

## International Journal of Advanced Biochemistry Research



ISSN Print: 2617-4693  
 ISSN Online: 2617-4707  
 IJABR 2024; 8(5): 828-840  
[www.biochemjournal.com](http://www.biochemjournal.com)  
 Received: 13-03-2024  
 Accepted: 18-04-2024

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## Bio-herbicides: An eco-friendly approach for integrated weed management

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DOI: <https://doi.org/10.33545/26174693.2024.v8.i5j.1201>

### Abstract

Bio-herbicides offer a promising eco-friendly alternative to chemical herbicides, addressing concerns over environmental impact and human health risks. These bio-controls, like Devine, Collego, and Biomal, are effective, economically feasible, and non-toxic to non-target organisms. They can significantly reduce weed-related crop yield losses, which account for 37% of overall pest-induced losses. However, the development of bio-herbicides requires careful consideration of formulation, fermentation systems, and strategic evaluation to ensure their effectiveness and commercial viability. To enhance the adoption and efficacy of bio-herbicides, researchers must address key constraints, including environmental, biological, technological, and commercial challenges. These constraints hinder the widespread use of bio-herbicides and limit their effectiveness as a sole weed management solution. While not a total replacement for chemical herbicides, bio-herbicides can serve as a vital component of integrated weed management systems, particularly as advancements in target selection, formulation, and marketing are made to overcome current limitations.

**Keywords:** Augmentative, bioherbicides, pathogen, commercialization, adjuvants and myco-herbicides

### Introduction

Weeds pose significant challenges in agriculture by competing with crops for essential resources like water, nutrients, and light. They can also serve as hosts for pests and diseases, further impacting crop health and yield (Nichols, 2015) <sup>[129]</sup>. Weeds can cause significant yield losses in various crops, including: Direct-seeded rice: 15 to 66%, Maize: 18 to 65%, Soybean: 50 to 76% and Groundnut: 45 to 71% (Gharde, 2018) <sup>[56]</sup>. The extent of crop yield losses caused by weeds varies significantly and is influenced by several factors, including the type of crop, weed management approaches employed, weed species present, duration of weed infestation, and environmental factors such as climate and soil characteristics (Oerke *et al.*, 2006) <sup>[131]</sup>. Weed management is a critical practice in agriculture. Due to labor shortages, there is a growing global trend towards increased use of herbicides to control weed populations (Aktar, 2009) <sup>[4]</sup>. Inevitably, the constant use of herbicides on the same field to control weeds over a prolonged period has been shown to cause herbicide resistance, residue in crops, ecological imbalance between harmful and beneficial organisms, and environmental pollution. However, time constraint, advances in pest control technology, as well as a continuous 'enticement' from the current agricultural system have encouraged farmers to keep using conventional herbicides which have been found to be effective and time- and cost-efficient. The application of synthetic herbicides for effective weed control has thus become indispensable despite the unwelcome side effects. Organic fruits, vegetables, dairy products, and beverages have been increasingly popular worldwide, especially in developed nations. This trend reflects a growing consumer preference for organic food (Somasundram, 2016) <sup>[174]</sup>. Organic products represent a small portion of the food industry, yet their rapid expansion has sparked significant interest among consumers, businesses, and researchers alike. In 2013, there were nearly two million organic producers worldwide. Asia accounted for 36% of global organic farmers, followed by Africa at 29% and Europe at 17% (Somasundram, 2016) <sup>[174]</sup>. Organic product sales have consistently increased over the last decades (Willer, 2015) <sup>[194]</sup>. To meet increasing consumer demand, farmers are transitioning from chemical-dependent conventional agriculture to sustainable, environmentally friendly

practices. This shift has led to the emergence of more sustainable weed control alternatives. Sustainable weed management focuses on preventing weed spread rather than controlling them after they have already become problematic. Sustainable weed management includes various practices like crop rotation, intercropping, crop competitiveness, tillage, mulching, biological control agents, and bioherbicides. These methods aim to control weeds without relying on chemical herbicides. Biological weed control involves using natural enemies, substances, or agents to limit weed populations to a manageable level economically. The application methods for bioherbicides are similar to conventional herbicides, but with mycoherbicides, the pathogenic fungi are sprayed onto target weeds to inoculate them. Recently, bio herbicides have been regarded as a crucial weed control element (Hoagland, 2007) <sup>[65]</sup> albeit not as a total replacement but rather as an alternative to chemical herbicides (Singh, 2009 Raghuvanshi *et al.*, 2023a; Yadav *et al.*, 2023a, Singh *et al.*, 2022; Jha *et al.*, 2023, Kantwa *et al.*, 2019; Yadav *et al.*, 2023c; Singh *et al.*, 2013b) <sup>[168, 145, 197, 85, 91, 199, 167]</sup>. Sustainable weed management is characterized by its reliance on a variety of weed control techniques, rather than a single approach, as seen with synthetic herbicides in conventional agriculture. Therefore, to effectively control weeds, bio-herbicides should be used in conjunction with other weed management methods (Yadav *et al.*, 2023b; Patel *et al.*, 2023, Singh *et al.*, 2013a; Sahu *et al.*, 2022) <sup>[198, 137, 164, 152]</sup>. Bio herbicides that are thought to be safer and 'greener' have drawn attention, as scientific reports provide increasing evidence of their efficacy. However, their commercial presence in comparison to conventional herbicides is relatively new. Therefore, rigorous testing and validation is necessary to evaluate their efficacy and reliability for weed control (Tiwari *et al.*, 2011a; Tomar *et al.*, 2023a, Kumar *et al.*, 2022; Raghav *et al.*, 2023; Tanisha *et al.*, 2022) <sup>[186, 188-189, 104, 144, 179]</sup>. This review examines the influence of bio-herbicides and their limitations. The article aims to determine the role of bio-herbicides in weed control, including their incorporation into existing systems, to analyze factors influencing their effective use in both traditional and alternative management practices, to evaluate additional benefits beyond weed control, (Malviya *et al.*, 2012, Sahu *et al.*, 2022) <sup>[118, 152]</sup> and to assess the obstacles to the future advancement of bio-herbicides for efficient biologically-based weed management.

### Bio herbicides

Bioherbicides consist of microorganisms such as pathogens and other microbes or phytotoxins derived from microbes, insects, or plant extracts that act as a natural means of weed control. According to Bailey, bioherbicides are naturally originated products which can be used to control weeds. But one must remember that, although bioherbicides comprise nature-derived compounds, this is not to say they are completely harmless. Plants naturally produce toxins that can impact the health of non-plant organisms in the environment. These toxins can also affect certain bacteria, viruses, and fungi, potentially leading to health issues in animals and humans (Sekhar, 2012) <sup>[162]</sup>. Thus, it is crucial to manage these natural toxins carefully to prevent unintended harm to crops or beneficial organisms in the environment (Duke, 2000) <sup>[49-50]</sup>. The first evidence of bioherbicide development was documented in the mid-

1970s with the discovery of mycoherbicides. Since then, numerous bioherbicides have been registered and become available in the global market (Zeng, 2020) <sup>[200]</sup>. The earliest bioherbicide project involved simply the application of *Fusarium oxysporum* Schlecht, a fungus, against *Opuntia ficus-indica* (L.) Mill, (Pacanoski, 2015) <sup>[134]</sup>. In the 1950s, the parasitic weed *Cuscuta* spp. was controlled with *Alternaria cucurbitaceae* Rudakov (Pacanoski, 2015) <sup>[134]</sup>. Meanwhile, the utilization of both registered and unregistered bio-herbicides has risen notably. In the late 1960s, an ambitious effort was made to discover pathogens from *Rumex* spp. in the United States (Inman, 1971) <sup>[70]</sup> and *Rubus* spp. in Chile (Oehrens, 1977) <sup>[130]</sup> for weed control.

### Steps in developing a bioherbicide

Certainly! Here's a more detailed explanation of the three essential stages in the development of organic or natural herbicides:

#### Detection Stage

- a) **Assembly of Unhealthy Flora:** Researchers identify and collect plant pathogens or microbes that have the potential to act as herbicidal agents.
- b) **Separation of Underlying Creatures:** Once potential pathogens are collected, they are isolated from other microorganisms to ensure purity.
- c) **Application of Koch's Hypotheses:** Koch's postulates are applied to confirm that the isolated pathogen is the causative agent of disease in plants.
- d) **Pathogen Recognition:** The specific pathogen is identified and characterized for its herbicidal properties.
- e) **Pathogen Cultivation on Synthetic Medium:** The isolated pathogen is grown in a controlled synthetic medium to produce sufficient biomass for further studies.
- f) **Maintenance of Crops:** The cultivated crops are maintained for short-term or long-term storage, depending on the requirements of the research.

#### Phase of Progression

- a) **Creating Suitable Conditions for Spore Growth:** Optimal conditions are provided for the pathogen to produce spores, which are the infectious agents.
- b) **Suitable Conditions for Contamination and Progression of Infection:** Conditions that promote the spread of the pathogen and its infection of target weed species are identified and optimized.
- c) **Assessing Host Range with Exposure to Pathogen Exploitation Appliances:** The range of plant species that can be affected by the pathogen is determined through controlled experiments.

#### Stage of Placement

- a) **Close Association Between Non-industrial and Developed Sectors:** Collaboration between academia, research institutions, and industry is established to facilitate the development, field assessment, and commercialization of the bioherbicide.
- b) **Growth:** The production process is scaled up to produce the bioherbicide in larger quantities.
- c) **Field Assessment:** The efficacy and safety of the bioherbicide are tested in field trials under real-world conditions.

**d) Publication of Phases of the Bioherbicide Production Marketing Cycle:** The successful development and marketing of the bioherbicide are publicized to promote its adoption in sustainable agriculture practices.

#### Some commercially available bioherbicides

Certainly! Here's a detailed explanation of the bioherbicides we mentioned:

##### *Phytophthora palmivora* (Devine)

Developed by Abbott Laboratories, USA, it was the first mycoherbicide derived from the fungus *Phytophthora palmivora*. It targets *Morrenia odorata*, causing deadly root rot and collar rot. It can survive in the soil for extended periods. It was introduced in 1981 (Rao, 2000) [147].

##### *Colletotrichum gloeosporioides* (Collego)

This mycoherbicide was developed from the anthracnose

fungus *Colletotrichum gloeosporioides* f. sp. *Aeschynomene*. It is effective against *Aeschynomene virginica* in rice and soybean crops. It was the first mycoherbicide commercially available for controlling annual wild plants in crops with over 90 percent efficacy. Named in 1982 (Boyette *et al.*, 2012) [26].

***Colletotrichum gloeosporioides* (Biomal):** Developed by Philom BiosInc., Canada, it is another mycoherbicide based on *Colletotrichum*. It targets *Malva pusilla*. It includes C-Spores. Successfully developed by Collego.

***Streptomyces viridochromogenes* (Bialaphos and Glufosinate):** Bialaphos and Glufosinate are derived from *Streptomyces viridochromogenes*. It is a by-product obtained through fermentation. It is the active component of bialaphos, obtained from non-phytopathogenic *Streptomyces*. These bioherbicides are available internationally. (Carbonari *et al.* 2016) [29].

**Table 1(a):** Some commercially available bio herbicides are available in market

Trade name	Pathogen	Weed host	Country	Reference
Collego®	<i>Colletotrichum gloeosporioides</i> f. sp. <i>aeschynomene</i>	Northern joint vetch	USA	Bowers, 1986 [22]; Smith, 1982, 1991 [171-172]
BioMal®	<i>Colletotrichum gloeosporioides</i> f. sp. <i>malvae</i>	Round leaved mallow	USA; Canada	Boyetchko, 1998 [24]; Mortensen, 1998 [126]; Mortensen and Makowski, 1997 [125]
Hakatak™	<i>Colletotrichum gloeosporioides</i>	Silky Hakea	South Africa	Morris <i>et al.</i> , 1999 [205]
De vine®	<i>Phytophthora citrophthora</i> p.v. <i>palmivora</i>	Strangler vine	USA	
CASST™	<i>Alternaria cassiae</i>	Sicklepod, senna	Brazil	Charudattan <i>et al.</i> , 1982 [30]
Smolder®	<i>Alternaria</i> sp.	Dodder	USA	Bewick <i>et al.</i> , 2000 [206]
ABG-5003	<i>Cercospora rodmanii</i>	Water hyacinth	USA	Charudattan, 1991, 2001 [31-32]
Dr. BioSedge®	<i>Puccinia canaliculata</i>	Yellow nutsedge	USA	-
Woad Warrior	<i>Puccinia thlaspeos</i>	Isastis tinctoria	USA	-
Camperico®	<i>Xanthomonas Campestris</i>	Annual bluegrass	Japan	-
Mycotech™	<i>Chondrostereum purpureum</i>	Various broad leaved trees	Canada;USA	-
Chontrol™Ecoclear™	<i>Chondrostereum purpureum</i>	Alders and Hard wood	USA	Barton <i>et al.</i> , 2005 [19]
Sarritor	<i>Sclerotinia minor</i>	Dandelion	Australia	Abu-Dieyeh and Watson 2009 [207]

#### Characteristics of Bioherbicides

Bio-herbicides in culture produce durable and plentiful inoculae. They are cultivated by fermentation for obtaining large masses or for obtaining large yields that are active up to a few days

- Bioherbicides are living inoculum of plant pathogen mainly fungi since the potential of bacteria is hardly explored and viruses proves difficult to handle on the ground of their host specificity and dependence on vectors.
- They are capable of in-vitro culturing in artificial media and mass production.
- They are applied directly to the target weed to kill or reduce the population and growth and they are commercially formulated and spray like herbicides over crops and weeds in the field.

#### Advantages and Disadvantages of Bioherbicides

The increasing public interest in safe and environmentally friendly products has led to the emergence of numerous new options for pest control, including weed management. Bioherbicides, derived from plant extracts, phytopathogenic microorganisms, or microbial phytotoxins (known as mycoherbicides), represent a valuable approach to weed control (Lamberth & Cai, 2016; Boyetchko, 2004) [115, 23]. These bioherbicides offer a sustainable and ecologically sound alternative to conventional synthetic herbicides. They are often biodegradable, with lower toxicity levels, and can

be part of integrated weed management strategies that minimize environmental impact. However, their effectiveness can vary depending on factors such as weather conditions, target weed species, and application methods. Ongoing research and development are essential to improve the efficacy and broaden the spectrum of bioherbicides available for sustainable weed management practices. Bioherbicides typically lack persistent characteristics, meaning they do not remain active in the environment for extended periods. This characteristic reduces the likelihood of soil and water contamination and minimizes adverse effects on non-target organisms. Bioherbicides derived from allelochemicals are particularly benign to both ecosystems and human health (Soltys, 2013) [173]. Additionally, some allelochemicals are water-soluble, simplifying their application without the need for surfactants (Dayan, 2009; Vyvyan, 2002) [44, 192]. This combination of traits underscores the potential of bioherbicides as a safe and sustainable alternative for weed management, aligning with the growing demand for environmentally friendly solutions. However, further research is necessary to optimize their effectiveness and application methods, ensuring they can be integrated seamlessly into sustainable agriculture practices. Allelochemicals are characterized by their environmentally friendly chemical structures, contrasting with the often complex and potentially harmful structures of synthetic herbicides. Bioherbicides derived from allelochemicals typically exhibit short-lived environmental persistence and



low toxicity, making them safer for the environment and human health (Bailey, 2014) <sup>[16]</sup>. Moreover, these bioherbicides often employ multiple modes of action, which can help reduce the risk of herbicide resistance in weed populations.

Given these characteristics, allelochemicals are considered excellent candidates for the development of bioherbicides, as well as antimicrobial agents and growth regulators. Their natural origins and environmentally friendly attributes make them attractive for various agricultural applications, aligning with the growing demand for sustainable and eco-friendly solutions.

Despite their numerous benefits in sustainable weed control, bioherbicides have limitations that can make them less suitable than synthetic herbicides, especially at the field scale. One significant drawback is their relatively short environmental half-life. While this characteristic reduces environmental toxicity, an effective herbicide needs to persist long enough to achieve the desired effect on weed species (Manahan, 2017) <sup>[119]</sup>.

Another challenge is the variability in the quantity and content of secondary metabolites produced by plants from the same area or taxonomic group. This variability means that not all plants will exude the same amount or quality of allelochemicals, potentially affecting the consistency and efficacy of bioherbicides (Cheema & Imatomi, 2013; Albert, 2012) <sup>[39, 5]</sup>.

Additionally, many allelochemicals are structurally complex and costly to produce, limiting their practicality as agrochemicals. The expense associated with these compounds can hinder their widespread adoption in agriculture despite their potential benefits (Cheema & Imatomi, 2013; Albert, 2012) <sup>[39, 5]</sup>. For example, the cyclic tetrapeptide toxin is an excellent herbicide, but it is very expensive (Imatomi, 2013) <sup>[69]</sup>. Some natural products that exhibit high phytotoxicity can also be highly poisonous to mammals. For example, AAL-toxins are known to be fairly toxic to mammalian cells (Charudattan, 2001) <sup>[31-32]</sup>. Due to these characteristics, the interest in developing these natural phytotoxins into herbicides for weed management has been reduced.

### Currently Marketed Products

Since their introduction in 1980, bioherbicides have been part of a broader category of biopesticides that also includes bioinsecticides, biobactericides, biofungicides, and bionematicides. While several biopesticides have been introduced globally, bioherbicides constitute less than 10% of the total market share (Hintz, 2007) <sup>[64]</sup>.

Most registered bioherbicides are derived from microorganisms (Zeng, 2020) <sup>[200]</sup>. By 2012, seven bioherbicides were registered in the USA, six in Canada, and one each in Ukraine and Japan (Bailey). In 2016, there were thirteen bioherbicides marketed globally, with nine derived from fungal microorganisms, three from bacterial microorganisms, and only one from plant extracts. By 2020, six commercial bioherbicides derived from essential oils and/or their compounds were registered and available in the USA (Verdeguer *et al.* 2020) <sup>[190]</sup>.

### Approaches to biological control of weeds

#### Classical/Inoculative Approach

Classical / Inoculative approach involves the release of bio-agents (insects, fungi, nematodes, fish and other biological

system) just for once in the belief that it will readily adapt to the prevailing climate and multiply enough to keep pace with the multiplication rate of weed. The main principle of classical approach is “The regulation of population of an individual pest below the economic threshold”. Therefore, biological control exercise to limit their infestation in such a density/level at which they will not be highly damaging to the crops (Jha *et al.*, 2008; Jha *et al.*, 2011; Pahade *et al.*, 2023.; Kumhar *et al.*, 2022; Kumbhare *et al.*, 2023; Sahu *et al.*, 2023 and Patidar *et al.*, 2023) <sup>[87, 81, 135, 111, 113, 109, 151, 153, 139]</sup>.

#### Augmentative/Inundative Approach

Several inocula such as fungi, bacteria, parasitic nematodes even viruses having tested control ability over a weed species may be applied by inundative approach. certain philosophy like “Myco-herbicides/bioherbicides is sprayed like herbicides does not match with insects, which are released but not sprayed. Therefore, better “bio-herbicides” should include only microorganisms e.g. fungi, bacteria, nematodes viruses but not insects for which classical / Inoculative approach is the main domain.

#### Comparison between Inoculative and Inundative approach

Economic risk is a critical factor influencing the acceptance of a biocontrol program among farmers and consumers. Farmers, in particular, tend to demonstrate risk-averse behavior, seeking to minimize wide fluctuations in income from year to year (Auld and Tisdell, 1987) <sup>[14]</sup>. This risk consideration is more pertinent to the inundative approach than the classical approach in biocontrol.

The classical biocontrol approach typically involves the release of controlled and monitored biocontrol agents by government departments or regulatory bodies. This level of oversight and management helps mitigate economic risks associated with the introduction of biocontrol agents. In contrast, the inundative approach involves the release of large numbers of biocontrol agents, which can lead to greater uncertainty and potential economic risks for farmers. Consequently, farmers may be more cautious in adopting inundative biocontrol methods, preferring the more regulated and controlled classical approach for its predictability and lower economic risk. Bioherbicides differ from classical biocontrol agents in that consumers, such as farmers, are responsible for evaluating their economic benefits compared to chemical alternatives (if available). This evaluation necessitates farmer education programs provided by advisory and extension workers, as well as effective marketing strategies from the manufacturers and distributors of bioherbicides.

The importance of host specificity differs sharply between the classical and inundative approaches. Classical biological control insists upon specificity. The U.S. Environmental Protection Agency (EPA), along with equivalent agencies in other countries like the Australian Plant Quarantine section of the Department of Primary Industries and Energy, imposes stringent and extensive host-range tests on classical biocontrol agents. These tests are designed to ensure that these agents do not attack non-target plants before they are deemed safe for importation and release (Charudattan, 1982) <sup>[30]</sup>.

Despite these precautions, there is still cause for concern because the behavior of an exotic pathogen or parasite is

challenging to predict accurately (Evans, 1986) [53]. This unpredictability was demonstrated by the rapid adaptation of *Puccinia xanthii* Schaw. to both *Helianthus annuus* and *Calendula officinalis* in Australia following an accidental introduction in 1975 (Alcorn, 1976) [6]. Prior screening trials in Europe had not indicated that sunflowers were susceptible to this rust, and indeed, sunflowers had been continuously exposed to the fungus in America without issue.

### Formulation of bioherbicides

The formulation of bioherbicide active substances is a critical factor in determining their efficacy and success in weed control. Bioherbicides based on microbial metabolites and natural products must be formulated to protect their chemical nature and enhance their entry into plants. Therefore, formulations need to be developed to optimize deployment and efficacy in the field (Morra *et al.*, 2018; Hynes and Boyetchko, 2006) [124, 67].

Traditionally, formulation development has focused on adapting ingredients from the chemical pesticide industry, often without considering compatibility with microorganisms or natural products. Successful formulation development has been achieved through collaboration with industries such as food processing and pharmaceuticals, leading to formulations compatible with bioherbicide substances (Hynes and Boyetchko, 2006) [67].

The primary functions of bioherbicide formulations should include preparing the weed for infection by the pathogen and protecting the pathogen against environmental constraints while promoting disease development (Charudattan, 2001) [31-32]. Foliar and stem fungal pathogens, for example, require specific humidity (dew periods) and temperature ranges for full effectiveness. Unique formulations have been developed for mycoherbicides to ensure efficacy after agent delivery in the field. Factors critical in bioherbicide formulation technology, as outlined by Boyetchko *et al.* (1998) [24], include maintaining or enhancing the effectiveness of the biocontrol agent while ensuring compatibility with conventional field application practices.

Popular formulations for bioherbicides include various emulsions, organosilicon surfactants, hydrophilic polymers, and encapsulated granules made from alginate, starch, or cellulose. Each formulation type has its advantages and disadvantages in promoting the virulence and efficacy of biotic agents and in terms of ease of application (Charudattan, 2001; Hallett, 2005) [31-32].

### Bio herbicides in integrated weed management

Integrated Weed Management (IWM) is a comprehensive and long-term approach to weed control that utilizes all available strategies, including tillage, cultural practices, herbicides, allelopathy, and biological control (Sinodiya and Jha (2014) [169], Jha and Kewat (2013) [79-80], Tiwari *et al.* (2013) [185], and Tiwari *et al.* (2011a) [186] Kewat *et al.*, 2009) [96]. The goal of IWM is to reduce the weed seed bank in the soil, prevent weed emergence, and minimize competition from weeds growing alongside desired plants (Aldrich and Kremer, 1997) [97].

Similar to chemical herbicides, bioherbicides are often most effective when used as part of an integrated approach rather than as a standalone tactic. This integrated approach offers the most promising scenario for bioherbicides to be a

practical management option in cropping systems. When viewed as a three-part system, weed management provides several opportunities for integrating bioherbicides at critical stages during weed development: targeting seeds in the soil, controlling growing and competitive plants, and managing seed production (Aldrich and Kremer, 1997) [97].

### Integrating bioherbicides with chemical herbicides

Integrating bioherbicides into weed management strategies can enhance overall control, especially when dealing with multiple weed species. Since most biological control agents are specific to single weed species and fields are often infested with several predominant weeds, combining bioherbicides for one species with herbicides for others makes sense (Kumar *et al.*, 2023, Shri *et al.*, 2014 Jha and Soni 2011 and Sanodiya *et al.* 2013, Kantwa *et al.* 2019, Jha *et al.* 2007 and Kumbhare *et al.*, 2023) [105, 107, 163, 81, 159, 91, 88, 109].

For instance, the bioherbicide *Fusarium solani f. sp. cucurbitae*, which targets Texas gourd (*Cucurbita texana*), a problematic weed in soybean and cotton fields in the southern United States, was found to be compatible with trifluralin. This compatibility allows for integration into a broader weed management strategy within the crop (Weidemann and Templeton, 1988) [193].

Moreover, combining reduced rates of herbicides with mycoherbicides can improve the effectiveness of the latter against weeds. For example, *Phoma proboscis* was more effective in controlling field bindweed (*Convolvulus arvensis*) when combined with sublethal doses of 2,4-D compared to when applied alone (Heiny, 1994) [63]. Similarly, applying a sublethal dose of the pathogen *Pyricularia setariae* along with one-fourth the rate of three herbicides achieved complete control of green foxtail, demonstrating a synergistic effect between the pathogen and herbicides (Peng and Byer, 2005) [140-141].

In another example, the fungus *Colletotrichum gloeosporioides f. sp. malvae*, which naturally controls round-leaved mallow, achieved adequate control (about 75% kill) when used alone as a bioherbicide (Grant *et al.*, 1990) [58]. However, since several chemical herbicides are effective only on round-leaved mallow at the early seedling stage, tank mixtures of the fungus with metribuzin or imazethapyr at recommended rates significantly improved control and reduced biomass production compared to using the fungus or herbicide alone. These examples underscore the effectiveness of combining different weed control strategies and highlight the need for integrated approaches in managing weed infestations.

### Integrating bio herbicides with cultural practices

Cultural practices are adaptable for the delivery and integration of bioherbicides in cropping systems. Practices such as crop rotation, tillage, organic amendments, reduced chemical inputs, and crop varietal selection can encourage the development of specific inhibitory microorganisms in the soil and on roots. This can be achieved by enhancing indigenous disease-suppressive rhizobacteria (DRB) or by applying selected cultures along with primary and cover crops.

Combined management activities in long-term cropping systems can select and stimulate soil organisms that directly affect weed growth, leading to high populations of

deleterious soil microorganisms that contribute to natural weed suppression (Kremer and Li, 2003) <sup>[102]</sup>. Higher proportions of indigenous rhizobacteria inhibitory to downy brome and jointed goatgrass were detected under either conventional or reduced tillage compared to no-till, suggesting that application of selected DRB during tillage may be effective in integrated weed management (Kremer and Kennedy, 1996) <sup>[101]</sup>.

Lindquist *et al.* (1995) <sup>[116]</sup> reported that a natural population of the fungal pathogen *Verticillium sp.* significantly suppressed velvet leaf growth in soybeans under reduced tillage. Addition of composted swine manure to soil inhibited germination and seedling emergence of three weed species, possibly by enhancing soilborne weed-suppressive microorganisms (Menalled *et al.*, 2005) <sup>[121]</sup>.

Selecting highly competitive and allelopathic soybean varieties, matched with compatible bioherbicides, may provide early-season weed suppression and require only minimal subsequent postemergence weed control (Rose *et al.*, 1984) <sup>[105]</sup>.

Cover crops and mulches are vital for improving soil health and restoring soil productivity in agricultural management systems. They are well-suited for integrating bioherbicides by delivering the agents on seeds and promoting their establishment in soils for an attack on weed seeds and seedlings before planting the main crop. At planting, several cover crop species inoculated with a DRB bioherbicide maintained DRB rhizosphere populations, which transferred to and promoted root colonization of giant foxtail seedlings that emerged later with the primary crop after cover crop termination (Kremer, 2000) <sup>[99]</sup>. The combined effects of DRB and allelopathic cover crop residues suppressed weed growth. Formulated bioherbicides are applied at planting to attack weed seeds and seedlings by delivering microbial agents to the soil through direct inoculation of crop seeds or promoting crop root colonization (Skipper *et al.*, 1996) <sup>[170]</sup>. Crop roots not only deliver microbial agents to adjacent weed roots but may also maintain or even enhance biotic agent density for seedling attack later in the season. Combining bioherbicides with other control methods can enhance weed control effectiveness. For example, the efficacy of a bioherbicide on hemp sesbania (*Sesbania exaltata*) was increased by combining selected bacteria with the fungal pathogen *Colletotrichum truncatum* (Schisler *et al.*, 1991) <sup>[160]</sup>. Similarly, combining a *Colletotrichum sp.* bioherbicide with a naturally occurring rust fungus allowed the bioherbicide to infect the weed host (*Xanthium sp.*) through rust lesions, leading to the plant's death (Morin *et al.*, 1993) <sup>[123]</sup>.

Furthermore, combining a seed-feeding insect with seed-attacking fungi significantly reduced velvet leaf seed viability and seedling emergence while increasing seed infection compared to using either the insect or fungus alone (Kremer and Spencer, 1989) <sup>[103]</sup>. This approach can effectively manipulate and reduce weed seed banks in soil. Another strategy involves using soil-applied detrimental bacteria and/or fungi in combination with root-feeding insects, which has been shown to accelerate the growth decline of leafy spurge and knapweed weeds under continuous insect larvae attack (Caesar, 2005) <sup>[27]</sup>. These examples demonstrate how synergistic interactions between bioherbicides and other control methods can enhance weed management efforts.

## Synergism between bio-herbicides and chemical herbicides

Acifluorfen and bentazon were the most effective synergists and provided significant control in several weed/pathogen combinations.

## Achievements, Developments and Future Challenges

Recent developments have led to over 22 different bioherbicide formulations being registered for weed control, with the global bioherbicide market expected to reach USD 4.14 billion by 2024. However, barriers to their widespread adoption remain. Environmental conditions such as humidity, soil type, temperature, and UV light influence bioherbicide efficacy and formulation. Climate change may also impact weed population dynamics and herbicide resistance, emphasizing the need for improved weed management strategies like bioherbicides.

Formulating and commercializing bioherbicides is challenging due to the need to maintain the viability and stability of living biotic agents. Costs associated with formulation and registration, as well as regulatory hurdles, also hinder their adoption. Overcoming these barriers requires increased education about bioherbicides, technological advancements, and collaboration between governments and agencies to address uncertainties and promote their long-term use. Farmers are more likely to adopt low-cost solutions for weed management, highlighting the importance of addressing economic factors in the adoption of sustainable weed management practices.

## Conclusion

It seems like we've summarized the key points quite effectively. Bio-herbicides offer a targeted and environmentally friendly approach to weed management, and integrating them into existing practices can enhance overall effectiveness. Overcoming challenges such as cost and availability will require a concerted effort from various stakeholders, including governments, research institutions, and agricultural producers. Education and technological advancements will play crucial roles in promoting the long-term use of bio-herbicides.

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