated 4-times in complete randomized design. Five ml of drupe pulp aqueous extract was applied per petri dish on first day and afterwards distilled water was applied on alternate day to keep the filter paper moist till the end of experiment (15 days for bell pepper and 5 days for green gram) (21). Distilled water was used as control. Seeds were considered germinated after the emergence of 1 mm long radicle. The germinated seeds were counted daily till 15 days for bell pepper and 5 days for green gram from the date of application of aqueous solution and germination (%) was calculated.

### **Pot culture**

Pot experiments were done to investigate the effects of *M. dubia* drupe pulp powder on germination, initial growth and biomass of test crops. Fifty seeds of each test crop were sown (on 15 January, 2020) in plastic pots (25 cm dia., 20 cm height) containing 5 kg red soil: vermicompost in 2:1 ratio. Nitrogen, phosphorous, potassium and organic carbon at the time of start and after completion of study (in all treatments) are presented in Table 1. The seedlings were uprooted on 6 February, 2020 for growth and biomass measurements.

The drupe pulp powder at 25 g (T<sub>2</sub>), 50 g (T<sub>3</sub>), 75 g (T<sub>4</sub>) and 100 g (T<sub>5</sub>) was mixed in the upper soil layer in pots (21). The control treatment was without pulp powder (T<sub>1</sub> control). The treatments were replicated four times in complete randomized design. The pots were irrigated with tube well water (pH 7.71 and electrical conductivity 1.75 dS/m) and test crop seeds were sown 24 h after irrigation. The pots were kept in green house (75 % light penetration) and were irrigated as and when required. The data on emerged seedlings were counted daily upto 15 days. Seedling radical and plumule growth and biomass were recorded 20 days after sowing.

To evaluate the influence of drupe pulp powder for longer duration in pot soil on test crop growth, biomass and yield till maturity, a separate experiment was done with same procedure as adopted in above pot experiment. Each pulp powder treatment (as per pot experiment above) was replicated 4-times (10 plants per replication @ 1 plant/pot). Bell pepper seedlings at 3-4 leaf stage were transplanted in pots. Green gram was sown directly (2 seeds per pot and 15 days old seedlings were thinned to single plant). Plant height, collar diameter, root length, number of leaves, branches and biomass of both test

crops were recorded after 2 months of drupe pulp powder treatment (MADPT). In addition, number of flowers and pods as well as grain yield was recorded for green gram.

**Nutritional attributes of *V. radiata*:** Protein, phosphorus, potassium, calcium, magnesium and sulphur content were analyzed to know the effects of drupe pulp powder of *M. dubia* on nutritional value of green gram seeds.

### Statistical analysis

The experimental data of all characters studied were subjected to the statistical analysis for interpretation. The data was analyzed following completely randomized design (CRD) (33). Further, Duncan's multiple range test (DMRT) was used to compare the sets of means of each treatment using WASP (Web Agri Stat Package) (17).

## RESULTS AND DISCUSSION

Addition of mulch or other plant parts in crops enhance the organic matter, decomposition rate and other soil activities increase the plant growth. Apparently, pre- and post harvest analysis of pot soil in this study (Table 1) showed significant differences in soil fertility status of control and *M. dubia* drupe pulp applied at 25 to 100 g/pot. Hence, resulted in better growth, biomass and yield of test crops in this study. Such incorporations, significantly increases the total organic, total extractable, humified and non-humified C forms, and available K contents increased the yield of receiver crops (14,24). Some allelochemicals influence the availability of nutrients in the soil. Phenolics are an important group of allelopathic compounds, which activates the solubilization and release of Fe, P and other nutrients, thereby, helping their uptake by plants (12,30). Therefore, such allelopathic compounds might have improved the nitrogen use efficiency in our study and resulted in synergetic and stimulatory effects on test crops in pot culture.

Table 1. Effects of applied *M. dubia* drupe pulp powder on the fertility of pot soil after harvest at 60 days

Drupe pulp Powder dose (g pot <sup>-1</sup> )	Nitrogen (N) (kg/ha)		Phosphorus (P <sub>2</sub> O <sub>5</sub> ) (kg/ha)		Potassium (K <sub>2</sub> O) (kg/ha)		Organic carbon (%)	
	<i>C. annuum</i>	<i>V. radiata</i>	<i>C. annuum</i>	<i>V. radiata</i>	<i>C. annuum</i>	<i>V. radiata</i>	<i>C. annuum</i>	<i>V. radiata</i>
	Before seedling transplanting/seed sowing							
*Fertility	264.32		54.88		425.60		2.16	
	After harvest at 60 days							
0 (Control)	278.88 <sup>b</sup>	284.48 <sup>c</sup>	46.48 <sup>c</sup>	51.52 <sup>c</sup>	435.68 <sup>c</sup>	437.36 <sup>c</sup>	1.93	1.91
25	284.48 <sup>b</sup>	287.84 <sup>c</sup>	48.72 <sup>c</sup>	54.32 <sup>c</sup>	441.84 <sup>c</sup>	443.52 <sup>bc</sup>	1.95	1.93
50	295.12 <sup>ab</sup>	300.05 <sup>bc</sup>	53.76 <sup>bc</sup>	58.24 <sup>bc</sup>	456.96 <sup>bc</sup>	459.20 <sup>bc</sup>	2.01	1.98
75	309.68 <sup>a</sup>	312.48 <sup>ab</sup>	59.92 <sup>ab</sup>	64.40 <sup>b</sup>	487.20 <sup>ab</sup>	482.72 <sup>ab</sup>	2.08	2.07
100	314.16 <sup>a</sup>	327.60 <sup>a</sup>	66.08 <sup>a</sup>	73.92 <sup>a</sup>	515.20 <sup>a</sup>	508.48 <sup>a</sup>	2.14	2.12
SE(m)±	7.65	7.57	2.30	2.61	11.90	12.42	0.01	0.01

\*Initial Fertility status.

### *M. dubia* drupe pulp phytochemicals

Through gas-chromatography mass-spectrometry (GC-MS), 24 phytochemical compounds were detected (Table 2). Cyclohexane, 4-hydroxy-2-hexanoic acid, anhydro-rotunol, Crinan-11-One and 22, 23-dihydrospirosteron were most abundant (Fig. 2). The detected compounds have been corroborated with available literature for their allelopathic

proclivity to interpret the results of laboratory and pot culture bioassays of the present study.

Table 2. *M. dubia* drupe pulp phytochemical compounds detected through Gas Chromatography–Mass Spectrometry (GC-MS) and their relative percentage

Sr. No.	Compound name	Molecular formula	Molecular weight	Area under curve	Relative area (%)
1.	1-Ethylcyclopentane	C <sub>7</sub> H <sub>12</sub>	96	438006	0.02
2.	Cyclobutane	C <sub>4</sub> H <sub>8</sub>	92	3162817	0.12
3.	2-florobenzoic acid	C <sub>14</sub> H <sub>7</sub> CL <sub>2</sub> FO <sub>3</sub>	312	4676422	0.17
4.	1-H-indane	C <sub>10</sub> H <sub>8</sub>	128	484138	0.02
5.	Hydroxylamine	C <sub>10</sub> H <sub>23</sub> NO	173	1594579	0.06
6.	4-acetoxy-3-methoxystyrene	C <sub>11</sub> H <sub>12</sub> O <sub>3</sub>	192	1232554	0.05
7.	2-Hydroxyl-1-octanol	C <sub>14</sub> H <sub>30</sub> O	214	12088899	0.45
8.	6-Floro-2-trifluoromethylbenzoicacid	C <sub>15</sub> H <sub>6</sub> CL <sub>2</sub> F <sub>4</sub> O <sub>3</sub>	280	2388561	0.09
9.	2-10-bornenidol	C <sub>10</sub> H <sub>18</sub> O <sub>2</sub>	170	2067453	0.08
10.	3-methyl-2-furan	C <sub>8</sub> H <sub>10</sub> O <sub>2</sub>	138	2363561	0.09
11.	Dimethyl-2-carboxy-cyclopropene	C <sub>10</sub> H <sub>14</sub> O <sub>6</sub>	230	88622961	3.28
12.	4-hydroxy-2-hexacanoic acid	C <sub>8</sub> H <sub>14</sub> O <sub>3</sub>	158	590580701	21.84
13.	Methyl-6,9-octadecadinonate	C <sub>19</sub> H <sub>34</sub> O <sub>2</sub>	294	22325709	0.83
14.	Bicyclo heptane	C <sub>12</sub> H <sub>22</sub>	166	95105995	3.52
15.	Cyclohexane	C <sub>20</sub> H <sub>36</sub>	276	894687640	33.09
16.	Oxirane	C <sub>8</sub> H <sub>16</sub> O	128	16354224	0.60
17.	5,10-Pentadecadenoic acid	C <sub>15</sub> H <sub>26</sub> O <sub>2</sub>	238	28415244	1.05
18.	1-Hexayl-2-Nitrocyclohexane	C <sub>12</sub> H <sub>23</sub> NO <sub>2</sub>	213	15726389	0.58
19.	Butylphosphonic acid	C <sub>28</sub> H <sub>59</sub> O <sub>5</sub> P	506	25196072	0.93
20.	Ethyl monate A	C <sub>10</sub> H <sub>32</sub> O <sub>7</sub>	372	46524512	1.72
21.	Crinan-11-One	C <sub>18</sub> H <sub>19</sub> NO <sub>6</sub>	345	217072225	8.03
22.	22,23-dihydrospinasteron	C <sub>29</sub> H <sub>48</sub> O	412	127494052	4.72
23.	Anhydro-rotunol	C <sub>15</sub> H <sub>20</sub> O	216	400803330	14.82
24.	Carbamic acid	C <sub>10</sub> H <sub>12</sub> CLNO <sub>3</sub>	229	104359100	3.86

#### A. LABORATORY BIOASSAY

The results of laboratory bioassays revealed that *M. dubia* drupe pulp aqueous extract (0, 25, 50, 75 and 100 %) significantly ( $P < 0.05$ ) inhibited the germination (%) of *C. annuum* L.; however, there was 100 % germination in *V. radiata* L. (Table 3). In bell pepper, germination inhibition was maximum at 75 and 100 % drupe pulp aqueous extract. The *C. annuum* germination inhibition (%) gradually increased with increase in aqueous extract concentration (Table 3).

Table 3. Inhibitory effects of *M. dubia* drupe pulp aqueous extract on germination and inhibition (%) of *C. annuum* (15 DAET) and *V. radiata* (5 DAET) in laboratory bioassays

Pulp aqueous extract concentration (%)	<i>C. annuum</i>	<i>V. radiata</i>
Control: Distilled Water	96.50 (84.17) <sup>a</sup> [-29.53]	100 <sup>a</sup>
25	68.00 (56.48) <sup>b</sup> [-36.27]	100 <sup>a</sup>
50	61.50 (51.81) <sup>bc</sup> [-53.37]	100 <sup>a</sup>
75	45.00 (42.04) <sup>cd</sup> [-74.61]	100 <sup>a</sup>
100	(29.56) <sup>d</sup> [-74.61]	100 <sup>a</sup>
SE(m)±	4.45	-

DAET : Days after extract treatment; SE(m)± : Standard error of treatments means; Values in round parenthesis are arc sign transformed values; Values in square parenthesis are percent reduction (-) over control; Means with different and same superscript letter in the same column indicate significant and insignificant difference ( $p < 0.05$ ) according to Duncan's Multiple Range Test, respectively.

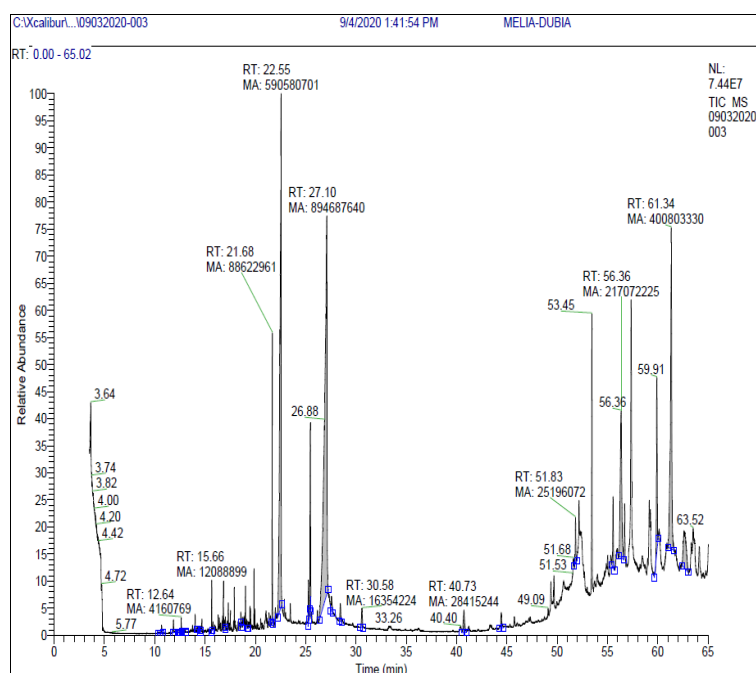


Figure 2. Chromatogram showing retention time and relative abundance of phytochemical compounds detected in *M. dubia* drupe pulp

## B. POT CULTURE

### (i). Germination and seedling growth

(a). *C. annuum* : The application of *M. dubia* drupe pulp powder doses 25, 50, 75 and 100 g per pot, stimulated and inhibited ( $P < 0.05$ ) the germination (Fig. 3; 15 DADPT) early seedling growth and biomass attributes (20 DADPT) of *C. annuum* seedlings

(Table 4). Application of 50 g drupe powder stimulated the seed germination (10.5 %) and also the shoot and root growth (10.56 and 4.75 cm, respectively) over the control. However, 75 and 100 g doses of drupe powder were inhibitory (Table 4). Shoot (53.49 %) and root length (24.02 %), and shoot, root and total biomass experienced maximum enhancement (77.78, 167.24 and 88.89 %, respectively) at 50 g powder dosages over control (Table 4; figures in square parenthesis). The 100 g drupe pulp inhibited the germination, initial growth of (20 DADPT) shoot and biomass. Higher doses were less stimulatory but rather inhibitory to *C. annuum*.

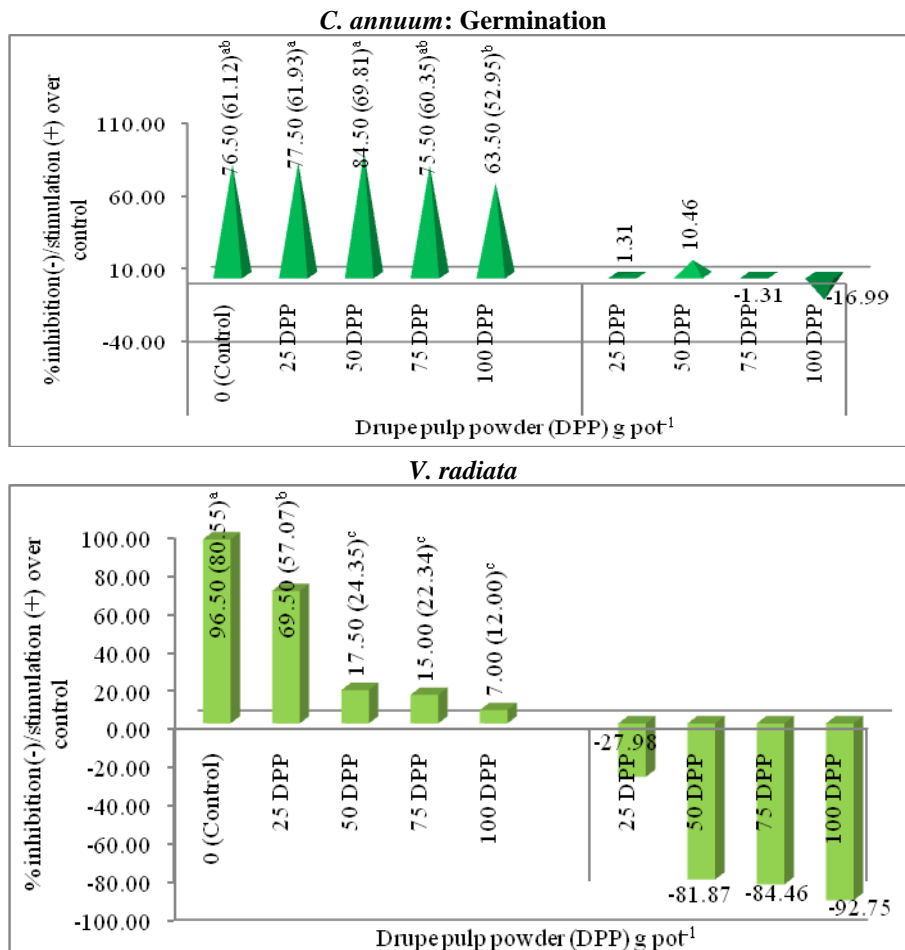


Figure 3. Effects of *M. dubia* drupe pulp powder on germination (%) of *C. annuum* and *V. radiata* 15 DADPT (Days after drupe pulp treatment) in pot culture. Means with different superscript letter in the bars indicate significant difference ( $p < 0.05$ ) according to Duncan's Multiple Range Test. Values in parenthesis are arc sign transformed values; SE(m)  $\pm 2.28$  and  $4.13$  for *C. annuum* and *V. radiata*, respectively)

Table 4. Influence of *M. dubia* drupe pulp powder on *C. annuum* seedling growth and biomass in pot culture at 20 DADPT

Drupe pulp powder (g pot <sup>-1</sup> )	Growth (cm)		Biomass (DM g/plant)		
	Shoot length	Root length	Shoot	Root	Total
0 (Control)	6.88 <sup>c</sup>	3.83 <sup>b</sup>	0.09 <sup>c</sup>	0.006 <sup>d</sup>	0.09 <sup>cd</sup>
25	7.99 <sup>b</sup> [+16.13]	3.69 <sup>b</sup> [-3.66]	0.10 <sup>b</sup> [+11.11]	0.009 <sup>b</sup> [+58.62]	0.11 <sup>b</sup> [+22.22]
50	10.56 <sup>a</sup> [+53.49]	4.75 <sup>a</sup> [+24.02]	0.16 <sup>a</sup> [+77.78]	0.016 <sup>a</sup> [+167.24]	0.17 <sup>a</sup> [+88.89]
75	8.00 <sup>b</sup> [+16.28]	3.25 <sup>c</sup> [-15.14]	0.09 <sup>c</sup> [0.00]	0.009 <sup>b</sup> [+46.55]	0.10 <sup>c</sup> [0.00]
100	6.48 <sup>c</sup> [-5.81]	4.67 <sup>a</sup> [+21.93]	0.07 <sup>d</sup> [-22.22]	0.007 <sup>c</sup> [+17.24]	0.07 <sup>d</sup> [-22.22]
SE(m) ±	0.24	0.13	0.0033	0.0004	0.004

DADPT : Days After Drupe Powder Treatment; DM: Dry Matter; SE(m)± : Standard error of treatments means; Values in parenthesis are arc sign transformed values; Values in square parenthesis are percent reduction (-)/ enhancement (+) over control; Means with different superscript letter in the same column indicate significant difference ( $p < 0.05$ ) according to Duncan's Multiple Range Test.

(b). *V. radiata*: In *V. radiata*, *M. dubia* drupe pulp powder inhibited the seeds germination and inhibitory effect increased with increase in drupe pulp dose (Fig. 3). On the contrary, pulp powder was stimulatory as well as inhibitory to growth and biomass of green gram (Table 5). The 25 g dose of *M. dubia* pulp stimulated the shoot and root length by 3.16 and 3.76 %, respectively over control. Likewise, the root biomass was also enhanced at 50, 75 and 100 g pulp powder application.

Table 5. Influence of *M. dubia* drupe pulp powder on *V. radiata* seedling growth and biomass in pot culture (20 DADPT)

Drupe pulp powder (g pot <sup>-1</sup> )	Growth (cm)		Biomass (DM g plant <sup>-1</sup> )		
	Shoot length	Root length	Shoot	Root	Total
0 (Control)	19.96 <sup>a</sup>	6.11 <sup>a</sup>	0.14 <sup>a</sup>	0.009 <sup>c</sup>	0.15 <sup>a</sup>
25	20.59 <sup>a</sup> [+3.16]	6.34 <sup>a</sup> [+3.76]	0.13 <sup>b</sup> [-7.14]	0.009 <sup>c</sup> [0.00]	0.13 <sup>b</sup> [-13.33]
50	9.89 <sup>b</sup> [-50.45]	4.80 <sup>b</sup> [-21.44]	0.03 <sup>d</sup> [-78.57]	0.011 <sup>b</sup> [+25.66]	0.04 <sup>d</sup> [-73.33]
75	10.39 <sup>b</sup> [-47.95]	4.35 <sup>c</sup> [-28.81]	0.09 <sup>c</sup> [-35.71]	0.009 <sup>c</sup> [+5.56]	0.09 <sup>c</sup> [-40.00]
100	8.25 <sup>c</sup> [-58.67]	6.18 <sup>a</sup> [+1.15]	0.03 <sup>d</sup> [-78.57]	0.019 <sup>a</sup> [+111.11]	0.05 <sup>d</sup> [-66.67]
SE(m)±	0.49	0.19	0.0033	0.0004	0.006

DADPT = Days After Drupe Powder Treatment; DM- Dry Matter; SE(m)± = standard error of treatments means; Values in parenthesis are arc sign transformed values; Values in square parenthesis are percent reduction (-)/ enhancement (+) over control; Means with different superscript letter in the same column indicate significant difference ( $p < 0.05$ ) according to Duncan's Multiple Range Test

These results showed the stimulatory and inhibitory effect of drupe pulp aqueous extracts/powder of *M. dubia* on germination, growth and biomass of test crops viz., *C. annuum* and *V. radiata*. The test crops behaved differently to lower and higher concentration of aqueous/powder extracts of *M. dubia*, in laboratory and pot culture studies. Pot culture proved that *M. dubia* drupe pulp powder had stimulatory and inhibitory effects on germination, seedling growth and biomass of test crops, which depended on the pulp powder quantity (concentration) and attribute of receptor crops.

In general, pulp powder at 25 and 50 g per pot stimulated the germination and growth of test crops, however, 75 and 100 g drupe powder was inhibitory to *C. annuum*. Several allelopathic studies with fruit (pulp extract) as donor in laboratory bioassays reported deleterious effects on test crops due to presence of phytochemical compounds like cyclohexane, 6-floro-2-trifluoromethyl benzoic acid (11,27,32). The phenolic acids and esters like 6-floro-2-trifluoromethyl benzoic acid from *Medicago arborea* (7), carbamic acid (40), oxirane and 3-methyl-2-furan (3), hydroxylamine, cyclohexane, derivatives of 3-methyl-2-furan (22) are allelopathic in nature. Similarly, 1,4- benzenedicarboxylic acid dimethyl ester, melianone, melianol and melianodinol detected in *M. dubia* drupe pulp in this study, proved allelopathic in laboratory bioassays (18). Hence, germination inhibition of *C. annuum* in our study may be ascribed to similar phytochemical compounds present in the *M. dubia* pulp aqueous extract.

Inhibitory effects of walnut fruit hull and leaf extracts (37), *Syzygium cumini* fruit pulp in both bioassay and pot culture (5), aqueous solution of fruits of *Eucalyptus camaldulensis*, *Melia azedarach* and *Sapindus mukorossi* (18), aqueous extract of *Sapindus saponaria* fruit (13) and *Phytolacca dioica* fruit aqueous extracts (6) on several test crops lend support to present findings. Different compounds are responsible to cause allelopathic effects in various plant species by affecting morphological, anatomical, physiological, cytological and biochemical processes (34). The restriction in water uptake and enzyme inhibition decreases the seed protease activity and play key role in protein hydrolysis during germination and results in germination inhibition (1,23,29).

On the other hand, several pot culture bioassays have reported stimulatory effect of fruit pulp extract as donor organ on receptor crops. Stimulatory effect of walnut fruit hulls and leaves (37) and *Morinda citrifolia* fruit extract (9) on various test crops agree with present findings. Further, stimulation of germination and growth of *Rhynchosia capitata* at lower extract concentrations and inhibition at higher concentrations in laboratory and soil-incorporated residues in pot culture bioassays (1), lends support to our findings. Leaf and fruit extracts of various other donor species have shown similar synergetic as well as inhibitory effects on different vegetable and pulses crops (15,34,19).

## (ii). Growth and yield

(a). *C. annuum* : The results indicated that, *M. dubia* drupe powder application (25, 50, 75, 100 g pot<sup>-1</sup>) significantly ( $P < 0.05$ ) affected the later (2 MADPT) growth and biomass (Fig. 4) in *C. annuum*.

Study revealed that drupe pulp powder dose of 50 g pot<sup>-1</sup> enhanced plant height (5.83% over control; Fig. 5), whereas, all other powder doses had inhibitory effects (2 MADPT). Collar diameter, root length, number of leaves per plant and total dry biomass experienced enhancement in response to all pulp powder doses over control. Maximum enhancement in most of the growth and biomass attributes was against 50 g pulp powder treatment (Fig. 5).

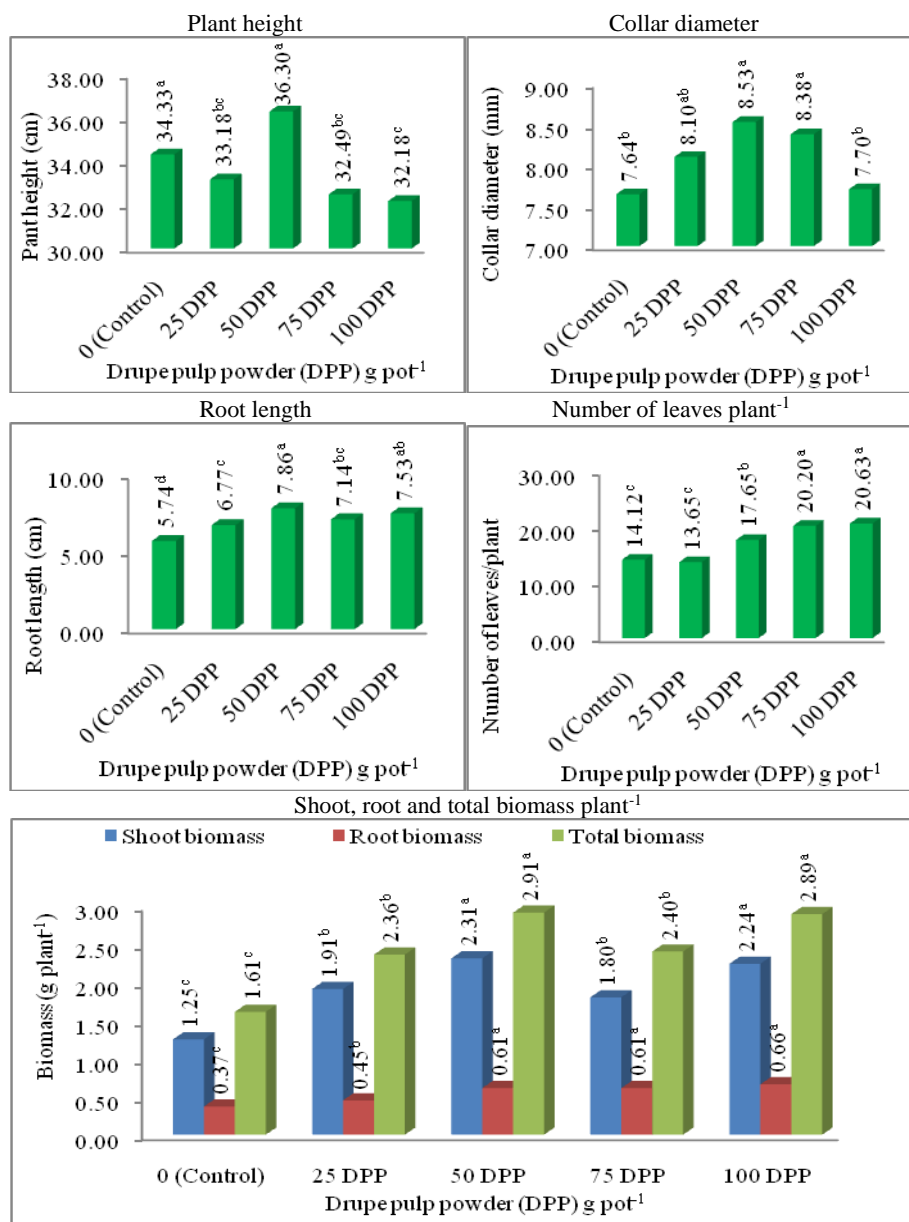


Figure 4. Effects of *M. dubia* drupe pulp powder (2 MADPT) on Plant height [SE(m) $\pm$ 0.63], Collar diameter [SE(m) $\pm$ 0.19], Root length [SE(m) $\pm$ 0.22], Number of leaves/plant [SE(m) $\pm$ 0.56] and Shoot-root and total biomass [SE(m) $\pm$ 0.07, 0.02 and 0.08, respectively] of *C. annuum* in pot culture. Means with different superscript letter in the bars indicate significant difference ( $p < 0.05$ ) according to Duncan's Multiple Range Test.

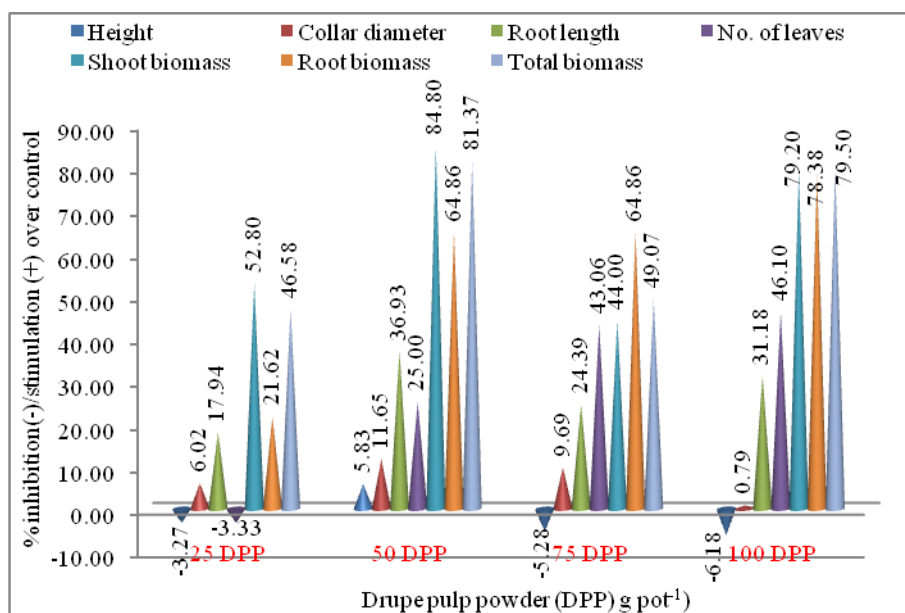


Figure 5. Effects of *M. dubia* drupe pulp powder on plant height, collar diameter, root length and number of leaves/plant (2 MADPT) of *C. annuum* in response to *M. dubia* drupe pulp powder applied in pot culture

(b). *V. radiata* : Apart from early stage, *M. dubia* drupe pulp powder application (25, 50, 75, 100 g pot<sup>-1</sup>) also showed significant ( $P < 0.05$ ) influence on the later (2 MADPT) growth, development and biomass in *V. radiata*. In contrary to *C. annuum*, the plant growth, biomass (Fig. 6), reproductive development and seed yield (Table 6) of *V. radiata* experienced stimulatory effects in response to all the pulp powder doses. The study evinced that magnitude of stimulation increased with increase in pulp powder quantity (Fig. 7, Table 6; numbers in square parenthesis), except in case of root length, number of branches, number of pods and plant dry biomass.

Table 6. Influence of *M. dubia* drupe pulp powder on reproductive growth and seed yield (2 MADPT) of *V. radiata* in pot culture

Drupe pulp powder (g/pot)	No. of flowers plant <sup>-1</sup>	No. of pods plant <sup>-1</sup>	Seed yield (g plant <sup>-1</sup> )
0	6.18 <sup>d</sup>	3.63 <sup>d</sup>	0.72 <sup>c</sup>
25	6.92 <sup>c</sup> [+11.97]	4.17 <sup>c</sup> [+14.88]	1.12 <sup>d</sup> [+55.56]
50	6.79 <sup>c</sup> [+9.87]	5.15 <sup>b</sup> [+41.87]	1.30 <sup>c</sup> [+80.56]
75	7.55 <sup>b</sup> [+22.17]	4.56 <sup>c</sup> [+25.62]	1.98 <sup>b</sup> [+175.00]
100	10.25 <sup>a</sup> [+65.86]	8.11 <sup>a</sup> [+123.42]	2.40 <sup>a</sup> [+233.33]
SE(m)±	0.15	0.16	0.05

Note: MADPT = Months after drupe powder treatment; DM- Dry matter; SE(m)± = standard error of treatments means; Values in square parenthesis are % reduction (-) / enhancement (+) over control; T<sup>1</sup>; Means with different superscript letter in the same column indicate significant difference ( $p < 0.05$ ) according to Duncan's Multiple Range Test.

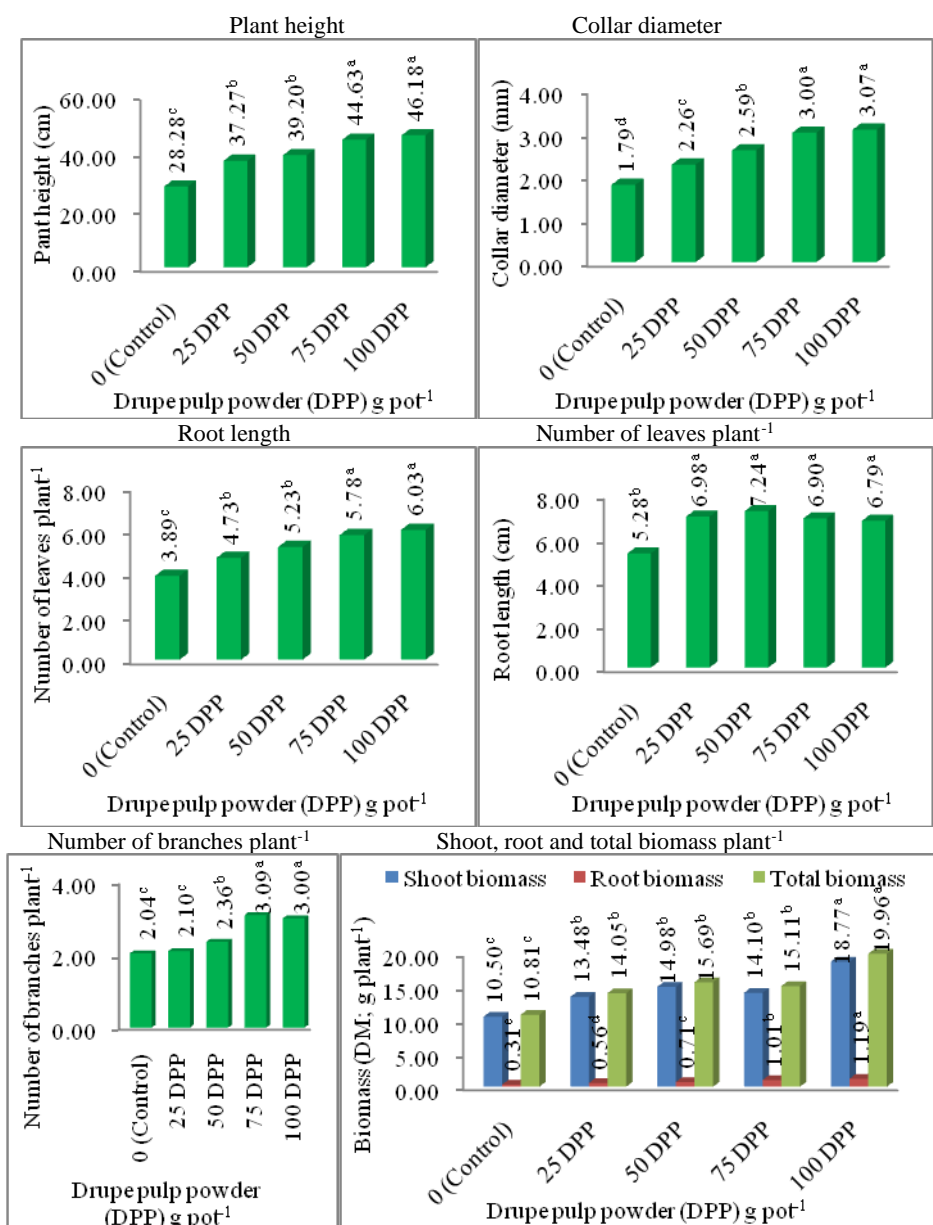


Figure 6. Effects of *M. dubia* drupe pulp powder on Plant height [SE(m) $\pm$ 1.05], Collar diameter [SE(m) $\pm$ 0.04], Number of leaves plant<sup>-1</sup> [SE(m) $\pm$ 0.18], Root length [SE(m) $\pm$ 0.20], Number of branches/plant [SE(m) $\pm$ 0.05] and Shoot-root and total biomass [SE(m) $\pm$ 0.58, 0.02 and 0.59, respectively] of *V. radiata* in pot culture (2 MADPT). Means with different

superscript letter in the bars indicate significant difference ( $p < 0.05$ ) according to Duncan's Multiple Range Test.

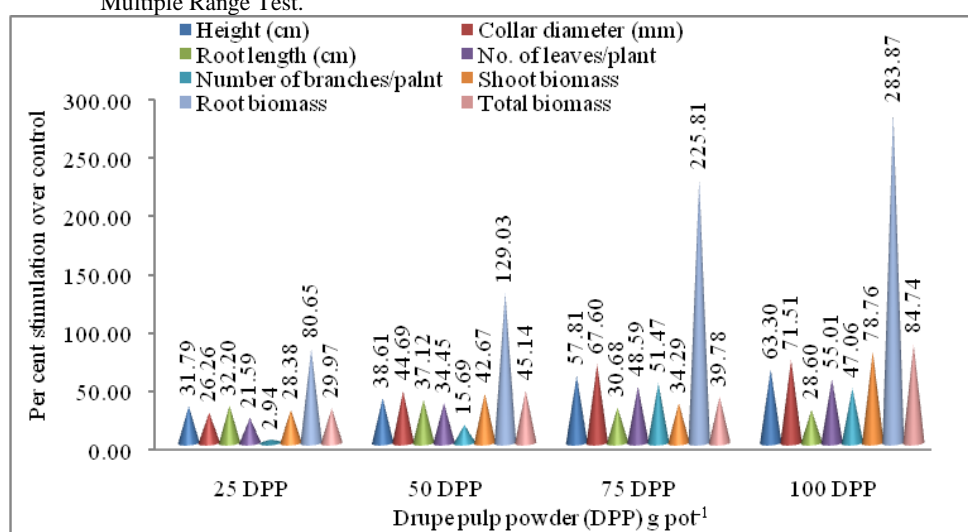


Figure 7. Stimulatory effects of *M. dubia* drupe pulp powder application on plant height, collar diameter, root length, number of leaves plant<sup>-1</sup>, number of branches plant<sup>-1</sup>, shoot-root and total biomass (2 MADPT) of *V. radiata* in pot culture

Apart from growth and biomass, nutritional attributes viz., protein, phosphorous (P), potassium (K), calcium (Ca), magnesium (Mg) and sulphur (S) content of *V. radiata* seeds were also analyzed. These nutritional attributes did not experienced any significant effect due to *M. dubia* drupe pulp powder application (Table 8). This showed that, incorporation of *M. dubia*, by and large, had no deleterious effects on nutritional parameters in *V. radiata*.

Table 7. Influence of *M. dubia* drupe pulp powder on nutrients contents of *V. radiata* grain nutritive attributes in pot culture

Drupe pulp powder (g/pot)	Protein content (%)	Phosphorous (%)	Potassium (%)	Calcium (%)	Magnesium (%)	Sulphur (%)
0 (Control)	1.65 <sup>a</sup>	0.27 <sup>a</sup>	0.93 <sup>a</sup>	0.16 <sup>a</sup>	0.11 <sup>a</sup>	0.12 <sup>a</sup>
25	1.67 <sup>a</sup>	0.25 <sup>a</sup>	0.94 <sup>a</sup>	0.16 <sup>a</sup>	0.10 <sup>a</sup>	0.12 <sup>a</sup>
50	1.66 <sup>a</sup>	0.26 <sup>a</sup>	0.92 <sup>a</sup>	0.16 <sup>a</sup>	0.10 <sup>a</sup>	0.12 <sup>a</sup>
75	1.67 <sup>a</sup>	0.25 <sup>a</sup>	0.91 <sup>a</sup>	0.15 <sup>a</sup>	0.11 <sup>a</sup>	0.11 <sup>a</sup>
100	1.66 <sup>a</sup>	0.25 <sup>a</sup>	0.93 <sup>a</sup>	0.15 <sup>a</sup>	0.11 <sup>a</sup>	0.11 <sup>a</sup>
SE(m)±	0.014	0.004	0.009	0.004	0.004	0.004

SE(m)± : Standard error of treatments means; Values prefixed '±' are SE(m); Means with same superscript letter in the same column indicate non-significant difference ( $p < 0.05$ ) according to Duncan's Multiple Range Test

In pot culture studies, crops growth till maturity, showed that *M. dubia* drupe pulp powder enhanced the later growth, development and yield of *V. radiata*. This may be attributed to nature of phytochemicals present in pulp powder and its behavior over the period of time in soil, effect of cultural practices followed after imposition of treatments,

soil physico-chemical properties before imposition of powder treatment and after harvest of crops (Table 1). Most of the allelopathic studies are restricted to petri dish bioassays or even if done in pot using soil are limited to a short period of one month or so. However, present study reported the response of test crops till their maturity (2 months). Similarly, many studies reported allelopathic effects in laboratory bioassays, however, no such effects were observed on plant growth, development and yield of receptor crops in pot studies done till crop harvest (21).

In field conditions, a simple compound may not be enough to affect the growth of the receiving plant and different allelochemicals act additively or synergistically (4,36). This may happen due to (i). Biomass decomposition, (ii). Leaching of allelochemicals due to frequent irrigations done to maintain the soil moisture in pots, (iii). Transient nature of allelochemicals and (iv). Their loss from soil through volatilization, especially phenols, (v). Other physiochemical processes, (vi). Microbial breakdown and (viii). Uptake by plants resulting in lower concentration of soil allelochemicals (16). Oxidation and sorption are the primary factors involved in the disappearance of allelochemicals in soil (39). These evidences lend support to our findings.

## CONCLUSIONS

The *M. dubia* drupe pulp powder chemicals have allelopathic/antagonistic effects on germination, growth and biomass of bell pepper and green gram. However, the effects were transitory as indicated in pot culture studies upto germination, seedling growth, crop maturity and harvest. Hence, vegetable and pulse crops can be grown with *M. dubia* trees without any harmful effects. The drupe pulp as by-product may be used to raise seedlings for commercial plantations, added to forest nurseries to enhance soil fertility and for higher plant growth and yield.

## DECLARATION

We declare that all authors of this Ms have made substantial contributions. We did not exclude any author who substantially contributed to this Ms. We have followed our ethical norms established by our respective institutions.

## CONFLICT OF INTEREST

The authors announce that they have no conflict of interest.

## ETHICAL APPROVAL

The authors declare that the study was carried out following scientific ethics and conduct. However, this study did not involve any use of animals, hence no ethical approval has been obtained from the concerned committee.

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