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Research Article

Evaluation of Herbicidal Potential of Selected Mycoherbicidal Strain against a Noxious Weed Cassia *otusifolia* L

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Abstract

The management of weeds by chemical herbicides creating adverse effect on human and animal health so there is urgent need for alternative weed control option to save environment by an environmental friendly and sustainable options. Bioactive compound of fungi is great source for the development of new herbicide and its offer an ecofriendly alternative to control weeds. Here We have evaluated mycoherbicidal activity of cell free culture broth (CFCF) of selected strains against C*assia obtusifolia*. Phytotoxic contents of natural metabolites present in CFCF of *Curvularia lunata* (FGCCW#33), *Phoma herbarum* (FGCCW#07), *Myrothecium roridum* (FGCCW#03) and *Fusarium roseum* (FGCCW#61) against *Cassia obtusifolia* were evaluated by seedling bioassays. In preliminary bioassay, the CFCF of FGCCW#03 of 28 days old were shown maximum phytotoxic effect on seedling of *Cassia obtusifolia* at 100% concentration. The symptoms were observed as browning and necrosis of weeds. With effective formulation and cheap mass production of this mycoherbicidal strains can possibly use as mycoherbicide for control of *Cassia obtusifolia* in future.

Keywords: Weeds; Natural Products; Mycoherbicide; *Cassia obtusifolia*; Bioassay

Introduction

A weed is a plant out of place, with a competitive and aggressive behavior, which is persistence and resistance to control and is useless, unwanted and undesirable. Weeds are ubiquitous and their management is required to maximize agricultural productivity. In somewhat they play major role in land and water resources but do noxious and deleterious impact to agriculture is very vast [2].

Cassia obtusifolia syn *Cassia tora* is an annual and herbaceous weed belong to the Leguminosae family. It is considered as obnoxious and aggressive weed. It is 1 m tall (rarely 2m) with slender tough stems which occurs as wastelands rainy season wild plant in India. It is a wild weed that grow in most parts of India and world as a weed. The presence of weed is reported in USA, Brazil, India and Kenya. They have also reported in other parts of world viz., Queensland, the northern territory and northern western Australia. It is reported as noxious weed in South Africa. It is seen in waste places, moist wood barnyards, rich lands, quite common and sub gregarious in waste ground, cultivated fields of soybean and groundnuts throughout country. It is observed that the heavy infestation in forest interfere with the regeneration of forest. Due to covering it creates competition for light and nutrition with seedlings, so growth of seedlings of forest plant is affected. In Madhya Pradesh, this weed is associated with sorghum, maize, soybean, jowar, arhar and affects the yields of the same (Gupta, 2000). It is reported as very troublesome weed in row crops and causes problems in various countries viz. southern united states and causes problems in India, Malaysia, Java, the Philippines and some Pacific islands. The control of this weed is very difficult and we can get success by using sustained combination method or with the application of new mycoherbicidal approach by applying natural product of fungi. The management of this weed by physical method will not control this weed but reduces plant vigour with a programme of top dressing and restricted grazing. The weed is susceptible to a num-

ber of chemical herbicides including glyphosate, dichlorprop, acidflurofen and picloram. The current strategies of weed control are associated with accidental poisoning and resistance problems in weeds. Mostly chemical herbicides are single site modes of action and prone to develop resistance among weeds. This weed grows on any type of soil and having wide range of habitats. Due reduction of application and resistance problems, there is urgent need for alternative management of weed control. As disused above, need for development of multiple weed management strategies, one of the alternatives for chemical pesticide is exploitation of microorganisms especially fungal strains as mycoherbicide. This approach has attracted the attention of a large number of workers worlds over [6-16,18,21-28]. Application of mycoherbicide is very useful and potential weed control in non-cropped as well as cropped areas, water bodies, grazing area and other public area [29,30]. Biocontrol of this weed is being control by the fungal pathogen *Alternaria cassia* applied as mycoherbicide have given better than 96% control of weed and increased soybean yield. The biodegradability of pesticide is the main approach for using herbicide. Due to this reason and solve above mentioned problems, we have evaluated Cell Free Culture Filtrate (CFCF) of strains of *Curvularia lunata* (FGCCW#33), *Phoma herbarum* (FGCCW#07), *Myrothecium roridum* (FGCCW#03) and *Fusarium roseum* (FGCCW#61) against *Cassia obtusifolia*.

Material and Methods

Selection of strains

We have selected four strains from previously recovered fungi namely *Myrothecium roridum* (FGCCW#03), *Phoma herbarum* (FGCCW#07), *Curvularia lunata* (FGCCW#33) and *Fusariumroseum* (FGCCW#61) maintained in Fungal Germplasm Collection Centre of Mycological Research Laboratory, Jabalpur, M. P. They were isolated earlier from infected leaf, seeds, seedlings and rhizospheric soil of the various weed.

Mass production of culture filtrate

The 500 ml of Erlenmeyer's flasks containing 250 ml Potato dextrose's broth were seeded with 5 mm disc of mycelial round blocks separated from 7 days old cultures grown on PDA medium. The inoculated flasks were incubated at 28 ± 2°C in a Bio-Oxygen Demand (BOD) incubator (Remi, India) and the cell free culture filtrate (CFCF) were filtered after 7, 14, 21 and 28 days of incubation [7,20].

Extraction of CFCF

The incubated metabolized broth was passed through Whatman#1 filter paper under aseptic conditions through a preweighed Whatman #01 filter paper [17]. The mycelial were thrown and the supernatant were filtered *in vacuo* by microfiltration using sterile microfilters, 0.45 µm pore size, Mininsart (Sartorius, Gottingen, Germany) making it cell free [20,32]. Then Cell Free Culture Filtrate were obtained and used for various bioassay.

Seedling Bioassay:

The weeds *Cassia obtusifolia* seedling was grown in pots containing Soil: Sand: Peat (1:1:1). The 2 weeks to 3 weeks old seedling were treated with different days old CFCF (7, 14, 21, 28 days) and also with different dilutions (25%, 50%, 75% and 100%) of the phytotoxic metabolites and were then incubated. The effect of CFCF was determined by the method of visual phytotoxicity rating score method of Abbas., *et al.* [1] on rating scale of 0-5.

Results and Discussion

The table 1 shown the results regarding herbicidal activity of CFCF of four strains of fungi under varied incubation period. It showed significant influence on weed seedling mortalities. The highest phytotoxicity of seedling was found in 28 days old culture broth followed by 21 and 14 days old fermented broth in all strains. Less phytotoxicity were observed in 7 days old broth. The spray of only culture filtrate of FGCCW#03 caused maximum phytotoxicity on *Cassia obtusifolia*. We observed damages such as yellowing, followed by sharp wilting and complete collapse of weed seedling. The culture filtrate of *Curvularia lunata* (FGCCW#33), *Phoma herbarum* (FGCCW#07) and *Fusarium roseum* (FGCCW#61) has shown less effect on weed.

When comparing the phytotoxicity of control with various concentration of CFCF in seedling bioassays, the maximum phytotoxicity in seedling shown by 75% and 100% concentration. Less phytotoxicity was recorded in 25% and 50% concentration of CFCF to weed seedlings for all four strains. The strain FGCCW#03 recorded maximum effect even at concentration of 25% CFCF when spray on weed seedling. It clearly shown initial browning of leaves with 24 hrs of application (Table 2). The 25% culture filtrate of FGCCW#03 caused significant injury and induces symptoms such as chlorosis, wilt and collapse of weeds.

Strains	Seedling Bioassay Phytotoxicity			
	7 days VSR*	14 days VSR*	21 days VSR*	28 days VSR*
Control A	0	0	0	0
Control B	0	0	0	0
FGCCW#03	1	2	4	5
FGCCW#07	1	2	2	3
FGCCW#33	1	1	1	2
FGCCW#61	1	1	2	2

Table 1: Evaluation of different incubation time effect on CFCF of four selected fungi against *Cassia obtusifolia***

Results taken after 24 hrs treatment.

Control A: Sterilized Distilled Water, Control B- Growth media.

*Visual Scoring Rate

0: No Symptoms; 1: Slight Chlorosis; 2: Slight Chlorosis and Necrosis; 3: Marked Chlorosis and Necrosis; 4: Marked Chlorosis and High Necrosis; 5: Complete Death of Seedling.

Parameter and condition used for CFCF production: Incubation days= 28 days, Incubation temperature = 28^0C#1^0C.

Studies are based on means of three experiments.

Concentration	Seedling Bioassay Damage Rating			
	FGC-CW#03	FGC-CW#07	FGC-CW#33	**FGC-CW#61**
Control A	0	0	0	0
Control B	0	0	0	0
25%	3	0	0	0
50%	4	2	3	2
75%	5	3	3	3
100%	5	4	3	3

Table 2: Evaluation of various concentration of four strains CFCF against *Cassia obtusifolia* weed**

**Results taken after 24 hrs treatment.

Control A: Sterilized Distilled Water, Control B- Growth media.

Visual Scoring Rate.

0: No Symptoms; 1: Slight Chlorosis; 2: Slight Chlorosis and Necrosis; 3: Marked Chlorosis and Necrosis; 4: Marked Chlorosis and High Necrosis; 5: Complete Death of Seedling.

Parameter and condition used for CFCF production: Incubation days= 28 days, Incubation temperature = 28^0C#1^0C.

Studies are based on means of three experiments.

This study characterizes the presence of herbicidal active in fungus fermented cell broth and observed seedling bioassay inhibition. It has potential for the development of mycoherbicide. The application of secondary metabolites from fungi was also examined by various researcher [1,3,7,28]. The phytotoxic effects in post emergence were brown spots, necrosis, chlorosis and collapse of leaves of weed plants. On the basis of above findings, it can be concluded that the secondary metabolites produced by *Myrothecium roridum* coded as FGCCW#03 fungi have remarkable herbicidal potency need further evaluation for its large-scale application. Variation in phytotoxicity due to toxin has also been recorded by other researchers [1,3,4,7,19,31]. Similar results were reported by Makowski [5]; Pandey., *et al.* [7] Winder and Watson [33], when evaluating fermented broth herbicidal activity of fungi on weed plants. These results showed the mycoherbicidal action of the fermented broth CFCF produced by selected fungal strains. The phytotoxic compound present in fermented fungi caused symptoms viz., chlorosis, wilt and collapsing of weeds. In addition, we have observed same symptoms in treated weed.

Conclusion

The fermented broth CFCF of FGCCW#03 has showing post emergence herbicidal action of *Cassia obtusifolia*. it has controlled the seedling in 100%-25% concentration of CFCF. It caused leaf spots, necrosis and chlorosis of weeds. So, in preliminary observation FGCCW#03 strains fermented broth CFCF has proved to be an alternative mycoherbicide for the control these weeds, with a view of chemical pesticide reduction and maintaining environment from pollution.

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