

Mycoherbicide Research for Controlling Weeds: Status, Opportunities and Future Needs

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Abstract: Mycoherbicide offer an innovative approach to the management of weeds using formulated fungal phytopathogens or their crude metabolite extracts would serve as an important component in integrated management strategy. The biological control of weeds by mycoherbicide (fungal weed pathogens/metabolites) has received considerable academic consideration, only a few commercial products are available. This review article explains the management of weeds with the native microbes and their metabolites isolated from their native weeds. The present weed management in agriculture mainly depends on synthetic herbicides, which cause environmental pollution, restricted choice of succeeding crops and long residual activity. The herbicide industry is continuously searching for identification and characterization of most effective, economical and environmentally safer synthetic herbicides by screening large number of synthetic organic molecules, synthesizing analogs of patent herbicides, designing new herbicide molecules based on target site approach and screening of natural products for herbicidal activity. The herbicidal properties of fungi can be exploited successfully as a tool for the management of weeds. Large number of secondary metabolites produced by fungi provides eco-friendly, diverse and challenging chemical structures. There are numerous reviews has published about mycoherbicide and mycoherbicidal agents were showing intense scientific and commercial interest in this field. Here, we attempt in this review to show that the mycoherbicide field has indeed grown significantly and future research needs.

Keywords: Fungi/Mycoherbicide/Biorational/Mass production/Formulation

1. Introduction

Weeds are undesirable vegetation directly or indirectly inferring with human welfare. There are several plants like *Cynodon dactylon*, *Hyptis suaveolens* and *Cyperus rotundus* etc. have significant medicinal, religious or economical value at some places, but creating severe problems in agriculture and other places of human uses. Thus, the definition of the term "Weed" is very subjective. They are an integral part of all cropping ecosystems and serve as a major biological constraint preventing crops from achieving their yield potentials. In addition, there are several plants viz., *Parthenium hysterophorus*, *Rhusradicans* *Ambrosia* spp., *Amaranthus spinosus*, *Argemone mexicana*, *Lantana camara*, *Xanthium strumarium* etc. which are responsible for major health problems to humans and animals. The Ministry of Environment and Forests, Govt. of India has collected information on invasive weeds in India. The major invasive weeds in India include *Chromolaena odorata*, *Lantana camara*, *Parthenium hysterophorus*, *Mikania micrantha*, *Mimosa diplotricha* var. *diplotricha*, *Acacia mearnsii*, *Ageratum conyzoides*, *Agertaina adenophora*, *Arundo donax*, *Cuscuta reflexa*, *Imperata cylindrica*, *Leucaena leucocephala*, *Merremia peltata*, *Prosopis juliflora*, *Pteridium aquilinum* and *Sphagneticolaa trilobata* (Sankaran & Suresh, 2013). Impact of some of these species on various ecosystems have been studied e.g., *Chromolaena odorata* and *Lantana camara*, *Ageratum conyzoides*, *Parthenium hysterophorus*, *Mikania micrantha*, *Prosopis juliflora* (Sw.) DC.; and *Ageratina adenophora* (Spreng.) (Sankaran et al., 2001).

Problems of weeds in agriculture, forestry, environment and health have extensively been discussed in many publications (Gupta, 1998; Pandey et al. 1995, 96ab, 2004; Pandey, 1999, 2000). Problems discussed above necessitated their efficacy and safer management. Manual methods of weed control

have earlier been considered as one of the most effective way to eradicate weeds. However, Industrialization has resulted in severe labour shortage and drastic increase in labour cost has significantly hampered this method. Synthetic chemical herbicides has no doubt played very crucial role in weed management since 1960s, however, due to indiscriminate and excessive use of these chemicals, several problems have arisen. Contamination of ground water, accumulation of residues, development of resistance, narrow spectrum of activity, injury to non target organisms, lack of residual effectiveness etc, are the major public concern nowadays. Therefore, there is a need to discover and develop new, economically and environmentally sustainable weed management technology. Biological control of weeds with plant pathogenic fungi and their metabolites offers opportunities for overcoming several of these inadequacies as evidenced by commercialization of many strains of fungi as Mycoherbicide (Aneja, 1998; Auld, 1990; Bhan et al., 1998; Boyette & Abbas, 1995; Pandey, 1999, 2000; Pandey et al. 1995-2003; Kovics et al., 2005). Therefore, the objectives if this review article is to highlight the potential of fungi and discuss the opportunities and challenges in their exploitation as mycoherbicide.

2. Status of Mycoherbicide Research

Out of 69000 species and genera of fungi recognized only very meager number of species has been evaluated for their mycoherbicidal potential. Most of the mycoherbicide candidates tested belong to Hyphomycetes, a large and varied class of conidial and non sporulating fungi. The Coelomycetes, which include *Colletotrichum*, *Septoria*, *Phomopsis*, *Phoma*, *Phaeoseptoria*, *Ascochyta*, *Mycleptodiscus* species etc. ranked second. Some pathogenic strains of Ascomycetes belong to *Sclerotinia*, *Balansia*, *Cochliobolus*, *Valsa*, *Cryptomycina*, *Ceratocystis* and *Nectria* have also been evaluated. Amongst Oomycetes

species only two genera *Pythium* and *Phytophthora* have been considered. *Puccinia*, *Uredo*, *Sphacelotheca* and *Sorosporium* are the frequently used fungi belong to Basidiomycetes (Charudattan, 1991). Thus, a wide number of candidate is being explored, although preferred pathogens appear to be those capable of causing some of the most destructive diseases such as anthracnose, wilts, blights and foliar spots. Due to partial symbiotic attitude, obligate parasites have less herbicidal potential while facultative parasite or saprophytes are usually considered as best candidates for their development as mycoherbicide (Templeton *et al.* 1986).

This is to be expected because the efficacy and performance standards for mycoherbicide dictate a high capacity for plant kill or damage (Charudattan, 1991). More than 500 isolates of fungi pathogenic to some major weeds of Central India such as *Parthenium hysterophorus*, *Lantana camara*, *Xanthium strumarium*, *Hyptis suaveolens* etc. have been recovered by a group of worker at Jabalpur. Rajak *et al.* (1990) isolated more than 25 fungi from different parts of *P. hysterophorus*. These include several fungi namely *Colletotrichum gloeosporioides* f .sp. *parthenii*, *C. dematium*, *Myrothecium roridum*, *Sclerotium rolfsii*, *Fusarium solani*, *F. oxysporum*, *Alternaria alternata*, *A. macrospora* which have shown significant pathogenic potential and satisfied most of the requirements desired for mycoherbicidal development (Pandey *et al.* 1990-1998; Farkya *et al.* 1994, 96, 2001; Farkya, 1994; Mishra, 1994; Mishra *et al.* 1994, 95, 96ab; Gayathri, 1998; Gayathri & Pandey, 1997; Pandey, 2004). *Lantana camara* is another problematic weed in Central India. Mycopathological survey undertaken at various places of the states of Madhya Pradesh and Chattisgarh yielded several fungi including a highly pathogenic strain of *Alternaria*, *Phoma*, *Fusarium* and *Curvularia* (Pandey, 2000, Pandey & Pandey, 2000; Saxena & Pandey, 2001; Saxena *et al.* 2001; 2002). More than 25 fungi have been isolated from different parts of the weed, *Hyptis suaveolens* from MP and Chattisgarh by Pandey (2004). Studies conducted on mycoherbicidal potential of these fungi yielded one each of highly effective strains of *C. dematium* and *S. rolfsii* (Pandey & Pandey, 2005; Pandey *et al.* 2002). Mycoherbicidal potential of various fungi isolated from Central India has extensively been discussed in many publications (Hasija *et al.* 1994; Pandey *et al.* 1995-2004; Pandey, 1999, 2000; Kovics *et al.* 2005). A comprehensive list of fungal strains commercialized/patented or under evaluation and development for weed management are listed in table I.

2.1 Mass production

Commercialization of mycoherbicides requires low cost, economically feasible and easily available large scale production technology for infective propagules of the agents. With few exceptions, the most suitable infective units are fungal spores. Fungi multiply through several types of spores, but asexually produced spores or conidia due to their enormous number, easy harvesting and efficient mechanism of dispersal, they are considered as best candidates as infective units of mycoherbicides. In some instances, especially when mycoherbicidal agents do not produce conidia, mycelial fragments and serve as infective unit

(Ridings *et al.*, 1975; Tute, 1969). However, mycelial fragments are harder to quantitative than spores, less readily separated from culture medium and often less virulent than spores (Tute, 1969). In addition, the durability, longevity and viability of mycelium generally much less than that of spores (Churchill, 1982). Analyses of various production methods have been the subject of several in depth review (Churchill, 1982; Jackson *et al.*, 1996; Jackson, 1997; Jenkins *et al.*, 1998; Daigle *et al.*, 1998; Ooijkaas *et al.*, 2000).

2.2 Liquid /Submerged fermentation:

Potential application of this method in pharmaceutical and food industries is well known. It has been the method of choice for the large scale production of many products including mycoherbicides. It is considered as a superior technique for those agents, which sporulate significantly in liquid culture over these which require additional steps to induce sporulation (available fermentation technology can be used to produce mycoherbicides at commercial scale. Various steps involved in this method have been extensively reviewed in many publications (Bowers, 1986; Boyette *et al.*, 1991; 96). Relatively low cost, automation and controlled parameters are the major advantages of liquid culture fermentation. Nutritional composition of the production medium has been shown to have a dramatic impact on propagules attributes such as biocontrol efficacy and desiccation tolerance (Jackson, 1997). Advances in fermentation technology have revolutionized this method and most of the commercially available mycoherbicides have been produced by submerged culture fermentation. In general submerged culture fermentation is considered the most cost effective production method and is being used to produce many registered mycoherbicides (Stowell, 1991; Jackson & Schisler, 1995). Singh (2004) recorded significant biomass yield by *C. dematium* in malt extract. Pandey (2004) obtained highest biomass yield and sporulation in *C. gloeosporioides* f sp *parthenii* when grown in Richard's medium supplemented with glucose and potassium nitrate as carbon and nitrogen sources. Pandey (2000) reported good biomass and conidial yield by *Alternaria alternata* strains effective mycoherbicidal agent against *Lantana camara* in Sabourands dextrose and Richard's broth respectively. Potato dextrose agar and Asthana & Hawkers medium were founded be highly suitable for biomass and conidial yield respectively by *Curvularia lunata*, potential mycoherbicidal agent against *Xanthium strumarium* (Shukla, 2001). Sabouraud's dextrose medium supported maximum spore/ as well as biomass yield in *Fusarium oxysporum* and *F. solani* (Farkya, 1994). PDA was also reported the best medium for *Sclerotium rolfsii* strain (Mishra, 1994).

2.3 Solid Substrate Fermentation

The strategy involves the use of solid nutritive, moist grains, agrowastes etc. soaked with water or liquid medium. The choice of substrate will depend on a number of factors including local availability, cost and isolate preference. Fungus infested substrates when incubated at optimal conditions of temperature and moisture, colonized rapidly by the agent. After depletion of nutrients, agent has produced

significant amount of spores on the surface. Due to low cost and simplicity of process, the method is of preferential choice for mycoherbicidal production especially in developing countries where labor is readily available. Additionally, solid substrate makes quantification and dispersal relatively easy and accurate. High labor costs, difficulties in maintaining sterility, storage, lack of control over fermentation conditions and recovery of spores/Infective propagules are the major inherent problems of this strategy. However, for those fungi which do not produce spores in liquid culture, it may be the only method for spore production. Hildebrand and McCain (1978) used wheat straw that was infested with *Fusarium oxysporum* f sp *cannabinus* to control *Canbis sativa* (Marijuana). Boyette (1982) used oat seed infested with *F. solani* f sp *cucurbitae* to control *Cucurbita texana* (Texas gourd). Gaythri (1998) and Pandey (1998) recorded very high growth and sporulation of *C. dematium* and *C. gloeosporioides* f sp *parthenii* on wheat porridge and wheat bran respectively. Out of 35 solid agrowastes tried by Singh (2004) for mass production of another strain of *C dematium* effective against *Parthenium*, maize cob, grits supported maximum conidial formation.

2.4 Biphasic Fermentation

It involves the production of biomass of the agent in liquid culture and then harvested, blended and spread on solid substrates and incubated under different cycling of light for sporulation. An advantage of this method is that fungi, which do not sporulate in submerged culture, can be included to sporulate after one growth cycle in liquid culture. *Alternaria macrospora*, a mycoherbicidal agent against *Anoda cristata* (Spurred anoda) was mass-produced by this method. This technique has also been used in production of mycoherbicidal agents viz., *Colletotrichum malvarum* against *Sida spinosa* (Prickly sida), *F. lateritium* against *A. cristata* and *Abutilon theoprastris* (Velvet leaf) and *A. cassiae* against *Cassia obtusifolia* (Sickle pod) etc. (Walker, 1980; 1981,a,b; Walker & Reley, 1982). Successful production of mycoherbicidal agents i.e., *A. crassa* (Boyette & Walker, 1982), *A. helianthi* (Quimby, 1989) and *Bipolaris sorghicola* (Van Dyke & Winder, 1985) have also achieved by this method. Singh (2004) obtained excellent sporulation in *C. domatiums* when biomass was bedded on wheat straw incubated at $28^{\circ}\text{C} \pm 1^{\circ}\text{C}$.

2.5 Formulations

Formulations of agents into cost effective products determines their level of success as mycoherbicide (Daigle et al., 1997). Advances have been made in the areas of formulations as a means to improve efficacy. For instance, the effectiveness of a pathogen can be improved by using hydrophilic polymers, emulsions, surfactant etc as adjuvant in bioherbicide formulations. The level of weed control can be improved by using several host specific pathogens in a "multiple-pathogen strategy". This strategy offers several advantages such as improved level of weed control, preventing possible buildup of host resistance, overcoming age related host resistance, assuring consistency in performance, improving the environmental latitude of activity and others. It is also possible to 'customize' the

pathogen mixture depending on the type of pathogens available for use in a given country or region. Possibilities of exploitation of various formulations to improve mycoherbicidal efficacy of the agents have been extensively discussed in many publications (Auld & Morin, 1985; Auld, 1993; Weidemann *et al.*, 1995; Boyette *et al.*, 1991, 1996; Fravel *et al.*, 1985; Green *et al.*, 1998; Daigle & Connick, 2002).

2.6 Liquid based formulation

With few exceptions, liquid formulations of mycoherbicide generally are best suited for post emergence weed control and are use primarily to incite leaf and stem diseases. Most of the mycoherbicide are applied with water mixture. However, presence of a waxy cuticle on surface prevents water based product from spreading evenly, which can result in unequal distribution of active ingredient (the infective propagule). Surfactants help to wet the plants and aid in dispersing the fungal spores through the spray mix. Because of spores of mycoherbicide agents are finite units, it is important that the surface area be covered with the materials as evenly and equally possible (Boyette *et al.*, 1996). Liquid based formulations commonly used in mycoherbicide are listed in **Table II**.

2.7 Solid based formulation

Solid based formulations are considered best for those mycoherbicidal agents which incite soil borne diseases in weeds. A variety of solid materials viz., fungus colourized grains, alginate beds, wheat flour(PESTA), non-ionic polymer beads, wood block etc have been extensively used as formulating materials (Daigle & Connick Jr., 2002). They have advantages over liquid formulations because

- 1) They provide a buffer from environmental extremes
- 2) They can serve as food base for the fungus, resulting longer period of persistence
- 3) They are less likely to wash away from the treated areas than are spores.

Some of the important solid based formulations are listed in **Table III**. Various aspects of these formulations have extensively been discussed in many publications (Boyette *et al.*, 1991, 1996; Daigle & Connick, 2002; Walker & Connick, 1983). Amongst them PESTA is one of the most important formulations used in many cases (Daigle *et al.*, 1997; Connick *et al.*, 1998).

2.8 Additives to improve formulation efficacy

Several adjuvant and amendments have been suggested to either improve or modify spore germination, virulence, viability, host specificity or environmental requirement (Boyette, 1994). Viability and virulence of macro conidia of *Fusarium lateritium* enhanced significantly at reduced moisture content when formulated in hydrated silica (Quimby, 1985). Encapsulation in alginate granules coated with oil absorbent also increased the efficacy of several mycoherbicidal agents (Quimby *et al.*, 1988). Simple addition of some additives viz., Sucrose, Soy dex, lecithin, sorbitol etc have also reported to enhanced biocontrol potential by modifying the agents viz., *Sclerotinia*

sclerotiorum, *Colletotrichum trunacatum* and *Alternaria macrospora* (Quimby *et al.*, 1988, 1993; Walker, 1980, 1981; Cardina *et al.*, 1988; Van Dyke & Winder, 1985; Winder & Van Dyke, 1989; Wymore & Watson, 1986, 1999). Addition of arginine in formulated spores significantly reduced the spread of *Sclerotinia* in environment after the death of the weed (Ligon, 2001). Significant control of Prickly pear cactus from some 60 million acres of land in Queensland and Northern South Wales have been achieved by combined effect of a soft rot bacterium (*Erwinia* sp.), anthracnose fungi (*Colletotrichum gloeosporioides*) Wilt fungi (*Fusarium oxysporum*) and cactus feeding mouth (*Cactoblastis cactorum*). Remarkable control of *Eupatorium adenophorum* (Croftus weed) in Queensland have been achieved with the gall fly (*Procecidochares utilis*) in association of leaf spot fungus, *Ceratospora eupatorii* (Templeton & Trujillo, 1981). Northern joint vetch and Winged water prirose have been successfully controlled by application of *C. gloeosporioides* f. sp. *aeschynomene* and *C. gloeosporioides* f. sp. *jussiae*. A mixture of these two pathogens with *C. malvarum* can effectively controlled the above weeds and also prickly sida (Boyette *et al.* 1991). Effective control of Sicklepod has been achieved by application of sub lethal doses of chemical herbicides viz., Linuron, Imaziquin and Lactofen to spore mixture of *A. cassiae*, (Hoagland, 1996). Sequential application of 2,4-D and *F. lateritium* significantly increased the control of velvet leaf (Boyette & Quimby, 1998). *C. coccides* and cotton defoliant Thiadiazuron also provide significantly control to velvet leaf. Almost complete control of *Cyprus esculentus* (yellow nut sedge) has been achieved by sequential application of Paraquat and *Puccinia canaliculata* (Boyette *et al.*, 1979; 1991). Khodayari *et al.*, (1987) demonstrated that it is possible to extend the weed control spectrum of CGA by mixing it with Aciflourfen a herbicide that control hemp sesbania, but not northern jointvetch. A mixture of the above can effectively control both the weed effectively. Host selectivity of *A. crassa* has been altered successfully either by addition of water soluble filtrates of Jimson weed or dilute fruit pectin to spore suspensions. Through proper timing and placement of inoculums, it is possible that this system could be used in a practical method to enhance the weed control spectrum of this pathogen (Boyette & Abbas, 1994). A mixture of CGA with Acifluorfen or Bentazone significantly enhanced the Bio-control of northern jointvetch and hemp sesbania (Smith 1986). The combination of Trifluorolin with a strain of *F. solani* enhances activity of both chemical and the pathogen for control of Texas gourd *Cucurbita texana*. Compatible formulation of chemical herbicide has expanded the spectrum of weeds controlled by a single application (Boyette, *et al.*, 1991). Some of the important activities enhanced by additives and listed in Table IV.

2.9 Other Improvement Method

Various technologies have been used and will continue to be used to enhance biological weed control (Cohen *et al.*, 2002). The protoplast fusion technique was used to create new strains using *Helminthosporium gramineum* subsp. *echinoclaoe* strain HM1 (high pathogenicity, low spore formation) and *Curvularia lunata* (low pathogenicity, high

spore formation) to create strains that effectively control barnyard grass and other weeds in rice production (Zhang *et al.*, 2007). Hypervirulence selection or manipulation may improve efficacy of biological control agents. Cohen *et al.* (2002) transformed genes of the indole-3-acetamide (IAM) pathway to cause an auxin imbalance that increased the virulence of *Fusarium oxysporum* and *F. arthrosporioides*, pathogenic on broomrape (*Orobanchae aegyptiaca*). Sands and Pilgeram (2009) outline the steps to enhance virulence of the bio-control agent using amino acid overproduction. They discuss control of the parasitic weeds *Orobanchae* and *Striga*, which are especially challenging to control due to the close relationship they develop with their hosts. Economic formulations and genetic manipulations to alter phenotype will assist in the understanding and development of microbial herbicides.

2.10 Regulatory for Mycoherbicide

In addition, as with all research and new products where there are safety concerns, buffer zones are often required to protect animal pastures and other non-target areas (Bourdote *et al.*, 2006). The risk of applying a microbial herbicide to the environment needs to be considered at the beginning and throughout the development of bio-control agents. With thorough host-range testing, very few, if any, detrimental effects occur from the release of fungal herbicides to control weeds (Barton, 2004). In a review of pre- and post-release records from 26 projects, Barton (2004) found that there were no reports of a fungal biological control agent striking an unintended plant species. Additional animal, avian, fish and daphnia testing are also required in many countries before bioherbicides can be registered. Rigorous testing is required prior to the release of a biological herbicide to ensure the safety of humans, animals and the environment. Host-range studies are needed to reduce potential risk and ensure that beneficial, non-target plant species are unaffected by the bio-control agent. However, the length of time needed to complete assessments of new biological herbicides adds to the costs and the length of time required before an agent can be released (Ghosheh, 2005). Non-host testing is important and the ranges of plant species tested depend on the areas of release, ecosystem variability and potential for dissemination of the bio-control agent by wind or water. Testing should cover all economically important plant species of the area, and those plants known to be involved in ecosystem maintenance. In agronomic ecosystems, the major crop species are of interest. The U.S. Environmental Protection Agency (EPA, 2011) published a list of the top 25 major agricultural crops. Plants were placed on this list because of their economic importance, ecosystem activity or total production values (EPA, 2011). In aquatic systems, several aquatic plants are suggested that include algae, aquatic bacteria, marine and freshwater diatoms. In rangeland ecosystems the non-target species would include native or near native plant species. It is recommended to test six species covering at least four families in the Dicotyledonae, and at least four species of at least two families in the Monocotyledonae. Testing must be performed on all plants of economic importance in agriculture, horticulture or rangeland systems or known to be beneficial to maintenance of the ecosystem that have any reasonable likelihood of serving as hosts. This selection of

additional plant species should be based upon a survey of plants closely related (same genus or, if not available, same family) to the target plant and a survey of known hosts of pathogens closely related to the microbial herbicide (EPA, 2011; Wapshere, 1974).

2.11 Opportunities & Future Needs

Inadequacies discussed earlier may be amenable to correction either by advances in formulation technology for biological or by advanced molecular techniques (Yoder, 1983; Yoder & Turgeon, 1985). They may also offer opportunities for biologically active metabolites with weed control potential. Mycoherbicides present suitable opportunities for return on investment from small market because the cost of developing them may be less than that for a chemical herbicide. Production technology already available in fermentation industries, thus capital investment for production is low. Registration costs could be significantly less than for synthetic herbicides. Time required for research and development of a potential agent through registration and commercial use may be substantially less than for herbicides, and this would represent a significant saving of developmental costs (Templeton *et al.* 1986). There is no doubt the extraordinary fungal diversity in ecosystem and thus, each pathogen must be considered as unique and must be thoroughly studied laboratory growth chamber or green houses to understand its disease cycle and potential as herbicide. The potential of particular genus as microbial herbicide can be obtained from knowledge about diseases of economic crops incited by other species or forms of the genus. Proper understanding of the disease cycle of a pathogen to be developed as mycoherbicides is very important step in a success of a programme. The interaction of the life cycles of the fungus and host plant must also be understood. Important facets include the source of primary inoculum, the method of dissemination of infectious propagules, the climatic parameters that favor rapid infection and disease development, the age and physiology of the host that favors or suppress plant infection, variation in genetic resistance of the host or virulence of the pathogen, the method and rapidity of secondary spread and the means of overwintering. Particular emphasis is placed on the climatic parameters, principally temperature and moisture that affect the disease cycle. With the above information together with knowledge of the climate in the geographic region where the weed grows and the growth stage during which the weed must be controlled, a fairly accurate assessment for the mycoherbicides potential of a particular fungus can be made. Unfortunately, many of the published reports that suggest specific fungi as potential mycoherbicide have not researched disease cycle or the weed biology adequately to make a definite judgment of the biological potential of a particular fungus (Templeton *et al.*, 1998). A wealth of knowledge about disease cycles can also be obtained with pathogens of economically important crops. However, this knowledge cannot be extrapolated too far because the crop pathogen relationship of disease is usually different than the weed pathogen relationship. Microorganisms specially fungi are known to produce variety of phytotoxic metabolites with herbicidal properties (Abbas & Duke, 1997; Culter, 1998; Duke, 1986 a,b; Hoagland, 1990, 1999, 2000, 2001). Still

only few have been screened. Therefore, lot of opportunities exists in their integration with mycoherbicidal agents.

Although, mycoherbicides have proved to be effective, but there is a need for technological improvement with chemical enhancer, by strain improvement or by combining fungi to increase the spectrum of weed control. Many fungal pathogens of weeds may be weed without additional technological improvement. However activity of many other fungal pathogens is supported by low virulence, stringent temperature and moisture requirement, wounding requirement or specific physiological requirement of the host plant. These fungi may have potential for particular weed problems if technological improvements can be made (Templeton *et al.*, 1986). Experience with Collego, Devine, Casst and Bio Mal leaves no doubt that mycoherbicides are effective and practical as weed control agents (Bannon 1988; Bowers 1986; Bowers 1982; Charudattan *et al* 1986; Kenney 1986., Ridings 1986., Ridings *et al* 1976; Smith 1982; Smith 1986; Templeton 1982; Walker and Riley 1982). The chemical industry is known to screen thousands of chemicals for every commercially feasible herbicide. When viewed in this light, mycoherbicides have had a remarkably high rate of return on scientific and monetary input. Experience with agents like *Alternaria cassiae*, *Cercospora rodamanii*, *Colletotrichum coccodes* and *C. gloesporioides* f. sp. *malvae* suggest that we are indeed witnessing this second phase of growth in mycoherbicides in which challenges, both scientific and commercial are being posed. The future direction of mycoherbicide is being influenced by current scientific, practical and government decisions (Charudattan, 1984).

On the research front following are emerging as major areas of importance:

- 1) **More mycoherbicide candidates of important weeds-** With each weed- pathogen system,, new conceptual and practical problems are bound to come to light. These in turn will provide a deeper understanding of mycoherbicide.
- 2) **Integration of mycoherbicide with chemical pesticides-** As an on going effort, the compatibility-incompatibility of mycoherbicide and chemicals should continue. This will be mandated by the fact that each weed –mycoherbicide- pest management system will be different and specific recommendations for the use of mycoherbicide will be needed.
- 3) **Integration of mycoherbicide and chemical plant growth regulators for improved weed control through decrease in weed growth and increase in mycoherbicide efficacy-** Weeds possessing high raters of vegetative growth and vegetative proliferation tend to be difficult to control with mycoherbicides. The ability to outgrow disease pressure is a characteristic of these weeds (Charudattan *et al* 1985; Winder & Dyke, 1989). In such cases the integration of mycoherbicides with plant growth regulators, which by themselves may not afford weed control, offer a useful solution (Charudattan, 1986).
- 4) **Extensive survey of literature-** It indicates that the role of weed pathogen interaction in weed management have neglected significantly. More knowledge is needed in this area alongwith phytoalexin production, defense protein

etc. Researches in these areas would definitely help in weed management.

- 5) **Development of suitable formulations to improve viability, efficacy and ease of application of mycoherbicides**- The need for optimum moisture and specific temperature regimes for infection pose problems in assuring mycoherbicide efficacy. The lack of proper epidemiological conditions for infections and disease development and the adverse effect of solar radiation on fungal propagules can be counted to an extent through formulation technology. Substances that improve moisture retention, reduce drying and UV-irradiation, dilute and evenly disperse the inoculums and provide better host- pathogen contact are being studied (Connick *et al* 1989).
- 6) **Fermentation technology**- Current industrial preference favours submerged liquid fermentation to produce mycoherbicides products (Churchill, 1982; Templeton *et al*, 1980). Although successful, cost effective and readily available, this technique is not suitable for fungi that do not sporulate in submerged culture. Solid substrate culturing and air-lift fermentation can offer solutions.
- 7) **Molecular genetic basis of virulence and host specificity**-Genetic improvement of mycoherbicide candidates through bioengineering for increased virulence and increased or decreased host specificity deserves research emphasis. With several mycoherbicide candidates the level of virulence is less than desirable. By incorporating genes for virulence factors such as host- specific toxins and phytotoxic metabolites or host receptors it should be possible to improve weed control ability of these candidates. On the other hand, several highly virulent and destructive pathogens exist that are suitable as mycoherbicides on account of their broad host range. Mutation- selection, gene cloning, interspecific and intragenic protoplast fusions, electroporation and other methods can be useful for this purpose.
- 8) **Use of biomolecule or phytotoxic secondary metabolites from pathogenic as well as non pathogenic fungi**: Discovery of host specific and non-specific herbicidal metabolites of fungal origin that could be used as virulence and host specificity factors for genetic engineering. Fungal compounds have commercial advantages over the living product/spores in having a longer shelf life, a requirement for yearly application, generally more predictable and uniform results and no possibility of spreading to non target organisms. Phytotoxic compound also used as new mode of action which is produced by many fungi as small peptides or other small molecule.
- 9) **Research Funding**: Increased public and private funding as well as administrative support for research and development of mycoherbicide.
- 10) **Education and Promotion of Product**:- Education of scientist unfamiliar with mycoherbicide and the user public, which is required for technology transfer- Mycoherbicide, like many other bio-control agents are sensitive to environmental conditions and need to be handled in strict accordance to the prescribed methods. They are usually slower in eliciting the desirable results. The more difficult challenge may be to convince the agricultural community that crop yield can be improved

without killing weeds (Auld & Morin, 1995). The users must therefore, be educated about the use and performance features of mycoherbicide.

Prospects for the development and utilization of mycoherbicide technology for major crops are in demand. The future of mycoherbicide is bright and full of possibilities with the many novel, successful fungi and their metabolites being studied. The advancements in genetics, cheap extraction and structural analysis work will help mycoherbicide control of weeds to move forward. Formulations are needed to increase shelf life of the living organisms to improve survival and efficacy. Research and development of mycoherbicide are needed so that stakeholders and industry buy in to the marketing, economics and time investments of this approach to weed management.

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Table I: List of various fungi evaluated for weed management worldwide

S. N.	Name of Weed	Name of organism	Country
1	<i>Abutilon theophrasti</i> Medic (Velvet leaf)	<i>Colletotrichum coccodes</i> *CA 11224055	USA
2	<i>A indicum</i> G Don	<i>Fusarium lateritium</i> *US4419120 <i>Puccinia abutili</i> <i>Puccinia heterospora</i> <i>Cercospora avicennae</i> <i>Cercospora mavacearum</i>	Canada India India India India
3	<i>Acacia meransii</i>	<i>Ceratocystis sp. cylindrobasidium</i>	S. Africa
4	<i>Acacia saligna</i>	<i>Uromycladium tepperioides</i>	S. Africa
5	<i>Acroptilon repens</i> (L.) DC	<i>Alternaria cichorii</i> <i>Puccinia acroptili</i> <i>Subanguina picridis</i>	Canada British USA, Canada
6	<i>Aeschynomene virginica</i> (L) BSP (Northern Jointvetch)	<i>Colletotrichum gloeosporioides f.sp aeschynomene</i> (Collego)	USA

	<i>A. indica</i> L	<i>Physoderma aeschynomensis</i>	India
7	<i>Ageratina riparia</i> (<i>Eupombrium reparaia</i>)	<i>Entyloma ageratinae</i> <i>Cercospora agertinae</i>	New Zealand, Hawaii
9	<i>Ageratina adenophora</i>	<i>Phaeoramularia eupatorii-odorati</i>	Australia, South Africa, New Zealand
10	<i>Alternanthera philoxeroides</i>	<i>Alternaria alternantherae</i> <i>Nimbya alternantherae</i>	USA, Canada
11	<i>A. adenophora</i>	<i>Phaeoramularia eupatorii-odorati</i>	Australia, South Africa, New Zealand
12	<i>Amaranthus</i> sp	<i>Phomopsis amaranthicola</i> <i>Alternaria alternata</i> <i>Trematophoma lignicola</i>	USA, Europe
13	<i>Ambrosia artemisiifolia</i>	<i>Albugo tragopogonis</i>	Russia
14	<i>Ambrosia trifida</i> (Giant ragweed)	<i>Puccinia xanthii</i> f sp <i>ambrosia-trifidae</i> <i>Protomyces gravidus</i>	USA, Canada
15	<i>Anoda cristata</i> (L) <i>Schlecht</i> (Spurred anoda)	<i>Alternaria macrospora</i>	
16	<i>Avena fatua</i>	<i>Drechslera avenacea</i>	Australia- Italy
17	<i>Baccharis halimifolia</i>	<i>Puccinia evadens</i>	Australia
18	<i>Calystyegia sepisum</i>	<i>Stagonospora convolvuli</i>	Europe
19	<i>Cannabis sativa</i> .(Marijuana)	<i>Fusarium oxysporum</i> f sp. <i>cannabis</i>	U. S.
20	<i>Capeonia palustris</i> St Hil	<i>Ampiphobotrytis ricini</i>	India
21	<i>Carduus tenuiflorus</i> Curt	<i>Alternaria zinniae</i> *US4636386 <i>Puccinia carduorum</i>	Brazil
22	<i>Carduus throermeri</i>	<i>Puccinia carduorum</i>	USA
23	<i>Carduus nutans</i>	<i>Puccinia carduorum</i>	U.S.A
24	<i>Cassia occidentalis</i> L. (Sickle pod) <i>C. obtusifolia</i> (L)	<i>Alternaria cassiae</i> *054390360 <i>Phyllactinia cortylea</i> f sp <i>sprialis</i> <i>Pseudocercospora nigricans</i>	USA India Australia
25	<i>Centaurea diffusa</i>	<i>Puccinia jaceae</i> <i>Puccinia cetaureae</i>	Canada Canada
26	<i>Chenopodium album</i> <i>Cispsium arvense</i>	<i>Ascochyta caulina</i> *EP296057 <i>Cercospora chenopodii</i>	Holland Netherlands
27	<i>Clidemia hirta</i>	<i>Colletotrichum gloeosporioides</i> f sp. <i>clidemiae</i>	Hawaii, USA
28.	<i>Clematis vitalba</i>	<i>Phoma clematidina</i>	New Zealand
29	<i>Chondrilla juncea</i> L	<i>Puccinia chondrillina</i> <i>Erysiphae cichoracearum leveillula taurica</i> f sp. <i>chondrillae</i>	Australia , USA Australia
30	<i>Cirsium arvense</i>	<i>Fusarium</i> sp. <i>Puccium punctiformis</i> <i>Botrytis</i> sp <i>Phoma</i> sp	India Australia, Canada India
31	<i>Convolvulus arvenis</i> (Field bindweed)	<i>Stagonospora convolvuli</i> <i>Phomopsis convolulus</i> <i>Erysiphae convoulus</i>	Europe USA USA
32	<i>Cryptostegia grandiflora</i>	<i>Maravalia cryptostegiae</i>	Australia
33	<i>Cucurbita texana</i> (A) Gray (Texas gourd)	<i>Fusarium solani</i> f sp <i>cucurbitae</i>	USA
34	<i>Cuscuta</i> sp <i>Cuscuta chinensis</i> (L) <i>Cuscuta iupiniiformis</i> Kroch	<i>Alternaria</i> sp. <i>Colletotrichum gloeosporioides</i> f sp <i>cuscutae</i> <i>Alternaria cucutacidae</i>	Fl., USA China USSR
35	<i>Cynodon dactylon</i> (L)	<i>Alternaria citis</i> <i>Bipolaris nodulosa</i> <i>Cercospora verroniae</i>	India India India
36	<i>Cyperus</i> sp	<i>Dactylaria higginsii</i>	Fl., USA
37	<i>Cyperus esculentus</i>	<i>Aschochyta cypricola</i> <i>Cintractia peribebuyensis</i> <i>Cercospora caricis oud</i> <i>Cercospora cyperi-roundi</i> <i>Curvularia tuber culata</i> <i>Puccinia canaliculat</i> *US4731104 <i>Puccinia cyperi</i> <i>Phytophthora cyperi-roundati</i> <i>Rhizoctonia solani</i> <i>Sclerotinia homoeocarpa</i>	USA USA USA India India USA USA Australia USA USA
38	<i>Cyperus rotundus</i> L	<i>Cercospora caricis</i> <i>Puccinia romagnoliana</i> <i>Dactylaria higginsii</i>	Brazil, Israel India, Israel USA, Israel
39	<i>Cytisus scoparius</i>	<i>Fusarium tumidum</i>	New Zealand
40	<i>Datura stramonium</i> L (Jimson weed)	<i>Alternaria cussiae</i>	India

41	<i>Dendrophthoe falcate</i> var <i>pubescen</i>	<i>Colletotrichum gloeosporioides</i>	India
42	<i>Diospyros virginiana</i>	<i>Acremonium diospyri</i> <i>Cercospora kaki</i> <i>Cephalosporium diospyri</i>	USA India USA
43	<i>Echium plantagineum</i>	<i>Cercospora echii</i>	Australia
44	<i>Echinochloa</i> sp. <i>Echinochloa crus-galli</i>	<i>Colletotrichum graminicola</i> <i>Exserohilum fusiforme</i> <i>Curvularia lunata</i>	Canada, South Korea Vietnam, Australia Nether lands
45	<i>Egeria densa</i> <i>E. najas</i>	<i>Fusarium</i> sp	Brazil
46	<i>Eichhornia crassipes</i> (Mart.) Solms.	<i>Acremonium zonatum</i> <i>Alternaria eichhorniae</i> <i>Bipolaris stenospila</i> <i>Cercospora rabmanii</i> *US4097261 <i>Cercospora piaropi</i> <i>Curvularia lunata</i> <i>Myrothecium roridum</i> f sp. <i>eichhorniae</i> <i>Phoma sorghina</i> <i>Rhizoctonia solani</i> <i>Uredo eichhorniae</i>	USA India USA USA South Africa India India Sudan USA USA
47	<i>Emex</i> spp.	<i>Cercospora tripolitana</i> <i>Peronospora rumicis</i>	Australia Australia
48	<i>Erigeron annuus</i>	<i>Phoma putaminum</i>	Italy
49	<i>Erythroxylum coca</i>	<i>F. oxysporum</i> f sp. <i>erythroxyli</i>	Coca producing region
50	<i>Euphorbia</i> sp	<i>Alternaria euphoricicola</i> *US4755208, *US4636386	USA
51	<i>Euphorbia cyprissias</i>	<i>Melampsora euphorbiae</i> <i>Uromyces scutellatus</i>	USA Switzerland
52	<i>Euphorbia heterophylla</i>	<i>Helminthosporium</i> sp	Brazil
53	<i>Euphorbia esula</i>	<i>Fusarium</i> spp., <i>Rhizoctonia</i> spp.,	USA
54	<i>Galega officinalis</i>	<i>Uromyces galegae</i>	Chile
55	<i>Galinsoga ciliate</i> <i>G. parviflora</i>	<i>Colletotrichum gloeosporioides</i>	Russia
56	Grass weeds	<i>Dreschlera</i> sp., ; <i>Exserohilum</i> sp <i>Pyrenophora sememnipreda</i>	Australia Fl., USA
57	<i>Hakear sericea</i>	<i>Colletotrichum caudatum</i>	Australia
58	<i>Hedychium gardnerianum</i>	<i>Ralstonia solanocerum</i>	Hawaii USA
59	<i>Heliotropium europaeum</i>	<i>Uromyces heliotropii</i> <i>Cercospora</i> sp.	S. Africa Australia
60	<i>Hydrilla verticillata</i>	<i>Fusarium roseum</i>	USA
61	<i>Hypericum androsaemum</i>	<i>Melampsora hypericorum</i>	Australia
62	<i>Imperata cylindrical</i>	<i>Colletotrichum caudatum</i> <i>Ascochyta</i> sp <i>Puccinia rufipes</i> <i>Colletotrichum graminicola</i> <i>Dibymeria</i> sp <i>Bipolaris sacchari</i> <i>Drechslera gigantea</i>	Malaysia Malaysia Malaysia Malaysia Malaysia USA USA
63	<i>Jussiaea decurrens</i>	<i>Colletotrichum gloeosporioides</i> f sp <i>jussiaeae</i>	USA
64	<i>Lantana camara</i> L.	<i>Cercospora lantanae-camarae</i> <i>C. guianensis</i> <i>C. lantanicola</i> <i>Mycovellosiela lantanae</i> <i>Prosopidium tubervaulatum</i> (= <i>Uredo tuberculata</i> = <i>Puccinia tuberculata</i>) <i>Puccinia lantanae</i> (= <i>Micropuccinia lantanae</i>) <i>Puccinia natalensis</i> <i>Phomopsis lantanae</i> <i>Phylosticta lantanicola</i> <i>Septoria lantanae</i> <i>Ramularia</i> sp <i>Acanthostigama</i> (= <i>Nematostoma</i>) <i>lantana</i> <i>Aecidium lantanae</i> <i>Aleurodiscus</i> sp <i>Alternaria</i> sp. <i>Alternaria alternata</i> <i>Amisphaeria lantanae</i> <i>Acremonium zonatum</i> <i>Calosphaeria lantanae</i> <i>Calospora lantanae</i>	India Guyana Colombia Brazil West Indies Brazil Ghana, Sudan India Zambia Puerto Rico Dominican Republic Dominican Republic Trinidad Dominican Republic Brazil Brazil India India India India

	<i>Capnodium sp</i>	India
	<i>Ceratobasidium lantanae-camarae</i>	India
	<i>Cercospora sp</i>	India
	<i>Cercospora canescens</i>	India
	<i>Cercospora guianensis</i>	Brazil
	<i>Cercospora lantanicola</i>	Mauritius
	<i>Ceratospaeria bicellula</i>	India
	<i>Cercospora lantanae</i>	India
	<i>Cladosporium oxysporum</i>	USA
	<i>Cochilobolus lunatus</i>	Colombia
	<i>Colletotrichum dematium</i>	India
	<i>Diatrype cryptostegiae</i>	India; Costa Rica
	<i>Diatrype chloroscarca</i>	India
	<i>Diatrype parvatae</i>	India
	<i>Didymosphaeria donacina</i>	India
	<i>Diplodia lantanicola</i>	Brazil
	<i>Epiphyma(=Botryosphaeria) nervisequens</i>	India
	<i>Eutypa aspera</i>	India
	<i>Eutypa lantanae</i>	India
	<i>Eutypella russodes</i>	Brazil
	<i>Fusarium oxysporum</i>	Venezuela
	<i>Gleospodium sp</i>	India
	<i>Godronia lantanae</i>	New Caledonia
	<i>Helicosporium</i>	India
	<i>Helminthosporium mauritianum</i>	USA
	<i>Helminthosporium velutinum</i>	Barbados
	<i>Hypoxyton notatum</i>	India
	<i>Hysterium lantanae</i>	India
	<i>Leptosphaeria conithyrium</i>	India
	<i>Leptosphaeria haemitites</i>	India
	<i>Leptosphaeria isocellula</i>	India
	<i>Leptosphaeria rajashtanensis</i>	India
	<i>Macrovalsaaria megalospora</i>	Brazil
	<i>Massarina mucosa</i>	India
	<i>Massarina tricellula</i>	India
	<i>Meliola sp</i>	India
	<i>Meliola ambigua</i>	India
	<i>Meliola cookeana</i>	India
	<i>Meliola durantae</i>	Brazil
	<i>Memmoniella echinata</i>	Ghana
	<i>Merulius corium Fr</i>	Sierra Leone
	<i>Metasphaeria abuensis</i>	Brazil
	<i>Microdiplodia minuta</i>	India
	<i>Mycovellosiella lantanae</i>	India
	<i>Myrothecium roridum</i>	India
	<i>Mysterographium multiseptum</i>	India
	<i>Nectria wegeliniana</i>	Cuba
	<i>Oidium sp</i>	Malaysia, India
	<i>Oospora sp.</i>	India
	<i>Ophiobolus lantanae</i>	Zambia, India
	<i>Ostropa indica</i>	India
	<i>Patellaria lantanae</i>	Brazil
	<i>Periconia cookie</i>	India
	<i>Perisporiopsis lantanae</i>	India
	<i>Phoma lantanae</i>	India
	<i>Phoma sp.</i>	India
	<i>Phoma multirostrata</i>	Puerto Rico
	<i>Phomopsis lantanae</i>	South Africa
	<i>Prospodium tuberculatum</i>	Myanmar, Zambia
	<i>Protostroma indica</i>	Zambia, India
	<i>Pseudocercospora formosana</i>	Portugal, India
	<i>Pseudocercospora natalensis</i>	N. America
	<i>Ramularia sp.</i>	India
	<i>Rhizoctonia sp.</i>	Hong Kong
	<i>Rhizoctonia solani</i>	Ghana
	<i>Rosencheldia paraguayana</i>	Trinidad
	<i>Sarcinella palwanensis</i>	Philippines
	<i>Sclerotium rolfsii</i>	USA
	<i>Scolecobasidium sp.</i>	S. America
	<i>Scolecopeltidium lantanae</i>	America

		<i>Septoria lantanae</i> <i>Septoria lantanifolii</i> <i>Spegazzinia sundra</i> <i>Sphaerulina sp.</i> <i>Stictis lantanae</i> <i>Strictis radiata</i> <i>Subramania poonensis</i> <i>Teichospora lantanae</i> <i>Teichosporella lantanae</i> <i>Torula harbarum</i> <i>Tryblidaria maharashtrensis</i> <i>Tryblidaria pongamiae</i> <i>Tryblidiella rufula</i> <i>Hysterium lantanae</i> <i>Leptosphaeria conithyrium</i> <i>Tubeufia helicomyces</i>	India Australia Brazil Brazil Brazil India Zambia Brazil N. Caledonia India India India
65	<i>Malva purilla</i> Sm	<i>Colletotrichum gloeosporioides</i> sp *EP218386	Canada
66	<i>Malva pusilla</i>	<i>Colletotrichum gloeosporioides</i> f sp malvae (BIOMAL)	Canada
67	<i>Mikania micrantha</i>	<i>Cercospora milaniicola</i>	Malaysia
68	<i>Mimosa pigra</i>	<i>Diabole cubensis</i>	Australia
69	<i>Morronea odorata</i> Lindl. (Stranglervine)	<i>Phytophthora palmivora</i> (DeVine)	USA
70	<i>Orbanche spp</i>	<i>Fusarium oxysporum</i> var .orrhoceras	USSR
71	<i>Oxalis</i> sp.	<i>Puccinia oxalidis</i>	France
72	<i>Parthenium hysterophorus</i> L	<i>Alternaria tenuis</i> , <i>Alternaria zinniae</i> <i>Alternaria alternata</i> , <i>Alternaria dianthi</i> <i>Alternaria macrospora</i> <i>Curvularia lunata</i> , <i>Curvularia senegalensis</i> <i>Colletotrichum gloeosporioides</i> <i>Colletotrichum capsici</i> <i>Cladosporium cladosporioides</i> <i>Cercospora partheni</i> <i>Dreschslera indica</i> <i>Fusarium equiseti</i> , <i>Fusarium oxysporum</i> <i>Myrothecium roridum</i> <i>Phoma herbarum</i> <i>Sclerotium rolfsii</i> <i>P. melampodii</i> <i>Puccinia abrupta</i> f sp parthenicola <i>Bremia lactucae</i> <i>Erysiphae cichoracearum</i> <i>Cercospora parthenicola</i> <i>Sphearotheca fulignae</i>	India India C. America UK Dominican R. India Cuba, Mexico India
73	<i>Passiflora tripartite</i> ; <i>P. mallissima</i>	<i>Septoria passiflorae</i>	Hawaii, USA
74	<i>Protulaca oleracea</i> L.(common purslane) <i>Pteridium aquilinum</i>	<i>Dichotomophthora protulacoceae</i> <i>Asochyta pteridis</i>	UK UK
75	<i>Pueraria lobata</i>	<i>Myrothecium verrucaria</i>	USA
76	<i>Prunus serotina</i>	<i>Chondrostereum purpureum</i>	Netherlands
77	<i>Quercus</i> spp	<i>Ceratocystis fagacearum</i>	USA
78	<i>Rosa multiflora</i>	Rose rosette disease	USA
79	<i>Rottboellia chochinensis</i>	<i>Sporisorium ophiuri</i> <i>Colletotrichum graminicola</i> <i>Puccinia rottboelliae</i>	UK Thailand, UK Thailand, UK
80	<i>Rubus</i> spp.	<i>Phragmidium violaceum</i>	Chile
81	<i>Rumex crispus</i> L	<i>Uromyces rumicis</i>	California
82	<i>Rumex</i> sp	<i>Uromyces rumicis</i> <i>Ramularia rubella</i>	Portugal Portugal
83	<i>Sagittaria</i> sp	<i>Rhynchosporiu malismatis</i>	Australia
84	<i>Senecio vulgaris</i> <i>S. jacobaea</i>	<i>Puccinia lagenosporae</i> <i>Puccinia expansa</i>	Australia Australia
85	<i>Senna optusifolia</i>	<i>Alternaria cassiae</i>	Brazil
86	<i>Sesbania exaltata</i> (Raf) cory (Hemp sesbania)	<i>Colletotrichum truncatum</i>	Miss., USA
87	<i>Sida spinosa</i> L	<i>Colletotrichum malvarum</i> <i>Fusarium lateritium</i>	USA USA
88	<i>Solanum elaeagnifolium</i>	<i>Nothanguina phyllobia</i>	USA
89	<i>Solanum ptycanthum</i> Dunn. (Eastern Black nightshade)	<i>Colletotrichum coccodes</i> *US4715881	USA
91	<i>Solanum viarum</i>	<i>Alternaria</i> sp.	Fl. USA
92	<i>Sorghum halepense</i> L Pers	<i>Bipolaris sorghicola</i>	Brazil

93	<i>Sphenocola zeyanica</i>	<i>Alternaria sp.</i> <i>Colletotrichum gloeosporioides</i>	Philippines Malaysia
94	<i>Striga hermonthica</i>	<i>Fusarium nygani</i> <i>Fusarium oxysporum</i> <i>Fusarium semitectum var majus</i>	Sudan, Germany W. Africa, Canada, Sudan, Africa
95	<i>Taraxacum officinale</i>	Fungal isolate MAC I	Canada
96	<i>Trianthema portulacastrum</i> L. (Horse purslane)	<i>Gibbago trianthemae</i>	India
97	<i>Ulex europaeus</i>	<i>Fusarium tumidum</i>	Germany
98	<i>Viola arvensis</i>	<i>Mycocentropora acerina</i>	New Zealand
99	Weed seed	<i>Pyrenophora semeniperda</i> <i>Chaetomium globosum</i> <i>Chaetomium spirate</i>	Australia UK UK
100	<i>Xanthium sp</i>	<i>Alternaria alternata</i> *JP6227897 <i>Alternaria zinniae</i> <i>Colletotrichum orbiculare</i>	India Australia, USA Australia
101	<i>Xanthium strumarium</i> L X. <i>spinosum</i> L	<i>Alternaria tenuissima</i> <i>Alternaria tenuis</i> <i>Alternaria zinniae</i> <i>Cercospora xanthicola</i> <i>Colletotrichum xanthi</i> <i>Cordiculare</i> <i>Odium xanthi</i> <i>Puccinia xanthi</i> <i>Sclerotium rolfsii</i>	India India India India Australia Australia India Australia India

Sources: Charudattan (1991), Hasija *et al.*, (1994), Pandey *et al.*, (1995, 1996a, b, 1997, 2001, 2004), Evans (1997).

Table II: Liquid based formulations of mycoherbicide

Weed host	Pathogen	Formulation reagents
Velvet leaf	<i>Fusarium lateritium</i>	Water+ Tween-20 Surfactant (0.02%) Experimental formulation-water
	<i>Colletotrichum coccodes</i>	Water + Sorbitol (0.075%)
Northern jointvetch	<i>Colletotrichum gloeosporioides</i>	Commercial formulation-component A; dried spores, component B; dehydrating agent + surfactant
Spurred anoda	<i>Alternaria macrospora</i>	Water+ nonoxynol surfactant (0.02%); Sucrose (5% w/v)
Giant ragweed	<i>Protomyces gravidus</i>	Water
Field bindweed	<i>Phomopsis convolvulus</i>	Water + Gelatin (0.1%)
Jimsonweed	<i>Alternaria cassiae</i>	Water + nonoxynol surfactant (0.04%);
Florida beggarweed	<i>Colletotrichum truncatum</i>	
Sicklepod	<i>Alternaria cassiae</i>	Water + nonoxynol surfactant (0.04%); paraffin wax mineral oil, soybean oil, corn syrup, lecithin
Common purslane	<i>Dichotomophthora portulacaceae</i>	Water + Tween-20 surfactant (0.02%)
Hemp sesbania	<i>Colletotrichum truncatum</i>	Water + nonoxynol surfactant (0.02%); paraffin wax, mineral oil, soybean oil, lecithin
Eastern Black nightshade	<i>Colletotrichum coccodes</i>	Water + Tween-20 surfactant (0.02%)
Strangler vine	<i>Phytophthora palmivora</i> (DeVine)	Commercial formulation; Chlamydo spores in water
Horse purslane	<i>Gibbago trianthemae</i>	Water + Tween-20 surfactant (0.02%)
Water hyacinth	<i>Cercospora rodmanii</i>	Mycelial fragment +wetable powder
Malva pusilla	<i>Colletotrichum gloeosporioides</i> f sp <i>malvae</i>	Spores + silica gel

Sources: Boyette *et al.*, (1991, 1996); Aneja *et al.*, (2000)

Table III: Solid Based Mycoherbicide Formulations

Weed host	Pathogen	Formulation reagents
Velvet leaf	<i>Fusarium lateritium</i>	Sodium alginate-kaolin granules
Spurred anoda	<i>Alternaria macrospora</i>	Vermiculite
Texas Gourd	<i>Fusarium solani</i>	Fungus infested oats; cornmeal/sand; sodium alginate-kaolin granules
Marijuana	<i>Fusarium oxysporum</i>	Fungus-infected wheat straw
Hemp sesbania	<i>Colletotrichum truncatum</i>	Fungus infected wheat gluten/ kaolin clay (PESTA)
Sicklepod	<i>Fusarium oxysporum</i>	Fungus infected wheat-gluten/ kaolin clay (PESTA)

Sources: Boyette & Abbas, (1994), Boyette *et al.*, (1991, 96)

Table IV: Additives to improve mycoherbicide efficacy

Fungi	Weed	Additives	Activity
<i>Alternaria macrospora</i>	<i>Spurred anoda</i>	Sucrose	• Increased severity
<i>A. cassia</i>	<i>Senna obtusifolia</i>	Lecithin	• Reduced dew requirement
<i>Colletotrichum truncatum</i>	<i>Desmodium tortuosum</i> <i>Sesbania exaltata</i>	Sucrose+ gum xanthum Lecithin,	• Increased severity & spore germination • Reduced dew requirement
<i>Bipolaris sorghicola</i>	<i>Sorghum halepense</i>	1% soya-Dox	• Severity of disease
<i>C. coccodes</i>	<i>Abutilon theopasti</i>	Sorbitol	• Viability of spores • Reduced dew requirement
<i>F. lateritium</i>	<i>Sida spinosa</i>	Hydrated silica	• Viability & virulence of spores
<i>Sclerotinia sclerotiorum</i>	<i>Many broad leaf</i>	Oil emulsion	• Shelf life
<i>C. orbiculare</i>	<i>Xanthium spinosum</i>	Vegetable oil	• Reduced moisture requirement

Sources: Boyette *et al.*, (1991)