

Outputs

Outputs

1. *Information on the mycobiota associated with the weed in India collected* National Research Centre for Weed Science, Jabalpur, India

Field Survey

A search was made for suitable plant pathogens for the biological management of *P. hysterophorus*. Surveys were conducted from March, 1996 to March, 1999. The areas surveyed were around Jabalpur, Katni, Seoni, Chargawa Road, Gadarwara, Mandla and NRCWS Farm. Some of the samples exhibited leaf spots and wilting of *P. hysterophorus* plants.

Isolation of pathogens

From infected samples of *P. hysterophorus*, the fungi, *F. pallidoroseum*, *Colletotrichum gloeosporioides*, *Alternaria alternata*, *Sclerotium rolfsii*, and *Sclerotinia sclerotiorum*, were isolated from Katni, Seoni, Chargawa Road, Gadarwara, Mandla, NRCWS Farm and around Jabalpur city. In the NRCWS Farm, *A. alternata* was found attacking the leaves, branches, and flowers of *P. hysterophorus*.

Fusarium pallidoroseum (Cooke). Sacc. (Hyphomycetes, Deuteromycotina)

The symptoms appeared as water soaked brown spots scattered on the leaf surface. These spots coalesced and formed larger brown spots. The seeds were shrivelled and small in size, however, no clear symptoms could be seen on the seeds. Under artificial inoculation the fungus infected seeds and seedlings, but no symptoms developed on leaves.

Culture pale to peach brown in reverse; aerial culture white, becoming somewhat compressed by the formation of effuse orange sporodochia due to the presence of conidial mass. Conidiophores formed on aerial culture which have loose branching structures with the formation of lateral and terminal conidiogenous cells. These may develop into polyblastic conidiogenous cells. Conidiophores formed in sporodochia are short and compressed and have a globose basal cell bearing a number of short, one celled branches which at the apex bear two to four short, cylindrical to pyriform phialides. Conidia hyaline, orange in mass, curved, basal cell septate. Chlamydospores, often sparse, are intercalary both in culture and conidia, globose, smooth; colonies fast growing on PDA (cover 9 cm petri dish in 5-6 days at 25°C).

Alternaria alternata (Fr.) Keissler - (Hyphomycetes, Deuteromycotina)

The symptoms appeared as small, oval discoloured lesions which were scattered on leaves. The spots became irregular in shape. When their size increased they turned brown to gray in colour. Sometimes concentric rings were formed by the yellow halo. Several such lesions may coalesce resulting in leaf drying. The symptoms also appeared on terminal branches and flowers. The colour of the flowers turned dark black and they aborted.

Culture light brown to grayish green, becoming black at maturity. Conidiophores light brown to golden brown, simple, branched, septate, straight or curved, smooth walled. Conidia light brown to olivaceous, borne long acropetal chains, ovoid or obclavate with a long or short beak, or ellipsoidal and without beak, smooth to echinulate, muriform with transverse and longitudinal septa. The beak, when present, is always smaller and lighter in colour than the conidial body.

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Sclerotium rolfsii Sacc. (Corticaceae, Basidiomycotina)

The affected plants appeared pale green and stunted. The infection occurred at the base of the plants around the collar region. The tissue of the infected portion softened and turned brown, leading eventually to leaf drop and plant death. White, fan-like mycelial growth was observed on the stem at the basal region. White to brown, sclerotia were also present.

S. rolfsii is a polyphagous fungus, and causes a number of rot diseases. In early stages of growth in pure culture the fungus culture is at first silky-white but gradually loses its lustre. Sclerotial initials are formed from hyphal strands which consist of 3-12 parallel hyphal strands. Mature sclerotia are dark-brown, about the size of a mustard seed, hard and usually round. Internal tissues of the sclerotium are white.

Sclerotinia sclerotiorum (Lib.) deBary (Sclerotiniaceae, Ascomycotina)

The infected plants first appeared pale green then wilted. Infection occurred at the base of the plants where a white cottony mycelial growth may be observed on the basal portion of the stem. On dissection, the stem exhibited black sclerotia within.

The sclerotia are white at first but later become black and hard on the outside and vary in size. They are more flattened and elongated than the spherical shape of *S. sclerotiorum*. Colonies are fast growing on PDA (cover 9 cm petridish in 4-5 days at 25°C).

Curvularia lunata (Wakker) Boedjin (Hyphomycetes, Deuteromycotina)

Culture at first hyaline, becoming brown. Conidiophores arise in tufts of 4-6 from subcultural stromata; erect, inflated at the base, dark brown, 3-10 septate, nodulose with spiral conidial scars. Conidia olive brown, usually curved, ellipsoid subcylindric, 3 septate, rounded at the base, 2 central cells, larger and darker than the two nearly hyaline end cells. Colonies fast growing on PDA (cover 9 cm petridish in 6-7 days at 25°C).

Trichoderma viride (Pers.) Fr.

Widespread in soil. Colonies fast growing on malt agar (cover 9 cm petri dish in four days at 20°C) and have a distinctive coconut odour when old.

Gliocladium virens Miller, Giddens & Foster

Often confused with *Trichoderma* spp. Colonies fast growing on PDA (cover 9 cm petridish in 7-8 days at 25°C).

Colletotrichum gloeosporioides (Penz) Sacc. (Coelomycetes, Deuteromycotina)

The symptoms appeared as irregular brown to deep brown spots of various sizes scattered all over the leaf surface. Under high humidity, the fungus grows rapidly forming elongated brown, necrotic areas. Infected leaves often exhibited shot hole symptoms. Disease incidence was higher on older leaves than the younger leaves.

The form genus *Colletotrichum* produces typically elongated, hyaline conidia with round ends. The conidia, which characteristically are slightly narrower in the middles than at the ends, are produced from phialides. Dark setae are often found in the acervuli of *Colletotrichum*, although this characteristic is variable, particularly under cultural conditions.

Outputs

Testing media for growth of different pathogens

For assessing the best medium for the growth of *F. pallidoroseum*, different media, such as potato dextrose agar, potato dextrose broth, sterilized moist soil, moist corn meal, soybean flour, moist cornmeal, moist soybean, fresh potato discs, pumpkin bits, moist gram, moist pea, moist jowar, moist wheat, were tested. Excellent growth of the fungus was obtained on potato dextrose agar, potato dextrose broth, arhar seed coat waste, pea seed coat waste and moist jowar. The virulence of the fungus was increased and death of *P. hysterothorus* seedling was more when the fungus was grown on pumpkin, soybean and wheat (Table 1).

Table 1: Comparative growth of *Fusarium pallidoroseum* on different media.

Sl. No	Name of test media	Comparative Growth
1.	Potato dextrose Agar	+++
2.	Potato dextrose broth	+++
3.	Potato sucrose Agar	+
4.	Neem oil cake	+
5.	Mustard oil cake	++
6.	Arhar seed coat waste	+++
7.	Pea seed coat waste	+++
8.	Moist maize	++
9.	Moist Rice	++
10.	Moist wheat	++
11.	Moist soybean	++
12.	Moist jowar	+++
13.	Maize Agar	+
14.	Potato chips	+
15.	Pieces pumpkin	+

+++ Excellent, ++ Good, + Moderate, - Nil

For determining the best medium for the growth of *Sclerotium rolfsii* and *A. alternata*, these were grown on 13 media. *F. pallidoroseum* was grown on sterilized moist rice, moist jowar, moist soybean and moist wheat. Excellent growth of *S. rolfsii* was obtained on potato dextrose agar, potato dextrose broth, potato sucrose agar, moist maize, moist wheat and moist soybean. (Tables 2 & 3).

Table 2 : Comparative growth of *Sclerotium rolfsii* on different media

Sl. No	Name of test media	Comparative (Growth)
1.	Potato dextrose Agar	+++
2.	Potato dextrose broth	+++
3.	Potato sucrose Agar	+++
4.	Neem oil cake	++
5.	Mustard oil cake	+
6.	Arhar seed coat waste	+++
7.	Pea seed coat waste	-
8.	Moist maize	+++
9.	Moist rice	+++
10.	Moist wheat	+++
11.	Moist soybean	+++
12.	Moist Agar	++
13.	Saw dust	-

+++ Excellent, ++ Good
+ Moderate, - Nil

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Neem oil cake, Mustard oil cake, Arhar seed coat waste, Pea seed coat waste and saw dust were tested after sterilization for the growth of *A. alternata*. Excellent growth of *A. alternata* was obtained on Potato Dextrose Agar and Potato Dextrose Broth (Table 3).

Table 3 : Comparative growth of *Alternaria alternata* on different media.

Sl. No.	Name of test media	Comparative (growth)
1.	Potato dextrose Agar	+++
2.	Potato dextrose broth	+++
3.	Neem oil cake	+
4.	Mustard oil cake	++
5.	Arhar seed coat waste	+
6.	Pea seed coat waste	+

Excellent growth of *S. sclerotiorum* was obtained on Potato Dextrose Agar, Potato Dextrose Broth, Arhar seed coat waste, Pea seed coat waste, moist maize, moist rice, moist wheat and moist soybean (Table 4).

Table 4 : Comparative growth of *Sclerotinia sclerotiorum* on different media

S.I. No.	Name of test media	Comparative growth
1.	Potato dextrose Agar	+++
2.	Potato dextrose broth	+++
3.	Neem oil cake	+
4.	Mustard oil cake	++
5.	Arhar seed coat waste	+++
6.	Pea seed coat waste	+
7.	Moist maize	+++
8.	Moist Rice	+++
9.	Moist wheat	+++
10.	Moist soybean	+++
11.	Saw dust	-

T. viride grows well on potato dextrose broth, neem oil cake, arhar seed coat waste, moist paddy, moist maize and moist rice (Table 5), whereas *Gliocladium virens* grows fastest on potato dextrose agar and potato dextrose broth (Table 6).

Table 5: Comparative growth of *Trichoderma viride* on different media

Sl. No.	Name of test media	Comparative (growth)
1.	Potato dextrose Agar	+++
2.	Potato dextrose broth	+++
3.	Potato sucrose Agar	+
4.	Neem oil cake	+++
5.	Mustard oil cake	+
6.	Arhar seed coat waste	+++
7.	Moist Paddy	+++
8.	Moist Wheat straw + Glucose	++
9.	Moist Maize	+++
10.	Moist Rice	+++
11.	Moist wheat	++
12.	Moist soybean	++
13.	Maize Agar	+

+++ Excellent, ++ Good
+ Moderate, - Nil

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Table 6 : Comparative growth of *Gliocladium virens* on different media.

SI No	Name of test media	Comparative growth
1.	Potato dextrose Agar	Excellent
2.	Potato dextrose broth	Excellent
3.	Neem oil cake	Good

An experiment was conducted in the laboratory to determine growth of fungi on different substrates. Neem oil cake, Mustard oil cake, Arhar seed coat waste, Pea seed coat waste and saw dust were tested after sterilization for the growth of *A. alternata*, *F. pallidoroseum*, *Sclerotium rolfsii*, *S. sclerotiorum* and *T. viride*. *T. viride* grew rapidly on neem oil cake. The fungi *A. alternata*, *F. pallidoroseum*, *S. rolfsii* and *S. sclerotiorum* grow better in all the test substrates except saw dust (Table 7).

It is evident from Table 7 that Arhar seed coat waste induced least growth of *S. rolfsii*, *S. sclerotiorum* and *T. viride*. Pea seed coat waste was better for *F. pallidoroseum* and Mustard oil cake for *A. alternata*. Neem oil cake and pea seed coat waste were equally as good for the growth of *T. viride*.

Table 7 : Comparative efficacy of five media for the growth of fungi isolated from weeds

Name of test fungus	Neem oil cake	Mustard oil cake	Arhar seed coat waste	Pea seed coat waste	Saw dust
<i>Trichoderma viride</i>	+++	+	+++	+++	-
<i>Sclerotinia sclerotiorum</i>	+	++	+++	+	-
<i>Sclerotium rolfsii</i>	++	+	+++	-	-
<i>Fusarium pallidoroseum</i>	+	++	++	+++	-
<i>Alternaria alternata</i>	+	++	+	+	-

+++ Excellent, ++ Good, + Moderate, - Nil

Growth of *Gliocladium virens* was studied on four broth media. Culture weight was measured after 10, 15 and 20 days of inoculation of inoculated flasks. The results presented in Table 8 reveal that maximum culture weight (1.145 gm/ml) was recorded on Richard's media 15 days after inoculation. It was superior to Richard's 0.941 gm/ml and Czapek's media 0.606 gm/ml after 20 days. Among the four media, minimum weight of 0.028 gm/ml was observed on Asthana and Hawker's medium.

Table 8 : Effect of different culture media on culture growth of *Gliocladium virens* at 25±1°C

S. No.	Test media	Culture weight (gm/ml)		
		10th Day	15th Day	20th Day
1.	Asthana & Hawker's	0.028	0.182	0.195
2.	Czapek's media	0.602	0.603	0.606
3.	Potato Dextrose Broth	0.247	0.262	0.246
4.	Richard's	0.582	1.145	0.941

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1. *Information on the mycobiota associated with the weed in India collected* Tamil Nadu Agricultural University, Coimbatore, India

Survey for Parthenium weed

Investigations were carried out on the biological control of Parthenium weed, using potential fungal pathogens prevalent in the cropping and non-cropping area of Tamil Nadu. Six major diseases were noticed and 21 fungal pathogens (18 foliar and 3 root pathogens) were isolated from diseased plant parts. Of those, *L. theobromae*, *F. pallidoroseum* and *Oidium parthenii* were shown to be highly pathogenic.

Among the 28 districts surveyed, Parthenium weed infestation was found to be moderate (26-50%) in Coimbatore and Vellore districts, low (10-25%) in Dharmapuri, Karur, Perambalur, Trichy and Virudhunagar districts and very low in the remaining districts of Tamil Nadu.

Survey for *Parthenium hysterophorus* diseases

Leaf blight, leaf spot, tip drying, powdery mildew, phyllody and wilt incidence were found to be associated with *P. hysterophorus* plants. In total, 21 fungal pathogens were isolated from the various diseased plant parts of Parthenium weed. Among the 21 pathogens, *O. parthenii* had a maximum distribution in Tamil Nadu followed by *M. phaseolina*, *D. australiensis*, *R. solani* and *A. alternata* (Table 9).

Table 9 : Occurrence of fungal pathogens associated with various types of *Parthenium hysterophorus* diseases in Tamil Nadu

S. No.	Fungal flora	IMI Number	Type of symptom	Distribution in different districts of Tamil Nadu	Per cent distribution
1.	<i>Alternaria alternata</i> Link.	-	Leaf blight	Coimbatore, Cuddalore, Dharmapuri, Erode, Kanchipuram, Kanyakumari, Namakkal, Nagapattinam, Perambalur, Salem, Sivagangai, Tiruvallure, Tiruvannamalai, Tuticorin, Theni, Vellore and Villupuram	60.71
			Leaf spot	Cuddalore and Dharmapuri	7.14
2.	<i>Alternaria zinniae</i> M.B. Ellis	378930	Leaf blight	Cuddalore, Dindigul, Kanchipuram, Nagapattinam, Namakkal, Tiruvallure, Tuticorin and Virudhunagar	28.57
			Leaf spot	Erode	3.57
3.	<i>Curvularia lunata</i> R.R. Nelson and F.A. Hassis	378925	Leaf spot	Kanyakumari, Karur, Madurai, Pudukottai, The Nilgiris, Tiruvannamalai, Tiruvarur, Tirunelveli and Tuticorin	32.14
4.	<i>Curvularia pallescens</i> (Tsuda and Viyama) Sivan.	379991	Leaf spot	Coimbatore, Dharmapuri, Namakkal, Pudukottai, Tirunelveli, Trichy and Vellore	25.00
5.	<i>Curvularia verruculosa</i> Tandon and Bilgrami ex. M.B. Ellis	379993	Leaf spot	Coimbatore, Karur, Kanyakumari, Nagapattinam, Ramanathapuram, Tiruvannamalai and Theni	25.00
6.	<i>Colletotrichum dematium</i> (Pers. Fr.) Grove.	378928	Leaf spot	Coimbatore, Dindigul, Dharmapuri, Erode, Karur, Madurai, Perambalur, Salem, Trichy, Theni and Vellore	39.29

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S. No.	Fungal flora	IMI Number	Type of symptom	Distribution in different districts of Tamil Nadu	Per cent distribution
7.	<i>Drechslera australiensis</i> (Tsuda and Viyama) Alcorn	378924	Leaf blight	Coimbatore, Cuddalore, Dindigul, Erode, Kanchipuram, Karur, Madurai, Namakkal, Perambalur, Sivagangai, Salem, Tanjavur, The Nilgiris, Tiruvallore, Tiruvannamalai, Tiruvarur, Trichy, Tuticorin, Theni and Villupuram	71.43
			Leaf spot	Coimbatore, Erode, Madurai and Salem	14.29
			Tip drying	Coimbatore, Cuddalore, Nagapattinam, Namakkal, Perambalur, Salem, Theni, Vellore and Villupuram	32.14
8.	<i>Drechslera hawaiiensis</i> Alcorn.	379990	Leaf blight	Coimbatore, Cuddalore, Dindigul, Karur, Madurai, Salem, Sivagangai, Tanjavur, Tiruvallore, Tiruvarur, Trichy, Theni and Villupuram	46.43
			Leaf spot	Coimbatore, Dindigul, Erode, Madurai, Salem, Tirunelveli and Vellore	25.00
			Tip drying	Coimbatore, Cuddalore, Dindigul, Dharmapuri, Erode, Karur, Madurai, Nagapattinam, Tiruvarur and Vellore	35.71
9.	<i>Fusarium equiseti</i> (Corda) Sacc.	379998	Leaf blight	Coimbatore, Karur, Pudukottai, Trichy, Tuticorin, Theni and Vellore	32.14
			Tip drying	Pudukottai and Tuticorin	7.14
10.	<i>Fusarium moniliforme</i> Sheld.	-	Leaf blight	Coimbatore, Dharmapuri, Kanchipuram, Karur, Perambalur, Pudukottai, Ramanathapuram, Tanjavur, Tiruvallore and Theni	35.71
			Tip drying	Coimbatore, Dharmapuri, Karur, Perambalur, Ramanathapuram and Theni	21.42
11.	<i>Fusarium oxysporum</i> Sch. Ex. Fries	-	Leaf blight	Cuddalore, Dharmapuri, Kanyakumari, Perambalur, Theni and Vellore	21.43
			Tip drying	Cuddalore, Dharmapuri, Kanyakumari and Vellore	14.29
12.	<i>Fusarium pallidoroseum</i> (Cooke) Sacc.	378923	Leaf blight	Coimbatore, Cuddalore, Erode, Karur, Madurai, Nagapattinam, Pudukottai, Ramanathapuram, Salem, Sivagangai, Tanjavur, Theni, Vellore, Villupuram and Virudhunagar	53.57
			Tip drying	Coimbatore, Cuddalore, Madurai, Theni and Vellore	17.86
13.	<i>Fusarium solani</i> (Martius) Sacc.	379992	Leaf blight	Kanchipuram, Madurai, Perambalur, Pudukottai, Ramanathapuram, Tiruvarur and Theni	25.00
			Leaf spot	Cuddalore and Nagapattinam	7.14
			Tip drying	Kanchipuram, Tiruvarur and Theni	10.71
14.	<i>Macrophomina phaseolina</i> (Tassi) Goid.	-	Root rot	Coimbatore, Cuddalore, Dindigul, Dharmapuri, Kanchipuram, Karur, Madurai, Nagapattinam, Namakkal, Perambalur, Pudukottai, Ramanathapuram, Salem, Tirunelveli, Tiruvallore, Tiruvarur, Tiruvannamalai, Trichy, Tuticorin, Theni, Vellore and Villupuram	82.14
15.	<i>Oidium parthenii</i> S & U	-	Powdery mildew	All districts	100.00
16.	<i>Phoma sorghina</i> (Sacc.) Boerema	378931	Leaf blight	Coimbatore, Dharmapuri, Salem, Theni and Vellore	17.86
17.	<i>Phomopsis</i> sp.	378987	Leaf blight	Coimbatore, Dharmapuri, Madurai and Theni	14.29
18.	<i>Rhizoctonia solani</i> Khun.	-	Root rot	Coimbatore, Dindigul, Erode, Kanchipuram, Karur, Madurai, Namakkal, Perambalur, Ramanathapuram, Salem, Tiruvannamalai, Tirunelveli, Tiruvallore, Trichy, Tuticorin, Theni, Vellore and Villupuram	64.29
19.	<i>Sclerotium rolfsii</i> (Sacc.)	-	Root rot/ collar rot	Coimbatore, Karur, Madurai, Sivagangai, Tanjavur, Tirunelveli and Villupuram	25.00
20.	<i>Syncephalastrum racemosum</i> Cohn. Ex. J. Schrot.	378926	Leaf blight	Coimbatore and Nagapattinam	7.14
21.	Newly Isolated Fungus (NIF)	-	Leaf blight	Coimbatore, Dindigul, Erode, Salem, Theni and Vellore	21.43
			Tip drying	Coimbatore, Theni and Vellore	10.73

Outputs

1. *Information on the mycobiota associated with the weed in India collected* Project Directorate of Biological Control, Bangalore, India

Out of the total 27 districts, 16 were covered very intensively (Fig. 1) over different periods. The districts where Parthenium weed was not a serious problem were not visited during 1997-99.

However, the list of fungal pathogens provided in this report does not represent all the pathogenic mycobiota associated with Parthenium weed in Karnataka, because a number of weak and opportunistic pathogens are not included. However, as is evident from the identifications, a number of the pathogens hitherto not known to be associated with the weed in India or anywhere in the world have come to light.

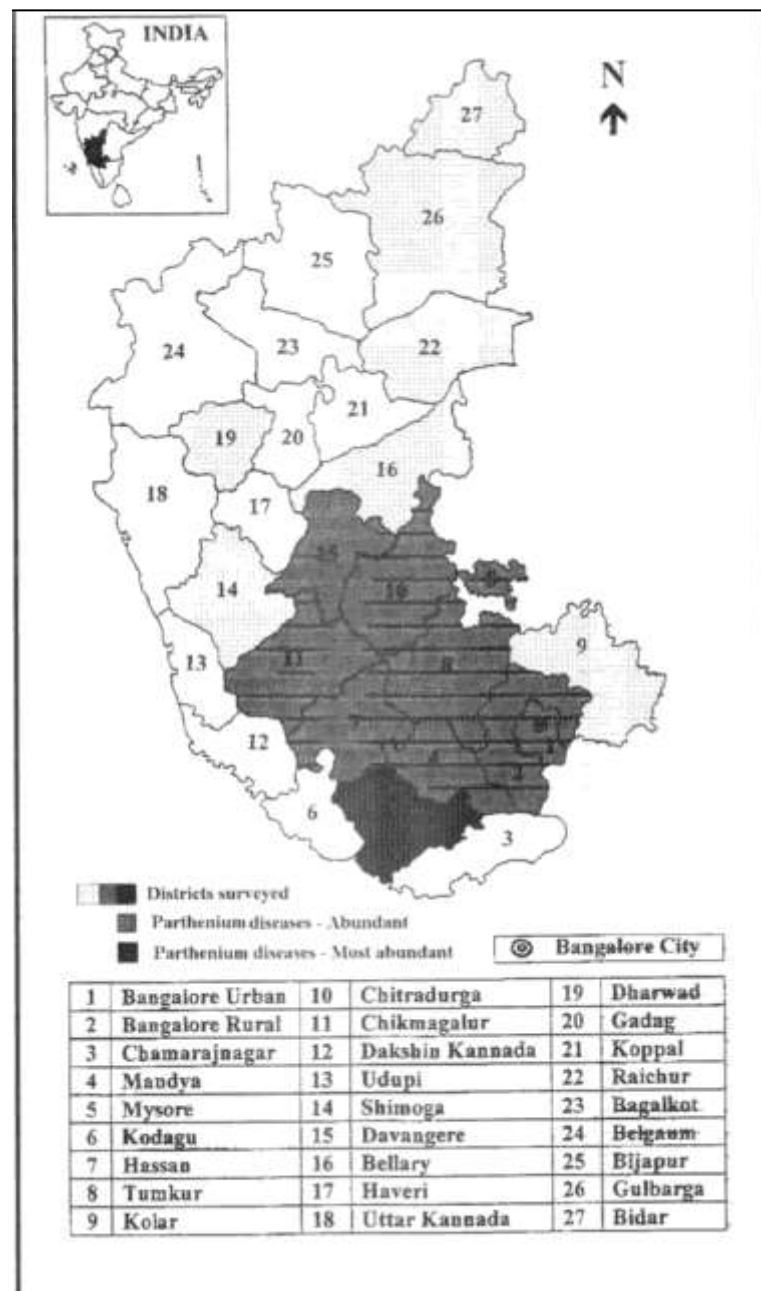


Fig. 1: Map of Karnataka State indicating the districts surveyed and those found to be abundant in parthenium diseases during 1997-99

Outputs

A total of 14 isolates of some of the most damaging fungal pathogens of Parthenium weed were identified and confirmed at CABI Bioscience, UK Centres (Ascot and Egham) (Table 10). Also, many of the commonly occurring fungal pathogens of Parthenium weed in Karnataka collected during 1997-99 were identified at least up to the genus level and documented (Table 11). Identifications of the cultures revealed that some of the pathogens like *Cryptosporiopsis* sp., *Phoma sorghina*, *Lasiodiplodia theobromae*, *Fusarium equiseti*, *Khuskia oryzae* and the suspected *Alternaria tenuissima* are new records for Parthenium weed.

Table 10 : Details of the most damaging isolates of some important fungal pathogens of *Parthenium hysterophorus* identified and confirmed at CABI Bioscience, UK Centres (Ascot and Egham).

PDBC No.	IMI No.	Pathogen	Symptoms	Date of collection	Place of collection
WF(Ph)1	378921	<i>Fusarium pallidroseum</i> (Cooke) Sacc. (= <i>F. semitectum</i> Berk. & Rav.)	Leaf spot/blight	1 January 1997	Hebbal, Bangalore, Bangalore North Taluk, Bangalore Urban District.
WF(Ph)3	378270	<i>Cryptosporiopsis</i> sp.	Leaf spot/blight	14 August 1997	Azadnagar, Hunsur, Hunsur Taluk, Mysore District.
WF(Ph)4	-	<i>Fusarium pallidroseum</i>	Leaf spot/blight	14 August 1997	Azadnagar, Hunsur, Hunsur Taluk, Mysore District.
WF(Ph)5	378480	<i>Rhizoctonia</i> sp.	Stem and bases of branches	14 August 1997	Chikkahunsur, Hunsur, Hunsur Taluk, Mysore District.
WF(Ph)6	378271	<i>Phoma sorghina</i> (Sacc.) Boerema, Dorenb. & Kesteren	Leaf spot/blight	10 December 1997	Gangenahalli, Bangalore, Bangalore North Taluk, Bangalore Urban District.
WF(Ph)7	378918	<i>Alternaria zinniae</i> M. B. Ellis	Leaf spot/blight	8 January 1998	Dandupalya, Hoskote Taluk, Bangalore Rural District.
WF(Ph)8	378919 a	<i>Lasiodiplodia theobromae</i> (Pat.) Griffon & Maubl. (= <i>Botryodiplodia theobromae</i> Pat.)	Leaf spot/blight	31 December 1997	Siddeswaranadurga, Challakere Taluk, Chitradurga District.
WF(Ph)9	378920	<i>Alternaria zinniae</i> M. B. Ellis	Leaf spot/blight	8 January 1998	Hoskote, Hoskote Taluk, Bangalore Rural District.
WF(Ph)10	379988	<i>Fusarium equiseti</i> (Corda) Sacc.	Leaf spot/blight	29 April 1998	Bharamasagar, Davangere Taluk, Davangere District.
WF(Ph)11	379979	<i>Khuskia oryzae</i> H. J. Huds <i>Nigrospora oryzae</i> (Berk. & Broome) Petch	Leaf spot/blight	29 April 1998	Harihara, Harihara Taluk, Davangere District.
WF(Ph)12	379980	<i>Alternaria</i> sp. (? <i>A. tenuissima</i> (Kunze ex Pers.) Wiltshire	Leaf spot/blight	30 April 1998	Shimoga, Shimoga Taluk, Shimoga District.
WF(Ph)13	379981	<i>Alternaria</i> sp.	Leaf spot/blight	1 May 1998	Mallechennahalli, Tarikere Taluk, Chikmagalur District.
WF(Ph)14	379982	<i>Fusarium equiseti</i> Sacc.	Leaf spot/blight	1 May 1998	Kadur, Kadur Taluk, Chikmagalur District.
WF(Ph)31	378922	<i>Colletotrichum gloeosporioides</i>	Leaf spot/blight	14 August 1997	Azadnagar, Hunsur, Hunsur Taluk, Mysore District.

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Table 11: Other pathogenic fungal flora commonly recorded on *Parthenium hysterophorus* in Karnataka State during 1997-99

Pathogen	Plant parts affected	Symptoms	Districts
<i>Alternaria</i> spp.	Leaf	Spot/blight	Mysore, Mandya, Bangalore Urban and Bangalore Rural
<i>Cercospora</i> spp.	Leaf	Spot	Bangalore Rural, Tumkur and Raichur
<i>Colletotrichum capsici</i> (Syd.) Butler & Bisby	Leaf	Spot	Mysore, Mandya and Gulbarga
<i>Curvularia lunata</i> (Walker) Boedjin	Leaf	Spot	Mysore and Bangalore Rural
<i>Dechslera</i> sp.	Leaf	Spot	Bangalore Rural
<i>Fusarium</i> spp.	Root	Wilt/rot	Mysore, Hassan and Bangalore Rural
<i>Macrophomina</i> sp.	Root	Rot	Mysore and Bangalore Rural
<i>Nigrospora sphaerica</i> (Sacc.) Mason	Leaf	Spot/blight	Bangalore Rural
<i>Oidium parthenii</i> Satyaprasad & Usharani	Leaf	Powdery mildew	All
<i>Pestalotia</i> sp.	Leaf	Spot	Bangalore Rural and Bidar
<i>Phoma chrysanthemicola</i> Holls	Leaf	Spot/blight	Hassan
<i>Phoma eupyrina</i> Sacc.	Leaf	Spot/blight	Chikmagalur
<i>Rhizoctonia solani</i> Kuhn	Leaf	Blight	Hassan and Bangalore Rural
<i>Rhizoctonia</i> sp.	Leaf	Blight	Chikmagalur and Bangalore Rural
<i>Sclerotinia sclerotiorum</i> (Lib.) de Bary	Stem and bases of branches	Wilt and rot	Bangalore Rural and Kolar
<i>Sclerotium rolfsii</i> Sacc.	Collar region	Rot	All

A *Cryptosporiopsis* sp. was found to be inciting severe leaf spots, which in turn resulted in severe blighting and necrosis of the leaves, at many places in and around Hunsur town in Mysore district. The isolate WF(Ph)3 (IMI 378270) collected in August 1997 at Azadnagar near Hunsur has been found to be highly virulent to *Parthenium* weed. This pathogen was noticed to be severely affecting *P. hysterophorus* plants in a coconut grove. Although no species has been so far described on *P. hysterophorus*, there is one previous record at IMI for this host from Tamil Nadu State in India. However, the published literature does not mention *Cryptosporiopsis* sp. as a pathogen on *Parthenium* weed.

Although several species of *Alternaria* including *A. dianthi*, *A. alternata* and *A. macrospora* have been reported to be pathogenic to *Parthenium* weed in India, the occurrence of *Alternaria zinniae* on *Parthenium* weed is a new record for India. The pathogen was consistently isolated from leaf spots collected in many places in Hoskote taluk of Bangalore Urban district in January 1998. This pathogen is known to be cause leaf spots on *Parthenium* weed in Mexico (Evans, 1997a).

Another pathogen that was ubiquitous in many districts of Karnataka was *Fusarium pallidoroseum* (= *F. semitectum*). This was first reported to affect *Parthenium* weed in Andhra Pradesh (Rao & Rao, 1987). It was consistently isolated from leaf spots collected in different districts, particularly Bangalore Urban and Mysore, during the surveys. The frequency of occurrence and the virulence of many of the isolates prompted studies on its potential as a

Outputs

mycoherbicide for *P. hysterophorus*. Preliminary host-specificity tests at PDBC have given promising results.

In addition to *F. pallidoroseum*, a number of *Fusarium* spp. were encountered during the surveys in almost all the districts visited. *F. equiseti* could be singled out for its ubiquitous presence on Parthenium weed. Leaf spots/blights collected in a number of places showed its widespread occurrence across Karnataka.

The collar rot pathogen, *Sclerotium rolfsii*, was seen attacking Parthenium weed in almost all the regions of the State. In fact this was one of the first pathogens to be recorded on the weed in India. It was first observed to be causing wilting and death of *P. hysterophorus* plants in and around groundnut plots in Dharwad district (Siddaramaiah *et al.*, 1984).

Oidium parthenii, the incitant of powdery mildew, was rampant in and around Bangalore and in many other districts of Karnataka. Since *Erysiphe cichoracearum* has been reported to cause powdery mildew on rosette leaves resulting in greyish-white, irregular necrosis on *P. hysterophorus* plants in Mexico (Evans, 1987a), the exact identity of the powdery mildew in India needs to be resolved.

Species of *Colletotrichum*, *Drechslera*, *Curvularia*, *Phoma*, *Nigrospora*, *Rhizoctonia*, *Pestalotia* were found associated with leaf spots and blights of Parthenium weed in many sites. Isolations from root and stem damages yielded *Macrophomina* sp., *Sclerotinia sclerotiorum* and *Rhizoctonia* sp. on several occasions.

Regular collection of disease samples resulted in the occasional identification of a number of non-fungal diseases of Parthenium weed as well (Table 12). In spite of the fact that these diseases were not included in further investigations, they were documented to get an overall picture of the guild of pathogens associated with the weed. Certain leaf spots collected in Bangalore Rural and Mysore districts yielded a *Xanthomonas* sp. Even though *Xanthomonas campestris* pv. *parthenii* nov. is reported to be causing a blight disease on Parthenium weed (Ramesh Chand *et al.*, 1995), the present isolate could be identified only up to the genus level. Similarly a wilt/rot pathogen, *Pseudomonas* sp. was also noticed to be damaging Parthenium weed in several areas surveyed. *Parthenium hysterophorus* has been reported to be a collateral host for *Pseudomonas solanacearum* (Ram Kishun & Ramesh Chand, 1987). Phyllody, caused by phytoplasmas (MLO's), and mosaic and leaf curl were as rampant as any other disease on Parthenium weed in all the districts surveyed.

Table 12 : Some non-fungal diseases commonly recorded on *Parthenium hysterophorus* in Karnataka State during 1997-99

Pathogen	Plant parts affected	Symptoms	Districts
Phytoplasma (=MLO's)	Leaf and inflorescence	Phyllody/ Witches' broom	All
Virus (es)	Leaf	Mosaic and curl	All
<i>Xanthomonas</i> sp.	Leaf	Spot/blight	Mysore and Bangalore Rural
Bacterium (? <i>Pseudomonas</i> sp.)	Collar region	Wilt/rot	Bangalore Rural and Kolar

Outputs

1. *Information on the mycobiota associated with the weed in India collected Kurukshetra University, India*

Collection of *Parthenium hysterophorus* seed

During the surveys, seeds of 10 “isolates” of *Parthenium* weed collected from different places such as Chandigarh (Isolate 1), Patiala (Isolate 2), Uttar Pradesh (Isolate 3) and Kurukshetra (Isolates 4-7), were sent to the UK for screening against two rusts *Puccinia abrupta* var. *partheniicola* and *P. melampodii* in order to test their virulence.

Collection of seeds of cereals/vegetables/fodder and oil yielding crops

Seeds of various cereals/vegetables/pulses/oil yielding crops (Table 13), collected from and typical of this region, were sent to CABI Bioscience (UK) for testing host specificity of the two rusts, *Puccinia abrupta* var. *partheniicola* and *P. melampodii*.

Table 13: List of seeds of cereals, vegetables, pulses and oil yielding crops collected from Haryana

S.No.	Common Name	Species Name	Variety
A. CEREALS			
1.	Wheat	<i>Triticum aestivum</i>	HD2428, HD2329,
2.	Hybrid maize	<i>Zea mays</i>	203492
3.	Paddy rice	<i>Oryza sativa</i>	P 44
4.	Paddy rice	<i>O. sativa</i>	P. Basmati
B. VEGETABLES			
5.	Radish	<i>Raphanus sativus</i>	P. Rashmi
6.	Cauliflower	<i>Brassica oleracea</i> var. <i>botryti</i>	P. Deepali
7.	Red chilli	<i>Capsicum annuum</i>	P. Jawala
8.	Pea	<i>Pisum sativum</i>	
9.	Bottle gourd	<i>Lagenaria vulgaris</i>	
C. PULSES			
10.	Gram	<i>Cicer arietinum</i>	P-256
11.	Gram	<i>C. arietinum</i>	P-267
12.	Moong	<i>Vigna radiata</i>	P. Bashaki
13.	Cowpea	<i>Vigna unguiculata</i>	
14.	Pigeon pea	<i>Cajanus cajan</i>	P. 855
D. OIL YIELDING			
15.	Sunflower	<i>Helianthus annuus</i>	PSFH-67, DK 3890, Sungene 80, Jawalamukhi PR 459
16.	Mustard	<i>Brassica campestris</i>	
17.	Kala till	<i>Guizotia abyssinica</i>	

Mycobiota associated with *Parthenium* weed

Of the over 30 diseased specimens collected during the surveys, 27 were leaf spots, 1 anthracnose and 2 powdery mildews (Table 14).

Diseased specimens were examined critically and symptoms were recorded. Disease specimens were processed and air-mailed CABI Bioscience (UK) for maintenance, processing and for confirmation of identification (at Egham Centre).

Outputs

Isolation of fungal pathogens from the diseased specimens followed two methods: Incubation in moist chambers (sterilized and unsterilized tissue) and direct isolation from infected tissue on various media (PDA, PDAY, PeDA and PeDAY) supplemented with and without streptopenicillin. The following fungal genera were recorded: *Alternaria* spp., *Cercospora* sp., *Colletotrichum* sp., *Curvularia* sp., *Fusarium* sp. and *Myrothecium* sp., which were later identified, in the UK, as *Alternaria zinniae*, *A. alternata*, *Curvularia lunata*, *Cercospora partheniiphila* (Table 14). On the basis of sporulating structures produced on the live diseased *P. hysterophorus* leaves, the following fungal pathogens were identified: *Pseudocercospora* sp. and *Erysiphe cichoracearum* D.C. (*Oidium* state). The symptoms and cultural characteristics recorded are as follows:

Table14: Mycobiota associated with Parthenium weed in northern India

S.No.	Date of Collection	Place of Collection	Plant Disease/s	Fungus	IMI No.
1.	2/2/97	KUK	Leaf Spot	<i>Cladosporium cladosporioides</i>	
2.	23/3/97	Panipat	Leaf Spot	<i>Alternaria alternata</i>	
3.	26/3/97	KKR	Leaf Spot	<i>Alternari alternata</i>	
4.	5/4/97	KUK	Leaf Spot	<i>Alternaria</i> sp.	
5.	5/4/97	Ambala	Leaf Spot	<i>Alternaria</i> sp.	
6.	12/5/97	KKR	Leaf Spot	<i>Alternaria</i> sp.	
7.	15/5/97	KUK	Powdery mildew	<i>Erysiphe cichoraceraum</i> (<i>Oidium</i> state)	377841
8.	15/6/97	UP	Leaf Spot	<i>Alternaria alternata</i>	
9.	15/6/97	UP	Powdery mildew	<i>Erysiphe cichoraceraum</i> (<i>Oidium</i> state)	377842
			Leaf spot	<i>Cuvularia lunata</i>	
10.	10/6/97	KUK	Leaf Spot	<i>Pseudocercospora</i> sp.	375237
11.	12/6/97	KUK	Leaf Spot	<i>Cercospora partheniiphila</i>	375238
12.	18/6/97	KUK	Leaf Spot	<i>C. partheniiphila</i>	377833
13.	22/6/98	UP	Leaf Spot	<i>Alternaria alternata.</i>	
14.	12/7/97	KUK	Leaf Spot	<i>Myrothecium</i> sp. *	
15.	28/7/97	KUK	Leaf Spot	<i>C. partheniiphila</i>	377834
16.	9/8/97	Yamuna Nagar	Leaf Spot	<i>Alternaria alternata,</i> <i>Fusarium</i> sp. *	
17.	12/8/97	KUK	Leaf Spot	<i>C. partheniiphila</i>	377836
18.	18/8/97	KUK	Anthraxnose	<i>Colletotrichum</i> sp. *	
19.	22/8/97	KUK	Leaf Spot	<i>C. partheniiphila</i>	377835
20.	22/8/97	Jyotisar	Leaf Spot	<i>C. partheniiphila</i>	377837
21.	25/8/97	Jyotisar	Leaf Spot	<i>C. partheniiphila</i>	377839
22.	4/9/97	KUK	Leaf Spot	<i>C. partheniiphila</i>	377839
23.	10/9/97	Karnal	Leaf Spot	<i>C. partheniiphila</i>	377840
24.	9/10/97	Patiala	Leaf Spot	<i>Alternaria</i> sp.	
25.	8/11/97	Karnal	Leaf Spot	<i>Alternaria alternata</i>	
26.	22/11/97	KKR	Leaf Spot	<i>Alternaria alternata</i>	
27.	10/1/98	Ambala	Leaf Spot	<i>Alternaria zinniae</i>	378914
28.	7/2/98	UP, KKR	Leaf Spot	<i>Alternaria zinniae</i>	378915
29.	20/2/98	KKR	Leaf Spot	<i>Alternaria zinniae</i>	378916
30.	16/3/98	KKR	Leaf Spot	<i>Fusarium</i> sp. *	

*Tentatively identified, diseased specimen and cultures are maintained in our laboratory for further investigation

1. *Alternaria zinniae* Pape

On living leaves of Parthenium weed symptoms are characterized as dark brown, irregular marginal spots. Colonies dark grey to black. Conidiophores solitary, rarely in groups (1-3 in no.), brown, straight to geniculate with 1- 4 scars, upto 190 um long, 5.7-9.5 um thick.

Outputs

Conidia mostly solitary, rarely in chains of 2, obclate, rostrate, pale to golden brown, smooth to minutely verrucose, with 5-9 transverse and several longitudinal septa, body 72-106x17.1-26.6 μm , beak hyaline, filiform, septate, straight to geniculate some times swollen at the apex, often much longer than the body of the spore, 55-165 μm long and 1.9-3.8 μm thick.

Three herbarium specimens and a live fungal culture has been deposited at CABI Bioscience, (Egham) as IMI 378914, 378915 and 378916.

The leaf spot disease due to *A. zinniae*, recorded on young *P. hysterophorus* plants, is widespread in distribution and it has been recorded from Kurukshetra, Ambala (Haryana) and Ghaziabad (Uttar Pradesh). The pathogen has been recorded mainly during the winter months i.e. January & February (Table 14), thus indicating its adaptation to the higher rainfall of the winter season.

2. *Cercospora partheniiphila*

On the basis of variations in disease symptoms and morphology of conidiophores and conidia in *C. partheniiphila*, authors have categorized it into five isolates (Table 15).

Table 15 : Comparison of five isolates of *Cercospora partheniiphila* recorded from different parts of northern India

Isolate/no	Symptoms	Conidiophores	Conidia
Isolate 1 IMI 375238	Round to oval, light brown central spots with grey coloured centre	Brown, in clusters, upto 236 μm long, 4 μm wide, with a scar at the tip.	Hyaline, filiform variable in shape and size, 60-200x3.8-4.0 μm
Isolate 2 IMI 377833	Irregular, round/oval light to dark brown, sometimes ash centred, present in centre occasionally on margins	Light brown, in groups (5-12) straight to flexuous, septate upto 691 μm long , 4.5-6.8 μm thick, 1-5 scars	Hyaline, tapering ends 125-488 x 1.5-5.7 μm
Isolate 3 IMI 377836	Dark brown (burned appearance), crescent marginal spots	Mid pale brown, straight and bent septate, in groups (5-12), 2-3 scars upto 209 μm long and 6.84 μm thick	Hyaline, septate, curved, tapering ends 121-247 x 1.9-3.04 μm
Isolate 4 IMI 377839	Brown, round and irregular marginal & central spots	Light brown, straight septate, in groups (2-5), each with 1-3 scars, upto 592 μm long, 5.7 μm thick	Hyaline, septate, straight, with blunt ends shape and size variable 57-224 x 3.8-5.7 μm .
Isolate 5 IMI 377840	Dark brown, irregular and round spots distributed on all over the leaf	Light yellow, straight, septate In groups (3-7) each with 2-5 scars, upto 627 μm long and 5.7 μm thick.	Hyaline, straight septate, shape and size variable 68-209 x 1.9-3.8 μm .

3. *Pseudocercospora* sp.

On living leaves of Parthenium weed, symptoms are characterized as circular to irregular, light yellow spots at the centre as well as on margins. Conidiophores fasciculate, each arising from a stroma (1-7 conidiophores/stroma), conidiogenous cells truncate, 38 μm long, 4-5 μm thick. Conidia hyaline, septate, short, with blunt ends, 11-16 x 3.0-3.9 μm .

Diseased specimen has been deposited as IMI 375237.

4. *Erysiphe cichoracearum* D.C. (*Oidium* state)

Symptoms are characterized by the presence of white powdery growth on the adaxial surface of leaf. Mycelium grows externally on host. Conidiophores upright, simple 95.0-136.8 x 11.4-15.2 μm . Conidia cylindrical, hyaline, 1 celled, 22.8- 30.4 x 15.2-19.0 μm .

Diseased specimens have been deposited as IMI 377841-377842.

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5. *Alternaria alternata* (Fr.) Keissler

Symptoms are characterized by the presence of dark brown marginal spot on living leaves of Parthenium weed. Colonies greyish black on PDAY at $25 \pm 2^{\circ}$ C. Conidiophores simple, straight, septate, branched, dark brown, up to 80 μm long, 2-3 μm thick. Conidia catenate (2-4), straight, obclavate, smooth walled, golden to dark brown, with short beak 24.7-41.8 x 9.5-15.2 μm , 5-8 transverse and 1-3 oblique septa.

6. *Colletotrichum* sp.

Symptoms are characterized as dark brown, irregular leaf spots, with yellow margins, acervuli irregularly scattered, conidiophores simple, light coloured and septate, formed from the upper cells of the fructification. Conidia hyaline, unicellular, oblong, 11.4-19.00 x 1.4 μm .

7. *Myrothecium* sp.

Symptoms are characterized as large, pale brown, circular spots on leaves of Parthenium weed, which eventually drop out giving a shot-hole effect. Sporodochia sessile up to 1.8mm diameter, phialides 10-12 x 1-2 μm . Conidia unicellular, cylindrical with slightly round ends, hyaline, 8-9 x 1.9-2.8 μm .

8. *Curvularia lunata* (Walker) Boedijn

Symptoms are characterized as dark coloured spots initiating from the margins, gradually spread towards the centre and finally becoming irregular in outline. Conidiophores mononematous, brown, smooth upto 228 μm long. Conidia curved, 3 septate, pale to dark brown, 19-33x8-16 μm .

9. *Fusarium* sp.

Symptoms are characterised as, dark, irregular, marginal and central spots. Colonies on PDA+Y cottony, light pink colour. Conidiophores simple, short, branched bearing whorl of phialides, grouped into sporodochia. Conidia hyaline, sickle shaped with pointed ends, 2-3 celled.

Looking into the wide distribution of the three leaf spot diseases caused by *Alternaria zinniae*, *A. alternata* and *Cercospora partheniiphila* on Parthenium weed in northern India, these pathogens offer the possibility for development and exploitation as biocontrol agents for reducing *P. hysterophorus* growth.

Outputs

1. Information on the mycobiota associated with the weed in India collected CABI Bioscience, UK

Isolates which had shown potential as control agents were forwarded to the UK for identification at the Egham Centre (Table 17).

Table 17: Isolates sent from India for identification

Code	Preliminary identification	Location	IMI number	Identification
Kurukshetra University				
1	<i>Cercospora</i> sp.	Chandigarh	375237	<i>Pseudocercospora</i> sp.
2	<i>Cercospora</i> sp.	Patiala	375238	<i>Cercospora partheniiphila</i>
3	<i>Alternaria</i> sp.	Uttar Pradesh	375238	<i>Alternaria zinniae</i>
4	<i>Alternaria</i> sp.	Kurukshetra 1		<i>Alternaria alternata</i>
5	<i>Alternaria</i> sp.	Kurukshetra 2		<i>Alternaria alternata</i>
6	<i>Alternaria</i> sp.	Kurukshetra 3		<i>Alternaria alternata</i>
7	<i>Cladosporium</i> sp.	Kurukshetra 4		<i>Cladosporium cladosporioides</i>
9	<i>Cercospora</i> sp.	Haryana State	377833	<i>Cercospora partheniiphila</i>
10	<i>Cercospora</i> sp.	Haryana State	377834	<i>Cercospora partheniiphila</i>
11	<i>Cercospora</i> sp.	Haryana State	377835	<i>Cercospora partheniiphila</i>
12	<i>Cercospora</i> sp.	Haryana State	377836	<i>Cercospora partheniiphila</i>
13	<i>Cercospora</i> sp.	Haryana State	377837	<i>Cercospora partheniiphila</i>
14	<i>Cercospora</i> sp.	Haryana State	377838	<i>Cercospora partheniiphila</i>
15	<i>Cercospora</i> sp.	Haryana State	377839	<i>Cercospora partheniiphila</i>
16	<i>Cercospora</i> sp.	Haryana State	377840	<i>Cercospora partheniiphila</i>
17	<i>Alternaria</i> sp.	Haryana State		<i>Alternaria alternata</i>
18	<i>Cercospora</i> sp.	Haryana State		<i>Cercospora partheniiphila</i>
19	Powdery Mildew	Haryana State	377841	<i>Erysiphe cichoracearum</i>
20	Powdery Mildew	Ghaziabad, UP	377842	<i>Erysiphe cichoracearum</i>
21	<i>Cercospora</i> sp.	Haryana State		
21		Haryana State		<i>Alternaria zinniae</i>
22		Haryana State		<i>Alternaria zinniae</i>
23		Haryana State		<i>Alternaria zinniae</i>
Project Directorate of Biological Control				
1	<i>Fusarium semitectum</i>	Bangalore Urban Dist.	378917	<i>Fusarium pallidoroseum</i>
3		Mysore District	378270	<i>Cryptosporiopsis</i> sp.
4		Mysore District	378921	<i>Fusarium pallidoroseum</i>
5		Mysore District	378480	<i>Rhizoctonia</i> sp.
6		Bangalore Urban Dist.	378271	<i>Phoma sorghina</i>
7		Bangalore Rural Dist.	378918	<i>Alternaria zinniae</i>
8		Chitradurga District	378919	<i>Lasiodiplodia theobromae</i>
9		Bangalore Rural Dist.	378920	<i>Alternaria zinniae</i>
10		Davangere District	379978	<i>Fusarium equiseti</i>
11		Davangere District	379979	<i>Khuskia oryzae</i>
12		Shimoga District	379980	<i>Alternaria</i> sp.
13		Chickmagalur Dist.	379981	<i>Alternaria</i> sp.
14		Chickmagalur Dist.	379982	<i>Fusarium</i> sp.
31		Mysore District	378922	<i>Glomerella cingulata</i>
National Research Centre for Weed Science				
1		Jabalpur	378932	<i>Fusarium equiseti</i>
2*		Jabalpur	378933	<i>Phoma chrysanthemicola</i>
2°		Jabalpur	378934	<i>Phoma sorghina</i>
2 ^x		Jabalpur	378935	<i>Phoma sorghina</i>

Outputs

Code	Preliminary identification	Location	IMI number	Identification
3I		Jabalpur	378936	<i>Fusarium</i> sp.
6II		Jabalpur	378937	<i>Fusarium pallidoroseum</i>
6III		Jabalpur	378938	<i>Trichoderma hamatum</i>
7		Jabalpur	378939	<i>Fusarium pallidoroseum</i>
8		Jabalpur	378940	<i>Fusarium pallidoroseum</i>
Tamil Nadu University				
C		Coimbatore District	378924	<i>Cochliobolus australiensis</i>
D		Coimbatore District	378930	<i>Alternaria zinniae</i>
G		Coimbatore District	378928	<i>Colletotrichum dematium</i>
F		Coimbatore District	378927	<i>Cochliobolus lunatus</i>
Hd		Coimbatore District	378923	<i>Fusarium pallidoroseum</i>
H		Coimbatore District	378925	<i>Cochliobolus lunatus</i>
J ₁		Coimbatore District	378926	<i>Syncephalastrum racemosum</i>
J ₂		Coimbatore District	378929	Sterile
J ₃		Coimbatore District	378931	<i>Phoma sorghina</i>
P1a		Coimbatore District	379984	<i>Glomerella cingulata</i>
P2a	<i>Alternaria</i> sp.	Coimbatore District	379985	<i>Corynespora</i> sp.
P3a	<i>Phoma</i> sp.	Coimbatore District	379986	<i>Phoma</i> sp.
P3b	<i>Phoma</i> sp.	Coimbatore District	379987	<i>Phomopsis</i> sp.
P4a	<i>Alternaria</i> sp.	Coimbatore District	379988	<i>Fusarium</i> sp.
P4b	<i>Alternaria</i> sp.	Coimbatore District	379989	<i>Alternaria alternata</i>
P5a	<i>Fusarium</i> sp.	Coimbatore District	379990	<i>Cochliobolus hawaiiensis</i>
P6a		Coimbatore District	379991	<i>Cochliobolus pallescens</i>
P7a	<i>Fusarium</i> sp.	Coimbatore District	379992	<i>Fusarium solani</i>
P8a	<i>Curvularia</i> sp.	Coimbatore District	379993	<i>Curvularia verruculosa</i>
P8b	<i>Curvularia</i> sp.	Coimbatore District	379994	<i>Eurotium chevalieri</i>
P9b	<i>Fusarium</i> sp.	Coimbatore District	379995	<i>Fusarium pallidoroseum</i>
P10a		Coimbatore District	379996	<i>Lasiodiplodia theobromae</i>
P11a	<i>Fusarium</i> sp.	Coimbatore District	379997	<i>Alternaria</i> sp.
P11b	<i>Fusarium</i> sp.	Coimbatore District	379998	<i>Fusarium equisiti</i>
P12a	<i>Monilia</i> sp.	Coimbatore District	379999	<i>Curvularia</i> sp.
P12b	<i>Monilia</i> sp.	Coimbatore District	380000	<i>Sagenomella alba</i>

Outputs

2. Accurate data on the socio-economic impact of the weed in India obtained, particularly the affect on human affairs in peri-urban situations.

Tamil Nadu Agricultural University, Coimbatore, India

Once the health hazards associated with Parthenium weed are known or experienced, labourers are reluctant to come forward for weeding Parthenium weed. It is the major problem as reported by 86.6 % of the farmers sampled (Table 18). Labourers are also demanding extra wages for weeding *P. hysterophorus*-infested land, and it was the major weed reported by 70 % of the sample farmers in Vellore district, and 53.3 % in Coimbatore district.

Table 18 : Problems in weeding *Parthenium hysterophorus*

Details	Vellore		Coimbatore	
	No. of farmers reported	% to total	No. of farmers reported	% to total
Labourers not willing	26	86.6	26	86.6
Fear of health hazards by farmers	2	30.0	20	66.6
Demanding extra wages	21	70.0	16	53.3

Health hazards due to Parthenium weed infestation were reported throughout the study area. Dermatitis caused by pollen grains of *P. hysterophorus* was reported by almost all the respondents excepting a few. Allergy was reported in high *P. hysterophorus*-infested regions viz., Vellore and Coimbatore districts. Other hazards, like reddening of eyes, fever, and headache, were also reported in Vellore and Coimbatore districts as shown in Table 19.

Table 19 : Health hazards due to *Parthenium hysterophorus* infestation

Sl.No	District	Dermatitis	Reddening of eyes	Allergy	Fever	Headache	Eye sight problems	Swelling
1	Virudhunagar	28 (93.33)	-	-	-	-	-	-
2	Vellore	25 (83.33)	3 (10.00)	15 (50.00)	6 (20.00)	2 (6.66)	1 (3.33)	-
3	Ramanathapuram	30 (100.00)	-	-	-	-	-	-
4	Salem	23 (76.66)	-	-	-	-	-	-
5	Trichirapalli	21 (70.00)	-	-	-	-	-	-
6	Coimbatore	29 (96.66)	11 (36.66)	17 (56.66)	-	1 (3.33)	-	4 (13.33)

Figures in parentheses indicate percentage to number of farmers sampled in the district concerned

To avoid health problems, farmers and labourers resorted to different methods as a precaution after weeding *P. hysterophorus*: taking a bath immediately after weeding was followed by the majority of people as shown in Table 20. It was followed by washing with soap, applying coconut oil, avoiding exposure to sun after weeding by sample farmers in Vellore and Coimbatore i.e high infested area.

Table 20: Precautions after weeding *Parthenium hysterophorus*

Sl. No	District	No. of farmers reported					
		Washing with soap	Applying coconut oil	Washing with sand	Avoid exposure to sun	Weeding before 1 P.M	Taking bath
1	Virudhunagar	26	1	5	-	-	2
2	Vellore	25	15	1	4	-	10
3	Ramanathapuram	-	-	-	-	-	30
4	Salem	6	-	-	-	-	19
5	Trichirapalli	-	-	-	-	-	29
6	Coimbatore	19	10	9	4	20	8

Outputs

2. Accurate data on the socio-economic impact of the weed in India obtained, particularly the affect on human affairs in peri-urban situations.

Kurukshetra University, India

During the extensive surveys conducted between 1997 and 1999 in the northern parts of India *P. hysterophorus* infestation was recorded in various agricultural/vegetable/fodder crops (Table 21), causing losses ranging between 10 and 80% as in wheat (*Triticum aestivum*, 30-40%), sugarcane (*Saccharum officinarum*, 40-60%), jowar (*Sorghum vulgare*, 20-30%), barseem (*Trifolium alexandrianum*, 50-60%), sunflower (*Helianthus annuus*, 30-80%), sarson (*Brassica campestris*, 30-40%), taramira (*Eruca sativa*, 70-80%), gram (*Cicer arietinum*, 10-20%), potato (*Solanum tuberosum*, 30-50%), onion (*Allium cepa*, 30-40%), garlic (*Allium sativum*, 25-30%), lady's fingers (*Abelmoscus esculentus*, 20-30%), Arvi (*Colocasia* sp., 25-30%), metha (*Trigonella foenum-graceum*, 20-40%). The infestation of Parthenium weed has also been seen in one timber crop, Poplar (*Populus alba*, 30-75%). An interesting observation made is that once Parthenium weed had infested an agricultural/vegetable/fodder crop it occurred in successive crops, if suitable control measures had not been applied. For example, it was found infesting sugarcane crop followed by wheat in succession. In another case, this weed was recorded in taramira crop followed by sunflower in the same field. No infestation of this weed was seen in the crops such as rice (*Oryza sativa*), pea (*Pisum sativum*), walaite kaddu (*Cucurbita maxima*), kerala (*Momordica charantia*), kakri (*cucumis melo* var. *utilissima*) and khira (*Cucumis sativus*). The worst hit crops by *P. hysterophorus* infestation in this region is sugarcane (*Saccharum officinarum*), mustard (*Brassica campestris*) and barseem (*Trifolium alexandrianum*).

Table 21: Data on infestation of Parthenium weed in various agricultural/vegetable/fodder/oil yielding crops of northern India

Sr. No.	Crop	No. of Fields visited	No. of Fields Weed	% Occurrence of the weed	% Losses in Yield*
1.	<i>Saccharum officinarum</i>	200	75	40%	40-60%
2.	<i>Brassica campestris</i>	100	30	30%	30-40%
3.	<i>Trifolium alexandrium</i>	100	30	30%	50-60%
4.	<i>Oryza sativa</i>	200	0	0	0
5.	<i>Helianthus annuus</i>	100	20	20%	30-80%
6.	<i>Triticum aestivum</i>	200	10	10%	30-40%
7.	<i>Sorghum vulgare</i>	100	25	25%	20-30%
8.	<i>Eruca sativa</i>	30	5	16%	70-80%
9.	<i>Cicer arietinum</i>	20	3	15%	10-20%
10.	<i>Pisum sativum</i>	170	0	0	0
11.	<i>Solanum tuberosum</i>	100	5	5%	30-50%
12.	<i>Allium cepa</i>	75	8	11%	30-40%
13.	<i>Allium sativum</i>	40	3	8%	25-30%
14.	<i>Colocasia</i> sp.	35	8	23%	25-30%
15.	<i>Abelmoscus esculentus</i>	100	5	5%	20-30%
16.	<i>Trigonella foenum-graceum</i>	30	4	14%	20-40%
17.	<i>Cucurbita maxima</i>	100	0	0	0
18.	<i>Momordica charantia</i>	100	0	0	0
19.	<i>Cucumis melo</i>	95	0	0	0
	var. <i>utilissima</i>				
20.	<i>Cucumis sativus</i>	100	0	0	0
23.	<i>Populus alba</i>	50	20	40%	30-75%

*% Losses in yield has been calculated on the basis of farmers report/comments and personal observations

Outputs

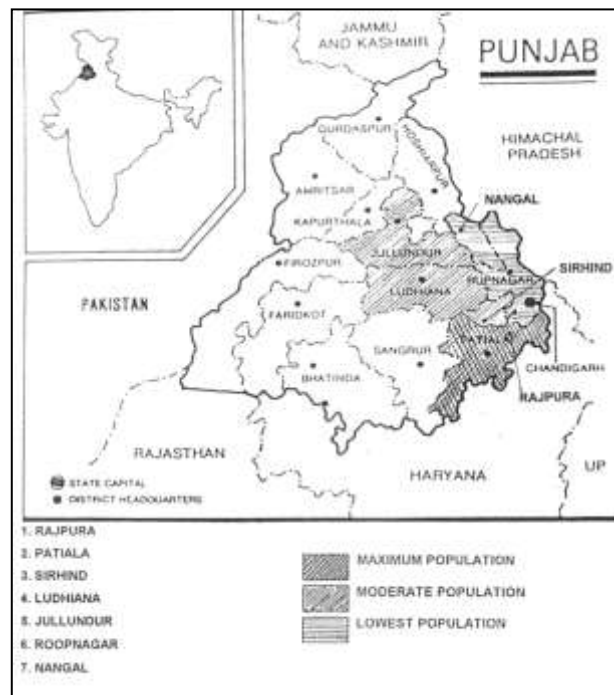
Surveys in Northern India – Distribution of Parthenium weed

In Haryana state, Parthenium weed was recorded along the road sides, railway tracks, in strips along village linking roads, pastures, gardens, vacant plots in urban areas, boundaries of the agricultural fields and infestation of some crops. Vacant plots in the Urban Estates were having the most luxuriant growth of the weed. In fact there was not one plot visited which was not completely occupied by the Parthenium weed. Kurukshetra district had the maximum population of this weed followed by Karnal, Ambala, Panchkula, Kalka, Yamuna Nagar and Panipat (Fig. 2). In Punjab, Rajpura was the worst affected by Parthenium weed followed by Patiala, Sirhind, Ludhiana, Jalandhar and Sirhind, Ropar and Nangal (Fig. 3). In fact, weed populations occurred along the whole of the Amritsar - Delhi stretch of Sher Shah Suri Marg (National Highway). Chandigarh, the capital of Punjab and Haryana states is the worst hit by Parthenium weed (Fig. 4). It was found growing in every nook and corner of the city throughout the year. In Himachal Pradesh, this weed was recorded from the foothills, i.e. Parwanoo, the place adjoining Kalka (Haryana) to Solan and Mandi; but it has still not reached Simla the capital of Himachal Pradesh, which is at a height of 600-700m above sea level (Fig. 5). In western Uttar Pradesh, Parthenium weed was observed in Ghaziabad, Modinagar, Meerut, Muzaffar Nagar, Roorki, Hardwar, Rishikesh and Saharanpur. Ghaziabad having the highest population of the weed followed by Saharanpur (Fig. 6). Delhi, the capital of India, is also badly hit by this weed and it was observed throughout the state (Fig. 7). Parthenium weed has been observed to be flowering and fruiting throughout the year, while the maximum growth in north India takes place during May to September, when the relative humidity is at its highest. The weed reaches a height of 2m. during the August to September months. The area occupied by this weed alone is calculated at between 8 and 10 million hectares in this region.

Fig. 2



Fig. 3



Outputs

Fig. 4



Fig. 5

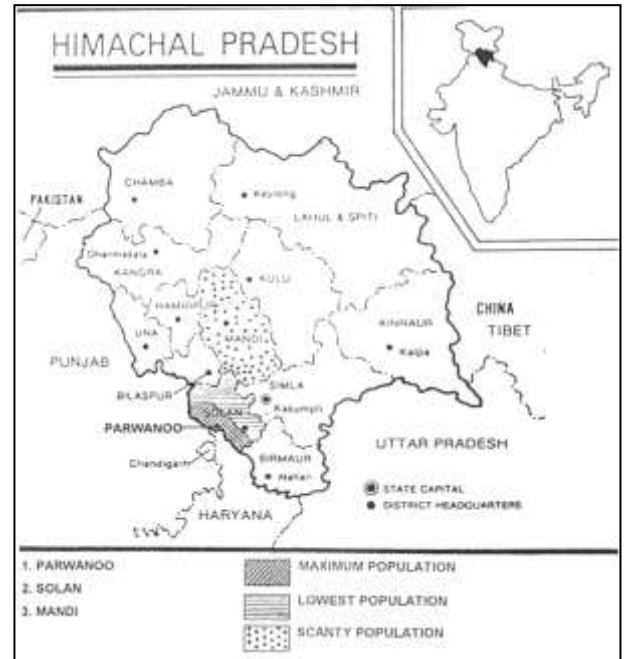


Fig. 6



Fig. 7



Outputs

The observations made by the authors during the surveys reveal that *Parthenium* weed is the most dominant weed of this region and should be treated as number one troublesome terrestrial weed. Moreover, the area of infestation of this weed is increasing not only from year to year but day by day. Worst hit directly by *Parthenium* weed are grazing animals e.g. buffaloes, cows and sheep, as the pastures are being occupied by *Parthenium* weed completely eradicating fodder grasses. If suitable control measures are not immediately adopted, production of milk and meat in India is likely to be reduced considerably resulting in a hike in the prices of these commodities in the years to come which put them beyond the reach of the masses.

Health hazards due to *Parthenium hysterophorus* infestation

Preliminary data collected on health hazards due to *P. hysterophorus* infestation in Kurukshetra district during 1998-1999 (Table 21) reveal that the health hazards caused by *P. hysterophorus* infestation in human beings include ABCD, asthma, irritation of the throat, headache and itching/reddening of eyes. Of these, the commonest disorders observed were itching and dermatitis.

Table 21: Health hazards due to *Parthenium hysterophorus* infestation in Kurukshetra district

S. No	Places	No. of patients interviewed	ABCD/ Skin diseases	Asthma	Throat irritation	Headache	Itching/ reddening eyes
1	Kurukshetra University Campus	25	8	2	6	5	4
2	Thaneshar	10	2	-	4	3	1
3	Amin	12	4	1	-	2	5
4	Khaspur	5	-	-	1	1	3
5	Jirbari	10	2	-	2	-	6
6	Umari	15	4	1	1	5	4
7	Kalamajara	6	-	-	1	1	4
8	Ratgal	6	1	1	-	-	4
9	Kishanpura	20	4	2	2	6	6
10	Pipli	10	1	1	3	3	2

Preliminary data collected from the Government and private hospitals of Kurukshetra District revealed that health hazards due to *P. hysterophorus* infestation were predominantly Air Borne Contact Dermatitis (ABCD). The daily turnover of patients suffering from this disease at LNJP Hospital, Kurukshetra, was 4-5. The commonest symptoms observed were itching, darkening and thickening of the skin, especially of the face. A private physician in Kurukshetra, specialising in skin, reported that 7 to 8 patients were visiting his clinic on a daily basis, coinciding with the peak flowering period of *Parthenium* weed. The University Health Centre's physician reported that several patients visited the centre suffering from dermatitis and asthma and that these had been predominantly engaged in mechanical removal of the weed. The most affected tend to be gardeners, labourers and farmers who come into frequent contact with the weed. At present, only temporary relief can be achieved through steroids and anti allergy therapy.

Outputs

3. *Biocontrol agent(s) identified and screened for release in India* CABI Bioscience, UK

All of the Indian varieties of *P. hysterophorus* were highly susceptible to both strains of *Puccinia melampodii*. This would indicate little genetic diversity within the Indian population of the weed, and the suitability of the *P. melampodii* rust as a biocontrol agent (Table 22). Initial symptoms on the host appeared as chlorotic spots within six to seven days of inoculation with sporulation occurring on the underside of the leaf two to three days later.

Inoculated test plant species were examined for the presence of any macroscopic symptoms of disease and results are summarised in Table 22.

Of the host range plants tested, only two showed signs of infection, *Calendula officinalis* and *Guizotia abyssinica*, both from the Asteraceae. On *C. officinalis*, (a New World plant) *P. melampodii* produced necrotic patches on all ages of leaves, in some of these areas a few teliospore could be identified. These teliospores were capable of producing basidiospores, the viability of these basidiospores was not tested, but in work with *C. officinalis* for Australia the basidiospores produced were able to infect *P. hysterophorus* (Tomley & Seier, 1999). *G. abyssinica* originates from Africa, a UK commercial cultivar was added to the screening due to the evidence of feeding of the *Zygogramma* beetles (Jayanth & Nagarkatti, 1987), the rust strain W1500 produced necrotic patches and limited sporulation on one of the replicate plants, which in turn produced basidiospores. As a result of this, seeds of *G. abyssinica* were requested from India, the plants grown from Karnataka seed produced no symptoms with either strain of the rust, those from Kurukshetra, developed discolouration on their leaves but no specific areas of infection nor sporulation.

Cultivars of *Helianthus annuus* showed generally chlorotic/necrotic spotting following inoculation with *P. melampodii*. However, sporulation of the rust was never observed.

Table 22 : Macroscopic results of host range screening of two strains of *Puccinia melampodii* from Mexico (W 1496 and W 1500)

<i>Species name</i>	Variety/cultivar/ commercial mix	W 1500 Comments	W 1496 Comments
<i>Parthenium hysterophorus</i>	Chandigarh	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Patiala	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Uttar Pradesh	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Kurukshetra 1	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Kurukshetra 2	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Kurukshetra 3	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Madhya Pradesh	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Kurukshetra (Kishanpura)	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Kurukshetra (field of <i>Eruca sativa</i>)	Infection level high, good sporulation	Infection level high, good sporulation

Outputs

<i>Species name</i>	Variety/cultivar/ commercial mix	W 1500 Comments	W 1496 Comments
<i>P. hysterophorus</i>	Tamil Nadu	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Karnataka (Bangalore)	Infection level high, good sporulation	Infection level high, good sporulation
<i>P. hysterophorus</i>	Kurukshetra STANDARD	Infection level high, good sporulation	Infection level high, good sporulation
Greengram	var. CO-4	Asymptomless	Asymptomless
Gourd		**	**
Bean	var. S9	Asymptomless	Asymptomless
<i>Abelmoschus esculentus</i>	var. Arka Abhay	**	**
<i>A. esculentus</i>	var. Varsha	Asymptomless	Asymptomless
<i>Amaranthus bicola</i>	var. Arka Suguna	Asymptomless	Asymptomless
<i>Arachis hypogaea</i>	var. CO-2	Asymptomless	Asymptomless
<i>A. hypogaea</i>	JL 24	Asymptomless	Asymptomless
<i>Aster</i>	Michaelmas daisies	Asymptomless	Asymptomless
<i>Aster</i>	Quadrille mixed	Asymptomless	Asymptomless
<i>Aster amellus</i>	Pot 'n' patio	Asymptomless	Asymptomless
<i>A. amellus</i>	Powder puffs mix	Asymptomless	Asymptomless
<i>Beta vulgaris</i>	Ruby-queen	Asymptomless	Asymptomless
<i>Brassica campestris</i>	PR 45 9	Asymptomless	Asymptomless
<i>Brassica juncea</i>		Asymptomless	Asymptomless
<i>Brassica oleracea</i>	var. Unnati	Asymptomless	Asymptomless
<i>Brassica oleracea</i> var. botryti	P Deepali	*	*
<i>Cajanus cajan</i>	P 855	Asymptomless	Asymptomless
<i>Calendula officinalis</i>	Touch red/yellow	+Chlorotic/ necrotic spotting; limited sporulation ¹	+Chlorotic/ necrotic spotting; limited sporulation ¹
<i>C. officinalis</i>	Touch red/orange	+Chlorotic/ necrotic spotting; limited sporulation ¹	+Chlorotic/ necrotic spotting; limited sporulation ¹
<i>Capsicum annuum</i>	(P Jawala)	Asymptomless	Asymptomless
<i>C. annuum</i>	var. LCG 4	Asymptomless	Asymptomless
<i>C. annuum</i>	var. LCG 5	Asymptomless	Asymptomless
<i>C. annuum</i>	var. LCA 206	Asymptomless	Asymptomless
<i>C. annuum</i>	var. LCA 235	Asymptomless	Asymptomless
<i>C. annuum</i>	var. LCA 960	Asymptomless	Asymptomless
<i>Carthamus tinctorius</i>	Goldtuft	Asymptomless	Asymptomless
<i>Cicer arietinum</i>	P-256	Asymptomless	Asymptomless
<i>C. arietinum</i>	P-267	Asymptomless	Asymptomless
<i>C. arietinum</i>		Asymptomless	Asymptomless
<i>Citrullus lanatus</i>	Madhu	Asymptomless	Asymptomless
<i>Colocasia esculenta</i>		Asymptomless	Asymptomless
<i>Cosmos bipinnatus</i>	Sensation mix	Asymptomless	Asymptomless
<i>C. bippinatus</i>	Sunny red	Asymptomless	Asymptomless
<i>Cucumis melo</i>	var. Arka Jeet	Asymptomless	Asymptomless
<i>Cucumis sativus</i>		Asymptomless	Asymptomless
<i>C. sativus</i>	Green long	Asymptomless	Asymptomless
<i>Cucurbita moschata</i>	var. Arka Chandan	Asymptomless	Asymptomless
<i>Cyamopsis tetragonoloba</i>	R98 BHSC10	Asymptomless	Asymptomless
<i>Daucus carota</i>	Early nantes	Asymptomless	Asymptomless
<i>Eleusine coracana</i>	INDAF 9	Asymptomless	Asymptomless
<i>E. coracana</i>	HR-911	Asymptomless	Asymptomless
<i>E. coracana</i>	GPU 28	Asymptomless	Asymptomless
<i>Gazania hybrida</i>	Sunshine mixed	Asymptomless	Asymptomless
<i>Glycine max</i>		Asymptomless	Asymptomless
<i>Guizotia abyssinica</i>	Karnataka	Minor chlorosis	Minor chlorosis

Outputs

<i>Species name</i>	Variety/cultivar/ commercial mix	W 1500 Comments	W 1496 Comments
<i>G. abyssinica</i>	Kurukshetra	Minor discolouration of leaves	Minor discolouration of leaves
<i>G. abyssinica</i>	UK source	Chlorosis; limited abnormal sporulation on 1 replicate ¹	Asymptomless
<i>Helianthus annuus</i>	var. PSFH-67	*	*
<i>H. annuus</i>	var. DK-3890	*	*
<i>H. annuus</i>	var. Jawala mukhi	Chlorotic/ necrotic spotting	Sometimes minor chlorosis
<i>H. annuus</i>	var. Sungene 25	Chlorotic/ necrotic spotting	Slight chlorosis
<i>H. annuus</i>		*	*
<i>H. annuus</i>	var. CO-4	Chlorotic/ necrotic spotting	Chlorotic/ necrotic spotting
<i>H. annuus</i>	var. Morden	Chlorotic/ necrotic spotting	Chlorotic/ necrotic spotting
<i>H. annuus</i>	var. KBSH-1	Chlorotic/ necrotic spotting	Chlorotic/ necrotic spotting
<i>H. annuus</i>	GAUSUF-15	Chlorotic/ necrotic spotting	Chlorotic/ necrotic spotting
<i>H. annuus</i>	PAC-1091	Chlorosis/ necrotic spotting	Chlorotic/ necrotic spotting
<i>H. annuus</i>	SH3322	Chlorotic/ necrotic spotting	Chlorotic/ necrotic spotting
<i>H. annuus</i>	Arun	Chlorotic/ necrotic spotting	Chlorotic/ necrotic spotting
<i>H. annuus</i>	var. MSF-17	Chlorotic/ necrotic spotting	Chlorotic/ necrotic spotting
<i>H. annuus</i>	var. EC-68414	Chlorotic spotting/ necrosis	Chlorotic/ necrotic spotting
<i>Lagenaria siceraria</i>		Asymptomless	Asymptomless
<i>L. siceraria</i>	var. Arka Bahar	Asymptomless	Asymptomless
<i>L. siceraria</i>	Khol Khol EW	Asymptomless	Asymptomless
<i>L. siceraria</i>	PSPL	Asymptomless	Asymptomless
<i>Luffa acutangula</i>	JL	Asymptomless	Asymptomless
<i>L. acutangula</i>	IAHS-1	*	*
<i>Lycopersicon esculentum</i>	var. Arka Saurabh	Asymptomless	Asymptomless
<i>L. esculentum</i>	var. Pusa Ruby	Asymptomless	Asymptomless
<i>L. esculentum</i>	var. Marutham	Asymptomless	Asymptomless
<i>L. esculentum</i>	Dwarf hybrid	Asymptomless	Asymptomless
<i>Oryza sativa</i>	var. Mandya Vijaya	Asymptomless	Asymptomless
<i>O. sativa</i>	P 44	Asymptomless	Asymptomless
<i>O. sativa</i>	P Basmati	Asymptomless	Asymptomless
<i>O. sativa</i>		Asymptomless	Asymptomless
<i>O. sativa</i>	var. Tellahamsa	Asymptomless	Asymptomless
<i>O. sativa</i>	var. Mangala	Asymptomless	Asymptomless
<i>O. sativa</i>	var. IR 64	Asymptomless	Asymptomless
<i>O. sativa</i>	var. Rasi	Asymptomless	Asymptomless
<i>O. sativa</i>	var. Jaya	Asymptomless	Asymptomless
<i>O. sativa</i>	var. Jyothi	Asymptomless	Asymptomless
<i>O. sativa</i>	BPT-5204	Asymptomless	Asymptomless
<i>O. sativa</i>	var. Vikramarya	Asymptomless	Asymptomless
<i>O. sativa</i>	var. T (N) 1	Asymptomless	Asymptomless
<i>O. sativa</i>	var. IET 8585	Asymptomless	Asymptomless
<i>O. sativa</i>	var. IET-9994	Asymptomless	Asymptomless
<i>Pennisetum typhoides</i>	var. PT-1890		
<i>Phaseolus mungo</i>	var. CO-5	Asymptomless	Asymptomless
<i>P. mungo</i>	var. T9	Asymptomless	Asymptomless
<i>Phaseolus vulgaris</i>		Asymptomless	Asymptomless
<i>P. vulgaris</i>	var. Arka Komal	Asymptomless	Asymptomless
<i>Pisum sativum</i>	var. Arkel	Asymptomless	Asymptomless
<i>Raphanus sativus</i>	var. P. Rashmi	Asymptomless	Asymptomless
<i>R. sativus</i>	var. Arka Nishant	Asymptomless	Asymptomless
<i>R. sativus</i>	var. Pusa Chetaki	Asymptomless	Asymptomless
<i>Rudbeckia speciosa</i>		*	*
<i>Setaria italica</i>	var. CO-6	Asymptomless	Asymptomless

Outputs

Species name	Variety/cultivar/ commercial mix	W 1500 Comments	W 1496 Comments
<i>Solanum melongena</i>	var. Arka Nidhi	Asymptomless	Asymptomless
<i>S. melongena</i>	var. Arka Sheel	Asymptomless	Asymptomless
<i>S. melongena</i>	var. Bhagyamathi	Asymptomless	Asymptomless
<i>S. melongena</i>	var. Shyamala	Asymptomless	Asymptomless
<i>S. melongena</i>	var. Brinjal Purple red	Asymptomless	Asymptomless
<i>S. melongena</i>	var. Pusa Purple long	*	*
<i>Solidago canadensis</i>	Golden baby	*	*
<i>Sorghum vulgare</i>	var. PC9	Asymptomless	Asymptomless
<i>S. vulgare</i>	var. CO-26	Asymptomless	Asymptomless
<i>Spinacia oleracea</i>		Asymptomless	Asymptomless
<i>Tagetes erecta.</i>	Calando mixed	Asymptomless	Asymptomless
<i>Tagetes patula</i>		Asymptomless	Asymptomless
<i>Trigonella focnum-graceum</i>		Asymptomless	Asymptomless
<i>Triticum aestivum</i>	HD 2009	Asymptomless	Asymptomless
<i>T. aestivum</i>	HD 2428	Asymptomless	Asymptomless
<i>T. aestivum</i>	HD 2329	Asymptomless	Asymptomless
<i>Triticum vulgare</i>		Asymptomless	Asymptomless
<i>Vigna mungo</i>	var. T9	Asymptomless	Asymptomless
<i>V. mungo</i>	var. LBG 402	Asymptomless	Asymptomless
<i>Vigna radiata</i>	P Basakhi	Asymptomless	Asymptomless
<i>Vigna sinensis</i>	var. Arka Garima	Asymptomless	Asymptomless
<i>Vigna unguiculata</i>		Asymptomless	Asymptomless
<i>V. unguiculata</i>	var. C-152	Asymptomless	Asymptomless
<i>V. unguiculata</i>	var. C-152	*	*
<i>Zea mays</i>	Hybrid 203492	Asymptomless	Asymptomless
<i>Z. mays</i>	var. CO-1	Asymptomless	Asymptomless
<i>Z. mays</i>	var. Kanchan	Asymptomless	Asymptomless
<i>Z. mays</i>	var. Ganga 11	Asymptomless	Asymptomless
<i>Z. mays</i>	var. C6	Asymptomless	Asymptomless
<i>Z. mays</i>	var. Himalaya 123	Asymptomless	Asymptomless
<i>Zinnia elegans</i>	Pulcino mix	Asymptomless	Asymptomless
<i>Z. elegans</i>	Candy cane mix	Asymptomless	Asymptomless

* No germination of seed

** Low germination of seed

+ Infection/Sporulation

¹ *P. melampodii* teliospores produced by the telia on *Calendula officinalis* and *Guizotia abyssinica* (UK source) were shown to produce basidiospores when incubated in the dew chamber at 20 °C.

The strain of *Puccinia abrupta* var. *partheniicola* tested was equally virulent on all strains of *P. hysterophorus* sent from India. Initial symptoms of chlorotic spots appeared seven to ten days after inoculation, sporulation occurred on the upper leaf surface three to four days later. Symptoms on the *H. annuus* cultivars were similar to those which developed on the Australian sunflowers (Tomley, 1990), no sporulation was visible. The results of the macroscopic examinations are summarized in Table 23.

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Table 23: Macroscopic results of host range screening of *Puccinia abrupta* var. *partheniicola* from Mexico (strain W1905)

<i>Species name</i>	Variety/cultivar/ commercial mix	Comments
<i>Parthenium hysterophorus</i>	Chandigarh	Good infection/sporulation
<i>P. hysterophorus</i>	Patiala	Good infection/sporulation
<i>P. hysterophorus</i>	Uttar Pradesh	Good infection/sporulation
<i>P. hysterophorus</i>	Kurukshetra 1	Good infection/sporulation
<i>P. hysterophorus</i>	Kurukshetra 2	Good infection/sporulation
<i>P. hysterophorus</i>	Kurukshetra 3	Good infection/sporulation
<i>P. hysterophorus</i>	Kurukshetra 4	Good infection/sporulation
<i>P. hysterophorus</i>	Madhya Pradesh	Good infection/sporulation
<i>P. hysterophorus</i>	Kurukshetra (Kishanpura)	Good infection/sporulation
<i>P. hysterophorus</i>	Kurukshetra	Good infection/sporulation
<i>P. hysterophorus</i>	Tamil Nadu	Good infection/sporulation
<i>P. hysterophorus</i>	Kurukshetra – STANDARD	Good infection/sporulation
<i>Helianthus annuus</i>	var. Jawala mukhi	Slight chlorosis
<i>H. annuus</i>	var. Sungene 25	Slight chlorosis
<i>H. annuus</i>	var. CO-4	Chlorotic/necrotic spotting
<i>H. annuus</i>	var. Morden	Chlorotic/necrotic spotting
<i>H. annuus</i>	var. KBSH-1	Chlorotic/necrotic spotting
<i>H. annuus</i>	GAUSUF-15	Chlorotic/necrotic spotting
<i>H. annuus</i>	PAC-1091	Chlorotic/necrotic spotting
<i>H. annuus</i>	SH3322	Chlorotic/necrotic spotting
<i>H. annuus</i>	Arun	Chlorotic/necrotic spotting
<i>H. annuus</i>	var. MSF-17	Chlorotic/necrotic spotting
<i>H. annuus</i>	var. EC-68414	Chlorotic/necrotic spotting

The macroscopic symptoms visible on the *Helianthus annuus* (chlorosis/necrosis) following inoculation with *P. melampodii*/*P. abrupta* var. *partheniicola* were evaluated microscopically, after tissue had been stained and cleared. These detailed evaluations were then ranked into 14 categories for both rust species, as listed in Tables 24 and 25, respectively. In the case of *P. melampodii*, the assessment categories refer exclusively to basidiospore behaviour, whilst for *P. abrupta* var. *partheniicola*, the assessment categories apply to urediniospores.

Results from staining are outlined in Tables 26 and 27 for *P. melampodii* strains, W1500 and W1496, respectively. Table 28 outlines results for strain W1905 of *P. abrupta* var. *partheniicola*.

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Table 24 : Assessment categories developed to classify micro-symptoms recorded for plants inoculated with *Puccinia melampodii*

1	=	basidiospore germination
2	=	no basidiospore germination; sometimes associated with plant defence reactions indicating inhibition of germination
3	=	basiospore germination with well-developed germ tube
4	=	abnormal basidiospore germination
5	=	successful basidiospore penetration
6	=	no attempted penetration
7	=	attempted but failed penetration
8	=	successful penetration, but no further internal development
9	=	limited internal development, usually associated with cellular plant defence reactions and chlorotic symptoms
10	=	haustoria formation
11	=	extensive internal development commonly associated with sori initiation
12	=	formation of underdeveloped telia and immature teliospores
13	=	abnormal sporulation with viable teliospores
14	=	abundant sporulation with well developed telia and viable teliospores

Table 25: Assessment categories developed to classify micro-symptoms recorded for plants inoculated with *Puccinia abrupta* var. *partheniicola* (after Tomley, 1990)

1	=	spore germination
2	=	appressoria formation over stomata
3	=	substomatal vesicle formation
4	=	short internal hyphae present
5	=	necrosis of guard cells under appressorium
6	=	necrosis around short internal hyphae
7	=	short internal hyphae with haustoria
8	=	longer internal hyphae with haustoria
9	=	callose around haustoria
10	=	host cell granulation
11	=	necrosis around longer internal hyphae
12	=	callose deposited on host cell walls around infection
13	=	sorus formation
14	=	sporulation

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Table 26 : Results of microscopic evaluation of inoculated plants revealing macroscopic symptoms following inoculation with *P. melampodii* (strain W1500)

Species	Macro/microsymptoms													
	Germination				Penetration			Colonization				Sporulation		
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
<i>Family Asteraceae</i>														
Sub-family Asteroideae														
Tribe Heliantheae														
<i>Helianthus annuus</i> vars.														
Morden	+	-	+	-	+	-	+	+	+	-	-	-	-	-
CO-4	+	-	+	+	+	-	+	-	-	-	-	-	-	-
EC-68414	+	-	+	-	+	-	-	+	+	-	-	-	-	-
KBSH-1	+	-	+	+	-	-	+	-	-	-	-	-	-	-
PAC-1091	+	-	+	-	+	-	-	+	-	-	-	-	-	-
Gausuf-15	+	-	+	-	+	-	-	-	+	-	-	-	-	-
MSF-17	+	-	+	-	+	-	-	-	+	-	-	-	-	-
Arun	+	+	+	-	+	-	+	-	-	-	-	-	-	-
SH3322	+	-	+	-	-	+	+	-	-	-	-	-	-	-
Jawala Mundhi	+	+	+	+	-	-	+	-	-	-	-	-	-	-
Sungene 25	+	-	+	+	+	-	-	+	-	-	-	-	-	-
<i>Guizotia abyssinica</i> (Karnataka)	+	+	+	-	-	+	+	-	-	-	-	-	-	-
<i>Guizotia abyssinica</i> (Kurukshehra)	+	-	+	+	-	+	-	-	-	-	-	-	-	-

Table 27 : Results of microscopic evaluation of inoculated plants revealing macroscopic symptoms following inoculation with *P. melampodii* (strain W1496)

Species	Macro/microsymptoms													
	Germination				Penetration			Colonization				Sporulation		
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
<i>Family Asteraceae</i>														
Sub-family Asteroideae														
Tribe Heliantheae														
<i>Helianthus annuus</i> vars.														
Morden	+	-	+	-	+	-	+	+	-	-	-	-	-	-
CO-4	+	-	+	+	-	-	+	-	-	-	-	-	-	-
EC-68414	+	-	+	-	+	-	-	-	+	-	-	-	-	-
KBSH-1	+	-	+	-	-	-	+	-	-	-	-	-	-	-
PAC-1091	+	-	+	+	-	-	+	+	-	-	-	-	-	-
Gausuf-15	+	-	+	+	-	-	-	+	-	-	-	-	-	-

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MSF-17	+	-	+	-	-	-	-	-	+	-	-	-	-	-
Arun	+	-	+	-	+	-	+	+	+	-	-	-	-	-
SH3322	+	+	+	+	-	-	-	-	+	-	-	-	-	-
Jawala Mundhi	+	+	+	-	-	-	+	-	-	-	-	-	-	-
Sungene 25	+	+	+	+	-	-	+	-	-	-	-	-	-	-
<i>Guizotia abyssinica</i> (Karnataka)	+	+	-	-	-	+	-	-	-	-	-	-	-	-
<i>Guizotia abyssinica</i> (Kurukshetra)	+	-	-	-	-	+	-	-	-	-	-	-	-	-

Table 28 : Results of microscopic evaluation of inoculated plants revealing macroscopic symptoms following inoculation with *P. abrupta* var. *partheniicola* (strain W1905)

Species	Macro/microsymptoms													
	SYMPTOMS OBSERVED													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
<i>Family Asteraceae</i>														
Sub-family Asteroideae														
Tribe Heliantheae														
<i>Helianthus annuus</i> vars.														
Morden	+	+	+	+	-	+	-	-	-	-	-	-	-	-
CO-4	-	-	-	-	-	-	-	-	-	-	-	-	-	-
EC-68414	+	+	-	-	+	-	-	-	-	-	-	-	-	-
KBSH-1	+	+	-	-	-	-	-	-	-	-	-	-	-	-
PAC-1091	+	+	+	-	+	-	-	-	-	-	-	-	-	-
Gausuf-15	+	+	+	+	+	+	+	-	-	-	-	-	-	-
MSF-17	+	+	+	+	-	+	-	-	-	-	-	-	-	-
Arun	+	+	-	-	-	-	-	-	-	-	-	-	-	-
Sh3322	+	+	-	-	-	-	-	-	-	-	-	-	-	-
Jawala Mundhi	+	+	+	+	+	-	-	-	-	-	-	-	-	-
Sungene 25	+	-	-	-	-	-	-	-	-	-	-	-	-	-

Discussion

Macroscopic symptoms observed on the sunflower varieties were generally found to be comparable for the two strains of *P. melampodii* (W1500 and W1496). The susceptibility of individual sunflower varieties to W1500 and W1496, based on the extent of development of the pathogen observed microscopically, revealed minor differences in resistance levels; for example, varieties Morden, EC-68414 and Gausuf-15 were consistently high scoring in the assessment categories (Table 26, microsymptom 9) for both strains, whilst varieties CO-4, KBSH-1 and Jawala Mundhi were less susceptible to both strains (Table 27, microsymptom 7).

Overall, the rust was not able to complete its life cycle on any of the sunflower varieties. Internal development was restricted to limited hyphal growth associated with localised cell necrosis and the initiation of sori or teliospore development was never recorded.

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P. melampodii strain W1500 was able to sporulate on *Guizotia abyssinica*, however this was limited to a single replicate out of the three tested and plants inoculated with W1496 showed no signs of sporulation. Repeated inoculations using two Indian varieties of *G. abyssinica* revealed that basidiospore penetration was frequently unsuccessful and associated with plant cell defence reactions such as cell wall apposition or thickening. No internal mycelium or sori initiation was recorded (Tables 26 and 27).

Similarly, there was no evidence to show that *P. abrupta* var. *partheniicola* could develop sufficiently to produce pustules in any of the sunflower varieties. Internal development was limited to the formation of short hyphae whose further development was restricted by host cell necrosis.

The results of this initial screening demonstrate the restricted host range of the two rusts, *P. melampodii* (strains W1500 and W1496) and *P. abrupta* var. *partheniicola* (strain W1905). The rusts failed to sporulate on the majority of host species tested, despite the optimal conditions for spore germination and infection provided in the greenhouse. Both rusts were found to be highly virulent towards *Parthenium hysterophorus* from all localities. The susceptibility to *P. melampodii* was shown to vary depending on the variety of the test species, for example *H. annuus* and *Guizotia abyssinica* (see also Output 7, CABI Bioscience) and on those plant species where sporulation was recorded (*G. abyssinica* and *C. officinalis*), pustule size and abundance were generally abnormal, indicating these species are not natural hosts of the rusts.

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3. Biocontrol agents identified and screened for release in India

National Research Centre for Weed Science, Jabalpur, India

Pathogenicity

It was confirmed that *Fusarium pallidoroseum*, *Sclerotium rolfsii*, *Sclerotinia sclerotiorum* and *Alternaria alternata* were found to be the most effective pathogens against Parthenium weed.

Host specificity testing

Host specificity testing of *Fusarium pallidoroseum* on crops

Out of 13 crops, only cowpea, cucumber, jowar and paddy were resistant. Brinjal and lady's finger was moderately resistant and chili, cauliflower, coriander, maize, radish and tomato were susceptible (Table 29).

Table 29: Effect of seed treatment with *Fusarium pallidoroseum* on seed germination of different crops (Blotter method).

Sl No	Crop	Treated			Control		
		no. of seeds sown/ plates	Germinated seeds	Death after germination	no. of seeds sown/ plates	Germinated seeds	Death after germination
1	Brinjal	100	60	60	100	54	-
2	Chilli	100	76	08	100	72	-
3	Cowpea	100	94	12	100	88	-
4	Cauliflower	100	87	10	100	100	-
5	Cucumber	100	78	78	100	100	-
6	Coriander	100	17	17	100	100	-
7	Jowar	100	82	00	100	100	-
8	Lady's finger	100	100	00	100	100	-
9	Maize	100	8	00	100	100	-
10	Paddy	100	29	06	100	100	-
11	Radish	100	94	10	100	100	-
12	Soybean	100	00	00	100	100	-
13	Tomato	100	75	75	100	100	-

Seeds of different crops/host were treated with spore suspensions of *F. pallidoroseum* and plated on filter paper in petri dishes. Germination was performed at $25 \pm 1^\circ\text{C}$ and 90 RH in a BOD incubator. After 10 days, the seedlings were planted in pots. The results indicate that the cowpea, cucumber, jowar and paddy are resistant to this pathogen, but crops such as brinjal, chilli, cauliflower, coriander, lady's finger, maize, radish and tomato are susceptible and that this pathogen is not safe to use in such cropping systems (Table 30).

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Table 30: Effect of *Fusarium pallidoroseum* seed treatment of different crop/non crop host

Sl No	Crop of name	Treated			Control		
		No. of seedling transplanted	Established	Death	No. of seedling transplanted	Established	Death
1	Brinjal	25	10	15	25	14	11
2	Chilli	25	05	20	25	17	8
3	Cowpea	25	25	00	25	25	0
4	Cauliflower	25	07	18	25	09	16
5	Cucumber	25	25	05	25	25	0
6	Coriander	25	05	20	25	04	21
7	Jowar	25	25	00	25	25	0
8	Ladyfinger	25	12	13	25	25	0
9	Maize	25	00	25	25	14	11
10	Paddy	25	20	05	25	17	8
11	Radish	25	00	25	25	04	21
12	Tomato	25	03	22	25	03	22

Effect of *Fusarium pallidoroseum* on seed germination and seedling mortality in petridishes.

Seed inoculation with *F. pallidoroseum* caused nearly 35% seed rot and 65% seedling mortality (Table 31). The fungus grew on the surface of the seed and became established within 36 hrs of inoculation, infecting roots of the plant. Root growth was inhibited and abnormal seedlings developed. The seedlings survived on the reserve food of the seed and died within 15 days of inoculation. The fungus also attacked the growing point of the seedling which turned light brown in colour and could affect all the tissues of the seedling.

Table 31: Effect of *Fusarium pallidoroseum* on seed germination and seedling mortality of *Parthenium hysterophorus*

Treatment	% germination	% seedling died
Inoculated seeds	65	100
Control seeds	86	0

Effect of seed treatment with *Sclerotium rolfsii* on *Parthenium hysterophorus* seed

The results indicated that *S. rolfsii* could inhibit seed germination of *P. hysterophorus* by 60 to 77 % as compared to the control. The fungus could also infect the basal portion of the stems and roots and the seedlings died within 10 to 12 days. This is a very virulent fungus, killing both the *P. hysterophorus* seeds and seedlings.

Effect of *Fusarium pallidoroseum* on *Parthenium hysterophorus* seed germination seedling infection at different days of sowing (in petri dishes)

For determining the critical time to spray *Fusarium pallidoroseum* for management of *Parthenium* weed, through reduction in the seed population of *Parthenium* weed, spore suspensions were sprayed on seed of *P. hysterophorus* 0, 1, 2, 3, 4, 5, 6 & 7 days after sowing. Only water was sprayed on the seeds for control treatment. The results indicated that out of 100 seeds sprayed, the fungus could colonise the seeds in the range of 6 to 19 %. The maximum colonization was obtained when seeds were sprayed 0 to 7 days after sowing. In case of spraying 0 to 3 days after sowing, germination was reduced by 57 to 100 %. This fungus could enhance the seed germination of *P. hysterophorus* by 2 to 3 days as compared to the control. In cases where the surface of the seed was completely covered by the growth of

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the fungus, there was no germination, where the surface was only partially covered, the seeds could germinate but the seedling subsequently died.

Effect of *Gliocladium virens* on *Parthenium hysterophorus*

Effect of *Gliocladium virens* and Neem oil on *Parthenium hysterophorus* seed germination

Seed germination of *P. hysterophorus* was tested against *G. virens* its culture filtrate, neem oil and their various combinations using different methods and the results are presented in Tables 32 and 33. It is clear from this data that the germination was inhibited in all the treatments compared to the control in which germination was 80 %. Highest percent inhibition of seed germination (86.41%) was observed in *G. virens* (culture filtrate) + Neem oil (10 %) combination which was greater than (67.90%) neem oil 10%, (62.02 %) *G. virens* (Culture filtrate +culture) +Neem oil 10% and (13.58) *G. virens* (culture filtrate).

Table 32 : Effect of *Gliocladium virens* (culture filtrate) and Neem oil on *Parthenium hysterophorus* seed germination (Blotter Paper soaked)

S.No	Treatment (Spray)	inoculated seeds	Germinated seed	% inhibition of germination	Root length/plant (cm)	Shoot length/plant (cm)	% inhibition of Root Shootlength length (cm)	
1.	<i>Gliocladium virens</i>	100	70	13.58	0.35	1.91	78.3	40.12
2.	Neem oil (10%)	100	26	67.90	1.12	2.25	30.86	29.46
3.	<i>Gliocladium virens</i> + Neem oil(10%)	100	11	86.41	0.30	1.75	81.48	45.14
4.	Control	100	81		1.62	3.19	-	-

Table 33 : Effect of *Gliocladium virens* (culture & culture filtrate) and Neem oil on *Parthenium hysterophorus* seed germination (Blotter Paper soaked)

S.No.	Treatment (Spray)	inoculated seed	No. of germinated seed	% inhibition of germination	Rootlength/ plant (cm)	Shoot length/plant (cm)	% inhibition of	
							Root length	length Shoot
1.	<i>Gliocladium virens</i>	100	76	3.79	0.35	1.95	83.56	26.96
2.	Neem oil (10%)	100	26	67.08	0.53	1.61	89.20	45.31
3.	<i>Gliocladium virens</i> + Neem oil (10%)	100	30	62.02	0.23	1.46	75.11	39.70
4.	Control	100	79	-	2.13	2.67	-	-

Effect of *Gliocladium virens* and Neem oil spray on *Parthenium hysterophorus* seed on soil

Root length and shoot length were also reduced in all the treatments compared to the control. Higher reduction in root, shoot and seed germination was obtained by the culture filtrate + oil treatment as compared to the culture + oil. Seed germination of *P. hysterophorus* was also tested with similar treatments on soil plates. Seed germination was 70% in the control as compared to 52% for *G. virens*, 50% for Neem oil and 50% for oil + *G. virens* respectively. Maximum inhibition of germination was in oil + *G. virens*. (Table 34).

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Table 34 : Effect of *Gliocladium virens* and Neem oil spray on *Parthenium hysterophorus* seed on soil.

Treatment	No. of inoculated seed	No. of germinated seeds	% inhibition of germination
<i>Gliocladium virens</i>	100	52	22.38
Neem oil	100	58	13.43
Oil + <i>Gliocladium virens</i>	100	50	25.37
Control	100	67	

Effect of Culture filtrate, Culture of *Gliocladium virens* and Neem oil on *Parthenium hysterophorus* seed germination

Table 35 compares the various treatment on seed germination using two methods. Both methods revealed that Neem oil + culture filtrate highly inhibited germination i.e. 96% in seed dip method and 74% in filter paper soaked method.

Table 35 : Effect of Culture filtrate, culture of *Gliocladium virens* and Neem oil on *Parthenium hysterophorus* seed germination (seed dip & Blotter Paper soaked)

S. No.	Treatments	No. of Inoculated Seed	No. of germinated seed		% inhibition of germination	
			Seed dip method	BPS method	Seed dip method	BPS method
1.	Control	100	50	43	0	
2.	Neem oil	100	11	16	78	62.00
3.	Culture filtrate (<i>G. virens</i>)	100	8	14	84	67.00
4.	Culture (<i>G. virens</i>)	100	46	26	08	39.00
5.	Neem oil + Culture filtrate	100	02	11	96	74.00
6.	Neem oil + Culture	100	14	14	72	67.00

Effect of *Gliocladium virens* and Neem oil spray on *Parthenium hysterophorus* seedlings (blotter paper)

Data presented in Table 36 shows the mortality of *P. hysterophorus* seedlings against treatments. Mortality was higher in all treatments as compared to the control. Maximum mortality seen on *Gliocladium virens* treatment (77%) as compared to Neem oil + *Gliocladium virens* (74%).

Table 36 : Effect of *Gliocladium virens* and Neem oil spray on *Parthenium hysterophorus* seedlings (blotter paper).

Treatment	No. of inoculated seedling	No. of dead seedling	No. of Healthy seedling
<i>Gliocladium virens</i>	100	77	33
Neem oil	100	53	47
Oil + <i>Gliocladium virens</i>	100	74	26
Control	100	-	100

Effect of Culture, Culture filtrate of *Gliocladium virens*, Thiophen and Neem oil on *Parthenium hysterophorus* seed germination .

The data on the influence of Marigold root extract (thiophen) on seed germination of *P. hysterophorus* (Table 37) revealed that there was maximum inhibition of seed germination (86.04%) on blotter soaked with thiophene (1%) + *Gliocladium virens* (culture filtrate) followed by neem oil + culture filtrate (74.41%). It was evident that culture filtrate plays a major role in inhibiting germination with thiophen.

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Table 37 : Effect of Culture, Culture filtrate of *Gliocladium virens*, Thiophen and Neem oil on *Parthenium hysterophorus* seed germination (Blotter paper)

S. No.	Treatments	No. of inoculated seed	No. of germinated seed	% inhibition of germination
1.	Control	100	43	
2.	Thiophen 1%	100	25	41.86
3.	Neem oil	100	16	62.79
4.	Culture filtrate (<i>Gliocladium virens</i>)	100	14	67.44
5.	Culture (<i>Gliocladium virens</i>)	100	26	39.53
6.	Culture filtrate + Thiophen	100	06	86.04
7.	Culture+Thiophen	100	28	34.88
8.	Neem oil +Culture filtrate	100	11	74.41
9.	Neem oil +Culture	100	14	67.44

Host Range Studies

Pathogenicity of *Fusarium pallidoroseum* on different vegetable crops

Twenty vegetable crops, as indicated Table 38, were tested for their susceptibility to *F. pallidoroseum*. It is clear from the Table that all the crops remained healthy (free from infection) and indicates that this fungus is safe to use against *P. hysterophorus* when applied in spray form.

Table 38: Effect of *Fusarium pallidoroseum* spray on different vegetable crops

Sl No	Crop	Germinated	Death of plant
1	Carrot	20	0
2	Radish	18	0
3	Turnip	17	0
4	Cabbage	15	0
5	Cauliflower	16	0
6	Ladyfinger	22	0
7	Coriander	21	0
8	Palak	20	0
9	Methi	24	0
10	Pea	17	0
11	Tomato	16	0
12	Brinjal	19	0
13	Onion	21	0
14	Ridgegourd	8	0
15	Cowpea	15	0
16	Chilli	18	0
17	Cucumber	7	0
18	Bottle gourd	6	0
19	Bean	13	0
20	Pumpkin	7	0

Effect of *Sclerotium rolfsii* as a pathogen on different crops.

Ten crops were tested for their susceptibility to *S. rolfsii* by applying a spray to seeds (Table 39). All showed some degree of inhibition, ranging from 25-42 % in the most susceptible crops (wheat, mung, lentil).

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Table 39 : Effect of *Sclerotium rolfsii* spray on germination of different crops seed .

Sl No.	Crop	Treated seeds	control (untreated)	% inhibition
1	Maize	14.66	16.66	12.00
2	Chickpea	19.00	22.00	13.63
3	Wheat	13.00	20.66	37.10
4	Urad	14.66	19.66	25.42
5	Moong	11.33	19.33	42.10
6	Lentil	13.00	18.33	29.08
7	Linseed	22.00	21.33	4.68
8	Pea	23.33	23.66	1.40
9	Mustard	16.00	22.00	18.18
10	Arhar	15.00	20.00	25.00

Testing Efficacy of Selected Pathogens as Weed Control Agents

Effect of *Fusarium pallidroseum* on seedling mortality of *Parthenium hysterophorus* in pots.

In pot experiments, seed inoculation, soil inoculation and direct spray to the seed caused seed rot and seedling mortality (Table 40). Maximum reduction in seed germination (56%) was obtained by soil inoculation method as compared to spray and seed inoculation (45 and 37%) respectively. But all the methods could significantly reduce seed germination as compared to the control and thus *F. pallidroseum* has some potential as a bio-control agent for *P. hysterophorus*.

Table 40: Effect of different methods of inoculation of *Fusarium pallidroseum* on seed germination and seedling mortality of *Parthenium hysterophorus* (100 seed sown).

Sl No	Treatment	Treated no. of seed germination	Control no of seed germination	% reduction in seed germination	% seedling died
1	Spray on seeds	17	31	45	29
2	Seed inoculation by mixing	15	24	37	20
3	Soil inoculation	16	37	56	25

Effect of *Fusarium pallidroseum* on *Parthenium hysterophorus*.

Data presented in Table 41 shows the effect of two sprays of *Fusarium pallidroseum*. Culture filtrate at the six leaf stage kills *P. hysterophorus* plants within four days, and culture sprays at the six leaf stage kill *P. hysterophorus* plants within six days.

Table 41 : Effect of *Fusarium pallidroseum* culture filtrate on *Parthenium hysterophorus*

Sl. No.	Treatments	No. of inoculated plants	No. of leaf/ plant	Symptoms appeared DAS	No. of wilted leaf/plant
1	Culture + water (1 spray)	16	6	6	2
2	Culture + water (2 spray)	16	6	CPW	CPW
3	Culture filtrate (1 spray)	16	6	4	3
4	Culture filtrate (2 spray)	16	6	CPW	CPW

CPW - Complete plant wilt

Effect of *Gliocladium virens* and Neem oil on different growth stage of *Parthenium hysterophorus*

One spray of *G. virens* with Neem oil at the 4 leaf stage kill *P. hysterophorus* within a week. At the 6 leaf stage 2 sprays are required to kill *P. hysterophorus* within 10 days. At the flowering stage, 3 sprays are required to kill *P. hysterophorus* within 15 days and after flowering 3 sprays can kill *P. hysterophorus* within 15 days. The total mortality in all treatments was 100%. (Table 42).

Outputs

Table 42 : Effect of *Gliocladium virens* and Neem oil on different growth stage of *Parthenium hysterophorus*.

S. No.	Stage of plant	No. of inoculated plants	No. of spray required to kill plant	Symptoms appeared days after	No. of days required for complete wilt	Per cent mortality
1.	4 leaf	16	1	3	8	100
2.	6 leaf	16	2	3	10	100
3.	Before flowering	16	3	8	15	100
4.	After flowering	16	3	20	15	100
5.	Control	16	-	-	-	-

Effect of spray of culture and culture filtrate of *Gliocladium virens* and Neem oil on *Parthenium hysterophorus*

Spray of *G. virens* culture filtrate mixed with Neem oil (5%) kills *P. hysterophorus* plants and *G. virens* culture mixed with Neem oil (5%) kills within seven days and Neem oil also reduces no. of flowers of *Parthenium* weed (Table 43).

Table 43 : Effect of spray of culture and culture filtrate of *Gliocladium virens* and Neem oil on *Parthenium hysterophorus*.

S. No.	Treatment (Spray)	No. of inoculated plants	No. of leaf/plant	No. of wilted leaf/plant	Symptoms appeared days after
1.	Culture spray	16	8	4	7
2.	Culture filtrate spray	16	6	16	4
3.	Control	16	8	-	-

Field Experiment

Effect of spray of *Fusarium pallidoroseum* at different days after sowing of *Parthenium hysterophorus*.

The experiment was conducted with 7 treatments, the results in figure 8, show that spraying 100 gm culture/l. of water could reduce plant height, number of branches and number of flowers/plant. Maximum reduction in height (15.78%) followed spraying 21 days after sowing. Maximum number of branches/plant was observed with a spray 15 days after sowing. Maximum number of branches/plant was observed with a spray 15 days after sowing and maximum reduction in number of flowers/plant (22.67 %) followed a spray 30 days after sowing, as compared to the control. *Fusarium* was sprayed @ 200 gm/l. of water the results are given in Figure 9. All the treatment reduced (8-75 DAS) height of plant, number of branches and number of flowers/plant as compared to the untreated control. Spraying 8-30 days after sowing reduced height of plant, no. of branches/plant, no. of flowers/plant significantly as compared to sprays applied 60 and 75 days after sowing.

Outputs

Figure 8. Effect of spray of *Fusarium pallidoroseum* at different days sowing of *Parthenium hysterophorus*

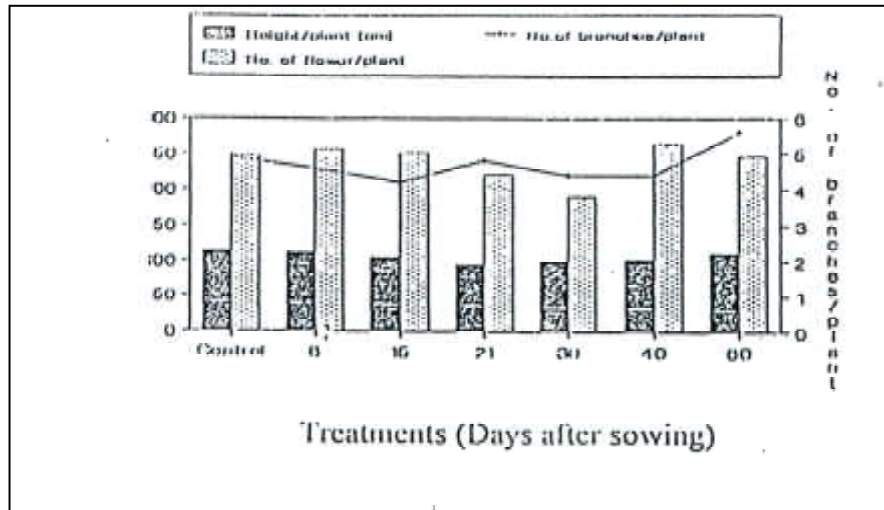
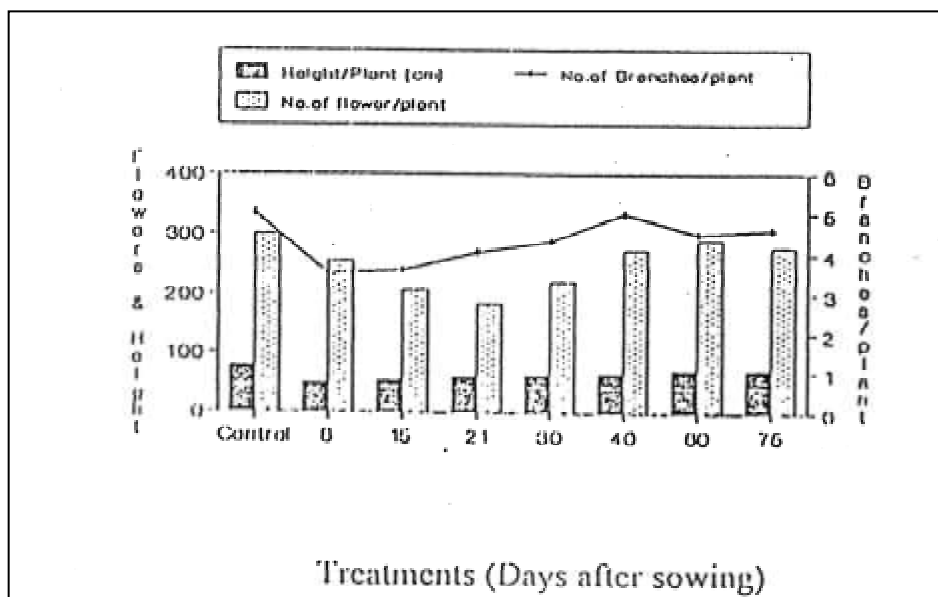


Fig. 9 Effect of spray of *Fusarium pallidroseum* at different days after sowing of *Parthenium hysterophorus*



Outputs

Effect of different amount of inoculum of *Fusarium pallidoroeseum* applied at different growth stages of *Parthenium hysterophorus*.

To evaluate the effect of spraying different amounts of inoculum of *F. pallidoroeseum* at the vegetative, pre-flowering and post-flowering stages. It is revealed from the data recorded in Table 44, that spraying 100, 150 & 200 gm wet culture/litre of water at the vegetative stage and before flowering could reduce plant height, no. of branches and flowers/plant but maximum reduction in these parameters was obtained when *Fusarium* was sprayed at 150-200 g/l of water at the vegetative and pre-flowering stages. Spraying after flowering killed both flowers and seeds.

Table 44 : Effect of different amount of inoculum of *Fusarium pallidoroeseum* at different growth stage of *Parthenium hysterophorus*.

Treatment	Height/ Plant (cm)	No. of Branch/ Plant	No. of Flowers/ Plant
VEGETATIVE STAGE			
100 gm Culture/l	65.33	4.40	167.33
150 gm Culture/l	63.33	4.46	149.66
200 gm Culture/l	57.93	3.60	138.00
Control	67.70	4.66	204.33
BEFORE FLOWERING STAGE			
100 gm Culture/l	50.86	3.60	169.66
150 gm Culture/l	57.13	3.67	165.00
200 gm Culture/l	61.06	3.53	188.00
Control	63.13	4.20	198.66
AFTER FLOWERING STAGE			
100 gm Culture/l	65.13	3.73	212.00
150 gm Culture/l	56.13	3.16	191.33
200 gm Culture/l	54.20	3.46	162.00
Control	67.60	4.06	250.00

Effect of methods of application of *Fusarium pallidoroeseum* on the germination of *Parthenium hysterophorus* seeds.

It is clear from Figure 10 that all three methods could reduce the germination of *P. hysterophorus* seeds as well as its growth. Different methods performed best at different months. Seed and soil treatment resulted best during June and January. While effect of spray was best in July, September, November and December. Highest reduction in seed germination by all methods was obtained during August and October.

Effect of spray of fungal suspension of *Sclerotium rolfsii* for the control of *Parthenium hysterophorus*.

In the present investigation, the effect of *S. rolfsii* on different growth stages of *P. hysterophorus* was determined. Table 45 shows that the spray of the fungus from 0 to 75 DAS could reduce plant height, number of branches/plant and no. of flowers/plant. Maximum reduction in height, number of branches/plant and no. of flowers/plant was obtained after spraying 0-30 DAS.

Outputs

Table 45 : Effect of spray of fungal suspension of *Sclerotium rolfsii* for the control of *Parthenium hysterophorus* .

Treatment	Percent Germination	Height/ Plant (cm)	Branching/ Plant	No. of Flowers/ Plant
0 DAS	68.53	54.16	4.13	269.27
8 DAS	57.33	49.13	3.20	257.30
15 DAS	54.00	55.20	3.60	209.50
21 DAS	60.33	60.93	4.06	185.06
30 DAS	52.66	60.83	3.73	220.60
40 DAS	63.66	67.13	5.00	273.60
60 DAS	77.26	72.33	4.50	263.00
75 DAS	72.53	72.93	4.80	277.46
CONTROL	89.33	78.80	5.00	299.46
CD at 5%	-	7.37	1.04	33.26

Efficacy of different methods of application of *Sclerotium rolfsii* on the germination of *Parthenium hysterophorus*

Treatment of seed and soil and spraying of plants reduced seed and plant growth (Table 46). Soil and spray treatment with *S. rolfsii* gave the best results were obtained in December.

Table 46: Efficacy of different methods of application of *Sclerotium rolfsii* on the germination (%) of *Parthenium hysterophorus*

Month	Seed Treatment	Soil Treatment	Spray Treatment	Control
July	48.33	30.66	65.66	61.00
August	42.66	18.00	62.66	70.66
September	49.66	27.66	81.33	72.66
October	38.00	18.33	54.33	43.00
November	21.66	32.33	25.66	22.33
December	21.66	12.00	12.00	26.66
January	22.66	22.33	13.00	26.66
C.D.	18.84	13.52	21.91	25.99

Effect of spraying of *Sclerotinia sclerotiorum* mycelial suspension for the control of *Parthenium hysterophorus* .

In the present studies it was observed that spraying of a mycelial suspension from 0 DAS to 15 DAS, could cause maximum reduction in height/plant, number of branches/plant and number of flower/plant (Fig. 11).

Outputs

Figure 10. Effect of methods of application of *Fusarium pallidoroseum* on germination of *Parthenium hysterophorus* seeds

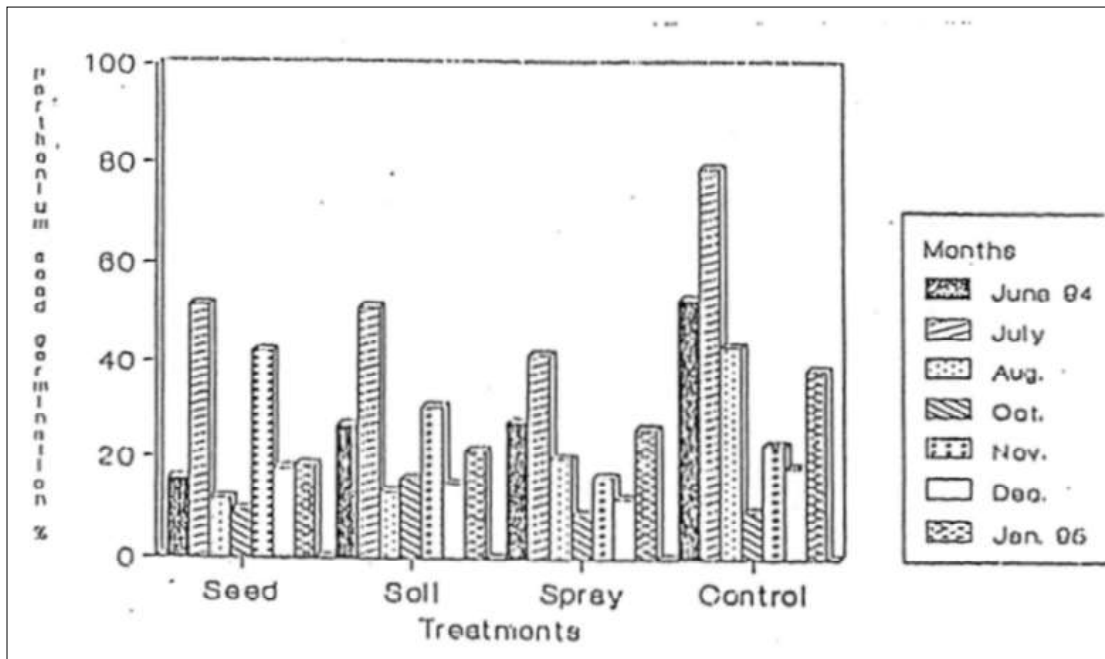
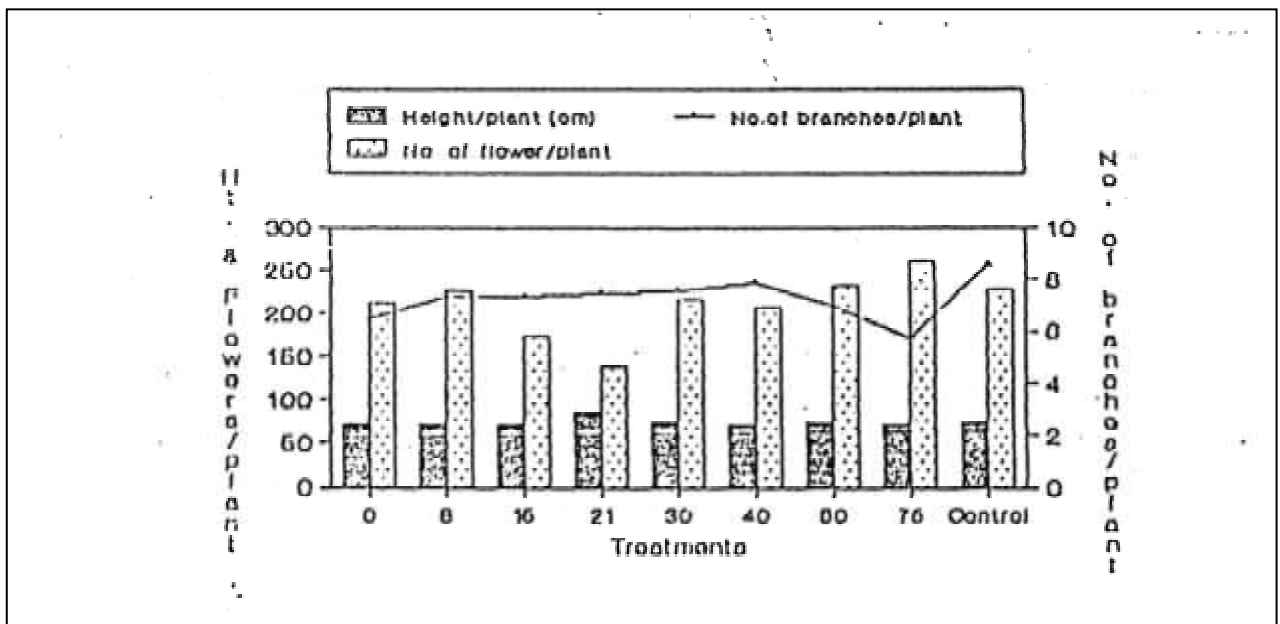


Figure 11. Effect of spray of *Sclerotinia sclerotiorum* mycelial and spore suspension on *Parthenium hysterophorus*



Outputs

Effect of *Trichoderma viride* culture and spore spray to control *Parthenium hysterophorus*

To establish the critical stage of *P. hysterophorus* at which *T. viride* can kill the plant, 200 g wet culture was macerated in a waring blender in 1 litre of sterilized water. The fungal suspension was sprayed on the soil in 3x2 sq.m. area at intervals of 0, 8, 15, 21, 30, 40, 60 & 75 DAS, in a randomised block design with three replications. Observations were recorded at plant maturity i.e., plant height, number of branches and flowers.

From these studies it was observed (Table 47) that spraying the fungal suspension from 0 DAS to 75 DAS could reduce plant height, number of branches/plant and number of flowers/plant. Maximum reduction in height, number of branches/plants and number of flower/plant was obtained in plots sprayed from 0-30 DAS and much less at 60-75 DAS.

T. viride fungus has a better inhibitory effect on *P. hysterophorus* at the early stages of plant growth.

Table 47 : Effect of *Trichoderma viride* culture and spore suspension spray to control of *Parthenium hysterophorus* .

Treatment	Percent Germination	Height/ Plant (cm)	No. of Branches/ Plant	No. of Flowers /Plant
0 DAS	41.73	61.33	5.33	244.53
8 DAS	47.33	58.06	4.40	147.86
15 DAS	53.13	61.66	3.66	107.20
21 DAS	46.20	54.53	2.73	153.00
30 DAS	51.20	57.76	3.20	127.46
40 DAS	50.93	60.20	4.60	171.90
60 DAS	45.40	60.60	4.66	217.73
75 DAS	49.93	59.40	4.60	229.86
CONTROL	95.60	81.86	5.06	328.30
CD at 5%	-	10.50	1.12	58.10

Effect of Neem oil cake, cowdung and *Trichoderma viride* on recovery of *Parthenium hysterophorus*

In a field experiment, Neem oil cake, cowdung and *T. viride* were added to the soil and recovery of *P. hysterophorus*, number of branches per plant, plant height and number of flowers/plant were recorded. Data presented in Table 48 show that the recovery of *P. hysterophorus* in all treatment was similar to the control.

Table 48: Effect of Neem oil cake, cowdung and *Trichoderma viride* on recovery of *Parthenium hysterophorus*

Treatments	Recovery of Parthenium	No. of branches/ plant	Plant height (cm)	No. of flowers/ plant
2N:2C:1T 250 g	171.33	5.90	67.40	191.13
2N:2C:1T 500 g	219.66	5.96	69.00	198.80
2N:2C:1T 750 g	162.66	6.03	69.13	180.86
2N:2C:1T 1000 g	168.00	5.63	67.13	156.90
Control	208.66	6.26	71.66	255.90
SEm±	32.94	0.60	5.26	19.73
CD 5%	107.41	1.96	12.12	45.49

N - Neem oil cake C - Cowdung T - Trichoderma

Outputs

Competition between weeds and different crops

Effect of Marigold population on the growth and survival of *Parthenium hysterophorus*

In order to find out the number of marigold plants required to suppress 50 *P. hysterophorus* plants, a field experiment was carried out. The *P. hysterophorus* and marigold plants were grown in eight combinations i.e. 50:25 M, 50:50, 50:75, 50:100, 50:125, 50:150, 50:175, 50:200.

It was observed that *P. hysterophorus* plants were completely suppressed by marigold plants, and these were weak and fragile. This would appear to be a very cheap and effective method for the management of Parthenium weed in non-cultivated areas (Table 49).

Table 49 : Effect of Marigold population on the growth and survival of *Parthenium hysterophorus*

Treatment	Plant height (cm)	No. of branch/plant	No. of flowers/plant
50 P + 25 M	108.33	3.86	668.33
50 P + 50 M	102.40	3.00	392.73
50 P + 75 M	109.46	2.80	314.53
50 P + 100 M	106.00	1.86	319.06
50 P + 125 M	91.60	2.13	162.86
50 P + 150 M	103.20	2.60	246.26
50 P + 175 M	101.73	2.26	238.66
50 P + 200 M	93.46	1.80	177.33
Control	125.06	5.60	985.66
CD 5%	18.46	-	276.32

Effect of Sunnhemp plant population on *Parthenium hysterophorus*

To find out the number of Sunnhemp plant required to suppress 50 *P. hysterophorus* plants, a field experiment was carried out where both the plants were grown in different ratios i.e. 50 *P. hysterophorus*: 25 Sunnhemp, 50:50, 50:75, 50:100, 50:125, 50: 175, 50:200. The Sunnhemp crop inhibited seed germination, plant height and no. of flowers/plant (Table 50).

Table 50 : Effect of Sunnhemp plant population on *Parthenium hysterophorus*

Treatment	No. of Germinated Plants	Plant height(cm)	No. of branches/plant	No. of flowers/plant
50 P + 25 S	48.67	75.73	2.33	213.07
50 P + 50 S	36.00	69.67	1.80	133.87
50 P + 75 S	33.67	69.13	1.73	108.00
50 P + 100 S	30.33	76.88	2.07	135.13
50 P + 125 S	30.67	81.47	2.07	144.47
50 P + 150 S	31.67	75.13	1.99	84.27
50 P + 175 S	24.00	64.80	2.13	110.80
50 P + 200 S	24.33	65.20	2.53	121.10
control	50.00	98.44	4.40	660.87
CD 5%	11.17	16.91	1.39	251.66

Host Specificity testing

Host specificity testing of *S. rolfii* on different vegetable crops

Four vegetable crops were tested for their susceptibility to *S. rolfii* in the plots where the fungus was previously inoculated for control of Parthenium weed. During the year 1996-97 the fungus was used for control of *P. hysterophorus* and in the 1997-1998 season, crop plants were grown in these plots. Crop inhibition ranged from 5-35% (Table 51), showing that there was residual fungal activity which could affect following cultivation.

Outputs

Table 51 : Host specificity testing of *Sclerotium rolfsii* on different vegetable crops 1998-99.

S. No	Test crop	No. of germination <i>S. rolfsii</i>	No. of germination control	% inhibition of germination <i>S. rolfsii</i>
1.	Cowpea	63.32	56.00	-
2.	Tomato	11.32	13.32	15.01
3.	Guar	56.00	50.66	-
4.	Lady finger	34.00	36.00	5.55
5.	Chilli	14.66	22.66	35.30

Host specificity testing of *Sclerotium rolfsii* on different field crops

Five field crops (maize, rice, soybean, moong and jowar) were tested for their susceptibility to *S. rolfsii*. During the year 1996-97, the fungus was used for control of *P. hysterophorus*, in the following season the crops were sown in these test plots. No inhibition of germination was observed in the maize and moong crops. Inhibition of germination of other crops was in the range of 5-36% (Table 52), demonstrating again the ability of the pathogen to survive in the soil and pose a danger to crop species.

Table 52: Host specificity testing of *Sclerotium rolfsii* on different field crops 1998-99

S. No	Test crop	No. of germinating <i>S. rolfsii</i>	No. of germinating control	% inhibition of <i>S. rolfsii</i> germination
1.	Maize	53.66	50.00	-
2.	Rice	35.00	44.16	20.74
3.	Soybean	44.33	55.00	19.40
4.	Moong	70.00	65.66	-
5.	Jowar	21.10	33.33	36.69
6.	Maize	52.21	50.00	-
7.	Rice	30.33	44.16	31.31
8.	Soybean	47.33	55.00	13.94
9.	Moong	62.33	65.66	5.07
10.	Jowar	25.55	33.33	23.34

Host specificity testing of *Sclerotinia sclerotiorum* on different vegetable crops

Five vegetable crops (cowpea, tomato, guar, lady's finger, chilli) were tested for their susceptibility to *S. sclerotiorum*. During the year 1996-97, the fungus was used for control of *P. hysterophorus*, and the following year the crops were planted. No inhibition of germination was obtained in tomato crop but inhibition of germination of other crops ranged from 5-24% (Table 53).

Table 53: Host specificity testing of *Sclerotinia sclerotiorum* on different vegetable crops 1998-99.

S. No. No.	Test crop	No. of germinating <i>S. sclerotiorum</i>	No. of germinating controls	% inhibition of <i>S. sclerotiorum</i> germination
1.	Cowpea	56.00	63.32	11.56
2.	Tomato	14.00	13.32	-
3.	Guar	44.00	50.66	13.14
4.	Lady finger	27.32	36.00	24.11
5.	Chilli	21.32	22.66	5.91

Outputs

Host specificity testing of *Sclerotinia sclerotiorum* on different field crops

Five field crops (maize, rice, soybean, moong and jowar) were tested for their susceptibility to *S. sclerotiorum*, using the techniques described above. No inhibition of germination was obtained in the moong crop, but there was inhibition of germination of other crops, ranging from 6-33% (Table 54).

Table 54: Host specificity testing of *Sclerotinia sclerotiorum* on different field crops 1998-99

Sl. No.	Test crop	No. of germinating <i>Sclerotinia sclerotiorum</i>	No. of germinating control	% inhibition of <i>Sclerotinia sclerotiorum</i> germination
1	Maize	41.66	50.00	16.68
2	Rice	30.00	44.16	32.06
3	Soybean	52.60	55.00	4.25
4	Moong	62.66	65.66	4.56
5	Jowar	27.76	33.33	16.71
6	Maize	41.66	50.00	16.68
7	Rice	29.33	44.16	33.58
8	Soybean	51.33	55.00	6.67
9	Moong	67.33	65.66	-
10	Jowar	28.33	33.33	15.00

Outputs

3. Biocontrol agent(s) identified and screened for release in India

Tamil Nadu Agricultural University, Coimbatore, India

Effects of biocontrol agents on *Parthenium hysterophorus* plants

Foliar pathogens

Koch's postulate of all the microorganisms isolated from diseased plant parts were proved under laboratory and glasshouse conditions. Among these, *Lasiodiplodia theobromae* was found to be highly pathogenic to this weed by recording maximum plant mortality 30 days after spray, followed by *Oidium parthenii* and *Fusarium pallidoroseum* (Table 55).

Table 55 : Pathogenicity of foliar isolates associated with *Parthenium hysterophorus* under *in vitro* conditions

S.N o.	Treatment	initial symptom (Days)	Disease incidence* at 15 days after spray			Disease incidence* at 30 days after spray				
			PDI (%)	% Leaf infection	% Twig infection	% Plant mortality	PDI %	% Leaf infection	% Twig infection	% Plant mortality
1.	<i>Alternaria Alternata</i>	3	31.85 ^h (34.36)	51.00 ^{cd} (45.57)	34.77 ^{de} (36.12)	0.00 ^f (9.10)	36.29 ^g (37.04)	53.33 ^{cd} (46.91)	48.67 ^f (44.24)	0.00 ^f (9.10)
2.	<i>A. zinniae</i>	3	19.25 ^j (26.02)	35.43 ^{de} (36.51)	3.67 ⁱ (11.03)	0.00 ^f (9.10)	20.77 ^j (27.11)	37.50 ^{de} (37.76)	15.76 ^h (23.39)	0.00 ^f (9.10)
3.	<i>Colletotrichum Dematium</i>	3	27.03 ⁱ (31.32)	36.11 ^{cd} (36.93)	0.00 ^j (9.10)	0.00 ^f (9.10)	29.62 ⁱ (32.97)	40.28 ^{cde} (39.39)	0.00 ^j (5.74)	0.00 ^f (9.10)
4.	<i>Curvularia lunata</i>	4	4.44 ^o (12.10)	11.11 ^t (11.75)	0.00 ^j (9.10)	0.00 ^f (9.10)	6.29 ^o (14.51)	11.11 ^t (11.75)	0.00 ^j (5.74)	0.00 ^f (9.10)
5.	<i>C. pallescens</i>	4	16.66 ^k (24.09)	37.50 ^{de} (36.10)	6.33 ^j (14.56)	0.00 ^f (9.10)	19.25 ^k (26.02)	37.53 ^{de} (37.78)	7.67 ⁱ (16.07)	0.00 ^f (9.10)
6.	<i>C. verruculosa</i>	4	14.81 ⁱ (22.63)	34.72 ^{de} (36.10)	0.00 ^j (9.10)	0.00 ^f (9.10)	16.29 ⁱ (23.80)	34.72 ^{de} (36.09)	0.00 ^j (5.74)	0.00 ^f (9.10)
7.	<i>Drechslera Australiensis</i>	3	28.14 ⁱ (32.04)	40.98 ^{cde} (39.80)	24.93 ^g (29.95)	0.00 ^f (9.10)	32.59 ^h (34.81)	43.06 ^{cde} (41.00)	46.49 ^g (42.98)	0.00 ^f (9.10)
8.	<i>D. hawaiiensis</i>	3	68.14 ^d (55.64)	100.00 ^a (83.58)	40.96 ^c (39.79)	5.00 ^d (12.92)	71.81 ^c (57.93)	100.00 ^a (85.44)	82.00 ^b (64.90)	16.07 ^c (23.65)
9.	<i>Fusarium equiseti</i>	3	54.81 ^g (47.76)	82.00 ^b (64.93)	30.77 ^f (33.69)	3.33 ^e (8.62)	57.40 ^f (49.26)	84.05 ^b (66.46)	50.61 ^e (45.35)	5.00 ^e (2.93)
10.	<i>F. moniliforme</i>	3	59.62 ^f (50.55)	86.47 ^b (68.45)	34.20 ^e (35.79)	6.67 ^d (14.75)	62.96 ^e (52.51)	87.85 ^b (69.62)	64.12 ^d (53.20)	8.33 ^{de} (16.60)
11.	<i>F. oxysporum</i>	3	61.48 ^e (51.64)	90.73 ^b (72.44)	36.18 ^d (36.98)	8.33 ^d (16.59)	67.03 ^d (54.97)	92.83 ^b (74.53)	80.54 ^c (63.82)	10.00 ^d (18.44)
12.	<i>F. pallidoroseum</i>	3	80.70 ^c (66.19)	100.00 ^a (83.58)	54.46 ^b (47.58)	50.00 ^c (45.00)	87.03 ^b (68.90)	100.00 ^a (85.44)	82.00 ^b (64.90)	66.67 ^b (54.78)
13.	<i>F. solani</i>	3	53.70 ^g (47.12)	57.17 ^c (49.12)	30.72 ^f (33.66)	8.33 ^d (16.60)	57.40 ^f (49.26)	58.34 ^c (49.80)	51.70 ^e (45.98)	10.00 ^d (18.44)
14.	<i>Oidium parthenii</i>	3	86.59 ^a (68.53)	100.00 ^a (83.58)	100.00 ^a (80.90)	54.00 ^b (47.29)	99.99 ^a (89.43)	100.00 ^a (85.44)	100.00 ^a (80.90)	68.00 ^b (55.55)
15.	<i>Phoma sorghina</i>	5	11.85 ^m (20.13)	11.11 ^t (11.75)	0.00 ^j (9.10)	0.00 ^f (9.10)	12.22 ⁿ (20.46)	13.33 ⁱ (21.42)	0.00 ^j (5.74)	0.00 ^f (9.10)
16.	<i>Phomopsis</i> sp.	5	12.96 ^m (21.10)	27.10 ^c (31.34)	0.00 ^j (9.10)	0.00 ^f (9.10)	13.33 ^m (21.42)	38.89 ^{def} (38.58)	0.00 ^j (5.74)	0.00 ^f (9.10)
17.	<i>Syncephalastrum Raceinosum</i>	5	5.55 ⁿ (13.58)	6.67 ^{fg} (8.86)	0.00 ^j (9.10)	0.00 ^f (9.10)	5.92 ^o (14.03)	7.67 ⁱ (16.07)	0.00 ^j (5.74)	0.00 ^f (9.10)
18.	<i>Lasiodiplodia Theobromae</i>	3	83.33 ^b (65.88)	99.00 ^a (89.43)	100.00 ^a (80.90)	83.67 ^a (63.01)	87.77 ^b (70.54)	100.00 ^a (85.44)	100.00 ^a (80.90)	87.00 ^a (68.87)
19.	Control	-	0.00 ^p (5.74)	0.00 ^g (2.63)	0.00 ^j (9.10)	0.00 ^f (9.10)	0.00 ^p (5.74)	0.00 ^g (2.43)	0.00 ^j (5.74)	0.00 ^f (9.10)

* Mean of three replications. (Data in parentheses are arcsine transformed values. In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Outputs

Root pathogens

Application of sand/maize inoculum of test pathogens viz., *M. phaseolina*, *R. solani* and *S. rolfisii* @ 5 per cent level individually into the sterilized soil prior to sowing of *P. hysterophorus* seeds, resulted in 100 % inhibition of seedling emergence. The same organisms applied to the root zone of 30 day-old seedlings resulted in complete plant mortality 15 days after application (Table 56).

Table 56 : Pathogenicity of wilt/root rot pathogens

S.No.	Pathogens	15 days after inoculation*	
		Seedling emergence (%)	Wilt incidence (%)
1.	<i>Macrophomina phaseolina</i>	0.00	100.00
2.	<i>Rhizoctonia solani</i>	0.00	100.00
3.	<i>Sclerotium rolfisii</i>	0.00	100.00
4.	Control	80.00	-

* Mean of four replications

Effect of test pathogens on seed germination

Parthenium hysterophorus seeds soaked individually in cultures of *D. hawaiiensis*, *F. moniliforme*, *F. oxysporum*, *F. pallidoroseum*, *M. phaseolina*, *S. rolfisii*, *R. solani* and *L. theobromae* and a spore suspension of *O. parthenii* for one hr, failed to germinate (100 % inhibition) under *in vitro* conditions (Table 57).

Table 57 : Effect of test pathogens on *Parthenium hysterophorus* seed germination

S.No.	Treatment	%Seed* germination	%Reduction over control	Shoot length* (cm)	Reduction over control (%)	Root length* (cm)	Reduction over control (%)	Vigour Index*	Reduction over control (%)
1.	<i>Alternaria alternata</i>	4.00 ^b (11.54)	94.87	1.35 ^b	73.79	0.63 ^c	85.11	9.00 ^b	98.78
2.	<i>A. zinniae</i>	6.00 ^c (14.18)	92.31	1.85 ^d	64.08	0.80 ^d	81.09	17.00 ^c	97.70
3.	<i>Colletotrichum dematium</i>	10.00 ^d (18.43)	87.18	2.00 ^e	61.17	1.03 ^e	75.65	32.00 ^d	95.68
4.	<i>Curvularia lunata</i>	21.00 ^e (27.27)	73.08	2.65 ^g	48.54	1.33 ^f	68.56	85.00 ^e	88.51
5.	<i>C. pallescens</i>	50.00 ^h (45.00)	35.90	4.65 ^j	9.71	2.23 ^h	47.28	344.00 ^h	53.51
6.	<i>C. verruculosa</i>	48.00 ^g (43.85)	38.46	4.48 ⁱ	13.01	1.88 ^g	55.56	308.00 ^g	58.38
7.	<i>Drechslera australiensis</i>	6.00 ^c (14.18)	92.31	2.08 ⁱ	59.61	1.00 ^e	76.36	20.00 ^c	97.30
8.	<i>D. hawaiiensis</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
9.	<i>Fusarium equiseti</i>	25.00 ⁱ (30.00)	67.95	4.05 ^h	21.36	1.85 ^g	56.26	149.00 ⁱ	79.86
10.	<i>F. moniliforme</i>	0.00 ^a (2.87)	100.00	0.00 ^h	100.00	0.00 ^a	100.00	0.00 ^a	100.00
11.	<i>F. oxysporum</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
12.	<i>F. pallidoroseum</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
13.	<i>F. solani</i>	4.00 ^b (11.54)	94.87	1.43 ^c	72.23	0.50 ^b	88.18	8.00 ^b	98.92
14.	<i>Macrophomina phaseolina</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
15.	<i>Oidium parthenii</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
16.	<i>Phoma sorghina</i>	67.00 ^j (54.94)	14.10	5.10 ^{kl}	0.90	3.84 ⁱ	9.22	599.00 ^j	19.05
17.	<i>Phomopsis</i> sp.	65.00 ⁱ (53.73)	16.67	5.08 ^k	1.36	3.83 ⁱ	9.46	588.00 ⁱ	20.54
18.	<i>Rhizoctonia solani</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
19.	<i>Sclerotium rolfisii</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00

Outputs

S.No	Treatment	%Seed* germination	%Reduction over control	Shoot length* (cm)	Reduction over control (%)	Root length* (cm)	Reduction over control (%)	Vigour Index*	Reduction over control (%)
20.	<i>Syncephalastrum raceinosum</i>	72.00 ^k (58.05)	7.69	5.10 ^{kl}	0.90	4.20 ^j	0.70	670.00 ^k	9.50
21.	<i>Lasiodiplodia theobromae</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
22.	Control	78.00 ^l (62.03)	-	5.15 ^l	-	4.23 ^j	-	740.00 ^l	-

* Mean of four replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Efficacy of culture filtrates of test pathogens on *Parthenium hysterophorus* seed/plants

Cell free culture filtrates of *D. hawaiiensis*, *F. moniliforme*, *F. oxysporum*, *F. pallidoroseum*, *M. phaseolina*, *R. solani*, *S. rolfsii* and *L. theobromae* completely inhibited seed germination when they were individually used to soak *P. hysterophorus* seeds, one hr prior to germination test (Table 58).

Table 58 : Effect of culture filtrates of test pathogens on *Parthenium hysterophorus* seed germination

S.No	Treatment	Seed* germination (%)	Reduction over control (%)	Shoot length* (cm)	Reduction over control (%)	Root length* (cm)	Reduction over control (%)	Vigour Index*	Reduction over control (%)
1.	<i>Alternaria alternata</i>	4.75 ^b (12.66)	93.97	1.50 ^b	71.04	0.68 ^b	84.88	10.00 ^b	98.65
2.	<i>A. zinniae</i>	7.25 ^d (15.68)	90.79	2.13 ^c	58.88	0.88 ^c	80.44	22.00 ^c	97.04
3.	<i>Colletotrichum dematium</i>	10.25 ^e (18.72)	86.98	2.20 ^c	57.53	1.23 ^d	72.66	35.00 ^d	95.28
4.	<i>Curvularia lunata</i>	21.75 ^f (27.83)	72.38	2.80 ^e	45.95	1.50 ^e	66.67	93.00 ^e	87.47
5.	<i>C. pallidescens</i>	50.50 ⁱ (45.29)	35.87	4.90 ^h	5.41	2.40 ^g	46.66	369.00 ^h	50.27
6.	<i>C. verruculosa</i>	48.75 ^h (44.31)	38.10	4.68 ^g	9.65	2.08 ^f	53.17	329.00 ^g	55.66
7.	<i>Drechslera australiensis</i>	6.75 ^c (15.12)	91.43	2.33 ^d	55.02	1.23 ^d	72.67	24.00 ^c	96.77
8.	<i>D. hawaiiensis</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
9.	<i>Fusarium equiseti</i>	25.75 ^g (30.53)	67.30	4.10 ^f	20.85	2.13 ^f	52.67	160.00 ^f	78.44
10.	<i>F. moniliforme</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
11.	<i>F. oxysporum</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
12.	<i>F. pallidoroseum</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
13.	<i>F. solani</i>	4.00 ^b (11.54)	94.92	1.60 ^b	69.11	0.63 ^b	86.00	10.00 ^b	98.65
14.	<i>Macrophomina phaseolina</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
15.	<i>Phoma sorghina</i>	67.25 ^j (55.12)	14.60	5.15 ^j	0.50	4.15 ⁱ	7.78	625.00 ^j	15.77
16.	<i>Phomopsis</i> sp.	67.75 ^j (55.43)	13.97	5.03 ⁱ	2.90	3.25 ^h	27.78	564.00 ⁱ	23.99
17.	<i>Rhizoctonia solani</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
18.	<i>Sclerotium rolfsii</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
19.	<i>Syncephalastrum raceinosum</i>	74.00 ^k (59.34)	6.03	5.13 ^{ij}	0.90	4.08 ⁱ	9.33	681.00 ^k	8.22
20.	<i>Lasiodiplodia theobromae</i>	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
21.	Control	78.75 ^l (62.58)	-	5.18 ^j	-	4.50 ^j	-	742.00 ^l	-

* Mean of four replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Outputs

Culture filtrates of both foliar and root pathogens individually exhibited the characteristic symptoms of the disease, and damaged 30 day-old seedlings similar to the culture spray. Spraying of culture filtrates of *M. phaseolina* and *R. solani* on 30 day-old seedlings resulted in complete plant mortality after 15 days. The culture filtrates of *L. theobromae* and *F. pallidoroseum* also had a high virulence, with 86.00 and 63.33 % plant mortality after 30 days (Table 59).

Table 59 : Efficacy of culture filtrates of test pathogens on symptom expression

S.No.	Treatment	initial symptom (Days)	Symptom expression* at 15 days after spray				Symptom expression* at 30 days after spray			
			PDI (%)	Leaf infection (%)	Twig infection (%)	Plant mortality (%)	PDI (%)	Leaf infection (%)	Twig infection (%)	Plant mortality (%)
1.	<i>Alternaria alternata</i>	3	30.37 ^f (33.44)	52.09 ^{cde} (46.20)	34.72 ^{ef} (36.08)	6.67 ^{fg} (14.76)	35.55 ^f (36.60)	53.30 ^{de} (46.89)	49.39 ^f (44.65)	8.33 ^g (16.44)
2.	<i>A. zinniae</i>	3	18.14 ^g (25.21)	37.12 ^{def} (37.54)	5.00 ^j (12.93)	5.00 ^g (12.93)	20.40 ^g (26.85)	41.11 ^{ef} (39.80)	16.02 ^h (23.59)	6.67 ^h (14.89)
3.	<i>Colletotrichum dematium</i>	3	25.92 ^f (30.60)	40.62 ^{cdef} (39.60)	0.00 ^k (9.10)	0.00 ^h (9.10)	28.14 ^f (32.04)	39.66 ^{ef} (39.03)	0.00 ^j (5.74)	0.00 ⁱ (9.10)
4.	<i>Curvularia lunata</i>	4	2.96 ⁱ (5.77)	10.43 ^h (11.34)	0.00 ^k (9.10)	0.00 ^h (9.10)	3.30 ⁱ (6.11)	11.10 ^h (11.75)	0.00 ^j (5.74)	0.00 ⁱ (9.10)
5.	<i>C. pallescens</i>	4	15.92 ^{gh} (23.51)	33.30 ^{efg} (35.24)	6.33 ⁱ (14.57)	0.00 ^h (9.10)	16.44 ^{gh} (24.09)	37.20 ^f (37.58)	13.00 ⁱ (21.12)	0.00 ⁱ (9.10)
6.	<i>C. verruculosa</i>	4	14.07 ^{gh} (22.03)	32.63 ^{fg} (34.84)	0.00 ^k (9.10)	0.00 ^h (9.10)	14.44 ^{gh} (22.34)	35.59 ^f (36.62)	0.00 ^j (5.74)	0.00 ⁱ (9.10)
7.	<i>Drechslera australiensis</i>	3	27.03 ^f (31.32)	41.41 ^{cdef} (40.05)	25.75 ^h (30.48)	0.00 ^h (9.10)	31.85 ^f (34.36)	43.05 ^{ef} (41.01)	46.34 ^g (42.90)	0.00 ⁱ (9.10)
8.	<i>D. hawaiiensis</i>	3	67.40 ^c (55.18)	100.00 ^a (83.58)	41.06 ^d (39.85)	8.33 ^{ef} (16.60)	70.77 ^c (57.27)	100.00 ^a (85.44)	80.33 ^b (63.67)	10.00 ^f (18.43)
9.	<i>Fusarium equiseti</i>	3	52.59 ^{de} (46.49)	83.14 ^d (65.76)	30.87 ^g (33.75)	5.00 ^g (12.93)	55.55 ^e (48.19)	83.17 ^c (65.78)	51.10 ^e (45.62)	6.67 ^h (14.89)
10.	<i>F. moniliforme</i>	3	57.40 ^{de} (49.26)	88.04 ^b (69.77)	34.05 ^{ef} (35.70)	10.00 ^{de} (18.44)	61.85 ^{de} (51.86)	88.31 ^{bc} (70.01)	63.83 ^d (53.03)	11.67 ^e (20.00)
11.	<i>F. oxysporum</i>	3	60.77 ^{cd} (51.22)	92.09 ^b (73.72)	35.89 ^e (36.81)	11.67 ^d (19.89)	66.29 ^{cd} (54.51)	92.21 ^b (73.86)	79.89 ^b (63.36)	13.33 ^c (21.39)
12.	<i>F. pallidoroseum</i>	3	79.66 ^b (63.19)	100.00 ^a (83.58)	54.03 ^b (49.29)	50.00 ^c (45.00)	84.07 ^b (66.48)	100.00 ^a (85.44)	80.67 ^b (63.92)	63.33 ^c (52.71)
13.	<i>F. solani</i>	3	51.14 ^e (45.66)	57.42 ^c (49.27)	32.71 ^{fg} (34.88)	5.00 ^g (12.93)	56.66 ^e (48.83)	58.58 ^d (49.94)	51.16 ^e (45.66)	6.67 ^h (14.89)
14.	<i>Macrophomina phaseolina</i>	3	99.99 ^a (89.43)	100.00 ^a (83.58)	100.00 ^a (80.90)	100.00 ^a (80.90)	99.99 ^a (89.36)	100.00 ^a (85.44)	100.00 ^a (80.90)	100.00 ^a (80.90)
15.	<i>Phoma sorghina</i>	5	10.73 ^h (19.12)	19.41 ^g (26.13)	0.00 ^k (9.10)	0.00 ^h (9.10)	10.77 ^h (19.16)	19.67 ^g (26.31)	0.00 ^j (5.74)	0.00 ⁱ (9.10)
16.	<i>Phomopsis</i> sp.	5	11.48 ^{gh} (19.80)	20.00 ^g (26.57)	0.00 ^k (9.10)	0.00 ^h (9.10)	11.85 ^h (20.13)	20.00 ^g (26.57)	0.00 ^j (5.74)	0.00 ⁱ (9.10)
17.	<i>Rhizoctonia solani</i>	3	99.99 ^a (89.43)	100.00 ^a (83.58)	100.00 ^a (80.90)	100.00 ^a (80.90)	99.99 ^a (89.43)	100.00 ^a (85.44)	100.00 ^a (80.90)	100.00 ^a (80.90)
18.	<i>Sclerotium rolfsii</i>	4	32.20 ^f (34.58)	54.30 ^c (47.47)	51.00 ^c (45.51)	50.00 ^c (45.00)	37.03 ^f (37.48)	64.75 ^d (53.57)	73.46 ^c (58.99)	58.33 ^d (49.78)
19.	<i>Syncephalastrum raceinosum</i>	5	2.59 ⁱ (5.39)	10.43 ^h (11.34)	0.00 ^k (9.10)	0.00 ^h (9.10)	2.59 ⁱ (5.39)	10.43 ⁱ (11.34)	0.00 ^j (5.74)	0.00 ⁱ (9.10)
20.	<i>Lasiodiplodia theobromae</i>	3	83.00 ^b (65.65)	100.00 ^a (83.58)	100.00 ^a (80.90)	83.00 ^b (65.65)	86.67 ^b (68.61)	100.00 ^a (89.81)	100.00 ^a (80.90)	86.00 ^b (68.03)
21.	Control	-	0.00 ^j (5.74)	0.00 ⁱ (2.63)	0.00 ^k (9.10)	0.00 ^h (9.10)	0.00 ^j (5.74)	0.00 ⁱ (2.43)	0.00 ^j (5.74)	0.00 ⁱ (9.10)

* Mean of three replications. (Data in parentheses are arcsine transformed values).

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Outputs

Standardisation of culture media for the selected biocontrol agents

Richard's medium was found to support maximum growth and biomass production of *L. theobromae*. None of the other media induced sporulation. Coon's medium was found to be best for growth, biomass production and sporulation of *F. pallidroseum* (Table 60).

Table 60 : Standardization of suitable media for the growth, biomass production and sporulation of selected pathogenic isolates

S.No.	Treatment (medium)	<i>L. theobromae</i>			<i>F. pallidroseum</i> *		
		Mycelial growth ($\bar{\theta}$ mm)	Mycelial dry wt. (g)	Sporulation	Mycelial growth ($\bar{\theta}$ mm)	Mycelial dry wt. (g)	Sporulation
1.	Brown's	84.3 ^c	0.963 ^{bcdef}	-	41.3 ^g	0.837 ^k	+
2.	Coon's	80.3 ^d	0.860 ^{def}	-	89.0 ^a	1.867 ^a	+++++
3.	Czapek's	88.3 ^{ab}	1.997 ^a	-	89.0 ^a	1.517 ^d	++++
4.	Glucose nutrient	89.0 ^a	1.238 ^{bcd}	-	85.0 ^h	1.120 ⁱ	++
5.	Host extract	87.7 ^b	2.248 ^a	-	86.7 ^b	1.120 ⁱ	++++
6.	Host extract pectin	88.0 ^{ab}	1.227 ^{bcd}	-	87.7 ^{ab}	1.223 ^h	++++
7.	Lima bean	82.3 ^c	0.780 ^{ef}	-	72.0 ^f	0.663 ^m	+
8.	Malt extract	89.0 ^a	2.011 ^a	-	88.3 ^a	1.263 ^g	++
9.	Molasses yeast	21.3 ^h	0.463 ^g	-	26.0 ^j	0.123 ^p	++
10.	Potato dextrose	89.0 ^a	2.220 ^a	-	89.0 ^a	1.680 ^b	++++
11.	Potato sucrose	89.0 ^a	2.120 ^a	-	89.0 ^a	1.593 ^c	++++
12.	Richard's	89.0 ^a	2.345 ^a	-	88.7 ^a	1.360 ^f	++++
13.	Sabourauds dextrose	87.7 ^b	1.207 ^b	-	81.3 ^d	1.007 ^j	++
14.	Spezieller	38.0 ^e	0.673 ^{fg}	-	89.0 ^a	1.410 ^e	++++
15.	Starch	32.7 ^f	0.490 ^g	-	37.7 ^h	0.323 ⁿ	+
16.	V-8 juice	87.7 ^b	1.123 ^{bcd}	-	80.0 ^e	0.780 ^l	+
17.	Yeast extract	23.7 ^g	0.927 ^{bcdef}	-	31.0 ⁱ	0.213 ^o	+

* Mean of three replications.

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

+++++ : Very good ($> 40 \times 10^4$ conidia /ml)

++++ : Good ($30-40 \times 10^4$ conidia /ml)

+ : Very poor ($<10 \times 10^4$ conidia /ml) -

+++ : Medium ($20-30 \times 10^4$ conidia /ml)

++ : Poor ($10-20 \times 10^4$ conidia /ml)

- : No sporulation

Virulence of selected biocontrol agents grown on different culture media

On seed germination

Parthenium hysterophorus seeds soaked for one hr in a culture or culture filtrate of *L. theobromae*, prepared from both Richard's and host extract medium, failed to germinate completely seven days after inoculation (Table 61 & 62).

Outputs

Table 61 : Evaluation of virulence of *L. theobromae* grown on different selected media on *Parthenium hysterophorus* seed germination

S.No	Treatment medium	Seed* germination (%)	Reduction over control (%)	Shoot length* (cm)	Reduction over control (%)	Root length* (cm)	Reduction over control (%)	Vigour Index*	Reduction over control (%)
1.	Czapek's	45.50 ^e (42.42)	42.41	3.68 ^e	28.96	2.75 ^e	33.73	292.00 ^d	60.38
2.	Glucose nutrient	50.75 ^f (45.43)	35.76	3.85 ^f	25.68	3.03 ^f	26.99	349.00 ^e	52.64
3.	Host extract	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
4.	Malt extract	40.00 ^d (39.23)	49.37	3.15 ^f	39.19	2.08 ^d	49.88	209.00 ^c	71.64
5.	Potato dextrose	4.25 ^c (11.84)	94.62	1.00 ^c	80.69	0.95 ^c	77.11	8.00 ^b	98.91
6.	Potato sucrose	1.00 ^b (5.75)	98.73	0.85 ^b	83.59	0.43 ^b	89.64	1.00 ^{ab}	99.86
7.	Richard's	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
8.	Control	79.00 ^g (62.73)	-	5.18 ^g	-	4.15 ^g	-	737.00 ^f	-

* Mean of four replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Table 62 : Evaluation of virulence of culture filtrates of *L. theobromae* grown on different media on *Parthenium hysterophorus* seed germination

S.No	Media	Seed* germination (%)	Reduction over control (%)	Shoot length* (cm)	Reduction over control (%)	Root length* (cm)	Reduction over control (%)	Vigour Index*	Reduction over control (%)
1.	Czapek's	47.00 ^e (43.28)	40.13	3.85 ^e	25.68	2.88 ^e	30.27	316.00 ^d	56.71
2.	Glucose nutrient	51.25 ^f (45.72)	34.71	4.03 ^f	22.20	3.15 ^f	23.73	368.00 ^e	49.59
3.	Host extract	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
4.	Malt extract	47.77 ^d (39.81)	47.77	3.35 ^d	35.33	2.25 ^d	45.52	230.00 ^c	68.49
5.	Potato dextrose	5.50 ^c (13.55)	92.99	1.15 ^c	77.80	1.03 ^c	75.06	12.00 ^b	98.36
6.	Potato sucrose	1.50 ^b (6.93)	98.09	0.88 ^b	83.01	0.55 ^b	86.68	2.00 ^a	99.73
7.	Richard's	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
8.	Control	78.50 ^g (62.38)	-	5.18 ^g	-	4.13 ^g	-	730.00 ^g	-

* Mean of four replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Outputs

On symptom expression at different ages of *Parthenium hysterophorus*

Spraying of *P. hysterophorus* plants individually with the cultures or culture filtrates of *L. theobromae*, prepared from Richard's medium grown cultures, caused maximum plant mortality after 15 days (Tables 63 & 64).

A total inhibition of *P. hysterophorus* seed germination was observed when the seeds were soaked individually for one hr either in a fungal culture or culture filtrate of *F. pallidoroseum* prepared from Coon's medium (Tables 65 & 66).

Outputs

Table 63 : Evaluation of virulence of *L. theobromae* grown on selected media on symptom development at different ages of *Parthenium hysterophorus*

S.No.	Media	Age of <i>Parthenium hysterophorus</i> *														
		PDI (%)					Leaf infection (%)					Plant mortality (%)				
		15 DAS	30 DAS	45 DAS	60 DAS	Mean	15 DAS	30 DAS	45 DAS	60 DAS	Mean	15 DAS	30 DAS	45 DAS	60 DAS	Mean
1.	Czapek's medium	27.03 (31.32)	18.14 (25.21)	12.96 (21.10)	17.40 (24.65)	18.88 (25.77)	46.72 (43.11)	44.76 (41.99)	21.20 (27.41)	44.40 (41.78)	39.27 (38.82)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
2.	Glucose nutrient medium	25.92 (30.61)	16.29 (23.80)	11.48 (19.80)	15.92 (23.51)	17.40 (24.65)	43.76 (41.41)	38.21 (38.18)	20.08 (26.62)	38.12 (38.13)	35.04 (36.27)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
3.	Host extract medium	95.18 (77.33)	84.81 (67.06)	47.03 (43.30)	83.70 (66.19)	77.68 (61.82)	100.00 (80.90)	100.00 (80.90)	75.42 (60.28)	100.00 (80.90)	93.86 (75.70)	98.33 (85.69)	90.00 (71.57)	23.33 (28.78)	93.33 (77.71)	76.25 (60.87)
4.	Malt extract medium	29.62 (32.97)	21.11 (27.35)	17.40 (24.65)	20.74 (27.09)	22.22 (27.97)	56.75 (48.88)	43.57 (41.30)	27.51 (31.63)	43.52 (41.28)	42.84 (40.86)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
5.	Potato dextrose medium	75.92 (60.61)	66.29 (54.51)	38.14 (38.12)	65.92 (54.28)	61.57 (51.71)	100.00 (80.90)	86.48 (68.44)	59.50 (50.47)	85.77 (67.84)	82.94 (65.57)	91.67 (73.40)	76.67 (61.23)	5.00 (10.45)	76.67 (61.23)	62.50 (52.24)
6.	Potato sucrose medium	89.62 (71.21)	82.59 (67.70)	42.22 (40.52)	81.85 (64.79)	74.07 (59.41)	100.00 (80.90)	98.78 (83.67)	65.60 (54.09)	97.93 (81.84)	90.58 (72.15)	93.33 (75.24)	83.33 (66.14)	15.00 (22.60)	80.00 (63.44)	67.92 (55.49)
7.	Richard's medium**	99.99a (89.43)	92.22 (73.83)	52.96 (46.70)	91.48 (73.04)	84.16 (66.58)	100.00 (80.90)	100.00 (83.54)	84.96 (67.18)	100.00 (85.44)	96.21 (78.76)	100.00 (80.90)	97.00 (80.03)	40.00 (39.23)	95.00 (77.08)	85.00 (67.21)
8.	Control	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (2.87)	0.00 (2.63)	0.00 (2.43)	0.00 (2.36)	0.00 (2.57)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
	Mean	55.41 (48.10)	47.68 (43.68)	25.00 (30.00)	47.13 (43.34)		68.40 (55.80)	63.98 (53.13)	44.28 (41.73)	63.72 (52.95)		47.92 (43.80)	43.75 (41.44)	10.42 (18.81)	43.75 (41.44)	

CD(P=0.05)

Days	1.61	1.53	1.41
Medium	2.00	1.98	1.83
Days x Medium	1.32	1.20	1.20

* Mean of three replications. (Data in parentheses are arcsine transformed values).

** Complete death of the plant on 7 days after spray at 15 days old seedlings.

Outputs

Table 64 : Evaluation of virulence of culture filtrates of *L. theobromae* grown in selected media on symptom expression at different ages of *Parthenium hysterophorus*

S.No.	Media	Age of <i>Parthenium hysterophorus</i> *														
		PDI (%)					Leaf infection (%)					Plant mortality (%)				
		15 DAS	30 DAS	45 DAS	60 DAS	Mean	15 DAS	30 DAS	45 DAS	60 DAS	Mean	15 DAS	30 DAS	45 DAS	60 DAS	Mean
1.	Czapek's medium	25.55 (30.33)	15.55 (23.18)	11.11 (19.46)	15.55 (23.18)	16.94 (24.27)	45.12 (42.19)	43.90 (41.50)	19.51 (26.21)	43.90 (41.50)	38.11 (38.12)	0.00 (9.10)	0.00 (9.10)	0.00 (0.19)	0.00 (9.10)	0.00 (9.10)
2.	Glucose nutrient medium	23.33 (28.86)	14.44 (22.30)	10.00 (18.43)	14.44 (22.30)	15.55 (23.26)	43.08 (41.03)	37.80 (37.94)	19.05 (25.91)	37.50 (37.76)	37.36 (37.70)	0.00 (9.10)	0.00 (9.10)	0.00 (0.19)	0.00 (9.10)	0.00 (9.10)
3.	Host extract medium	94.44 (76.31)	82.22 (65.05)	44.44 (41.78)	80.00 (63.43)	75.28 (60.20)	100.00 (80.90)	100.00 (83.54)	73.00 (58.69)	100.00 (85.44)	93.25 (75.00)	93.33 (75.00)	93.33 (75.00)	32.33 (35.24)	93.33 (75.00)	78.33 (62.24)
4.	Malt extract medium	28.88 (32.52)	20.00 (26.57)	15.55 (23.18)	19.99 (26.57)	21.11 (27.35)	56.25 (48.62)	42.50 (40.69)	25.61 (30.40)	41.25 (39.99)	41.40 (40.05)	0.00 (9.10)	0.00 (9.10)	0.00 (0.19)	0.00 (9.10)	0.00 (9.10)
5.	Potato dextrose medium	73.37 (58.95)	64.44 (53.37)	36.66 (37.29)	63.33 (53.31)	59.45 (50.48)	100.00 (80.90)	86.25 (68.28)	57.00 (49.02)	85.00 (67.21)	82.06 (64.97)	90.00 (71.57)	70.00 (57.00)	3.33 (6.15)	60.00 (50.77)	55.83 (48.33)
6.	Potato sucrose medium	87.77 (69.47)	80.00 (63.43)	41.11 (39.87)	79.99 (63.36)	72.22 (58.18)	100.00 (80.90)	98.00 (81.87)	63.75 (53.01)	97.50 (80.90)	89.81 (71.37)	90.00 (71.57)	80.00 (63.44)	10.00 (18.44)	73.33 (59.00)	63.33 (52.71)
7.	Richard's medium**	99.99 (89.43)	91.11 (72.64)	50.00 (45.00)	90.00 (71.57)	82.78 (65.50)	100.00 (80.90)	100.00 (83.54)	83.17 (65.80)	100.00 (85.44)	95.79 (78.17)	100.00 (80.90)	96.66 (83.86)	33.33 (35.22)	93.33 (77.71)	80.83 (63.01)
8.	Control	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (2.87)	0.00 (2.63)	0.00 (2.43)	0.00 (2.36)	0.00 (2.57)	0.00 (9.10)	0.00 (9.10)	0.00 (0.19)	0.00 (9.10)	0.00 (9.10)
	Mean	54.17 (47.41)	45.97 (42.71)	26.11 (30.72)	45.41 (42.36)		68.06 (55.61)	63.56 (52.89)	42.61 (40.74)	63.14 (52.59)		46.67 (43.11)	42.50 (40.69)	10.00 (18.43)	40.00 (39.23)	

CD(P=0.05)

Days	1.63	1.14	1.54
Medium	1.81	1.48	2.00
Days x Medium	1.20	1.09	1.38

* Mean of three replications. (Data in parentheses are arcsine transformed values).

** Complete death of the plant on 7 days after spray at 15 days old seedlings.

Outputs

Table 65 : Evaluation of virulence of *F. pallidoroeseum* grown on different selected media on *Parthenium hysterophorus* seed germination

S.No	Media	Seed* germination (%)	Reduction over control (%)	Shoot length* (cm)	Reduction over control (%)	Root length* (cm)	Reduction over control (%)	Vigour Index*	Reduction over control (%)
1.	Coon's	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
2.	Czapek's	5.75 ^c (13.86)	92.72	1.08 ^c	79.03	0.93 ^c	77.59	11.00 ^b	98.50
3.	Malt extract	24.75 ^f (29.83)	68.67	2.63 ^f	48.93	1.83 ^f	55.90	110.00 ^e	85.03
4.	Potato dextrose	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
5.	Potato sucrose	1.50 ^b (6.93)	98.10	0.83 ^b	83.88	0.23 ^b	94.46	2.00 ^a	99.73
6.	Richard's	16.50 ^e (23.96)	79.11	2.08 ^e	59.61	1.75 ^e	57.83	63.00 ^d	91.43
7.	Spezieller Nahrstoffarmer	11.50 ^d (19.82)	85.44	1.60 ^d	68.93	1.00 ^d	75.90	30.00 ^c	95.92
8.	Control	79.00 ^g (62.73)	-	5.15 ^g	-	4.15 ^g	-	735.00 ^f	-

* Mean of four replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Table 66 : Evaluation of virulence of culture filtrates of *F. pallidoroeseum* grown on different selected media on *Parthenium hysterophorus* seed germination

S.No	Treatment	Seed* germination (%)	Reduction over control (%)	Shoot length* (cm)	Reduction over control (%)	Root length* (cm)	Reduction over control (%)	Vigour Index*	Reduction over control (%)
1.	Coon's	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
2.	Czapek's	6.25 ^c (14.47)	92.06	1.20 ^c	76.83	1.03 ^c	75.06	14.00 ^b	98.09
3.	Malt extract	26.25 ^f (30.82)	66.67	2.78 ^f	46.33	1.95 ^e	52.78	124.00 ^e	83.06
4.	Potato dextrose	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
5.	Potato sucrose	2.75 ^b (9.52)	96.51	0.90 ^b	82.63	0.45 ^b	89.10	4.00 ^a	99.45
6.	Richard's	17.25 ^e (24.53)	78.10	2.18 ^e	57.92	1.88 ^d	54.48	70.00 ^d	90.44
7.	Spezieller Nahrstoffarmer	11.75 ^d (20.04)	85.08	1.83 ^d	64.67	1.03 ^c	75.06	33.48 ^c	95.49
8.	Control	78.75 ^g (62.55)	-	5.18 ^g	-	4.13 ^f	-	732.00 ^f	-

* Mean of four replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Cultures and culture filtrates of *F. pallidoroeseum* were found to be highly aggressive to *Parthenium* weed (Tables 67 & 68).

Spraying of 15 day-old *P. hysterophorus* plants with either a culture or a culture filtrate of *L. theobromae* exhibited complete plant mortality, seven days after spray treatment and a similar trend was noted with *F. pallidoroeseum*.

Outputs

Table 67 : Evaluation of virulence of *F. pallidoroseum* grown in different media on disease incidence at different age of *Parthenium hysterophorus*

S.No.	Media	Age of <i>Parthenium hysterophorus</i> *														
		PDI (%)					Leaf infection (%)					Plant mortality (%)				
		15 DAS	30 DAS	45 DAS	60 DAS	Mean	15 DAS	30 DAS	45 DAS	60 DAS	Mean	15 DAS	30 DAS	45 DAS	60 DAS	Mean
1.	Coon's medium	88.14 (69.86)	82.59 (65.34)	38.14 (38.12)	81.85 (64.79)	72.68 (58.50)	100.00 (80.90)	100.00 (83.54)	68.43 (55.81)	100.00 (85.44)	92.11 (73.68)	70.00 (56.79)	63.33 (52.78)	13.33 (21.34)	60.00 (50.77)	51.67 (46.26)
2.	Czapek's medium	75.55 (60.37)	69.25 (56.32)	20.74 (27.09)	68.51 (55.86)	58.51 (49.89)	92.05 (73.63)	87.62 (69.40)	42.07 (40.44)	87.20 (69.03)	77.24 (61.48)	13.33 (21.33)	11.67 (19.89)	0.00 (9.10)	0.00 (18.44)	8.75 (17.26)
3.	Malt extract medium	31.85 (34.36)	30.36 (33.44)	7.77 (16.16)	29.25 (32.73)	24.81 (29.87)	61.79 (51.58)	59.19 (50.30)	24.12 (29.41)	58.86 (50.10)	50.99 (45.57)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
4.	Potato dextrose medium	84.81 (67.06)	80.74 (63.97)	30.36 (33.44)	77.25 (62.91)	68.79 (56.04)	100.00 (80.90)	100.00 (83.54)	64.37 (53.37)	100.00 (85.44)	91.09 (72.64)	60.00 (50.77)	53.33 (46.93)	10.00 (18.44)	50.00 (45.00)	43.33 (41.15)
5.	Potato sucrose medium	81.85 (64.79)	75.92 (60.61)	22.96 (28.63)	74.81 (59.88)	63.89 (53.07)	97.35 (80.65)	93.86 (75.65)	59.64 (50.56)	94.05 (75.89)	86.23 (68.19)	43.33 (41.15)	40.00 (39.23)	0.00 (9.10)	46.67 (43.08)	32.50 (34.76)
6.	Richard's medium	50.74 (45.42)	45.18 (42.24)	10.74 (19.13)	44.07 (41.59)	37.68 (37.88)	68.88 (56.10)	74.40 (61.03)	32.94 (35.03)	64.14 (53.21)	60.09 (50.83)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
7.	Spezieller Nahrstoffarmer medium	61.48 (51.64)	58.14 (49.68)	17.70 (24.93)	57.77 (49.47)	48.77 (44.31)	78.07 (62.07)	73.37 (58.94)	35.78 (36.74)	73.00 (58.69)	65.06 (53.79)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
8.	Control	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (2.87)	0.00 (2.63)	0.00 (2.43)	0.00 (2.36)	0.00 (2.57)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
	Mean	59.30 (50.36)	55.27 (48.04)	18.55 (25.55)	54.44 (47.52)		74.77 (59.87)	73.56 (59.08)	42.17 (40.51)	72.16 (58.18)		23.33 (28.86)	21.04 (27.27)	2.88 (9.80)	20.83 (27.13)	

CD(P=0.05)

Days	1.90	1.98	1.80
Medium	2.20	2.32	2.11
Days x Medium	1.58	1.72	1.64

*Mean of three replications. (Data in parentheses are arcsine transformed values).

Outputs

Table 68 : Evaluation of virulence of culture filtrate of *F. pallidoroseum* grown on different selected culture media on symptom expression at different age of *Parthenium hysterophorus*

S.No.	Media	Age of <i>Parthenium hysterophorus</i> *														
		PDI (%)					Leaf infection (%)					Plant mortality (%)				
		15 DAS	30 DAS	45 DAS	60 DAS	Mean	15 DAS	30 DAS	45 DAS	60 DAS	Mean	15 DAS	30 DAS	45 DAS	60 DAS	Mean
1.	Coon's medium	87.03 (68.90)	81.85 (64.79)	37.03 (37.48)	80.37 (63.70)	71.57 (57.80)	100.00 (80.90)	100.00 (83.54)	68.04 (55.57)	100.00 (85.44)	92.01 (73.57)	66.67 (54.78)	56.67 (48.85)	15.00 (22.60)	53.33 (46.92)	47.92 (43.80)
2.	Czapek's medium	75.18 (60.12)	68.14 (55.64)	19.99 (26.56)	67.03 (54.96)	57.59 (49.37)	90.05 (72.59)	86.80 (68.71)	41.59 (40.16)	84.68 (66.97)	75.78 (60.53)	10.00 (18.44)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	2.50 (9.10)
3.	Malt extract medium	31.48 (34.13)	29.62 (32.96)	7.03 (15.37)	27.03 (31.32)	23.79 (29.20)	60.49 (51.06)	58.46 (49.87)	23.77 (29.18)	57.60 (49.37)	50.08 (45.06)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
4.	Potato dextrose medium	84.07 (66.48)	79.67 (63.19)	29.62 (32.97)	78.51 (62.38)	67.97 (55.55)	100.00 (80.90)	100.00 (83.54)	62.82 (52.42)	100.00 (85.44)	90.71 (72.24)	60.00 (50.77)	50.00 (45.00)	10.00 (18.43)	40.00 (39.23)	40.00 (39.23)
5.	Potato sucrose medium	81.48 (64.51)	74.81 (59.88)	21.85 (27.87)	72.96 (58.67)	62.78 (52.42)	96.26 (78.95)	92.53 (74.14)	58.88 (50.11)	91.57 (73.12)	84.81 (67.05)	40.00 (39.23)	33.33 (35.22)	5.00 (12.92)	30.00 (33.21)	27.08 (31.37)
6.	Richard's medium	49.93 (44.99)	44.81 (42.02)	9.66 (18.10)	42.48 (40.67)	36.72 (37.29)	68.03 (55.57)	63.00 (52.53)	31.97 (34.43)	62.71 (52.36)	56.43 (48.68)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
7.	Spezieller Nahrstoffarmer medium	60.44 (51.02)	57.03 (49.04)	17.40 (24.65)	54.81 (47.76)	47.42 (43.51)	77.20 (61.48)	72.22 (58.19)	35.37 (36.49)	71.55 (57.77)	64.09 (53.19)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
8.	Control	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (5.74)	0.00 (2.87)	0.00 (2.63)	0.00 (2.43)	0.00 (2.36)	0.00 (2.57)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)	0.00 (9.10)
	Mean	58.70 (50.01)	54.49 (47.58)	17.82 (24.95)	52.90 (46.66)		74.00 (59.34)	71.63 (57.80)	41.56 (40.16)	71.01 (57.42)		22.08 (28.04)	17.50 (24.73)	3.75 (11.24)	15.42 (23.11)	

CD(P=0.05)

Days	1.83	1.91	1.87
Medium	2.10	2.00	2.08
Days x Medium	1.41	1.32	1.58

*Mean of three replications. (Data in parentheses are arcsine transformed values).

Outputs

Optimum age of inoculum

Young and actively growing cultures (7-15 days old) of *L. theobromae* and *F. pallidoroseum* were found to be more virulent than that of older cultures (>15 days old) (Tables 69 & 70).

Table 69 : Effect of age of fungus culture of selected pathogenic isolates on *Parthenium hysterophorus* seed germination

S.No.	Age of culture(days)	Seed germination (%)	Reduction over control (%)	Shoot length (cm)	Reduction over control (%)	Root length (cm)	Reduction over control (%)	Vigour Index*	Reduction over control (%)
<i>A. L. theobromae</i>									
1.	7	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
2.	14	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
3.	21	2.00 ^b (8.13)	97.47	0.43 ^b	91.62	0.40 ^b	90.31	2.00 ^b	99.73
4.	28	8.00 ^c (16.43)	89.87	0.98 ^c	80.90	0.63 ^c	84.75	13.00 ^c	98.22
5.	35	16.00 ^d (23.58)	79.75	2.86 ^d	44.25	2.00 ^d	51.57	78.00 ^d	89.32
6.	Control	79.00 ^e (62.73)	-	5.13 ^e	-	4.13 ^e	-	731.00 ^e	-
<i>B. F. pallidoroseum</i>									
1.	7	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
2.	14	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
3.	21	3.00 ^b (9.97)	96.20	0.48 ^b	90.64	0.43 ^b	89.59	3.00 ^b	99.59
4.	28	10.00 ^c (18.43)	87.34	1.12 ^c	78.17	1.00 ^c	75.79	21.00 ^c	97.13
5.	35	21.00 ^d (27.27)	73.42	2.91 ^d	43.27	2.43 ^d	41.16	112.00 ^d	84.68
6.	Control	79.00 ^e (62.73)	-	5.13 ^e	-	4.13 ^e	-	731.00 ^e	-

* Mean of three replications. (Data in parentheses are arcsine transformed values). In columns of A and B, means followed by a common letter are not significantly different at 5% level by DMRT.

Table 70 : Effect of age of fungus culture of selected pathogenic isolates on symptom expression

S.No.	Age of culture (days)	PDI (%)	IOC (%)	Leaf infection (%)	IOC (%)	Plant mortality (%)	IOC (%)
<i>A. L. theobromae</i> *							
1.	7	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00
2.	14	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00
3.	21	84.00 ^b (66.42)	84.00	86.00 ^b (68.03)	86.00	80.00 ^b (64.23)	80.00
4.	28	78.46 ^c (62.38)	78.46	80.00 ^c (63.43)	80.00	70.00 ^c (56.79)	70.00
5.	35	50.00 ^d (45.00)	50.00	60.00 ^d (50.77)	60.00	30.37 ^d (33.40)	30.37
6.	Control	0.00 ^e (5.74)	-	0.00 ^e (2.87)	-	0.00 ^e (9.10)	-

Outputs

S.No.	Age of culture (days)	PDI (%)	IOC (%)	Leaf infection (%)	IOC (%)	Plant mortality (%)	IOC (%)
B. <i>F. pallidoroseum</i>*							
1.	7	87.77 ^a (69.56)	87.77	100.00 ^a (80.90)	100.00	70.00 ^a (56.79)	70.00
2.	14	88.04 ^a (69.73)	88.04	100.00 ^a (80.90)	100.00	70.00 ^a (56.79)	70.00
3.	21	72.23 ^b (58.18)	72.23	82.00 ^b (64.90)	82.00	51.11 ^b (45.67)	51.11
4.	28	65.55 ^c (54.09)	65.55	74.00 ^c (59.34)	74.00	30.33 ^c (33.46)	30.33
5.	35	46.04 ^d (42.71)	46.04	52.00 ^d (46.14)	52.00	25.67 ^d (30.46)	25.67
6.	Control	0.00 ^e (5.74)	-	0.00 ^e (2.87)	-	0.00 ^e (9.10)	-

IOC – Increase over control

* Mean of three replications. (Data in parentheses are arcsine transformed values). In columns of A and B, means followed by a common letter are not significantly different at 5% level by DMRT.

The virulence of culture filtrates of *L. theobromae* and *F. pallidoroseum* increased with increase in days of incubation and 28 day-old culture filtrates were highly virulent (Tables 71 & 72).

Table 71 : Effect of age of culture filtrates of selected pathogenic isolates on *Parthenium hysterophorus* seed germination

S.No	Age of culture filtrate (days)	Seed Germination (%)	Reduction over control (%)	Shoot length (cm)	Reduction over control (%)	Root length (cm)	Reduction over control (%)	Vigour Index	Reduction over control (%)
A. <i>L. theobromae</i>*									
1.	7	79.00 ^d (62.73)	-	4.38 ^d	14.62	3.58 ^d	13.32	628.00 ^d	14.09
2.	14	21.00 ^c (27.27)	89.87	2.78 ^c	45.81	2.45 ^c	40.68	110.00 ^c	84.95
3.	21	8.00 ^b (16.43)	89.87	0.88 ^b	82.85	0.65 ^b	84.26	12.00 ^b	98.35
4.	28	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
5.	35	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
6.	Control	79.00 ^b (62.73)	-	5.13 ^e	-	4.13 ^e	-	731.00 ^e	-
B. <i>F. pallidoroseum</i>*									
1.	7	78.50 ^d (62.38)	0.32	4.65 ^e	9.36	3.90 ^e	5.56	671.00 ^e	8.20
2.	14	36.25 ^c (37.05)	53.97	3.08 ^d	39.96	2.55 ^d	38.26	204.00 ^d	72.09
3.	21	17.75 ^b (24.95)	77.46	1.63 ^c	68.23	1.18 ^c	71.43	50.00 ^c	93.16
4.	28	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
5.	35	2.00 ^a (8.13)	97.46	0.48 ^b	90.64	0.28 ^b	93.22	2.00 ^b	99.73

Outputs

6.	Control	78.75 ^e (62.55)	-	5.13 ^f	-	4.13 ^f	-	731.00 ^f	-
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* Mean of four replications. (Data in parentheses are arcsine transformed values). In columns of A and B, means followed by a common letter are not significantly different at 5% level by DMRT.

Table 72 : Effect of age of culture filtrates of selected pathogenic isolates on symptom expression

S.No.	Age of culture filtrate (days)	PDI (%)	IOC (%)	%Leaf infection	IOC (%)	%Plant mortality	IOC (%)
A. <i>L. theobromae</i>*							
1.	7	0.00 ^d (5.74)	-	0.00 ^d (2.87)	-	0.00 ^d (9.10)	-
2.	14	31.85 ^c (34.36)	31.85	59.06 ^c (50.22)	59.06	3.33 ^c (8.61)	3.33
3.	21	79.25 ^b (62.90)	79.25	91.31 ^b (72.85)	91.31	46.67 ^b (43.07)	46.67
4.	28**	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00
5.	35**	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00
6.	Control	0.00 ^d (5.74)	-	0.00 ^d (2.87)	-	0.00 ^d (9.10)	-
B. <i>F. pallidoroeseum</i>*							
1.	7	0.00 ^e (5.74)	-	0.00 ^e (2.87)	-	0.00 ^e (9.10)	-
2.	14	37.03 ^d (37.48)	37.03	67.40 ^d (55.18)	67.40	5.00 ^d (12.93)	5.00
3.	21	71.48 ^c (57.72)	71.48	91.57 ^c (73.13)	91.57	26.67 ^c (31.00)	26.67
4.	28	87.03 ^a (68.90)	87.03	100.00 ^a (80.90)	100.00	70.00 ^a (56.79)	66.67
5.	35	82.59 ^b (65.34)	82.59	96.27 ^b (78.87)	96.27	40.00 ^b (39.23)	40.00
6.	Control	0.00 ^e (5.74)	-	0.00 ^e (2.87)	-	0.00 ^e (9.10)	-

* Mean of three replications. (Data in parentheses are arcsine transformed values). In columns of A and B, means followed by common letter are not significantly different at 5% level by DMRT. ** Complete death of the plant on 7 days after spray

Evaluation of optimum concentration of inoculum

Fifteen % fungal cultures or 80 % culture filtrates of *L. theobromae* and *F. pallidoroeseum* caused maximum inhibition of seed germination (Tables 73, 74 & 75) and maximum *P. hysterophorus* mortality (Tables 76 & 77).

Table 73 : Effect of different concentrations of fungal cultures of selected pathogenic isolates on *Parthenium hysterophorus* seed germination

S.No	Fungal culture conc. (w/v) (%)	Seed Germination (%)	Reduction over control (%)	Shoot length (cm)	Reduction over control (%)	Root length (cm)	Reduction over control (%)	Vigour Index	Reduction over control (%)
A. <i>L. theobromae</i>*									
1.	5.0	2.00 ^b (8.13)	97.47	0.45 ^b	91.23	0.40 ^b	90.31	2.00 ^a	99.73
2.	10.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
3.	15.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
4.	20.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00

Outputs

S.No	Fungal culture conc. (w/v) (%)	Seed Germination n (%)	Reduction over control (%)	Shoot length (cm)	Reduction over control (%)	Root length (cm)	Reduction over control (%)	Vigour Index	Reduction over control (%)
5.	25.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
6.	30.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
7.	Control	79.00 ^c (62.73)	-	5.13 ^c	-	4.13 ^c	-	731.00 ^b	-
B. <i>Fusarium pallidoroseum</i>*									
1.	5.0	9.00 ^c (17.46)	88.61	1.73 ^b	66.28	1.00 ^b	75.79	25.00 ^c	96.58
2.	10.00	4.00 ^b (11.54)	94.93	0.74 ^b	85.58	0.44 ^b	89.35	5.00 ^b	99.32
3.	15.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
4.	20.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
5.	25.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
6.	30.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
7.	Control	79.00 ^d (62.73)	-	5.13 ^d	-	4.13 ^d	-	731.00 ^d	-

* Mean of four replications. (Data in parentheses are arcsine transformed values). In columns of A and B, means followed by a common letter are not significantly different at 5% level by DMRT.

Table 74 : Effect of different concentrations of fungal cultures of selected pathogenic fungi on disease incidence

S.No.	Fungal culture concentration (w/v) (%)	PDI (%)	IOC (%)	Leaf infection (%)	IOC (%)	% Plant mortality	IOC (%)
A. <i>L. theobronae</i>*							
1.	5.0	51.11 ^c (45.63)	51.11	80.00 ^b (63.43)	80.00	60.00 ^c (50.77)	60.00
2.	10.0	94.07 ^b (75.92)	94.07	100.00 ^a (80.90)	100.00	90.00 ^b (71.57)	90.00
3.	15.0**	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00
4.	20.0**	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00
5.	25.0**	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00
6.	30.0**	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00
7.	Control	0.00 ^d (5.74)	-	0.00 ^c (2.87)	-	0.00 ^d (9.10)	-
B. <i>F. pallidoroseum</i>*							
1.	5.0	30.36 ^c (33.40)	30.36	64.37 ^c (53.37)	64.37	10.00 ^c (18.44)	10.00
2.	10.0	64.07 ^b (53.17)	64.07	78.09 ^b (62.09)	78.09	36.67 ^b (37.22)	36.67
3.	15.0	87.77 ^a (69.53)	87.77	100.00 ^a (80.90)	100.00	70.00 ^a (56.79)	70.00
4.	20.0	87.77 ^a (69.53)	87.77	100.00 ^a (80.90)	100.00	70.00 ^a (56.79)	70.00
5.	25.0	88.14 ^a (69.86)	88.14	100.00 ^a (80.90)	100.00	70.00 ^a (56.79)	70.00
6.	30.0	88.51 ^a (70.19)	88.51	100.00 ^a (80.90)	100.00	70.00 ^a (56.79)	70.00

Outputs

7.	Control	0.00 ^c (5.74)	-	0.00 ^c (2.87)	-	0.00 ^c (9.10)	-
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IOC – Increase over control * Mean of three replications. (Data in parentheses are arcsine transformed values). In columns of A and B, means followed by a common letter are not significantly different at 5% level by DMRT. ** Complete death of the plant on 7 days after spray.

Table 75 : Effect of different concentrations of culture filtrates of *L. theobromae* on *Parthenium hysterophorus* seed germination

S. No	Culture filtrate conc. (%)	Seed germination (%)	Reduction over control (%)	Shoot length (cm)	Reduction over control (%)	Root length (cm)	Reduction over control (%)	Vigour Index	Reduction over control (%)
1.	10.00	71.25 ^g (57.58)	9.81	4.85 ^g	5.46	4.05 ^g	1.93	663.00 ^f	9.30
2.	20.00	60.25 ^f (50.91)	23.73	4.15 ^f	19.10	3.75 ^f	9.20	476.00 ^e	34.88
3.	30.00	57.25 ^e (49.07)	27.53	3.53 ^e	31.19	3.15 ^e	23.73	382.00 ^d	47.74
4.	40.00	27.50 ^d (31.62)	65.19	1.45 ^d	71.73	1.13 ^d	72.64	71.00 ^c	90.29
5.	50.00	10.50 ^c (18.90)	86.71	1.13 ^c	77.97	0.95 ^c	77.00	22.00 ^b	97.00
6.	60.00	2.50 ^b (9.05)	96.84	0.55 ^b	89.28	0.33 ^b	92.00	2.00 ^a	99.73
7.	70.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
8.	80.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
9.	90.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
10.	100.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
11.	Control	79.00 ^h (62.73)	-	5.13 ^h	-	4.13 ^h	-	731.00 ^g	-

* Mean of four replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Table 76 : Effect of different concentrations of culture filtrates of *F. pallidoroseum* on *Parthenium hysterophorus* seed germination

S.No	% Culture filtrate conc.	Seed germination (%)	Reduction over control (%)	Shoot length (cm)	Reduction over control (%)	Root length (cm)	Reduction over control (%)	Vigour Index	Reduction over control (%)
1.	10.00	78.50 ^g (62.38)	0.60	4.90 ^g	4.48	4.10 ^g	0.70	714.00 ^f	2.33
2.	20.00	78.50 ^g (62.38)	0.60	4.85 ^g	5.46	4.10 ^g	0.70	707.00 ^f	3.28
3.	30.00	70.50 ^f (57.10)	10.76	3.73 ^f	27.29	3.28 ^f	20.58	494.00 ^e	32.42
4.	40.00	64.50 ^e (53.43)	18.35	2.70 ^e	47.37	2.08 ^e	49.64	308.00 ^d	57.87
5.	50.00	50.50 ^d (45.29)	36.08	1.85 ^d	63.94	1.33 ^d	67.80	160.00 ^c	78.11
6.	60.00	20.75 ^c (27.09)	73.73	1.15 ^c	77.58	0.98 ^c	76.27	44.00 ^b	93.98
7.	70.00	2.25 ^b (8.59)	97.15	0.55 ^b	89.28	0.40 ^b	90.31	2.00 ^a	99.73
8.	80.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
9.	90.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00

Outputs

10.	100.00	0.00 ^a (2.87)	100.00	0.00 ^a	100.00	0.00 ^a	100.00	0.00 ^a	100.00
11.	Control	79.00 ^h (62.73)	-	5.13 ^h	-	4.13 ^h	-	731.00 ^g	-

* Mean of four replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Outputs

Table 77 : Effect of different concentrations of culture filtrates of selected pathogenic isolates on symptom expression

S.No.	Culture Filtrate concentration (%)	<i>L. theobromae</i> *						<i>F. pallidoroseum</i>					
		PDI (%)	IOC (%)	Leaf infection (%)	IOC (%)	Plant mortality (%)	IOC (%)	PDI (%)	IOC (%)	Leaf infection (%)	IOC (%)	Plant mortality (%)	IOC (%)
1.	10	7.03 ^h (15.37)	7.03	19.20 ^f (25.99)	19.20	0.00 ^d (9.10)	0.00	0.00 ^g (5.74)	0.00	0.00 ^e (2.87)	0.00	0.00 ^e (9.10)	0.00
2.	20	15.18 ^g (22.93)	15.18	37.67 ^e (37.86)	37.67	0.00 ^d (9.10)	0.00	0.00 ^g (5.74)	0.00	0.00 ^e (2.87)	0.00	0.00 ^e (9.10)	0.00
3.	30	21.48 ^f (27.61)	21.48	43.55 ^e (41.29)	43.55	0.00 ^d (9.10)	0.00	7.03 ^f (15.37)	7.03	23.96 ^d (29.31)	23.96	0.00 ^e (9.10)	0.00
4.	40	37.03 ^d (37.48)	37.03	58.21 ^d (49.73)	58.21	0.00 ^d (9.10)	0.00	22.22 ^e (28.13)	22.22	42.85 ^c (40.89)	42.85	0.00 ^e (9.10)	0.00
5.	50	59.25 ^d (50.33)	59.25	84.69 ^c (66.97)	84.69	8.33 ^c (16.60)	8.33	50.74 ^d (45.42)	50.74	83.17 ^b (65.78)	83.17	5.00 ^d (12.92)	5.00
6.	60	87.03 ^c (68.90)	87.03	93.35 ^b (76.55)	93.35	50.00 ^b (45.00)	50.33	70.74 ^c (57.25)	70.74	100.00 ^a (80.90)	100.00	13.33 ^c (21.14)	13.33
7.	70	98.51 ^b (83.09)	98.51	99.33 ^a (87.10)	99.33	90.00 ^a (71.57)	90.00	87.03 ^b (68.90)	87.03	100.00 ^a (80.90)	100.00	50.00 ^b (45.00)	50.00
8.	80**	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00	88.14 ^a (69.86)	88.14	100.00 ^a (80.90)	100.00	66.67 ^a (54.78)	66.67
9.	90**	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00	88.14 ^a (69.86)	88.14	100.00 ^a (80.90)	100.00	66.67 ^a (54.78)	66.67
10.	100**	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00	88.51 ^a (70.19)	88.51	100.00 ^a (80.90)	100.00	66.67 ^a (54.78)	66.67
11.	Control	0.00 ⁱ (5.74)	-	0.00 ^g (2.87)	-	0.00 ^d (9.10)	-	0.00 ^g (5.74)	-	0.00 ^e (2.87)	-	0.00 ^c (9.10)	-

IOC – Increase over control

* Mean of three replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

** Complete death of the plant on 7 days after spray .

Outputs

Age of susceptibility of *Parthenium hysterophorus* to powdery mildew

Sixty day-old plants were found to be highly susceptible to the powdery mildew and the highest disease incidence coincides with flower initiation and peak flowering stage (Table 78).

Table 78 : Effect of *Parthenium hysterophorus* age on susceptibility to powdery mildew

S.No.	Age of Parthenium plant (days)	PDI* (%)	IOC (%)	Leaf* infection (%)	IOC (%)	Twig* infection (%)	IOC (%)	Plant* mortality (%)	IOC (%)
1.	15	44.88 ^c (42.06)	44.88	62.28 ^b (52.11)	62.28	0.00 ^b (9.10)	0.00	0.00 ^d (9.10)	-
2.	30	86.44 ^b (68.36)	86.44	100.00 ^a (80.90)	100.00	100.00 ^a (80.90)	100.00	54.00 ^c (47.29)	54.00
3.	45	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (81.67)	100.00	68.00 ^b (55.55)	68.00
4.	60	99.99 ^a (89.43)	99.99	100.00 ^a (80.90)	100.00	100.00 ^a (82.60)	100.00	82.00 ^a (64.90)	82.00
5.	Control	0.00 ^d (5.74)	-	0.00 ^d (2.87)	-	0.00 ^b (9.10)	-	0.00 ^d (9.10)	-

IOC – Increase over control

* Mean of three replications. (Data in parentheses are arcsine transformed values). In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Physiological changes due to selected pathogenic isolates on *Parthenium hysterophorus*

A significant increase in biochemical constituents such as sugars and phenols and a decrease in protein content were recorded in the healthy *P. hysterophorus* leaves as the age advanced. Leaves infected by *L. theobromae* and *F. pallidoroeseum* showed a drastic decrease in sugar content and an increase in phenolic and protein content at all stages. Powdery mildew-infected leaves had less amount of total sugars and reducing sugars and increased quantities of non-reducing sugars, proteins and phenolic content, which was directly proportionate to the intensity of infection (Table 79).

Table 79 : Physiological changes in *Parthenium hysterophorus* due to infection with selected pathogenic isolates

S.No	Particulars (mg/g)	Age of plant (days)	Healthy*	Infected*		
				<i>L. theobromae</i>	<i>F. pallidoroeseum</i>	Powdery mildew
1.	Total sugars	15	19.51	3.90	7.54	14.95
		30	56.75	12.91	24.83	46.51
		45	172.37	113.90	136.69	132.29
		60	186.34	48.52	82.18	148.00
2.	Reducing sugars	15	11.66	1.87	4.62	5.00
		30	20.58	5.18	4.98	9.39
		45	57.97	37.68	46.57	17.37
		60	68.85	17.21	30.09	29.60
3.	Non-reducing sugars	15	7.85	2.03	2.92	9.95
		30	36.17	7.73	15.85	37.12
		45	114.40	76.22	93.12	114.92
		60	117.49	31.31	52.09	118.40

Outputs

S.No	Particulars (mg/g)	Age of plant (days)	Healthy*	Infected*		
				<i>L. theobromae</i>	<i>F. pallidoroseum</i>	Powdery mildew
4.	OD phenols	15	0.500	0.890	0.800	0.600
		30	1.080	1.860	1.660	1.621
		45	1.840	2.400	2.120	2.940
		60	1.910	3.250	2.880	3.240
5.	Total phenols	15	0.621	1.101	0.990	0.740
		30	1.210	2.082	1.864	1.811
		45	2.080	2.702	2.390	3.322
		60	2.240	3.804	3.404	8.800
6.	Proteins	15	592.20	610.08	608.22	593.41
		30	590.22	605.04	600.01	592.04
		45	586.90	588.62	587.50	594.20
		60	584.00	604.28	589.42	596.42

* Mean of three replications

Host range studies of selected pathogenic isolates

The fungal culture as well as culture filtrates of *L. theobromae* and *F. pallidoroseum* and spore suspension of *O. parthenii* did not affect seed germination and seedling vigour of any of the crop plants tested compared with the control (*P. hysterothorus*) in which seed germination was completely inhibited (Tables 80, 81, 82, 83 & 84).

Table 80 : Effect of fungus culture of *L. theobromae* on seed germination and seedling vigour of host crops under *in vitro* condition

S. No.	Host tested	Seed germination* (%)		Shoot length* (cm)		Root length* (cm)		Vigour Index*	
		UT	T	UT	T	UT	T	UT	T
1.	Beet root	100.00	100.00	7.55	7.50	7.75	7.73	1530.00	1523.00
2.	Bengal gram	84.00	84.00	4.70	4.68	14.70	14.70	1630.00	1628.00
3.	Bhendi	100.00	100.00	14.83	14.81	13.75	13.71	2858.00	2852.00
4.	Black gram	100.00	100.00	20.31	20.27	17.65	17.62	3796.00	3789.00
5.	Chilli	94.00	94.00	2.84	2.80	5.78	5.78	810.00	807.00
6.	Cowpea	95.00	95.00	25.80	25.77	20.00	20.00	4351.00	4348.00
7.	Cumbu	96.00	95.00	16.86	16.82	15.80	15.88	3135.00	3107.00
8.	French bean	94.00	94.00	20.04	20.01	19.50	19.50	3717.00	3714.00
9.	Green gram	100.00	100.00	21.11	21.10	17.80	17.71	3891.00	3881.00
10.	Groundnut	98.00	98.00	14.12	14.10	15.51	15.42	2904.00	2893.00
11.	Maize	100.00	100.00	23.00	23.00	29.00	28.98	5200.00	5198.00
12.	Peas	55.00	55.00	11.11	11.08	12.50	12.48	1181.00	1178.00
13.	Pumpkin	82.00	82.00	9.41	9.38	22.61	22.58	2626.00	2621.00
14.	Safflower	50.00	50.00	10.17	10.16	12.11	12.10	1025.00	1024.00
15.	Sesame	100.00	100.00	7.00	7.00	9.82	9.82	1682.00	1682.00
16.	Sorghum	98.00	98.00	18.42	18.42	20.52	20.52	3816.00	3816.00
17.	Soybean	70.00	70.00	18.31	18.28	18.50	18.47	1693.00	1691.00
18.	Sunflower	98.00	98.00	8.00	7.99	18.53	18.48	2600.00	2594.00
19.	Tomato	100.00	100.00	4.90	4.90	9.54	9.50	1444.00	1440.00
20.	Parthenium	79.00	0.00	5.10	0.00	4.10	0.00	727.00	0.00

*Mean of four replications. UT – Untreated; T – Treated

Outputs

Table 81 : Effect of culture filtrates of *L.theobromae* on seed germination and seedling vigour of host crops under *in vitro* conditions

S. No	Host tested	Seed germination* (%)		Shoot length* (cm)		Root length* (cm)		Vigour Index*	
		UT	T	UT	T	UT	T	UT	T
1.	Beet root	100.00	100.00	7.56	7.50	7.73	7.70	1529.00	1520.00
2.	Bengal gram	84.00	84.00	4.70	4.68	14.68	14.66	1628.00	1625.00
3.	Bhendi	100.00	100.00	14.82	14.80	13.74	13.72	2856.00	2852.00
4.	Black gram	100.00	100.00	20.32	20.20	17.64	17.61	3796.00	3781.00
5.	Chilli	94.00	94.00	2.82	2.81	5.75	5.74	806.00	804.00
6.	Cowpea	95.00	95.00	25.77	25.75	20.00	19.96	4348.00	4343.00
7.	Cumbu	96.00	96.00	16.84	16.82	15.79	15.76	3132.00	3128.00
8.	French bean	94.00	94.00	20.00	20.00	19.52	19.51	3715.00	3714.00
9.	Green gram	100.00	100.00	21.00	21.00	17.81	17.80	3881.00	3880.00
10.	Groundnut	98.00	98.00	14.11	14.10	15.50	15.49	3882.00	3880.00
11.	Maize	100.00	100.00	23.01	23.00	29.00	28.96	5201.00	5196.00
12.	Peas	55.00	55.00	11.11	11.08	12.50	12.48	1181.00	1178.00
13.	Pumpkin	82.00	82.00	9.41	9.39	22.60	22.60	2625.00	2623.00
14.	Safflower	50.00	50.00	10.15	10.15	12.10	12.10	1024.00	1024.00
15.	Sesame	100.00	100.00	7.02	7.01	9.81	9.80	1683.00	1681.00
16.	Sorghum	98.00	98.00	18.40	18.40	20.51	20.50	3814.00	3812.00
17.	Soybean	70.00	70.00	18.30	18.29	18.48	18.47	1692.00	1691.00
18.	Sunflower	98.00	98.00	8.00	7.98	18.53	18.46	2600.00	2591.00
19.	Tomato	100.00	100.00	4.91	4.90	9.54	9.52	1445.00	1442.00
20.	Parthenium	79.00	0.00	5.10	0.00	4.10	0.00	727.00	0.00

*Mean of four replications. UT – Untreated; T – Treated

Table 82 : Effect of fungal cultures of *F. pallidoroseum* on seed germination and seedling vigour of host crops under *in vitro* condition

S. No	Host tested	Seed germination* (%)		Shoot length* (cm)		Root length* (cm)		Vigour Index*	
		UT	T	UT	T	UT	T	UT	T
1.	Beet root	100.00	100.00	7.55	7.55	7.75	7.74	1530.00	1529.00
2.	Bengal gram	84.00	84.00	4.70	4.70	14.70	14.69	1630.00	1629.00
3.	Bhendi	100.00	100.00	14.83	14.82	13.75	13.73	2858.00	2855.00
4.	Black gram	100.00	100.00	20.31	20.30	17.65	17.64	3796.00	3794.00
5.	Chilli	94.00	94.00	2.84	2.84	5.78	5.77	810.00	809.00
6.	Cowpea	95.00	95.00	25.80	25.80	20.00	20.00	4351.00	4351.00
7.	Cumbu	96.00	96.00	16.86	16.84	15.80	15.80	3135.00	3133.00
8.	French bean	94.00	94.00	20.04	20.03	19.50	19.50	3717.00	3716.00
9.	Green gram	100.00	100.00	21.11	21.10	17.80	17.80	3891.00	3890.00
10.	Groundnut	98.00	98.00	14.12	14.11	15.51	15.50	3884.00	3882.00
11.	Maize	100.00	100.00	23.00	23.00	29.00	29.00	5200.00	5200.00
12.	Peas	55.00	55.00	11.11	11.11	12.50	12.50	1181.00	1181.00
13.	Pumpkin	82.00	82.00	9.41	9.40	22.61	22.60	2626.00	2624.00
14.	Safflower	50.00	50.00	10.17	10.17	12.11	12.10	1025.00	1024.00
15.	Sesame	100.00	100.00	7.00	6.99	9.82	9.81	1682.00	1680.00
16.	Sorghum	98.00	98.00	18.42	18.42	20.52	20.52	3816.00	3816.00
17.	Soybean	70.00	70.00	18.31	18.31	18.50	18.50	1693.00	1693.00
18.	Sunflower	98.00	98.00	8.00	8.00	18.53	18.53	2600.00	2600.00
19.	Tomato	100.00	100.00	4.90	4.90	9.54	9.54	1444.00	1444.00
20.	Parthenium	79.00	0.00	5.10	0.00	4.10	0.00	727.00	0.00

*Mean of four replications. UT – Untreated; T – Treated

Outputs

Table 83 : Effect of culture filtrates of *F. pallidoroseum* on seed germination and seedling vigour of host crops under *in vitro* condition

S. No	Host tested	Seed germination* (%)		Shoot length* (cm)		Root length* (cm)		Vigour Index*	
		UT	T	UT	T	UT	T	UT	T
1.	Beet root	100.00	100.00	7.55	7.54	7.75	7.73	1530.00	1527.00
2.	Bengal gram	84.00	84.00	4.70	4.70	14.70	14.69	1630.00	1629.00
3.	Bhendi	100.00	100.00	14.83	14.82	13.75	13.73	2858.00	2855.00
4.	Black gram	100.00	100.00	20.31	20.30	17.65	17.63	3796.00	3793.00
5.	Chilli	94.00	94.00	2.84	2.82	5.78	5.75	810.00	806.00
6.	Cowpea	95.00	95.00	25.80	25.80	20.00	20.00	4351.00	4351.00
7.	Cumbu	96.00	96.00	16.86	16.84	15.80	15.80	3135.00	3133.00
8.	French bean	94.00	94.00	20.04	20.02	19.50	19.50	3717.00	3715.00
9.	Green gram	100.00	100.00	21.11	21.09	17.80	17.80	3891.00	3889.00
10.	Groundnut	98.00	98.00	14.12	14.11	15.51	15.50	2904.00	2902.00
11.	Maize	100.00	100.00	23.00	23.00	29.00	29.00	5200.00	5200.00
12.	Peas	55.00	55.00	11.11	11.10	12.50	12.50	1181.00	1181.00
13.	Pumpkin	82.00	82.00	9.41	9.40	22.61	22.60	2626.00	2624.00
14.	Safflower	50.00	50.00	10.17	10.16	12.11	12.10	1025.00	1024.00
15.	Sesame	100.00	100.00	7.00	7.00	9.82	9.80	1682.00	1680.00
16.	Sorghum	98.00	98.00	18.42	18.41	20.52	20.51	3816.00	3814.00
17.	Soybean	70.00	70.00	18.31	18.30	18.50	18.50	1693.00	1693.00
18.	Sunflower	98.00	98.00	8.00	8.00	18.53	18.52	2600.00	2599.00
19.	Tomato	100.00	100.00	4.90	4.89	9.54	9.53	1444.00	1442.00
20.	Parthenium	79.00	0.00	5.10	0.00	4.10	0.00	727.00	0.00

*Mean of four replications. UT – Untreated; T – Treated

Table 84 : Effect of *Oidium parthenii* on cultivated crops under *in vitro* condition

S. No	Host tested	Seed germination* (%)		Shoot length* (cm)		Root length* (cm)		Vigour Index*	
		UT	T	UT	T	UT	T	UT	T
1.	Beet root	100.00	100.00	7.56	7.54	7.73	7.72	1529.00	1526.00
2.	Bengal gram	84.00	84.00	4.70	4.69	14.68	14.66	1628.00	1625.00
3.	Bhendi	100.00	100.00	14.82	14.81	13.74	13.72	2856.00	2853.00
4.	Black gram	100.00	100.00	20.32	20.30	17.64	17.63	3796.00	3793.00
5.	Chilli	94.00	94.00	2.82	2.81	5.75	5.74	806.00	804.00
6.	Cowpea	95.00	95.00	25.77	25.76	20.00	20.00	4348.00	4347.00
7.	Cumbu	96.00	96.00	16.84	16.83	15.79	15.78	3132.00	3031.00
8.	French bean	94.00	94.00	20.00	20.00	19.52	19.51	3715.00	3714.00
9.	Green gram	100.00	100.00	21.00	21.00	17.81	17.80	3881.00	3880.00
10.	Groundnut	98.00	98.00	14.11	14.11	15.50	15.49	3882.00	3881.00
11.	Maize	100.00	100.00	23.01	23.00	29.00	29.00	5201.00	5200.00
12.	Peas	55.00	55.00	11.11	11.10	12.50	12.48	1181.00	1179.00
13.	Pumpkin	82.00	82.00	9.41	9.40	22.60	22.60	2625.00	2624.00
14.	Safflower	50.00	50.00	10.15	10.13	12.10	12.10	1024.00	1023.00
15.	Sesame	100.00	100.00	7.02	7.01	9.81	9.80	1683.00	1681.00
16.	Sorghum	98.00	98.00	18.40	18.38	20.51	20.50	3814.00	3810.00
17.	Soybean	70.00	70.00	18.30	18.29	18.48	18.48	1692.00	1691.00
18.	Sunflower	98.00	98.00	8.00	8.00	18.53	18.52	2600.00	2599.00
19.	Tomato	100.00	100.00	4.91	4.90	9.54	9.53	1445.00	1443.00
20.	Parthenium	79.00	0.00	5.10	0.00	4.10	0.00	727.00	0.00

*Mean of four replications. UT – Untreated; T – Treated

Outputs

Forty-five day-old crop plants exhibited an immune reaction to the fungus cultures and culture filtrates of *L. theobromae* and *F. pallidoroseum* under *in vitro* condition. On the other hand, most of the crop plants at 15, 30 and 60 days old and the detached leaves showed a hypersensitive reaction to *L. theobromae*. Except for beet root, bhendi, chilli, safflower, sesame, sunflower and tomato, all the other crop plants showed an immune reaction to *F. pallidoroseum* at 15-60 days (Tables 85 & 86).

Table 85 : Reaction of cultivated crops to *L. theobromae* under *in vitro* condition

S.No.	Host tested	Fungal culture*					Culture filtrate*				
		Reaction of detached leaves	Reaction of host plants at				Reaction of detached leaves	Reaction of host plants at			
			15 DAS	30 DAS	45 DAS	60 DAS		15 DAS	30 DAS	45 DAS	60 DAS
1.	Beet root	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
2.	Bengal gram	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
3.	Bhendi	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
4.	Black gram	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
5.	Chilli	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
6.	Cowpea	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
7.	Cumbu	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	
8.	French bean	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
9.	Green gram	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
10.	Groundnut	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
11.	Maize	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	
12.	Peas	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	
13.	Pumpkin	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
14.	Safflower	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
15.	Sesame	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
16.	Sorghum	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	
17.	Soybean	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
18.	Sunflower	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
19.	Tomato	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^a	I ^b	
20.	Parthenium	HS	HS	HS	MS	HS	HS	HS	MS	HS	

* Mean of three replication

I^a – no viable symptom; I^b – Hypersensitive injury by the pathogen limited to small non damaging burning of leaves (flecking); SS – slightly susceptible (1-25% leaf area damaged); MS – Moderately susceptible (26-75% leaf area damaged) and HS – Highly susceptible (>75% leaf area damaged)

Table 86 : Reaction of cultivated crops to *F. pallidoroseum* under *in vitro* condition

S.No.	Host tested	Fungal culture*					Culture filtrate*				
		Reaction of detached leaves	Reaction of host plants at				Reaction of detached leaves	Reaction of host plants at			
			15 DAS	30 DAS	45 DAS	60 DAS		15 DAS	30 DAS	45 DAS	60 DAS
1.	Beet root	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^b	I ^a	I ^b
2.	Bengal gram	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
3.	Bhendi	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^b	I ^a	I ^b
4.	Black gram	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
5.	Chilli	I ^b	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^b	I ^a	I ^b
6.	Cowpea	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
7.	Cumbu	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
8.	French bean	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
9.	Green gram	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
10.	Groundnut	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
11.	Maize	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a

Outputs

S.No.	Host tested	Fungal culture*				Culture filtrate*					
		Reaction of detached leaves	Reaction of host plants at				Reaction of detached leaves	Reaction of host plants at			
			15 DAS	30 DAS	45 DAS	60 DAS		15 DAS	30 DAS	45 DAS	60 DAS
12.	Peas	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
13.	Pumpkin	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
14.	Safflower	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^b	I ^b	I ^a	I ^b
15.	Sesame	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^b	I ^b	I ^a	I ^b
16.	Sorghum	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
17.	Soybean	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
18.	Sunflower	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^b	I ^b	I ^a	I ^b
19.	Tomato	I ^b	I ^b	I ^a	I ^b	I ^b	I ^b	I ^b	I ^b	I ^a	I ^b
20.	Parthenium	HS	HS	HS	MS	HS	HS	HS	HS	MS	HS

* Mean of three replication

I^a – no viable symptom; I^b – Hypersensitive injury by the pathogen limited to small non damaging burning of leaves (flecking); SS – slightly susceptible (1-25% leaf area damaged); MS – Moderately susceptible (26-75% leaf area damaged) and HS – Highly susceptible (>75% leaf area damaged)

Efficacy of talc formulation of selected pathogenic isolates Seed germination assay under in vitro conditions

All the crop plants (15 day-old) were free from infection of *L. theobromae* and *F. pallidoroseum* under *in vivo* condition (Table 87).

Table 87 : Reaction of cultivated crops to selected pathogenic isolates from *Parthenium hysterophorus* under *in vivo* condition

S.No.	Host tested	Control	Symptom expression with*			
			<i>L. theobromae</i>		<i>F. pallidoroseum</i>	
			Fungus	Culture	Fungus	Culture
1.	Beet root	I ^a	I ^a	I ^a	I ^a	I ^a
2.	Bengal gram	I ^a	I ^a	I ^a	I ^a	I ^a
3.	Bhendi	I ^a	I ^a	I ^a	I ^a	I ^a
4.	Black gram	I ^a	I ^a	I ^a	I ^a	I ^a
5.	Chilli	I ^a	I ^a	I ^a	I ^a	I ^a
6.	Cowpea	I ^a	I ^a	I ^a	I ^a	I ^a
7.	Cumbu	I ^a	I ^a	I ^a	I ^a	I ^a
8.	French bean	I ^a	I ^a	I ^a	I ^a	I ^a
9.	Green gram	I ^a	I ^a	I ^a	I ^a	I ^a
10.	Groundnut	I ^a	I ^a	I ^a	I ^a	I ^a
11.	Maize	I ^a	I ^a	I ^a	I ^a	I ^a
12.	Peas	I ^a	I ^a	I ^a	I ^a	I ^a
13.	Pumpkin	I ^a	I ^a	I ^a	I ^a	I ^a
14.	Safflower	I ^a	I ^a	I ^a	I ^a	I ^a
15.	Sesame	I ^a	I ^a	I ^a	I ^a	I ^a
16.	Sorghum	I ^a	I ^a	I ^a	I ^a	I ^a
17.	Soybean	I ^a	I ^a	I ^a	I ^a	I ^a
18.	Sunflower	I ^a	I ^a	I ^a	I ^a	I ^a
19.	Tomato	I ^a	I ^a	I ^a	I ^a	I ^a
20.	Parthenium	I ^a	HS	HS	HS	HS

* Mean of three replications

I^a – no viable symptom; I^b – Hypersensitive injury by the pathogen limited to small non damaging burning of leaves (flecking); SS – slightly susceptible (1-25% leaf area damaged); MS – Moderately susceptible (26-75% leaf area damaged) and HS – Highly susceptible (>75% leaf area damaged)

Outputs

On crop plants and their germination

The powdery mildew pathogen, *Oidium parthenii*, did not cause infection on any the crop plants tested (Table 88).

Table 88 : Reaction of cultivated crops to *Parthenium hysterophorus* powdery mildew under *in vitro* and *in vivo* conditions

S.No.	Host tested	Detached Leaves*	<i>In vitro</i> reaction of *				<i>In vivo</i> reaction of host
			Host plants at				
			15 DAS	30 DAS	45 DAS	60 DAS	
1.	Beet root	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
2.	Bengal gram	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
3.	Bhendi	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
4.	Black gram	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
5.	Chilli	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
6.	Cowpea	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
7.	Cumbu	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
8.	French bean	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
9.	Green gram	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
10.	Groundnut	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
11.	Maize	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
12.	Peas	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
13.	Pumpkin	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
14.	Safflower	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
15.	Sesame	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
16.	Sorghum	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
17.	Soybean	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
18.	Sunflower	I ^a	I ^a	I ^a	I ^a	I ^a	I ^a
19.	Tomato	I ^a	I ^a	I ^a	I ^a	I ^a	I ^b
20.	Parthenium	MS	MS	HS	HS	HS	HS

* Mean of three replications

I^a – no viable symptom; I^b – Hypersensitive injury by the pathogen limited to small non damaging burning of leaves (flecking); SS – slightly susceptible (1-25% leaf area damaged); MS – Moderately susceptible (26-75% leaf area damaged) and HS – Highly susceptible (>75% leaf area damaged)

Mass multiplication of selected pathogenic isolates

All the crops recorded maximum per cent seed germination and an immune reaction with talc formulation of *L. theobromae* and *F. pallidoroseum*. *P. hysterophorus* showed a total inhibition of seed germination and maximum foliar damage 15 days after treatment (Tables 89 & 90).

Table 89 : *In vitro* studies of a talc formulation of selected fungal isolates on crop seed germination

S.No.	Host tested	Seed germination (%)*		
		Untreated	Treated	
			<i>L. theobromae</i>	<i>F. pallidoroseum</i>
1.	Beet root	100.00	100.00	100.00
2.	Bengal gram	84.00	84.00	84.00
3.	Bhendi	100.00	100.00	100.00
4.	Black gram	100.00	100.00	100.00
5.	Chilli	94.00	94.00	94.00
6.	Cowpea	95.00	95.00	95.00
7.	Cumbu	96.00	96.00	96.00
8.	French bean	94.00	94.00	94.00
9.	Green gram	100.00	100.00	100.00

Outputs

S.No.	Host tested	Seed germination (%)*		
		Untreated	Treated	
			<i>L. theobromae</i>	<i>F.pallidoroeseum</i>
10.	Groundnut	98.00	98.00	98.00
11.	Maize	100.00	100.00	100.00
12.	Peas	55.00	55.00	55.00
13.	Pumpkin	82.00	82.00	82.00
14.	Safflower	50.00	50.00	50.00
15.	Sesame	100.00	100.00	100.00
16.	Sorghum	98.00	98.00	98.00
17.	Soybean	70.00	70.00	70.00
18.	Sunflower	98.00	98.00	98.00
19.	Tomato	100.00	100.00	100.00
20.	Parthenium	79.00	0.00	0.00

*Mean of four replications

Table 90 : Microplot studies of talc formulations of selected fungal isolates on crop seed germination and symptom expression under *in vivo* condition

S.No.	Host tested	Soil application*			Foliar spray*		
		Seed germination (%)			Symptom expression with		
		Control	<i>L. theobromae</i>	<i>F.pallidoroeseum</i>	Control	<i>L. theobromae</i>	<i>F.pallidoroeseum</i>
1.	Beet root	88.00	88.00	88.00	I ^a	I ^a	I ^a
2.	Bengal gram	80.00	80.00	80.00	I ^a	I ^a	I ^a
3.	Bhendi	85.00	85.00	85.00	I ^a	I ^a	I ^a
4.	Black gram	88.00	88.00	88.00	I ^a	I ^a	I ^a
5.	Chilli	90.00	90.00	90.00	I ^a	I ^a	I ^a
6.	Cowpea	90.00	90.00	90.00	I ^a	I ^a	I ^a
7.	Cumbu	91.00	91.00	91.00	I ^a	I ^a	I ^a
8.	French bean	88.00	88.00	88.00	I ^a	I ^a	I ^a
9.	Green gram	88.00	88.00	88.00	I ^a	I ^a	I ^a
10.	Groundnut	90.00	90.00	90.00	I ^a	I ^a	I ^a
11.	Maize	94.00	94.00	94.00	I ^a	I ^a	I ^a
12.	Peas	46.00	46.00	46.00	I ^a	I ^a	I ^a
13.	Pumpkin	70.00	70.00	70.00	I ^a	I ^a	I ^a
14.	Safflower	40.00	40.00	40.00	I ^a	I ^a	I ^a
15.	Sesame	94.00	94.00	94.00	I ^a	I ^a	I ^a
16.	Sorghum	92.00	92.00	92.00	I ^a	I ^a	I ^a
17.	Soybean	60.00	60.00	60.00	I ^a	I ^a	I ^a
18.	Sunflower	80.00	80.00	80.00	I ^a	I ^a	I ^a
19.	Tomato	88.00	88.00	88.00	I ^a	I ^a	I ^a
20.	Parthenium	78.00	00.00	00.00	I ^a	HS	HS

* Mean of three replications.

3. Biocontrol agent(s) identified and screened for release in India

Project Directorate of Biological Control, Bangalore, India

Pathogenicity screening

Out of the fungal species tested from diseased *P. hysterophorus* plants collected in various districts within Karnataka State, only a small number of them turned out to be highly virulent in the course of preliminary trials. In particular, *Cryptosporiopsis* sp. (see pure culture, Fig. 12), a leaf-spotting pathogen, showed the most desirable characteristics for development as a mycoherbicide for Parthenium weed. The most pathogenic isolate [WF(Ph)3] (IMI 378270) of the fungus, collected in Mysore district, was selected for further study.



Fig. 12

Although there are at least 11 described species of *Cryptosporiopsis* (Sutton, 1980), no species has been so far described on Parthenium weed and there is only one previous record at CABI Bioscience for this host from Tamil Nadu, India. A dried preserved sample of this *P. hysterophorus* isolate has been placed in the IMI dried reference collection. The mycelium of the present isolate is immersed, branched, septate, hyaline to pale brown and produces no conidiophores.

Conidia are hyaline, thin-walled, guttulate or eguttulate, smooth, straight, apex obtuse, base abruptly tapered to a distinct truncate scar. Although Dr G. Kinsey, who identified the pathogen, opined that it is not possible to provide a species name until a modern revision of the genus is available, based on the literature search and on the results obtained in investigations with this fungus the tentative name *Cryptosporiopsis parthenii* sp. nov. can be proposed for this undescribed fungus.

Susceptibility of different *Parthenium hysterophorus* populations to *Cryptosporiopsis* sp.

All the *P. hysterophorus* isolates tested were susceptible to *Cryptosporiopsis* sp. (Table 91). Whereas the maximum percentage of susceptible plants was 100, the minimum was 70. The samples collected in Bangalore Rural, Mandya and Mysore districts were all susceptible. However, the study indicated there are not many resistant populations of Parthenium weed as far as the disease caused by *Cryptosporiopsis* sp. is concerned. However, more studies are needed with several isolates of the pathogen to arrive at a proper conclusion.

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Table 91 : Reaction of *Parthenium hysterophorus* populations from various districts of Karnataka to *Cryptosporiopsis* sp.

District (locality)*	No. of plants tested	Susceptible plants (%)
Bangalore Urban (Hebbal, Bangalore City)	50	92.00
Bangalore Rural (Hoskote)	50	94.00
Mandya	25	100.00
Mysore	25	100.00
Hassan	20	100.00
Tumkur	10	90.00
Kolar	8	75.00
Chitradurga	10	70.00
Chikmagalur	10	70.00
Shimoga	15	86.67
Davangere	12	91.67
Bellary	10	70.00
Dharwad	10	70.00
Raichur	10	80.00
Gulbarga	8	87.50
Bidar	15	93.33

* District and locality names are the same except where mentioned.

Host range screening

Species of *Cryptosporiopsis* have been reported as pathogens of only a few plant species in India: Leaf spot (*C. eucalypti*) of eucalyptus; anthracnose (*C. curvispora*), perennial canker (*C. perennans*) and storage/post-harvest rot (*Cryptosporiopsis* sp.) of apple; blight (*Cryptosporiopsis* sp.) of almond; and leaf spot (*C. citri*), of citrus are the diseases reported to be caused by these pathogens in India. Except for almond, all the other hosts (Table 92) were tested against the *P. hysterophorus* isolate of the pathogen. Injured apple fruits developed rotting symptoms after three days of inoculation with the mycelial inoculum of the pathogen. However, no definite damage was caused by the pathogen to uninjured apple fruits. A comparative study of the *P. hysterophorus* and apple strains would throw more light on cross-infection.

A total of 83 cultivars of economically important plants in 10 families, including Compositae, Papilionaceae, Solanaceae, Cucurbitaceae, Cruciferae, Malvaceae, Amaranthaceae, Chenopodiaceae, Umbelliferae and Poaceae were screened against *Cryptosporiopsis* sp. and found to be immune (Table 93). Both mycelial and conidial inoculations did not incite disease on any of the plant species screened. The preliminary host-range testing determined that all the crops, including related species of the Compositae, especially the 8 sunflower cultivars, were not susceptible to *Cryptosporiopsis* sp. In spite of the positive reaction of injured apples to the pathogen, the specificity of the isolate WF(Ph)3 may be sufficient for its possible use as a mycoherbicide for *P. hysterophorus*. However, the host range test needs to be wider and more systematic to confirm this.

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Table 92 : Host-specificity screening for *Cryptosporiopsis* sp.: (A) Reaction of plant species reported to be infected by different species of the genus.

Plant species	Family	Part tested	Reaction*
Apple (<i>Malus pumila</i> Mill.)	Rosaceae	Fruit	@
Blue-gum (<i>Eucalyptus globulus</i> Labill.)	Myrtaceae	Leaf	-
Lime (<i>Citrus aurantifolia</i> (Christm.) Swingle)	Rutaceae	Leaf	-
Pomelo (<i>Citrus decumana</i> L.)	Rutaceae	Leaf	-

@ Only injured fruits exhibited rotting symptoms after three days of inoculation.

* Results from tests with both detached leaves and intact plants.

Table 93 : Host-specificity screening for *Cryptosporiopsis* sp.: (B) Reaction of other economically important non-target plant species.*

Family	Test plant species / Cultivars	Reaction
Compositae	Sunflower (<i>Helianthus annuus</i> L.) KBSH1, Morden, MSF17, EC68414, PAC1091, GAUSUF15, Arun, SH3322	-
	Niger-seed (<i>Guizotia abyssinica</i> Cass.) Unknown cultivar	-
	Calendula (<i>Calendula officinalis</i> L.) Touch red-orange, Touch red-yellow	-
	Aster (<i>Aster amellus</i> L.) Pot 'n' patio, Powder puffs mix	-
	Zinnia (<i>Zinnia elegans</i> Jacq.) Cany cane mix, Pulcino mix	-
	Cosmos (<i>Cosmos bipinnatus</i> Cav.) Sensation mix, Sunny red	-
Papilionaceae	Groundnut (<i>Arachis hypogaea</i> L.) JL24	-
	Cowpea (<i>Vigna unguiculata</i> (L.) Walp.) C152, Arka Garima	-
	Blackgram (<i>Vigna mungo</i> (L.) Hepper) T9, LBG402	-
	French bean (<i>Phaseolus vulgaris</i> L.) Arka Komal	-
	Cluster bean (<i>Cyamopsis tetragonoloba</i> (L.) Taub. R8BHSC10	-
Solanaceae	Brinjal (<i>Solanum melongena</i> L.) Arka Nidhi, Arka Sheel, Bhagyamathi, Shyamala, Pusa Purple Long, Pusa Purple Round, Sourabha, PPL, CVK	-
	Tomato (<i>Lycopersicon esculentum</i> Mill.) Arka Saurabh, Pusa Ruby, Marutham, Dwarf Hybrid, S22	-
	Chillies (<i>Capsicum annuum</i> L.) LCG4, LCG5, LCA206, LCA235, LCA960, GA, X235	-
	Tobacco (<i>Nicotiana tabacum</i> L.) Unknown cultivar	-
Cucurbitaceae	Pumpkin (<i>Cucurbita moschata</i> (Duch.) Poir. Arka Chandan, Arka Surya	-
	Bottle gourd (<i>Lagenaria siceraria</i> (Molina)) Arka Bahar, PSPL	-
	Musk melon (<i>Cucumis melo</i> L.) Arka Jeet	-
	Water melon (<i>Citrullus lanatus</i> (Thunb.) Mansf.) Madhu	-
	Cucumber (<i>Cucumis sativus</i> L.) Green Long, Priya	-

Outputs

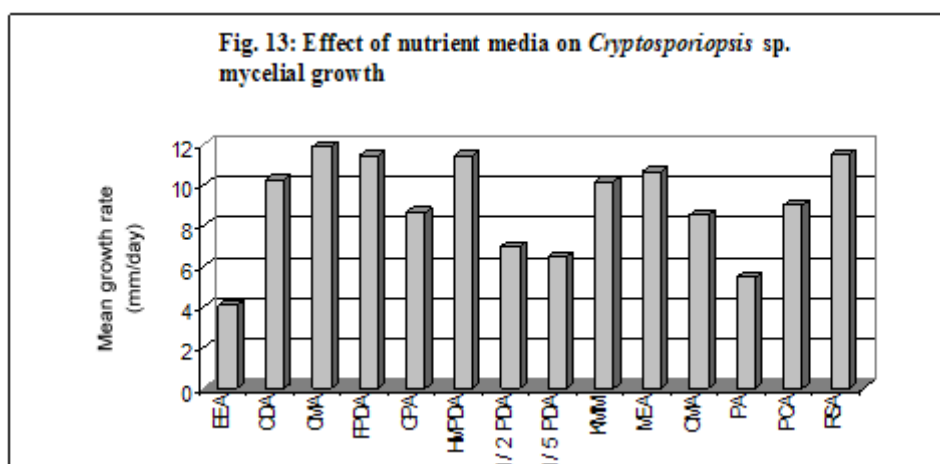
Family	Test plant species / Cultivars	Reaction
Cruciferae	Radish (<i>Raphanus sativus</i> L.) Arka Nishant, Pusa Chetaki, No.7, Pusa Cheti	-
	Cabbage (<i>Brassica oleraceae</i> L.) Unnati	-
	Knol Khol (<i>Brassica caulorapa</i> L.) EW	-
Malvaceae	Okra (<i>Abelmoschus esculentus</i> L.) Arka Abhay, Varsha	-
Amaranthaceae	Amaranthus (<i>Amaranthus viridis</i> L.) Arka Suguna	-
Chenopodiaceae	Beet (<i>Beta vulgaris</i> L.) Ruby Queen	-
Umbelliferae	Carrot (<i>Daucus carota</i> L.) Early Nantes	-
Poaceae	Finger millet (<i>Eleusine coracana</i> (L.) Gaertn HR911, Indaf9, GPU28	-
	Corn (<i>Zea mays</i> L.) Ganga11, C6, Himalaya23, Kanchan	-
	Rice (<i>Oryza sativa</i> L.) Rasi, Mangala, Tellahamsa, Jaya, IR64, Mandya Vijaya, Jyothi, T(N)1, Vikramarya, IET9994, IET8585, BPT5204	-

* Results from tests with both detached leaves and intact plants.

Growth studies

Effect of nutrient media on mycelial growth and conidial production

Variations in radial growth were evident on the different nutrient media (Fig 13). Whereas the maximum growth (11.51 mm/day) occurred on RSA, the minimum growth (4.06 mm/day) of the fungus was recorded on BEA. The growth rate of the fungus on both the versions of PDA, viz. HMPDA and FPDA, which are very commonly used, was found to be the same (11.43 mm/day). There was no conidial production on any of these media. The growth of the fungus was observed to be appressed on 1/2 and 1/5 PDA. Similarly, aerial growth was not observed on PCA, BEA and PA.



Effect of addition of *Parthenium hysterophorus* leaf decoction and yeast extract to nutrient media on mycelial growth and conidial production

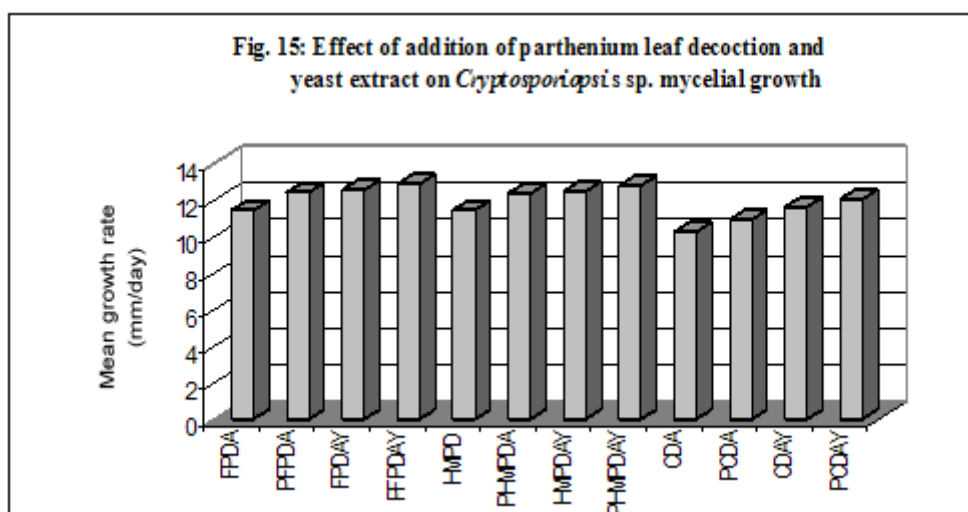
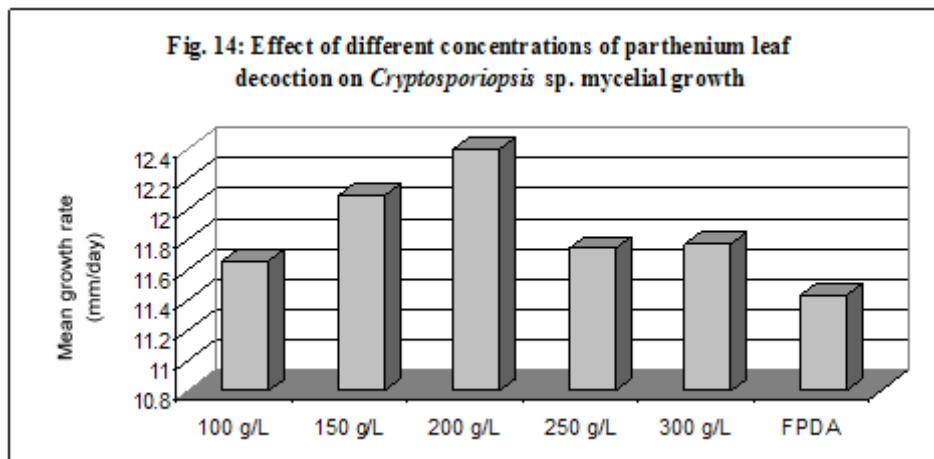
Sporulation was checked employing the same method as described above. (Fig. 14). *Cryptosporiopsis* sp. were determined following the procedures outlined above. Both *P.*

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hysterophorus leaf decoction and yeast extract showed positive effect on the growth of the fungus as additives to the three media tested (Fig. 15).

Supplementing the medium with *P. hysterophorus* leaf decoction stimulated the growth rate of *Cryptosporiopsis* sp. Among the five concentrations tested, 200g/L was the best in terms of the rate of growth (12.40 mm/day). Although there was an increasing trend observed from 100 to 200g, still higher concentrations actually depressed the growth rate of the fungus (11.77 mm/day).

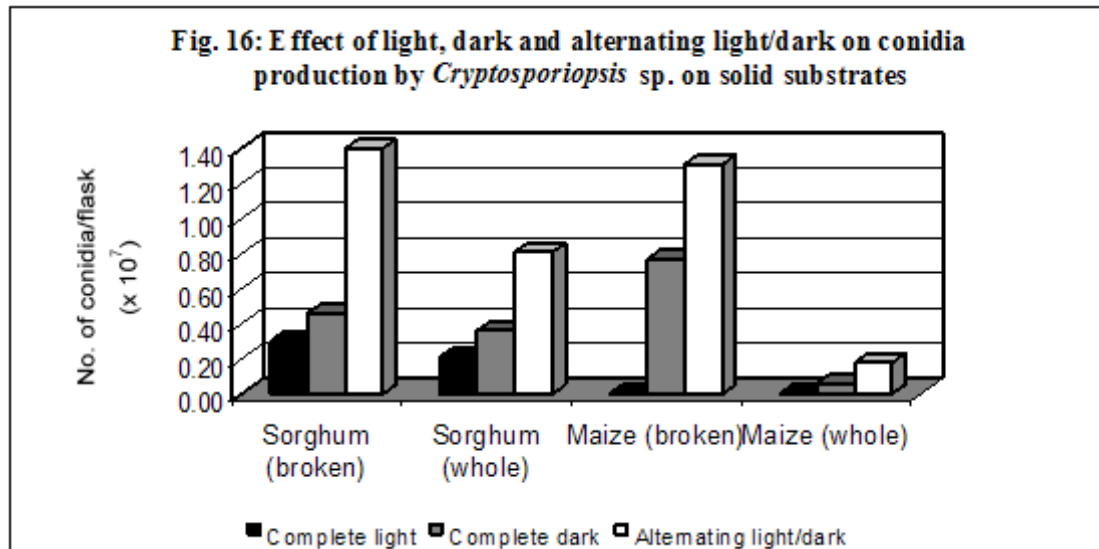
Almost the same pattern of growth rates was observed on all the supplemented media. Yeast was the better additive as evident from the results. The maximum growth (12.86 mm/day) of the fungus was obtained on PFPDAY. The second best was PHMPDAY (12.77 mm/day). Growth was also enhanced on CDA due to the addition of either leaf extract (10.89 mm/day), or yeast (11.6 mm/day) or both (12.03 mm/day), clearly indicating the growth promoting effects of these supplements. However, neither *P. hysterophorus* leaf decoction nor yeast helped in conidial formation.



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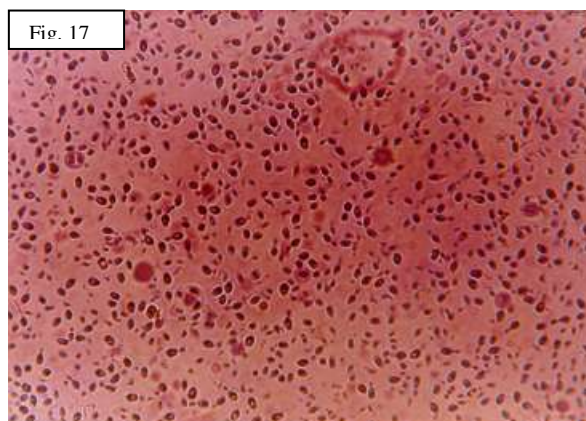
Effect of solid substrates on conidial production

Not all the solid substrates could induce conidial production by *Cryptosporiopsis* sp. (Fig. 16). Differences were observed in conidial production because of the light regimes followed. The best substrate was found to be broken sorghum grain. Under alternating light/dark, the maximum conidial production was observed (1.40×10^7). Even though under complete darkness and alternating light/dark conditions, both broken and whole grains supported conidial production, under complete light situation no sporulation occurred. Complete darkness was better than complete light in terms of conidial production. Except for sorghum and maize, none of the other substrates, viz. pearl millet, oats, barley, soybean, greengram, wheat, rice, finger millet and wheat bran induced sporulation.



Fermentation studies

MYM was found to be superior to PDB in terms of biomass as well as conidial production. After 7 days of fermentation (Fig. 17 shows conidia obtained after 7 days of fermentation) in



PDB and MYM, the conidial number obtained was 6.52×10^7 and 8.75×10^7 per every mL of the medium, respectively. Differences were evident between the two media in the total biomass (dry weight) yielded. Whereas PDB could produce an average of 25.0 g, MYM was able to produce 30.5 g in each run. A huge difference in dimensions were also noticed between the conidia produced in the two media. Conidia in PDB were found to be significantly bigger, measuring $10.15 \mu \times 4.50 \mu$, whereas, the conidia in MYM were smaller and measured

$4.60 \mu \times 2.82 \mu$. However, since it is known that there exist both macro- and micro-conidia in certain species of *Cryptosporiopsis*, it can be concluded that whereas PDB favoured the production of the former, MYM aided in the production of the latter.

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Evaluation of *Cryptosporiopsis* sp. as a mycoherbicide

Effect of conidial density on disease development

The severity of disease, expressed as necrotic leaf area, was dependent on the density of conidia applied to the plants (Table 94). Whereas the highest necrotic leaf area (98.80%) was obtained on plants sprayed with 10^{10} MYM conidia/mL, the lowest (35.00%) was recorded on plants treated with 10^5 PDB conidia/mL. The average necrotic leaf area over all the concentrations tested was 73.47%. At all the concentrations tested, MYM conidia performed better than PDB conidia. The average necrotic leaf areas with PDB and MYM conidia were 67.45% and 79.49%, respectively. With both the types of conidia, more than 90% severity of disease was achieved at a minimum concentration of 10^8 .

Table 94 : Effect of different conidial densities on the disease caused by *Cryptosporiopsis* sp. Produced on two media

Conidial density (conidia/mL)	Necrotic leaf area (%)	
	PDB	MYM
10^{10}	95.40 (77.63)	98.80 (85.12)
10^9	93.42 (75.14)	97.60 (81.12)
10^8	90.74 (72.31)	96.73 (79.66)
10^7	52.11 (46.21)	86.01 (68.04)
10^6	38.07 (38.09)	55.60 (48.22)
10^5	35.00 (36.27)	42.18 (40.50)
CD		
5%	1.04	2.97
1%	1.42	4.05

Note: Figures in parentheses are angular-transformed values.

Effect of plant growth stage on disease development

All the growth stages of *P. hysterophorus* were susceptible to the pathogen, younger plants being more susceptible than older ones (Table 95). The necrotic leaf area was greater than 65% in all the cases, the average being 86.17%. Plants at growth stages 3-5 and 6-9 were significantly more susceptible than those at growth stage 10-13. PDB and MYM conidia produced 94.09% and 98.40% necrotic area, respectively at the 3-5 growth stage. MYM conidia were found to be superior to PDB conidia in disease production at all stages of inoculation.

Table 95 : Effect of *Parthenium hysterophorus* growth stage on the disease caused by *Cryptosporiopsis* sp. grown on two media

Growth stage (No. of leaves)	Necrotic leaf area (%)	
	PDB	MYM
3-5	94.09 (75.96)	98.40 (83.51)
6-9	87.67 (69.45)	90.96 (72.51)
10-13	66.01 (54.34)	79.90 (63.37)
CD		
5%	1.71	3.30
1%	2.49	4.81

Note: Figures in parentheses are angular-transformed values.

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Effect of surfactants on the pathogenicity of *Cryptosporiopsis* sp. to *Parthenium hysterophorus*

Significant differences were evident among all the treatments in terms of pathogenicity of *Cryptosporiopsis* sp. (Table 96). The best of all the surfactants was Tween 80, which caused a necrotic leaf area of 94.80%. However Tween 20 (94.71%) did not differ significantly from the former. The least effective surfactant was Triton X-100 with a necrotic leaf area of 78.00%. Overall, the addition of surfactants resulted in increased pathogenicity of the fungus to *P. hysterophorus*.

Table 96 : Effect of certain surfactants on the pathogenicity of *Cryptosporiopsis* sp. to *Parthenium hysterophorus*

Surfactant	Necrotic leaf area (%)
Tween 20	94.71 (76.71)
Tween 80	94.80 (76.83)
Glycerol	87.43 (69.26)
Triton X-100	78.00 (62.09)
Control (sterile water)	75.00 (60.00)
CD	
5%	2.00
1%	2.74

Note: Figures in parentheses are angular-transformed values

Effect of hydrophilic substances on the pathogenicity of *Cryptosporiopsis* sp. to *Parthenium hysterophorus*

The performance of *Cryptosporiopsis* sp. was observed to be affected by the addition of hydrophilic substances to the inoculum (Table 97). The best among them was gum arabic, which produced the maximum necrotic leaf area of 97.16%. Sodium alginate (84.80%) was next only to gum arabic in increasing the pathogenicity of the fungus. However, polyacrylamide and carboxy methyl cellulose (CMC) were not able to enhance the severity of the disease brought about by *Cryptosporiopsis* sp.

Table 97: Effect of certain hydrophilic substances on the pathogenicity of *Cryptosporiopsis* sp. to *Parthenium hysterophorus*

Hydrophilic substance	Necrotic leaf area (%)
Gum arabic	97.16 (80.47)
Polyacrylamide	64.79 (53.61)
Carboxy methy cellulose	69.77 (56.65)
Sodium alginate	84.80 (67.06)
Control (sterile water)	75.00 (60.00)
CD	
5%	1.81
1%	2.49

Note: Figures in parentheses are angular-transformed values.

Outputs

3. Biocontrol agent(s) identified and screened for release in India Kurukshetra University, India

Evaluation of biocontrol agents for the management of *Parthenium hysterophorus* I Effect of light and media on growth and sporulation of *A. zinniae* and *C. partheniiphila*

It is evident from the results presented in Tables 98a and 98b that *A. zinniae* and *C. partheniiphila* showed varying growth on all the ten media tested. *A. zinniae* showed excellent growth under light on PSA followed by PDA> V₈ juice agar media. Growth was good on PeDA> PeDAY> PDAY> CDAY> MA while the growth was poorest on CDA (Table 98a & Fig. 18). However, under dark best growth of the fungus was observed on PSA> CDA> PeDA> PDAY> V₈ Juice agar> PeDAY> MA> CDAY> CDA> NA (Fig. 19). *C. partheniiphila* showed best growth on CDAY under light followed by MA> PDAY> PDA> PSA> PeDAY> NA> V₈> PeDA> CDA (Table 98b & Fig. 20). Under dark, best growth of *C. partheniiphila* was recorded on NA followed by PSA>CDAY medium (Fig. 21). The statistical analysis reveals that there was a significant difference in the growth of *A. zinniae* in light and dark conditions while in *C. partheniiphila* no significant difference was observed on different media .

Table 98a: Growth and sporulation of *Alternaria zinniae* on ten different media after 9 days post incubation

Sr No./ Media	Growth (diameter cm)		t value	(Sporulation/unit area)		t value
	Dark	Light		Dark	Light	
1. PDA	6.47±0.70	8.77±0.04	3.28*	-	1.00±0.82	1.21
2. PDAY	6.35±0.02	7.32±0.03	26.90*	-	-	
3. PeDA	6.47±0.08	8.45±0.08	17.50*	-	11.66±0.72	16.19*
4. PeDAY	5.52±0.06	7.52±0.07	21.69*	-	6.00±0.47	12.76*
5. CDA	3.13±0.03	3.78±0.09	6.85*	-	9.00±0.82	10.97*
6. CDAY	5.18±0.08	6.95±0.14	10.98*	-	-	
7. V ₈	6.33±0.04	8.60±0.04	39.77*	-	11.0±1.25	8.80*
8. MA	5.47±0.05	6.75±0.06	16.39*	-	6.33±0.72	8.79*
9. PSA	7.18±0.06	8.83±0.04	22.88*	-	3.33±0.27	12.33*
10. NA	3.30±0.08	6.63±0.27	11.83*	-	-	

* t value significant at P= 0.05; df=2

From the data it is concluded that growth of *A. zinniae* was affected by light and dark in all the media tested, light favouring the growth of the fungus. The sporulation was only seen in plates of *A. zinniae* that were kept under light, while in *C. partheniiphila* sporulation on the media PDAY, CDA, V₈ juice agar and MA was observed in both under light and dark conditions. In *A. zinniae* the best sporulation was reported on PeDA while in *C. partheniiphila* the sporulation was found best on CDA.

The results reveal that light has a stimulatory effect on growth and sporulation of *A. zinniae* while dark completely inhibits sporulation in this fungus. If we consider both the parameters, i.e. growth and sporulation which are the prerequisite of any mycoherbicide, for inoculum production, *A. zinniae* should be grown on PeDA or V₈ juice agar under light whereas *C. partheniiphila* should be grown on CDA either in light and/or dark.

Outputs

Table 98b: Growth and sporulation of *Cercospora partheniiphila* on ten different media after 9 days post incubation

Sr. No./ Media	Growth(diameter cm)		t value	Sporulation/unit area)		t value
	Dark	Light		Dark	Light	
1. PDA	2.93± 0.10	3.12± 0.06	1.63	-	-	-
2. PDAY	2.51± 0.005	3.10± 0.11	5.36*	6.00±1.25	5.33±1.19	0.27
3. PeDA	2.28± 0.08	2.40± 0.09	1.16	-	-	-
4. PeDAY	2.27± 0.06	2.82± 0.19	2.84*	-	-	-
5. CDA	1.88±0.11	2.37± 0.08	3.60*	9.33±2.88	8.67±1.44	0.20
6. CDAY	2.95±0.19	3.87± 0.19	3.42*	-	-	-
7. V ₈	2.13±0.02	2.50± 0.13	2.81*	3.33± 0.72	1.67±0.27	2.15
8. MA	2.65±0.15	3.18±0.01	3.52*	-	2.0±0.47	-
9. PSA	2.98±0.05	2.90±0.16	0.47	-	-	-
10. NA	3.63±0.36	2.52±0.10	2.90*	8.33± 2.23	-	-

* t value significant at P= 0.05; df=2

It is concluded that *C. partheniiphila* sporulates best on Czapek dox agar under dark while *A. zinniae* sporulates best on *P. hysterophorus* extract dextrose agar and V₈ juice agar media under light.

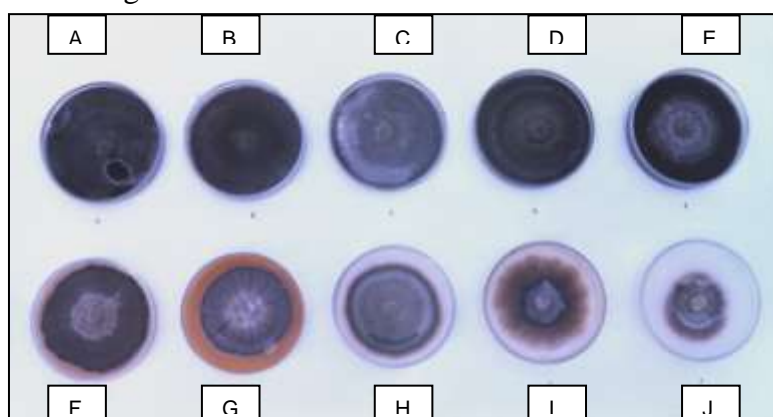


Fig. 18. Comparative growth characteristics of *A. zinniae* on various media after incubation at $25 \pm 1^\circ\text{C}$ for 9 days under light conditions (A-J). A: Potato Sucrose Agar, B: Potato Dextrose Agar, C: V₈ Juice Agar, D: Parthenium extract Dextrose Agar, E: Parthenium extract Dextrose Agar+Yeast extract, F: Potato Dextrose Agar+Yeast extract, G: Czapek Dox Agar+Yeast extract, H: Martin Agar, I: Nutrient Agar, J: Czapek Dox Agar.

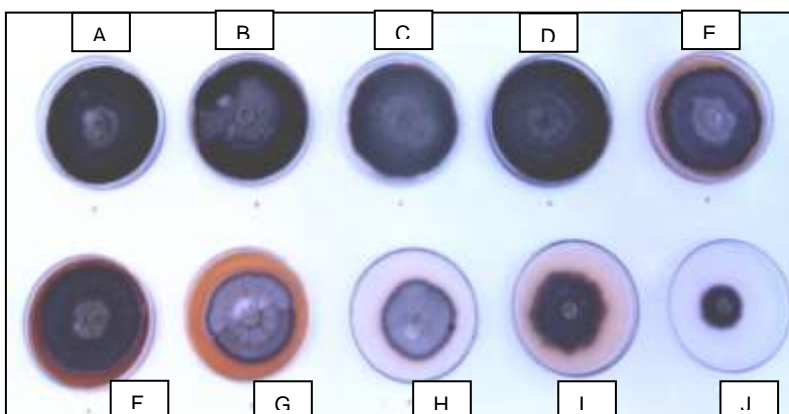


Fig. 19. Comparative growth characteristics of *A. zinniae* on various media after incubation at $25 \pm 1^\circ\text{C}$ for 9 days under dark conditions (A-J). A: Potato Sucrose Agar, B: Potato Dextrose Agar, C: Parthenium extract Dextrose Agar, D: Potato Dextrose Agar+Yeast extract, E: V₈ Juice Agar, F: Parthenium extract Dextrose Agar+Yeast extract, G: Martin Agar, H: Czapek Dox Agar +Yeast extract, I: Czapek Dox Agar, J: Nutrient Agar.

Note that the sequence of lettering follows the same pattern for Figs. 20 and 21.

Outputs

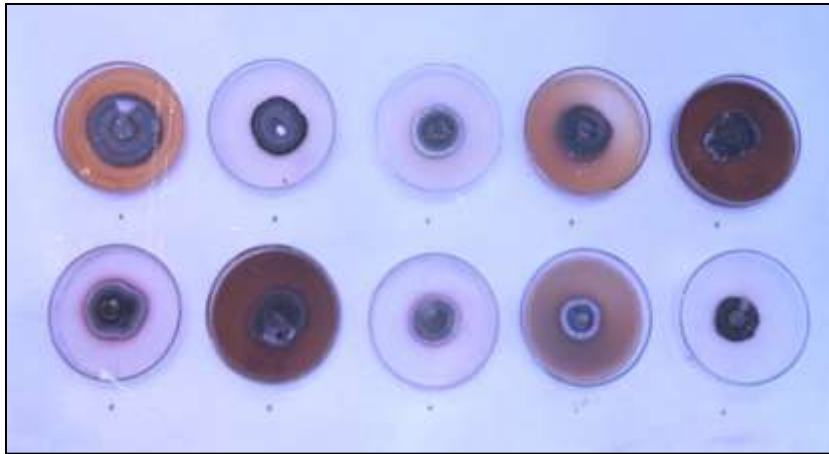


Fig. 20. Comparative growth characteristics of *C. partheniiphila* on various media after incubation at $25 \pm 1^\circ\text{C}$ for 9 days under light conditions (A-J). A: Czapek Dox Agar+Yeast extract, B: Martin Agar, C: Potato Dextrose Agar+Yeast extract, D: Potato Dextrose Agar, E: Potato Sucrose Agar, F: Parthenium extract Dextrose Agar+Yeast extract, G: Nutrient Agar, H: V_8 Juice Agar, I: Parthenium extract Dextrose Agar, J: Czapek Dox Agar.

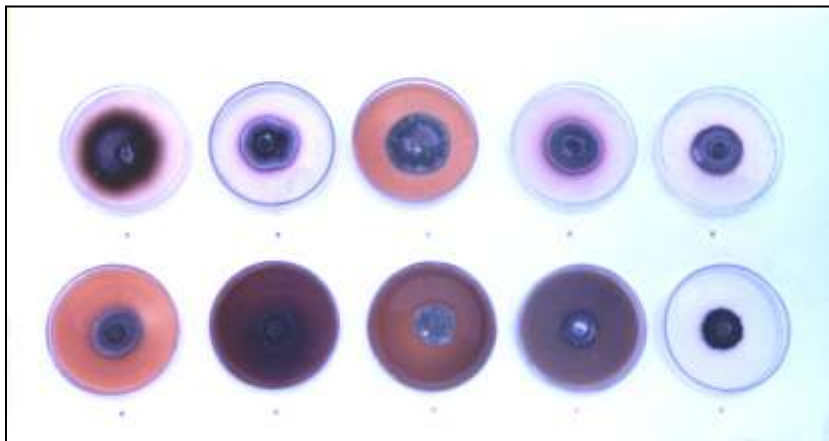


Fig. 21. Comparative growth characteristics of *C. partheniiphila* on various media after incubation at $25 \pm 1^\circ\text{C}$ for 9 days under dark conditions (A-J). A: Nutrient Agar, B: Potato Sucrose Agar, C: Czapek Dox Agar+Yeast extract, D: Potato Dextrose Agar, E: Martin Agar, F: Potato Dextrose Agar+Yeast extract, G: Parthenium extract Dextrose Agar, H: Parthenium extract Dextrose Agar +Yeast extract, I: V_8 Juice Agar, J: Czapek Dox Agar.

Outputs

Evaluation of biocontrol agents for the management of *Parthenium hysterophorus* II Host specificity of *A. zinniae* and *C. partheniiphila*

Host specificity of *A. zinniae* and *C. partheniiphila* isolated from diseased *P. hysterophorus* plants, was tested against plant species belonging to the families Solanaceae, Brassicacea (Cruciferae), Liliaceae, Asteraceae, Graminae and Papilionatae (Table 99). The plants were selected on the basis of their local economic importance.

Table 99: Response of various crops tested for susceptibility to *Alternaria zinniae* and *Cercospora partheniiphila*

Sr.No.	Crop/s	Host Response	
		<i>A. zinniae</i>	<i>C. partheniiphila</i>
1.	<i>Lycopersicon esculentum</i>	OO	OO
2.	<i>Phaseolus lunatus</i>	XX	OX
3.	<i>Solanum tuberosum</i>	OX	OO
4.	<i>Brassica oleracea</i> var. <i>capitata</i>	XX	OX
5.	<i>B. oleracea</i> var. <i>botrytis</i>	XX	OX
6.	<i>Allium sativum</i>	OO	OO
7.	<i>A. cepa</i>	OO	OO
8.	<i>Triticum aestivum</i>	OO	OO
9.	<i>Helianthus annuus</i>	OX	OO
10.	<i>Brassica campestris</i>	OX	OO

Reactions: XX=Susceptible, OX=Poor Infection, OO=No Infection

Out of the ten host plant species tested, *i.e.* tomato (*Lycopersicon esculentum*), lobia (*Phaseolus lunatus*), potato (*Solanum tuberosum*), cabbage (*Brassica oleracea* var. *capitata*), cauliflower (*B. oleracea* var. *botrytis*), garlic (*Allium sativum*), onion (*A. cepa*), wheat (*Triticum aestivum*), sunflower (*Helianthus annuus*) and mustard (*Brassica campestris*), 3 plant species *viz.* lobia, cabbage and cauliflower, were found to be susceptible to *A. zinniae*. In addition, some infection was also caused on potato, sunflower and mustard by *A. zinniae*. These results clearly indicate that *A. zinniae* has a wide host range, thus eliminating the possibility of developing it as a suitable mycoherbicide for the control of Parthenium weed.

None of the plants tested were found to be infected by *C. partheniiphila* (Table 99) thus indicating that all these crop species, namely tomato, lobia, potato, cabbage, cauliflower, garlic, onion, wheat, sunflower and mustard are immune to *C. partheniiphila*. However, some infection due to this fungus was seen in lobia, cabbage and cauliflower where the pathogen remains restricted to the penetration site and no further symptoms were produced on these hosts. The data clearly reveal the host specific nature of *C. partheniiphila*. Therefore, it seems to be safe to develop the present isolate of *C. partheniiphila* as a mycoherbicide for the control of Parthenium weed.

Evaluation of biocontrol agents for the management of *Parthenium hysterophorus* III Biocontrol potential

The pathogenicity and biocontrol potential of two fungal pathogens, were tested on *P. hysterophorus* plants grown in plastic pots. The pots contained a sand - soil mixture in a 1 : 1 ratio. The inocula of *A. zinniae* and *C. partheniiphila* were prepared on the *P. hysterophorus* extract dextrose agar and Czapek dox agar media, respectively since these were found to be best for growth and sporulation (Table 98). The conidial/mycelial suspensions for spraying on experimental plants were prepared in sterile distilled water as follows, spore suspension with and without 0.5% Tween 80. Control plants were sprayed with sterile distilled water + 0.5%

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Tween 80. Spraying was done on two types of plants, i.e. wounded (pricked) and unwounded which were kept under three different conditions : (i) uncovered; (ii) covered with polythene bags for 24 hrs only; and (iii) covered with polythene bags. Observations were made at four-day intervals for the development of the disease, i.e. onset of symptoms and percent area covered by the disease until death of the plants.

Biocontrol potential of *A. zinniae*

Fig. 22 *in vitro* *A. zinniae* disease symptoms on Parthenium leaf



Typical disease symptoms were observed on both wounded and unwounded leaves *in vitro* (Fig. 22) and *in vivo* and the pathogen was reisolated, thus confirming the pathogenicity of *A. zinniae* to *P. hysterophorus*.

P. hysterophorus leaves responded differently towards infection under different conditions.

Infection in covered pots which were artificially sprayed with inoculum of the pathogen (8×10^4 conidia+mycelium/ml), was higher than uncovered pots (Table 100a), these values were statistically significant at 0.05 level in both wounded and unwounded plants (Tables 100a & 100b). High infection in the covered pots can be due to retention of high moisture which ultimately increased the ability of conidia to germinate and infect. Moreover, *P. hysterophorus* leaves which were wounded

on the upper surface showed more infection suggesting a possible role for insects in causing wounds to allow the entry of the pathogens.

The present data also suggest that *A. zinniae* can be highly aggressive to Parthenium weed under certain conditions. The pathogen has characteristics that make it a desirable candidate as biological control agent of Parthenium weed, such as: easily cultured on natural host and hence can be mass produced at a lower cost and in a short time; easily disseminated and self-maintaining, but its broader host range (Table 99) is one of the major constraints regarding its suitability for development as mycoherbicide for controlling this weed in India.

Table 100a : Percent infection due to *Alternaria zinniae* on *Parthenium hysterophorus* leaves in experimental pots

Sr.No.	Days after inoculation	Covered		Uncovered	
		Injured	Uninjured	Injured	Uninjured
1.	4	-	-	-	-
2.	8	2.76± 0.54*	1.85± 0.50	-	-
3.	12	5.47± 1.06	3.30± 0.90	1.98± 0.79	0.32± 0.21
4.	15	19.03± 3.80	9.74± 2.76	7.88± 3.00	0.77± 0.56
5.	22		Death of the plants		

*Mean of 12 replicates

Outputs

Table 100b : Statistical analysis of percent infection of *Parthenium hysterophorus* plants by *Alternaria zinniae* 15 days post inoculation

Injured		t-value	Uninjured		t-value
covered	uncovered		covered	uncovered	
19.03± 3.8	7.88± 3.0	2.30*	9.74± 2.76	0.77± 0.56	3.18*

* Values significant at $p < .05$, $df = 11$

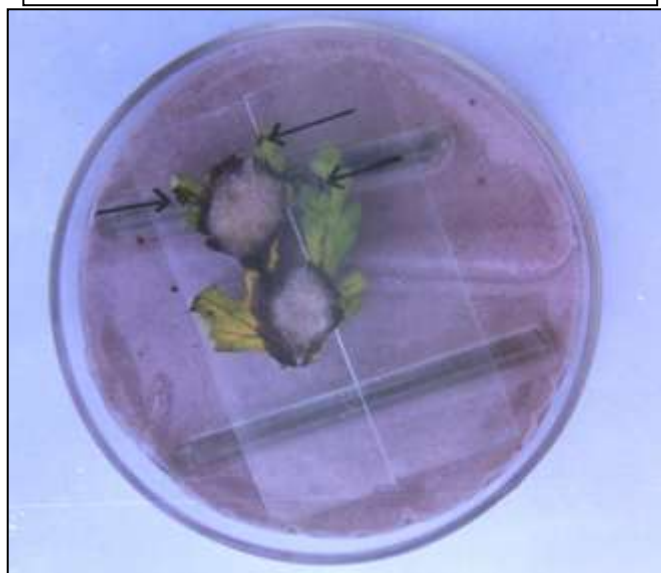
Table 100c: Statistical analysis showing the percent infection caused by *Alternaria zinniae* 15 days post inoculation between inoculated and uninoculated *Parthenium hysterophorus* plants

Treatment	Injured		Uninjured	
	Covered	Uncovered	Covered	Uncovered
Inoculated	19.03 ± 3.80	7.88 ± 3.00	9.74 ± 2.76	0.77 ± 0.56
Uninoculated	0	0.56 ± .15	0.46 ± 0.21	0.07 ± 0.05
t value	5.00*	2.43*	3.35*	1.24

* Values significant at $p < .05$, $df = 11$

Biocontrol potential of *C. partheniiphila*

Fig. 23. *In vitro* disease symptoms of *C. partheniicola* on *Parthenium* leaf



Typical disease symptoms were observed on both wounded and unwounded leaves *in vitro* (Fig. 23) and *in vivo* and the inoculated pathogen was reisolated thus confirming the pathogenicity of *Cercospora partheniiphila* to *Parthenium* weed. Pathogenicity tests conducted in the laboratory at room temperature ($28 \pm 1^{\circ}\text{C}$) showed that infection on leaves started after 3-4 days of inoculation as pin-point, light brown spots on the margins and centre of the leaves. The disease progressed with increasing incubation time and ultimately resulted in rotting of the plants within 20 days of inoculation with the pathogen.

P. hysterophorus plants responded differently towards infection under different conditions. Infection in covered pots which were artificially sprayed with the spore suspension (6×10^4 conidial+mycelial/ml) was higher than in uncovered pots (Table 100d), significant at 0.05 level, (Table 100e). Moreover, *P. hysterophorus* leaves which were pre-wounded on the

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upper surface showed high infection both in covered and uncovered pots (Tables 100d & e) suggesting a possible role of insects in causing wounds for the entry of the pathogen.

Table 100d: Percent infection due to *Cercospora partheniiphila* on *Parthenium hysterophorus* leaves 4, 8, 12, and 15 days post inoculation

Sr.No.	Days after inoculation	Covered		Uncovered	
		Injured	Uninjured	Injured	Uninjured
1.	4	-	-	-	-
2.	8	1.75± 0.50*	0.60± 0.16	0.21± 0.10	0.09± 0.06
3.	12	3.31± 0.93	1.13± 0.29	0.40± 0.20	0.71± 0.11
4.	15	9.34± 2.74	3.86± 1.00	1.36± 0.64	0.60± 0.38
5.	20	Death of the plants			

*Mean of 12 replicates

Table 100e: Statistical analysis of percent infection of *Parthenium hysterophorus* leaves by *Cercospora partheniiphila* 15 days post inoculation

Injured covered	Injured uncovered	t-value	Uninjured covered	Uninjured uncovered	t-value
9.34± 2.74	1.36± 0.64	2.83*	0.60± 0.15	0.09± 0.06	3.04*

* Values significant at $p < .05$, $df = 11$

Table 100f: Statistical analysis showing the percent infection caused by *Cercospora partheniiphila* 15 days post inoculation between inoculated and uninoculated *Parthenium hysterophorus* plants

Treatment	Injured		Uninjured	
	Covered	Uncovered	Covered	Uncovered
Inoculated	9.34 ± 2.74	1.36 ± 0.64	0.60 ± 0.15	0.09 ± 0.06
Uninoculated	0	0.12 ± 0.03	0	0.06 ± 0.03
t value	3.40*	1.93	4.00*	0.44

* Values significant at $p < .05$, $df = 11$

The present data suggest that *C. partheniiphila* can be highly aggressive towards *Parthenium* weed and has certain characteristics that make it as a desirable candidate as a biological control agent of *Parthenium* weed :

- (i) wide natural distribution;
- (ii) it sporulates well on Czapek dox agar (a simple and cheap culture medium), within ten days, and can thus be mass produced in a short time and at low cost;
- (iii) narrow host range;
- (iv) capable of limiting populations of the weed

Thus it should not be ruled out as a possible agent for development and exploitation as a mycoherbicide for this weed in India. Further studies on host screening and the time taken for complete control of this weed in field conditions, when used singly and in combination with other fungal pathogens and insects, are in progress.

Outputs

4. *Local capability in biocontrol promoted and developed*

Training course in the UK

The three collaborators who attended the course, left with a grounding on the handling of the exotic rust pathogens *Puccinia melampodii* and *Puccinia abrupta* var *partheniicola*. Subjects covered included inoculation with the rusts, methods for assessing environmental conditions for the rusts, host range screening methods and microscopic examination of symptoms.

5. *Scientific papers published including review article on Parthenium hysterophorus (through additional funding 1996/97).*

Review (add-on-funding) published in 1997:

“*Parthenium hysterophorus*: a review of its weed status and the possibilities for biological control”; *Biocontrol News and Information* **18**, 89-98. H.C. Evans

Two papers were presented at the First International Conference on Parthenium Management on 6-8 October 1997 at Dharwad, India and published in the proceedings.

The potential of neotropical fungal pathogens as classical biological control agents for management of *Parthenium hysterophorus* L. H.C. Evans

Safety testing of the rust *Puccinia melampodii* as a potential biocontrol agent of *Parthenium hysterophorus* L.. M.K Seier, J.L. Harvey, A. Romero and R.P. Kinnersley

Two posters were presented at the International Symposium the Future of Fungi in the Control of Pests, Weeds & Diseases on 5-9 April 1998 at University of Southampton, UK

Specificity testing of fungal biocontrol agents using a wind tunnel to simulate natural dispersal conditions. J.L. Harvey (CABI Bioscience); R.P. Kinnersley (Imperial College)

Mycoherbicidal properties of *Gliocladium virens* towards *Parthenium hysterophorus*. P. Sreerama Kumar (Bangalore, India; trainee Parthenium Project)

A review paper presented at the X International Symposium on the Biological Control of Weeds on 4-9 July 1999 in Montana, USA

The impact of Parthenium weed in India and the development of an integrated management strategy based on Australian experiences. M. Seier, D.Djeddour, J. Harvey, S. Doraiswamy, P. Sreerama Kumar, L.P. Kauraw and K.R. Aneja

A paper was published in Biological Suppression of Plant Diseases, Phytoparasitic Nematodes and Weeds.

Biological suppression of Parthenium with pathogens. P. Sreerama Kumar

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A poster was presented at the National Symposium on Development of Microbial Pesticides and Insect Pest Management on 12-13 November 1998 at Shivajinagar, India.

Initial efforts towards the development of mycoherbicides for the management of Parthenium, a serious weed in India. P. Sreerama Kumar

A paper was presented at the International Conference - From Ethnomycology to Fungal Biotechnology - Exploiting Fungi From Natural Resources For Novel Products 15-16 December, 1997, Holiday Home Simla, India.

Biotechnology for the Production and Enhancement of Mycopesticide Potential. K.R. Aneja

General publications (published or in press):

Aneja, K.R. (1999). Biotechnology for the production and enhancement of mycoherbicide potential. pp. 91-114. In: *From Ethnomycology to Fungal Biotechnology* (Eds. J. Singh and K.R. Aneja). Kluwer Academic/Plenum Publishers, New York, USA.

Aneja, K.R. (1999). Movement against the dangers of Congress grass - An Interview. p. 9. In: *Dainik Tribune*. March 18, 1999 (In Hindi Language)

Aneja, K.R. and Khan, S.A. (in press). Congress grass (*Parthenium hysterophorus* L.) - Its impact, infestation and biocontrol with fungal pathogens - An overview. In: *Glimpses in Plant Sciences*. Aneja, K.R., Charaya, M.U., Aggarwal, A.K. and Hans, N.K. (Eds.) Prgati Prakashan, Meerut, India.

Aneja, K.R. and Khan, S.A. (in press) Occurrence of *Erysiphe cichoracearum* causing a powdery mildew disease in India. *Journal of Mycopathological Research*.

Aneja, K.R. and Khan, S.A. (in press) Congress grass – its life history, impact and control. *Jeevanti*. Kurukshetra University, India.

Aneja, K.R. and Khan, S.A. (in press) Leaf spot disease of Congress grass *Parthenium hysterophorus* – A new disease record. *Tropical Pest Management*.

Evans, H.C. (1997) *Parthenium hysterophorus*: a review of its weed status and the possibilities for biological control. *Biocontrol News and Information* **18**, 89-98.

Evans, H.C. (1997) The potential of neotropical fungal pathogens as classical biological control agents for management of *Parthenium hysterophorus* L.. pp. 55-62. In: *First International Conference on Parthenium Management*. Mahadevappa, M. and Patil, V.C. (Eds.) University of Agricultural Sciences, Dharwad, Karnataka, India.

Evans, H.C. (1998) Major Indian weeds of neotropical origin and the possibilities for collaborative biocontrol projects. pp. 22-27. In: *Proceedings of the Fourth International Workshop on Biological Control and Management of Chromolaena odorata*. Ferrar, P., Muniappan, R. and Jayanth, K.P. (Eds.) Agricultural Experiment Station, University of Guam, Mangilao, Guam 96923, USA, Publication No. 216.

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Karauw, L.P., Chile, A. and Bhan, V.M. (1997) Evaluation of *Fusarium pallidoroseum* (Cooke) Sacc for the biocontrol of *Parthenium hysterophorus* L. pp.70-74. In: *First International Conference on Parthenium Management*. Mahadevappa, M. and Patil, V.C. (Eds.) University of Agricultural Sciences, Dharwad, Karnataka, India.

Karauw, L.P., Chile, A. and Bhan, V.M. (1997) Effect of Marigold (*Tagetes patula* Linn) populations on the growth and survival of *Parthenium hysterophorus* L. pp.39-40. In: *First International Conference on Parthenium Management*. Mahadevappa, M. and Patil, V.C. (Eds.) University of Agricultural Sciences, Dharwad, Karnataka, India.

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For full papers and abstracts see appendix 5.

6. Protocol for import of biocontrol agents developed

This output is addressed under output No 8.

7. Data obtained on the susceptibility of selected test species to *Puccinia melampodii* under field conditions; risk analysis prepared

The field trial was successfully completed within the 4-week period with sporulation of *Puccinia melampodii* being recorded on the control *P. hysterophorus* plants for all 5 blocks. In general, the symptoms observed on non-host species were consistent for all blocks. Where test plant species had been assessed at different growth stages - as for *Calendula officinalis* cvs. "Touch red/orange", "Yellow coronet", *Guizotia abyssinica* local cultivar, *Helianthus annuus* cv. "Morden", *Tagetes patula* cv. "Red Marietta", *Zinnia elegans* cv. "Pulcino mix" - symptoms were found to be independent of plant age. The distance of test plants from the inoculum source (1m, 3m, 5m) had no influence on the type of symptoms observed, but did affect their extent.

Out of all test plant species/cultivars assessed in the field trial, only *Calendula officinalis* showed macroscopic symptoms of attack similar to those observed during previous greenhouse inoculations with *Puccinia melampodii* (see Output 3, CABI Bioscience, UK). Abnormal telia were observed on the lower as well as on the upper leaf surface of both *Calendula officinalis* cultivars. However, only teliospores produced on the cultivar "Yellow

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Coronet“, but not those formed on the cultivar “Touch red/orange, subsequently germinated to form basidiospores. In contrast, the rust was found to produce viable teliospores on both cultivars of *Calendula officinalis* in the greenhouse test which was undertaken parallel to the field trial.

Chlorotic and/or necrotic spotting recorded from other test plant species/cultivars included in the field trial could not definitely be related to the pathogen. After the 4-week period, such symptoms was frequently recorded from a number of plants, however, this appeared to be predominantly the result of insect damage.

The subsequent detailed microscopic assessment of leaf samples showed fungal mycelium to be present in isolated necrotic spots on leaves of the sunflower cultivar “Morden“ (compare “Ouptut 3, CABI Bioscience” describing the same observation). Internal mycelium was also occasionally recorded from leaves of the *Guizotia abyssinica* cultivars “ootacamund” and local cultivar. Although not proven, this must be considered to originate from infection with *Puccinia melampodii*. The initiation of telia and/or formation of teliospores was never observed.

Internal mycelium was not observed in leaf samples of any of the other test plant species.

It has to be pointed out that, similar to *Helianthus annuus*, the susceptibility of the test species *Guizotia abyssinica* appears to be highly dependent on the cultivar, since in contrast to the Indian cultivars included in the field trial, Indian cultivars assessed under quarantine greenhouse conditions in the UK showed no signs of internal fungal mycelium (compare “Ouptut 3, CABI Bioscience”).

Risk analysis

The results of the host specificity testing undertaken for *Puccinia melampodii* in the greenhouse and in the field form the base of a risk analysis addressing the potential release of the pathogen into India as a classical biocontrol agent for *P. hysterophorus*.

From the results of the field and greenhouse assessments, it can be concluded that *Puccinia melampodii* shows an apparent higher host specificity under field conditions than in a greenhouse situation. This phenomenon of an artificial host range extension under greenhouse conditions has been widely reported for pathogens - as well as for insects -and is thought to be due to optimum conditions for spore germination and infection, as well as an extremely high inoculum load (Evans, 1995, Evans & Tomley, 1996, Watson, 1985). Nevertheless, *Puccinia melampodii* was shown to be able to attack *Calendula officinalis* in a field situation and internal mycelium has been found in leaves of the sunflower cultivar “Morden“ as well as of selected *Guizotia abyssinica* cultivars following exposure to the rust in the field .

Considerably caution has to be exercised when assessing the biocontrol potential of this rust agent for India, since this country has no experience of, and no protocol or legislation for the importation of plant pathogens. In India, there is a strong undercurrent against “alien organisms”, which has been fuelled by an extremely volatile press. By way of example, controversy still surrounds the release in India of a Mexican beetle (via the Australian Queensland Parthenium Weed Biocontrol Programme), which caused minor damage to neighbouring sunflower crops in several *P. hysterophorus*-infested areas, despite the subsequent findings of an independent scientific committee that the feeding behaviour was both transient and aberrant, and not as supposed a host “jump” (Jayanth *et al.*, 1993).

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Initially, therefore, the PRA (pest risk assessment) of *P. melampodii* has to be handled with high sensitivity until the Indian quarantine authorities, the legislation and decision makers, as well as the general public, are better informed of the principles and potential benefits of classical biological control so as not to prejudice other biocontrol programmes against alien weeds in India waiting in the wings (Evans, 1998).

Significantly, this situation is different in Australia which has a long experience in classical biological control and a well established legislation for the importation of exotic agents. In Australia, *Puccinia melampodii* has been recently approved by AQIS (Australian Quarantine and Inspection Service) for introduction as a classical agent for Parthenium weed, despite the fact that the rust can also induce advanced symptoms on several local Australian sunflower cultivars, as well as sporulate successfully, although never abundantly, on several related composite species, such as *Flaveria australasica*, which is considered as a minor weed in Queensland.

For the PRA, it was concluded that the symptoms on sunflower probably reflect the artificial nature of the greenhouse tests, rather than a susceptible reaction (Wapshere, 1989), whilst infection of the native weed host, if this should ever occur in a field situation, was viewed as a positive rather than a negative attribute. Thus, the PRA panel decided that the risks of not attempting to control the weed, especially the threat to human health (McFadyen, 1995), far outweighed the risks posed by introducing the rust.

The release of *Puccinia melampodii* in Australia in early 2000 allowed the field trial with India test plant species to be carried out and, in future, will give India the unique opportunity to benefit from the Australian experience with the rust in the open field situation.

8. Indian scientist develops an insight into the release and monitoring strategies for fungal biocontrol agents in the field; experience in the organisation of extension work gained. Similar insight into, and experience of, Australian quarantine procedures gained and model protocol for importation developed.

Attendance at the international course on “Biological Control of Tropical Weeds” gave the Indian collaborator from PDBC an insight into biological control programmes run by Australia, as well as Australian quarantine procedures. This is particularly relevant as in the long term India is likely to profit immensely from the Australian wealth of experience, particularly with respect to classical biological control.

During the subsequent “hands-on“ training to assess the host specificity of the classical agent *Puccinia melampodii* in the field in Australia, the Indian scientist gained knowledge and practice in the design of field experiments, as well as in the interpretation of symptoms observed on non-host plants under field conditions compared to greenhouse conditions. The training gave him the opportunity to critically analyse the behaviour of the rust and familiarize himself with risk assessment of classical biological control agents.

The protocol has been discussed in detail with ICAR/PDBC but a model has yet to be prepared. Towards the end of 2000 CABI Bioscience has an FAO consultancy to advise on a new quarantine unit for PDBC and this protocol will then be finalized.

Contribution of Outputs

a. Contribution to project goal

The project goal (Peri-Urban Interface Purpose 1 / Semi-Arid Purpose 2) is: “Volume, quality and seasonal availability of food and crop products improved through the reduction of economic and physical losses / Impact of significant pests on production from cereal (particularly sorghum) based systems minimised”.

The outputs have contributed to this goal in that the project purpose has been achieved i.e. a sustainable, environmentally-friendly and economic control method as a management strategy for *P. hysterophorus* in India has been promoted and two fungal pathogens have been identified for use as classical biocontrol agents in an integrated management approach for this weed. However, doubts still exist about the potential behaviour of one of these pathogens, the exotic rust *Puccinia melampodii*, once released into India. Since CABI also has additional weed pathology projects in India (NRI *Mikania micrantha* project, ZA0026/R6735), it is felt best to err on the side of caution until this project has been implemented in India. Unlike *P. hysterophorus*, it is considered that *Mikania micrantha* will be successfully controlled by a single biocontrol agent thus enhancing the status of classical biological control in India. The best strategy for Parthenium weed is to await these developments before considering introduction of the potentially more controversial rust *Puccinia melampodii*. However, it is probable that permission will be sought to introduce *Puccinia abrupta* var. *partheniicola* once the Mikania rust has been approved for release into India.

More specifically, the outputs have made the following contributions:

	Major Results	Contribution to Project Goal
1.	Comprehensive inventory of fungal pathogens associated with Parthenium weed in India collated	Mycobiota characterized providing essential data for the selection of potential biological control agents.
2.	Impact of Parthenium weed on human health and cropping systems quantified	Base-line data of socio-economic impact contribute to generate support for action across state and national research organisations e.g. resulting in the launch of the “International Parthenium Research News Group” website acting as a discussion forum
3.	<p>a) Pathogens associated with <i>P. hysterophorus</i> in India shown to have low potential as biological control agents</p> <p>b) Potential of two exotic fungal pathogens, the rusts <i>Puccinia abrupta</i> var. <i>partheniicola</i> and <i>Puccinia melampodii</i>, as biocontrol agents for <i>P. hysterophorus</i> in India established</p>	<p>a) Low potential of local agents established providing the base for consideration to introduce exotic agents</p> <p>b) Classical biological control can be regarded as one option in an integrated management approach; however, due to concerns addressed in the text preceding this table potential introductions of the two rusts, in particular <i>Puccinia melampodii</i>, as classical agents can only be considered at a later time</p>

	Major Results	Contribution to Project Goal
4.	Indian scientists trained in general and specific aspects of biological control as well as handling of exotic pathogens (as named under 3.b.) as biological control agents for <i>P. hysterophorus</i>	Local human resource capability established for handling exotic pathogens and India is now investing in a new containment facility in Bangalore to handle exotic pathogens
5.	Scientific papers including major review article published and/or presented at conferences	Project widely publicised and technical results provided to the scientific community, especially in India; support for biological control gained from relevant Indian authorities (e.g. Indian Council of Agricultural Research)
6.	<i>Puccinia melampodii</i> established to be more host specific under field conditions than under greenhouse conditions and data for risk analysis provided	Risk analysis incorporating greenhouse and field data provides tool for decision process for relevant Indian authorities on introduction of the rust as a classical agent
7.	Indian scientist trained in field evaluation and risk assessment of classical biological control agents and aspects of biological control from an Australian perspective	Local human resource capability developed for implementation and risk assessment of classical biological control agents

b. Promotion pathways to target institutions and beneficiaries

A strong connection has already been established with the target institutions (Tamil Nadu Agricultural University, National Research Centre for Weed Science - Jabalpur, PDBC – Bangalore and Kurukshetra University) through their collaboration in this project. With respect to a potential introduction of one or both rust fungi at a later stage, stronger links will be forged with PDBC which will act as the ICAR quarantine station for importation of exotic agents into India. Once released from quarantine, imported pathogens would then be made available to the collaborating institutions which would arrange the initial field releases.

c. Follow-up action/research

As spelt out under a), any follow up regarding the two exotic rusts, *Puccinia melampodii* and *Puccinia abrupta* var. *partheniicola* will be deferred until the Mikania rust has been approved for release into India.

However, discussions will be initiated with the Project Directorate of Biological Control (ICAR), Bangalore, concerning the possibility and benefit of introducing additional selected strains of *P. abrupta* var. *partheniicola* from Mexico into India. A dossier on this rust will be prepared to provide the base for these discussions.

Further research required will be:

- Characterization of the *P. abrupta* var. *partheniicola* strain currently present in India and assessment of its distribution and impact on Parthenium weed

- Assessment of pathogenicity and environmental requirements of selected strains of this rust from Mexico to identify the most suitable strain(s) for the situation in India, genetic characterization of this/these strain(s)
- Possibly restricted host specificity testing for additional rust strain(s), subject to requirement of Indian authorities

Subsequently, the discussions can be broadened to consider also *P. melampodii* for importation and assessment by Indian scientists in the new quarantine facility.

Meanwhile, each of the four collaborating Indian institutions has obtained funding to continue with selected aspects of research into the management of Parthenium weed. Tamil Nadu has on-going ACIAR funding and will be a major collaborator in a new ACIAR initiative (IPM/socio-economics) on Parthenium management. PDBC has ICAR funding to continue development of mycoherbicides against Parthenium weed. Jabalpur has ICAR funding on soil suppression and competitive plants. Kurukshetra is continuing with state funding on monitoring diseases and impact of Parthenium weed in Haryana and Punjab.