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RESEARCH PAPER

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Phytotoxic potential of *Lycium edgeworthii* miers against three cultivated test crop species

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Key words: Allelopathy, L. edgeworthii, T. aestivum, T. alexandrinum, L. sativa.

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Abstract

Allelopathy can play a crucial role in future weed management and crop productivity. The allelopathic compounds tend to be used as natural herbicides and alternative pesticides; they are less riotous of the global ecosystem than artificial agrochemicals. Therefore, the investigations were made to test the potential of L. edgeworthii by using aqueous extracts from various parts. The parts of plant were collected shade dried, grinded and different extracts were made. All the extracts significantly arrested the germination rate and overall growth of Triticum aestivum L., Lactuca sativa L. and Trifolium alexandrinum L. in laboratory trials. The aqueous extracts with higher concentration 10g/100ml were more noxious than lower concentration 5g/100ml. Likewise, the extracts obtained after 72 hours were more inhibitory than 24 and 48 hours. Bark extract was found to be more toxic than leaves extracts. The robust inhibition i.e. 40%, 44% and 36% showed by 10/100ml at 72 hours' duration of bark followed by leaves 28%, 24% and 20% in germination of T. aestivum, L. sativa and T. alexandrinum respectively, similarly same concentration and duration showed maximum inhibition in radicle and plumule length i.e bark showed 70.16% and 73.33% in Triticum aestivum respectively. Hot water extracts, litter and mulching experiments also ascertained to be inhibitory to all the test species. It was proposed that various evaluated parts of L. edgeworthii have robust allelopathic potential contrary to the nominated tested crop species. Further exploration is required to find the chemicals that triggered inhibition, which may provide basis for the development of novel herbicides of biological origin.

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Introduction

"Allelopathy is the influence of a plant (including microorganisms) on other plant (which live the vicinity) via the release of chemicals constituents into the environments" (Khan et al., 2011). Different types of chemicals that are release to the environment by plants which can cause stimulatory or inhibitory effect in alternative plants in their vicinity in the time of initial growth of that target plant by absorbing the chemicals from the environment, when positive effect produced by the plant, termed facilitation, and if negative effects prevail, the interaction results in competition or interference (AN et al., 1996; Vidal and Bauman, 1997; Javaid and Anjum, 2006; Padilla and Pugnaire, 2006; Fiorentino et al., 2007; Kong et al., 2008; Mutlu and Atici, 2009; Sisodia and Siddiqui, 2010; Gantayet et al., 2014). Allelopathy usually is a form of destructive biochemical communication that donor release while acceptor absorb (Mancini et al., 2009). The plants depend on each other for various life support requirements' that live in same niche (Ells and Mcsay, 1991; Ben-Hammouda et al., 2002; Quan et al., 2003; Khan et al., 2011). Different process such as stomata opening and closing, cell wall permeability, photosynthesis, respiration is adversely affected by the chemical that released by the plants in their vicinity (Nektarios et al., 2005; Javaid, 2010; Bouchikh-Boucif et al., 2014). Allelochemicals are usually stored by the plant cell in bound form therefore it is not noxious but become toxic when exonerate to the environment (Inderjit et al., 2008; Terzi, 2008; Kamal, 2011) deprived growth of the crops that can grow unceasingly in the same field year after year is due to the allelochemicals in the soil (Chen et al., 2011; Duke, 2015). Phenolic are well recognized to effects numerous physiological processes of plants (Siddiqui and Arif-Uz-Zaman, 2005; Li et al., 2010; Al-Watban and Salama, 2012). Flavonoids are polyaromatic compound having 15carbon skeleton and having about 10,000 types (Shaw et al., 2006) it can play a numeral role in plants (Taylor and Grotewold, 2005; Pourcel et al., 2006; Buer et al., 2010; Cesco et al., 2012; Weston and Mathesius, 2013). Vanillic, cis-ferulic acid, trans-pcoumaric, cis-p-coumaric, syringic, p-hydroxybenzoic

acid, trans-ferulic and 2, 4-dihydroxy-7-methoxy-1, 4-benzoxazin-3-one (DIMBOA) have been secluded from wheat shoot and roots (Qasim, 2001; Li *et al.*, 2010; Yadav and Singh, 2013).

Much agronomic importance can be accomplished by means of natural compound for weed managements such as, soil quality enhancement, to reduce environmental deterioration triggered by synthetic agrochemicals and crop diversity is increased through crop rotation which diminished the growth of pest and weeds (Sanchez et al., 2004; Khanh et al., 2005; MA et al., 2006; Machado, 2007; Araniti et al., 2012; Araniti et al., 2012; Saad and Abdelgaleil, 2014). Thousands of natural compounds have been sequestered and their structures elucidated but relatively few of these have been adequately tested for phytotoxicity (Duke et al., 2012; Pacanoski et al., 2014). Therefore, the current investigation was made to evaluate the phytotoxic potential of Lycium edgeworthii Miers in contradiction of Triticum aestivum L., Trifolium alexandrinum L., and Lactuca sativa L.

Material and methods

Healthy plants of *L. edgeworthii* were collected from Charsadda. Leaves and bark were separated and diverse parts were distinctly kept for shade drying at room temperature (20°C-25°C). The crushed material of each part was distinctly packed in airtight plastic bag for further research activities.

Aqueous extract bioassay

Leaves and bark powder in amount of 5g and 10g of the collected plant were soaked in 100ml distilled water separately for shorter 24 hours, medium 48 hours and longer 72 hours' durations at room temperature (20°C-25°C). The extracts were then filtered and adjust the pH at 6.5. All the extracts and distilled water as control were utilized in contradiction of test species viz. *L. sativa, T. alexandrinum* and *T. aestivum* in Petri dishes. Ten seeds were positioned in each Petri dish having two folds moistened filter paper with the respective aqueous extract, while distilled water was used as negative control.

Five replicates each having 10 seeds were taken for each handling. After 72 hours' duration the percent germination, radicle and plumule length in millimeter, dry and fresh weight in milligrams and moisture content were noted. 10 seedlings were selected indiscriminately for dry and fresh weight determination in each treatment.

Hot water extract bioassay

Ten-gram powder of leaves and bark of *L. edgeworthii* were separately boiled and filtered. The extracts were then cooled and were practiced in contradiction of the above mentioned test species.

Mulching bioassay

To find the effects of mulching 5g of plant powder were mixed discretely in sterilized and humidified sand in pots. 5 replicates per handling and five as a control were made, each with 10 seeds of the test species, *L. sativa*, *T. alexandrinum* and *T. aestivum*. Pieces of filter paper were used in control handling to intensify water holding capability of sand taken and keep the seeds humid.

Litter bioassay

Five grams' powder litter of *L. edgeworthii* placed discretely in Petri dishes, with a superficial single

filter paper dampened with 5ml water. A double layer of filter paper was used in control treatment. Five replicates for each treatment were made, each with 10 seeds of the test species.

The Petri dishes were incubated at 25°C for 7 days. After 7 days of germination the roots and shoots length, dry and fresh weight and percent germination were determined. All the collected data were statistically analyzed through one-way ANOVA. The analysis was done using SPPS/PC version 2017.

Results and discussion

Effect of aqueous extracts

Both the aqueous and hot water extracts significantly affected various growth parameters and germination of all the test species. Extreme inhibition caused by the bark extract at higher concentration 10g/100ml at longer soaking duration 72 hours as compared to leaves extract in all the test species. All the species *T. aestivum*, *L. sativa* and *T. alexandrinum* showed highest inhibition by bark extracts *viz.*, 40%, 44% and 36%, however hot water extracts of bark caused 30%, 52% and 40% inhibition in germination, while 28%, 24% and 20% by leaves extracts respectively (Table No. 1).

Table 1. Effect of aqueous extracts of *L. edgeworthii* on the germination of the test species.

Treatments/extracts	Test species				
	T. aestivum	L. sativa	T. alexandrinum		
Control	100±0.00	100±00.00	100±0.00		
5g/100ml leaf. 24 hrs.	82±3.74 ^{Ns}	86±4.00 ^{Ns}	90±3.16 ^{Ns}		
5g/100ml leaf. 48 hrs.	80±4.47 ^{Ns}	84±6.78 ^{Ns}	88±4.90 ^{Ns}		
5g/100ml leaf. 72 hrs.	78 ± 2.00^{Ns}	82±4.90 ^{Ns}	86±2.45 ^{Ns}		
10g/100ml leaf. 24 hrs.	76±6.00 ^{Ns}	80±3.16*	84±6.78 ^{Ns}		
10g/100ml leaf. 48 hrs.	74±6.00*	78±7.35*	82±3.74*		
10g/100ml leaf. 72 hrs.	72±7.35*	76±6.78*	80±3.16*		
5g/100ml bark. 24 hrs.	70±4.49*	70±5.48*	74±2.45*		
5g/100ml bark. 48 hrs.	68±8.60*	68±6.63*	72±3.74*		
5g/100ml bark. 72 hrs.	66±6.78*	66±5.10*	70±3.16*		
10g/100ml bark. 24 hrs.	64±5.10*	64±2.45*	68±2.00*		
10g/100ml bark. 48 hrs.	62±6.20*	60±2.00*	66±4.00*		
10g/100ml bark. 72 hrs.	60±7.75*	58±2.00*	64±2.45*		
10g/100ml H.W.L.E.	82±3.74 ^{Ns}	66±5.10*	78±5.83*		
10g/100ml H.W.B.E.	70±3.16*	48±3.74*	60±7.07*		

Key; H.W.L.E = Hot water leaves extracts, H.W.B.E = Hot water bark extract. Ns = Non-significant, * = Significant at $\alpha > 0.01$. Each value is the Grand mean and standard error of 5 replicates, each with 10 seeds.

This agrees with (Hussain and Ilahi, 2009; Hussain *et al.*, 2010) who's also proved that aqueous and hot water extract of *Cenchrus* and *Bothriochla* has the strong allelopathic behavior against test species. Our result also supported by (Lodhi and Nickell, 1973; Lodhi, 1976).

Likewise, in the length maximum inhibition was recorded as 61.9% for T. alexandrinum, 67.41% for L. sativa and 70.16% for T. aestivum while in case of plumule length T. aestivum showed highest reduction 73.33% by aqueous water extract of bark (Table No. 2).

Table 2. Effect of aqueous extracts of *L. edgeworthii* on the length of radicle and plumule of the test species.

Test species	T. aestivum		L. sativa		T. alexandrinum	
Treatments	Radicle	Plumule	Radicle	Plumule	Radicle	Plumule
Control	12.4±0.55	10.5±1.09	9.82±0.55	12.5±0.71	6.3±0.35	9.3±1.20
5g/100ml leaf. 24 hrs.	5.4±0.77*	4.1±0.68*	3.8±0.85*	5.0±0.11*	4.5±0.47*	6.7±1.50*
5g/100ml leaf. 48 hrs.	5.3±0.58*	4.0±0.53*	3.8±0.72*	5.0±0.23*	4.4±0.45*	6.6±0.93*
5g/100ml leaf. 72 hrs.	5.3±1.31*	4.0±0.84*	3.8±1.08*	4.9±0.32*	4.2±0.21*	6.4±0.64*
10g/100ml leaf. 24 hrs.	5.2±1.46*	3.8±0.79*	3.6±0.67*	4.8±0.56*	3.7±0.51*	6.3±0.49*
10g/100ml leaf. 48 hrs.	5.2±1.60*	3.6±1.12*	3.6±1.08*	4.8±0.89*	3.2±0.24*	6.2±0.22*
10g/100ml leaf. 72 hrs.	5.0±1.44*	3.6±1.25*	3.6±1.06*	4.8±0.49*	3.0±0.38*	6.2±0.73*
5g/100ml bark. 24 hrs.	4.3±1.25*	3.3±1.60*	3.7±1.07*	3.9±0.49*	2.9±0.38*	5.8±0.73*
5g/100ml bark. 48 hrs.	4.1±1.32*	3.2±1.34*	3.6±0.77*	3.9±1.08*	2.8±0.34*	5.6±0.71*
5g/100ml bark. 72 hrs.	4.0±1.29*	3.2±0.73*	3.6±0.72*	3.6±0.68*	2.8±1.03*	5.5±0.61*
10g/100ml bark. 24 hrs.	3.9±1.09*	3.2±0.84*	3.5±0.77*	3.5±0.46*	2.7±0.29*	5.5±1.11*
10g/100ml bark. 48 hrs.	3.8±1.58*	2.9±1.25*	3.4±0.96*	3.3±0.81*	2.7±0.60*	4.7±1.02*
10g/100ml bark. 72 hrs.	3.7±1.38*	2.8±1.10*	3.2±0.37*	3.0±0.44*	2.4±0.32*	4.7±0.63*
10g/100ml H.W.L.E.	3.9±0.56*	3.5±0.60*	3.8±0.49*	2.8±0.37*	4.7±0.54*	5.7±0.55*
10g/100ml H.W.B.E.	3.5±0.28*	2.6±0.40*	2.8±0.37*	2.0±0.63*	2.6±0.51*	2.8±0.37*

Key; H.W.L.E = Hot water leaves extracts, H.W.B.E = Hot water bark extract. Ns = Non-significant, * = Significant at α > 0.01. Each value is Grand mean and standard error of 5 replicates each having 10 seeds.

Likewise, all the test species less percent of control value i.e 59.2%, 30.9% and 43.9% in fresh weight and 53.7%, 18.7% and 13.0% in dry weight by *aqueous* bark extracts whereas 65.1%, 48.2% and 53.2% fresh weight and 59.5%, 43.7% and 21.0% dry weight by

leaves extracts, while in case of hot water bark extracts 50.5%, 41.8% and 58.8% in fresh weight and 46.3%, 14.6% and 30.9% in dry weight was found for *T. aestivum*, *L. sativa*, *T. alexandrinum* respectively (Table No. 3).

Table 3. Effect of aqueous extracts of *L. edgeworthii* on the fresh and dry weight of the test species.

Treatments	fresh weight (% of Control)			Dry weight (% of Control)			
	T. aestivum	L. sativa	T. alexandrinum	T. aestivum	L. sativa	T. alexandrinum	
5g/100ml leaf. 24 hrs.	66.2±0.91*	66.1 ±0.45*	59.8±0.63*	60.9±0.46*	63.5±1.73*	30.6±0.79*	
5g/100ml leaf. 48 hrs.	66.1±1.47*	62.7±0.49*	57.1±1.39*	60.9±1.29*	63.2±1.86*	30.1±1.82*	
5g/100ml leaf. 72 hrs.	65.4±1.67*	60.7±0.80*	56.3±2.73*	60.7±1.76*	59.8±1.88*	28.4±1.46*	
10g/100ml leaf. 24 hrs.	65.3±1.65*	57.4±0.92*	55.3±2.79*	60.2±1.34*	57.2±1.11*	27.4±1.15*	
10g/100ml leaf. 48 hrs.	65.3±1.91*	50.0±0.58*	55.0±1.56*	60.1±1.07*	44.6±0.87*	24.9±1.30*	
10g/100ml leaf. 72 hrs.	65.1±0.09*	48.2±0.32*	53.2±2.38*	59.5±0.72*	43.7±0.40*	21.0±0.12*	
5g/100ml bark. 24 hrs.	64.9±0.73*	46.4±3.37*	52.6±0.75*	57.3±2.75*	41.7±1.24*	20.0±0.67*	
5g/100ml bark. 48 hrs.	64.4±2.20*	38.0±1.39*	51.4±0.33*	56.7±0.19*	27.6±0.92*	17.8±0.73*	
5g/100ml bark. 72 hrs.	63.9±1.21*	34.6±1.16*	49.2±1.74*	56.5±1.92*	22.8±0.91*	16.4±0.94*	
10g/100ml bark. 24 hrs.	61.4±1.27*	33.3±2.40*	47.2±0.11*	56.0±1.37*	21.3±2.43*	15.4±1.70*	
10g/100ml bark. 48 hrs.	61.3±2.61*	31.2±1.74*	44.9±0.27*	55.9±0.63*	18.7±0.32*	14.7±2.29*	
10g/100ml bark. 72 hrs.	59.2±2.43*	30.9±1.09*	43.9±1.69*	53.7±2.03*	18.7±1.10*	13.0±1.21*	
10g/100ml H.W.L.E.	60.5±0.33*	53.1±0.60*	75.7±0.32*	56.5±2.21*	21.2±0.44*	41.4±0.40*	
10g/100ml H.W.B.E.	50.5±2.43*	41.8±0.94*	58.8±0.32*	46.3±3.66*	14.6±0.31*	30.9±0.30*	

Key; H.W.L.E = Hot water leaves extracts, H.W.B.E = Hot water bark extract. Ns = Non-significant, * = Significant at $\alpha > 0.01$. Each value is the Grand mean and standard error of 5 replicates each having 10 seeds.

In case of moisture contents the less percent of control value was recorded for longer 72 hours' duration and higher 10g/100ml concentration of bark extract in *L. sativa*, *T. alexandrinum* and *T. aestivum* i.e. 04.0%, 49.2% and 56.2% respectively (Table No. 4).

Table 4. Effect of aqueous extracts of *L. edgeworthii* on the moisture contents and seminal roots of the test species.

Treatments	T. aestivum	L. sativa	T. alexandrinum	T. aestivum	
	Moisture contents % of	Moisture contents %	Moisture contents %	Mean number of seminal	
	control	of control	of control	roots (% of control)	
5g/100ml leaf. 24 hrs.	63.7±0.11*	38.7±0.31*	63.0±0.05*	46.4±0.24*	
5g/100ml leaf. 48 hrs.	63.6±0.13*	28.0±0.19*	58.8±0.09*	42.9±0.24*	
5g/100ml leaf. 72 hrs.	62.8±0.12*	26.2±0.23*	58.6±0.01*	39.3±0.20*	
10g/100ml leaf. 24 hrs.	62.8±0.09*	18.9±0.74*	57.5±0.03*	35.7±0.00*	
10g/100ml leaf. 48 hrs.	62.7±0.08*	14.5±0.78*	58.5±0.08*	32.1±0.20*	
10g/100ml leaf. 72 hrs.	62.6±0.04*	09.5±0.64*	58.3±0.07*	28.6±0.24*	
5g/100ml bark. 24 hrs.	62.6±0.14*	06.9±0.56*	58.1±0.04*	25.0±0.24*	
5g/100ml bark. 48 hrs.	62.2±0.10*	06.3±0.45*	57.6±0.03*	21.4±0.20*	
5g/100ml bark. 72 hrs.	61.6±0.11*	06.0±0.30*	55.1±0.06*	17.9±0.00*	
10g/100ml bark. 24 hrs.	58.6±0.09*	05.6±0.88*	52.7±0.11*	14.3±0.20*	
10g/100ml bark. 48 hrs.	58.4±0.05*	05.0±0.98*	49.5±0.09*	10.7±0.24*	
10g/100ml bark. 72 hrs.	56.2±0.14*	04.0±0.46*	49.2±0.10*	07.1±0.24*	
10g/100ml H.W.L.E.	57.4±0.11*	65.2±0.34*	83.6±0.04*	57.1±0.49*	
10g/100ml H.W.B.E.	46.5±0.17*	48.2±0.44*	61.0±0.07*	46.4±0.51*	
Leaves mulching	86.0±0.07*	61.0±0.41*	57.5±0.09*	25.8±0.24*	
Bark mulching	71.0±0.08*	73.4±0.54*	41.5±0.03*	52.3±0.68*	
Litter	83.6±0.10*	03.2±0.53*	08.3±0.05*	50.0±0.24*	

Key; H.W.L.E = Hot water leaves extracts, H.W.B.E = Hot water bark extract. Ns = Non-significant,* = Significant at $\alpha > 0.01$. Each value is the Grand mean and standard error of 5 replicates each having 10 seeds.

Effect of mulching and litter

The mulching and litter of L. edgeworthii significantly inhibited the germination and overall growth of the selected test crop species. In case of mulching the bark caused maximum reduction in growth and germination of selected crop species followed by the leaves (Table No. 5). Our finding agrees with those of (Sher et al., 2011) used Populus euphratica, (Sarah et al., 2011) used Polypogon monspeliensis as mulching who's reported the same manners of allopathic potential of mulching and litter of the selected plant. The highest inhibition by mulch of the plant in germination was recorded for bark i.e 66%, 60% and 60%, similarly highest recorded value of inhibition in root (51.66%, 91.99% and 95.58%) and shoot (41.24%, 79.10% and 87.57%) in T. aestivum, L. sativa and *T*. alexandrinum

respectively. The less percent of control value in fresh weight and moisture contents was recorded for T. alexandrinum caused by bark mulch i.e 51.10% and 41.5%, whereas in dry weight 26.20% percent of control value was showed by leaves mulch in L. sativa. It was also noticed that L. sativa and T. alexandrinum were the most suppressive species than T. aestivum (Table No. 4, 5). Our results are in agreement with those of (Khan et al., 2005) who's also reported the allelopathic potential of Accacia nilotica (Maharian et al., 2007) Parthenium hysterophorus, (Khan et al., 2008) Eucalyptus camaldulensis, (Siddiqui et al., 2009) Prosopis juliflora, (Barkatullah et al., 2010) Dodonaea viscosa, and (Sher et al., 2011) Populus euphratica, which reduced the germination as well as overall growth of test species.

Table 5. Effect of mulching of *L. edgeworthii* on the of germination and overall growth of the test species.

	Test species							
Treatments	T. aestivum	L. sativa	T. alexandrinum	T. aestivum	L. sativa	T. alexandrinum		
			Germinat	ion				
Control	100.0±0.00	100.0±0.00	100.0±0.00	100±0.00	100±0.00	100±0.00		
Leaves	64.0±2.45*	46.0±4.00*	50.0±3.16*	84.0±2.45*	26.6±0.68*	38.3±0.28*		
bark	34.0±2.45*	40.0±3.16*	40.0±3.16*	-				
Root length								
Control	72.4±1.12	14.8±0.37	12.8±0.37	13.3±0.58	20.6±0.40	12.5±0.22		
Leaves	42.2±0.49*	7.2±0.48*	05.4±0.51*	03.6±0.41*	01.4±0.26*	02.7±0.31*		
Bark	35.8±0.73*	05.8±0.37*	03.2±0.20*	_				
			Shoot len	gth				
Control	106.2±0.60	47.4±0.51	31.2±0.58	07.6±0.24	14.7±0.38	23.4±0.24		
Leaves	70.2±0.80*	24.2±0.80*	15.4±0.51*	01.9±0.30*	01.7±0.13*	03.1±0.32*		
Bark	62.4±0.51*	22.2±0.58*	13.2±0.37*	-				
Fresh weight % of control								
Leaves	86.4±0.51*	65.9±3.82*	66.1±2.32*	91.5±1.52*	27.4±0.45*	14.4±0.29*		
Bark	72.2±0.37*	58.5±0.58*	51.1±1.00*	-				
Dry weight % of control								
Leaves	83.9±0.40*	46.2±0.40*	59.8±0.37*	96.7±0.98*	31.9±0.29*	24.5±0.25*		
Bark	72.6±0.86*	26.9±0.71*	39.9±0.37*	-				

Key; Ns = Non-significant, * = Significant at $\alpha > 0.01$. Each value is the Grand mean and standard error of 5 replicates each having 10 seeds.

Effect of all parameters on seminal roots growth and number of T. aestivum

The number of seminal roots of *T. aestivum* were also significantly inhibited by all the parameter including aqueous extracts, mulching and litter (Table No. 4). The inhibition in the seminal root number and growth was found in the same manner that higher concentration at longer soaking duration of bark aqueous extract caused maximum inhibition followed by the leaves extract. This agree with (Sarah *et al.*, 2011) reported the mulching, litter and aqueous extracts of *Polypogon monspeliensis* and (Ahmad *et al.*, 2014) reported *Celtis australis* as a allelopatic plant, which showed strong effect in case of germination, overall growth and seminal roots numbers.

Overall finding agrees with (El-Khatib *et al.*, 2004) who's reported *Chenopodium murale*. Leaf litter

leachates of Derris scandens, Cymbopogon citratus, Tamarindus indicia and Gliricidia sepium (Fritz et al., 2007) used Ethanolic extracts from Hypericum myrianthum which retarded the germination and overall growth of the species. This results also supported by (Nasrine et al., 2011) who's reported Euphorbia guyoniana, Retama retam, Bromus tectorum, Melilotus indica and Triticum aestivum. (Nekonam et al., 2014) reported Crocus sativum, Ricinus communis, Nicotine tabacum, Datura inoxia, Nerium oleander and Sorghum vulgare which showed significant inhibitory effect on overall growth of the test species.

Conclusion

From the present results, it was concluded that *L. edgeworthii* has strong allelopathic potential against the selected crop species. Further exploration is required for isolation of the active phytochemicals

that cause inhibition, which may provide basis for the development of novel herbicides of biological origin. Biological herbicides are environment friendly and produce minimal adverse effects in comparison to synthetic herbicides.

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