frontiers | Frontiers in Agronomy

TYPE Systematic Review
PUBLISHED 08 August 2025
DOI 10.3389/fagro.2025.1633565

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OPEN ACCESS

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RECEIVED 22 May 2025
ACCEPTED 22 July 2025
PUBLISHED 08 August 2025

CITATION
Zhang Z, Becerra-Alvarez A and Al-Khatib K (2025) Physiological action of bioherbicides in weed control: a systematic review. *Front. Agron.* 7:1633565.
doi: 10.3389/fagro.2025.1633565

Physiological action of bioherbicides in weed control: a systematic review

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Introduction: Bioherbicides are naturally derived substances that can be used to control weeds. Bioherbicide compounds can be alternatives to synthetic herbicides and are key resources for the discovery of novel molecules and modes of action (MOA) for weed control. To better understand the physiological action of bioherbicides, a systematic review was conducted with an emphasis on understanding the MOA of bioherbicides.

Methods: A systematic review screened 287 studies of published literature. The review retained seventeen studies that demonstrated evidence of bioherbicide mode of action.

Results: From our review, we found that bioherbicides are often a mixture of various substances and potentially have multiple MOAs. Compound mixtures present in bioherbicides intrinsically increase the difficulty level in elucidating the mechanistic causation for plant injury. The majority of empirical studies reported injury to weeds at the plant, tissue, or cell level - but were unable to define specific biological pathways affected by bioherbicide application. In total, seventeen studies had strong evidence for specific MOAs, including photosystem II inhibition, microtubule synthesis inhibition, carotenoid synthesis inhibition, cellular metabolism inhibition, and auxin mimics.

Discussion: Hypothesis driven research, chemical characterization, gene expression, and molecular *in-silico* modeling were important steps in identifying the MOA and should be considered in future studies. It was not uncommon to observe bioherbicide compounds with evidence for more than one MOA. With a better understanding of bioherbicides and their herbicidal action, increased efficacy can be achieved and catalyze novel product development.

KEYWORDS

bioherbicide, chemical characterization, organic agriculture, mode of action, site of action

Introduction

Weed's interference with cultivated crops is a major challenge in cropping systems and when unmanaged can cause significant yield reductions (Holm, 1971). The increased use of synthetic herbicides in conventional agricultural production has helped alleviate weed management challenges (Harker, 2013). However, the increasing resistance to synthetic herbicides has more recently caused reduced weed control with available herbicides; therefore, the need for new management options and new herbicidal compounds are necessary (Becerra-Alvarez et al., 2023; Harker, 2013; HRAC, 2024).

Organic agriculture has received increased popularity in recent years among producers and consumers because of premiums paid for organic products and zero use of synthetic chemicals in food production (Carlson et al., 2023). A challenge in organic production systems is the reduced number of available herbicides for weed control. Organic producers rely heavily on cultural or mechanical methods for weed control (Fennimore and Doohan, 2008). There is a need for alternative methods in weed control such as natural compounds that can be used in organic production systems.

Consequently, bioherbicides have gained notable traction in recent years (Cordeau et al., 2016). Bioherbicides are pesticide products derived from living organisms or their natural metabolites that can be used to control weeds (Hoagland et al., 2007). Bioherbicides currently represent less than 10% of the total herbicide market due to a variety of issues ranging from non-selectivity, to reduced efficacy, and pricing challenges (Cordeau et al., 2016). However, they have the potential to improve organic weed control, and can also be a starting point for synthetic herbicide development, especially for undiscovered molecules and unutilized modes of action (MOA) (Duke et al., 2014).

Additionally, there are co-benefits associated with bioherbicides, such as reduced environmental persistence that reduces the risk of environmental pollution and human health detriments, contributing to an integrated pest management approach by including an additional option of weed control (Cordeau et al., 2016). Furthermore, many robotics and precision technologies for weed control have increased as alternatives to synthetic herbicides; however, the technology can be expensive for many producers and even more expensive to maintain because of the lack of supporting infrastructure (Westwood et al., 2018). The infrastructure for applying chemicals in agriculture is already in place, and bioherbicides may be readily adopted if effective options are developed to be market-ready.

Bioherbicides have significant utility and there is a need to consolidate current knowledge of physiological action. While several extensive reviews have been conducted on the topic of bioherbicides (Cordeau et al., 2016; Radhakrishnan et al., 2018; Supplementary Table S1), no review utilized a systematic search methodology. Hence, with an urgent need to understand the physiological mode of action (MOA) of bioherbicides, we conducted a systematic review utilizing a transparent search methodology to fill this knowledge gap.

Methods

A systematic review method was utilized to identify research methods toward determining the MOA of bioherbicides using the Scopus and Web of Science databases (Munn et al., 2018). The following Boolean search term was used with no restriction to study date range: (TITLE-ABS-KEY (bioherbicide) OR TITLE-ABS-KEY (bio AND herbicide) OR TITLE-ABS-KEY (organic AND herbicide) AND TITLE-ABS-KEY (weed) AND TITLE-ABS-KEY (mode AND of AND action)) (Koutsos et al., 2019; Moher et al., 2009). Results were evaluated and presented using the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) (Moher et al., 2009).

The search terms yielded a total of 291 hits with four repetitions. In total, 287 studies were screened (Figure 1). The literature search only included published literature. The hits were screened using title, abstract, and full paper review. Studies were rejected for irrelevance to weed control or lack of connection to bioherbicides in the title and abstract screen, totaling 137 studies removed. Furthermore, studies were rejected in the full paper screen if they did not aim to understand or characterize physiological action, totaling 133 studies removed. Studies were reviewed for quality, and only replicated studies were retained. Studies removed included empirical studies of molecules with potential bioherbicidal effects, empirical studies on allelopathic effects, empirical studies on biological control of weeds, and reviews. The final list selected studies that demonstrated significant evidence for the MOA (Table 1). For a complete list of shortlisted and screened materials, refer to Supplementary Materials (Supplementary Tables S1, S2).

Results

Overview

This review retained 17 studies (Table 1; Figure 1). Many of the studies removed had a focus on characterizing the composition of various essential oils and phytotoxins; however, many did not pursue additional studies for MOA. It is noted that most molecules with MOA characterization studies were compounds in the market or were more market-ready (Table 1).

The central question for the review was to understand the MOA in bioherbicides. A key finding is that specific mechanisms for injury causation are not well understood. Most empirical studies examined damage at the whole plant or cellular level but failed to relate it to specific plant biological processes or site of action (Supplementary Table S1). This was very common among studies reviewed that studied plant essential oils, which highlighted many similar compounds like oxygenated terpenoids, phenolic acids or other volatile organic compounds with injury like oxidative stress, evidence of reactive oxygen species leading to tissue damage, reduced chlorophyll or cellular respiration (Ni et al., 2024; Kaur

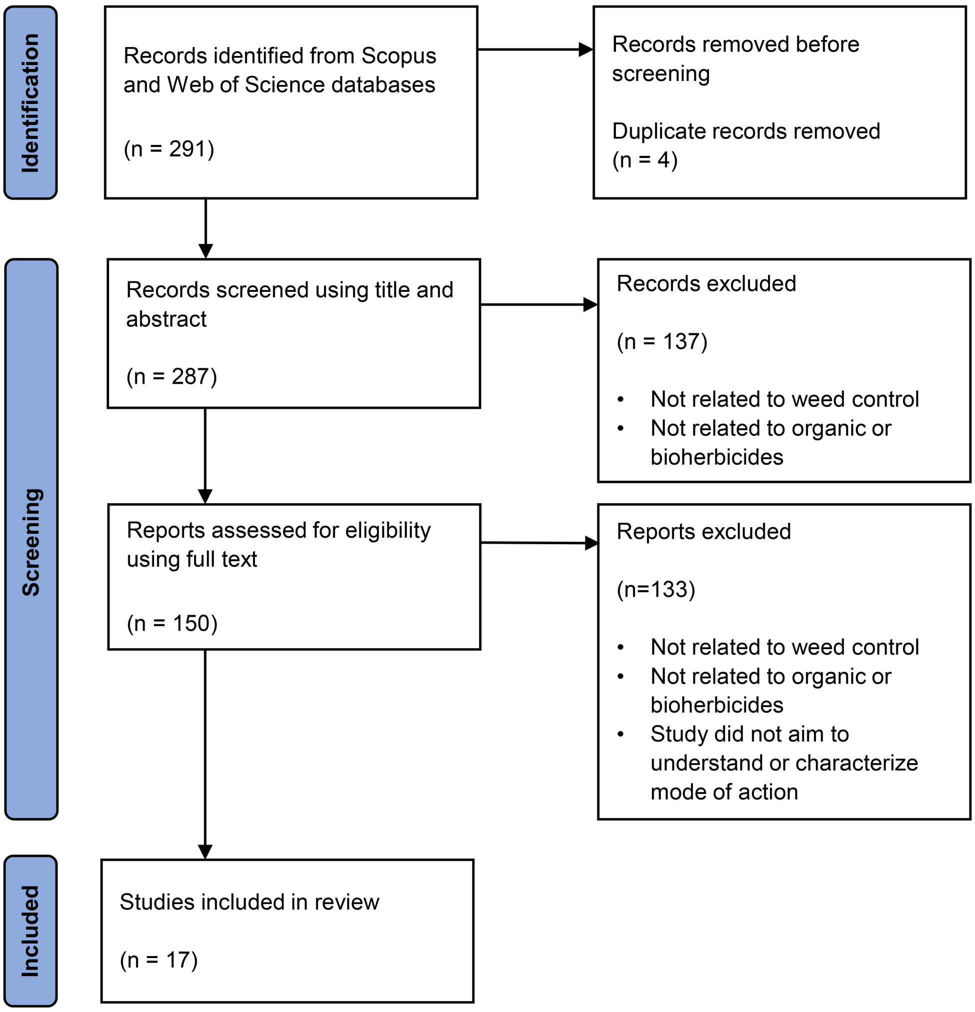

FIGURE 1
PRISMA flow diagram detailing systematic search workflow on the physiological action of bioherbicides for weed control.

et al., 2011; Anwar et al., 2023). Similarly, microbial phytotoxins demonstrated injury symptoms of a plant under stress with oxidative stress, and a reduction of chlorophyll or cellular respiration (Jiang et al., 2008; Guo et al., 2021; Yang et al., 2024). However, the injury descriptions are not enough to narrow down the MOA.

Bioherbicides can take different production routes to reach the final application (Figure 2) (Zhang, 2025). The commonality among all products examined was that the base material was naturally derived from microbes like fungi and bacteria or plants. The compounds can then either be directly used for application or further separated or processed before being applied for weed control (Figure 2). Microbes were at times cultured and directly applied to weeds (Radhakrishnan et al., 2016). Inoculation acted as a direct introduction of microbial populations and has a similar effect to disease damage in crops. Alternatively, microbes were cultured for the harvest of their secondary metabolites or phytotoxins to be used for application (de Almeida et al., 2020). The culturing process required specific media and formulation for maximum herbicidal effect. For example, de Almeida et al. (2020) tested a ratio of sucrose and corn steep liquor (CSL). The ratio of 13 g L^{-1} of sucrose and 15 g L^{-1} of CSL was demonstrated to be the optimum media for herbicide production by the fungus, *Phoma* sp (de Almeida et al., 2020). When plants were used as the base material, compounds were synthesized by extracting chemicals from various tissues such as leaves and seeds. Many techniques were used for extraction, including distillation, water-based extraction, and organic extractions (e.g. ethanolic extractions) (Ni et al., 2024). Weeds were either sprayed with the "raw" extract or had specific compounds further separated and formulated before application.

For both microbial and plant extracts, it was noted that the initial product obtained is not a pure substance but rather a "mixture" of different compounds (Ni et al., 2024). The application of a "raw" extract with multiple compounds thus has the potential to cause different types of injuries to weeds – possibly with multiple MOAs (Dayan et al., 2015). The mixtures of various compounds could be the reason for the unclear physiological action causing plant injury and observations of various plant injury

TABLE 1 Summary of studies that investigated the physiological action of bioherbicides on weed control and determined a mode of action.

Study	Control agent	Production process	Molecule (s) investigated	Type of organism derived from	Species	Evidence for mode of action[a]
Guo et al., 2021	Phytotoxins	Fermentation in apple products	Patulin	Fungi	*Penicillium, Aspergillus, and Byssochlamys*	PSII inhibitor
Yang et al., 2024	Phytotoxins	Culture and extract	Citrinin	Fungi	*Penicillium, and Aspergillus*	PSII inhibitor
Xiao et al., 2020	Phytotoxins	Culture and extract	Alamethicin	Fungi	*Trichoderma viride*	PSII inhibitor
Moura et al., 2020	Phytotoxins	Culture and extract	Methanolic extracts	Fungi	*Diaporthe phaseolorum, Penicillium simplicissimum and Trichoderma spirale*	PSII inhibitor
Alvarez-Rodríguez et al., 2023	Essential oils	Extract	Polyacetylene carlina oxide	Plant	*Carlina acaulis* L.	PSII inhibitor
Pouresmaeil et al., 2020	Essential oils	Extract	α-thujone, camphor, 1,8-cineole and β-thujone	Plant	*Artemisia fragrans* Willd	PSII inhibitor
Anwar et al., 2023	Phenolic acids	Extract	Ferulic acid (FA) and gallic acid (GA)	Plant	Various	PSII inhibitor
Dayan et al., 2015	Essential oils	Extract	Sarmentine	Plant	*Piper* sp.	PSII inhibitor, lipid peroxidation, fatty acid synthesis inhibitor
Jiang et al., 2008	Phytotoxins	Fermentation and extract	α,β-dehydrocurvularin	Fungi	*Curvularia eragrostidis*	Microtubule inhibitor
Chaimovitsh et al., 2017	Essential oils	Extract	17 types of monoterpenes	Plant	Various	Microtubule inhibitor
Hubbard et al., 2015	Phytotoxins	Culture and extract	Macrocidins	Fungi	*Phoma macrostoma*	Carotenoid synthesis PDS-inhibitor
Barickman et al., 2024	Essential oils	Extract	Water soluble B-triketone	Plant	Manuka tree	HPPD-inhibitor
Li et al., 2023	Essential oils	Extract	Monoterpenes, including eucalyptol, thujone, β-caryophyllene, borneol, and camphor	Plant	*Artemisia argyi*	HPPD-inhibitor
Tong et al., 2021	Essential oils	Extract	Bruceine D	Plant	*Brucea javanica* (L.)	Phenylpropanoid biosynthesis inhibitor
López-González et al., 2024	Essential oils	Extract	Pelargonic acid	Plant	*Pelargonium roseum* Willd	Auxin mimic
Bajsa-Hirschel et al., 2023	Phytotoxins	Culture and extract	Spliceostatin C	Bacteria	*Burkholderia rinojensis*	Auxin mimic
Radhakrishnan et al., 2016	Phytotoxins	Inoculation	The bacteria itself, unknown phytotoxins	Bacteria	*Enterobacter* sp. I-3	Auxin mimic and amino acids synthesis inhibitor[a]

[a]PSII, photosystem II; PDS, phytoene desaturase; HPPD, 4-hydroxyphenylpyruvate dioxygenase.

symptoms. At the tissue level, injuries such as lipid peroxidation or decreases in chlorophyll synthesis can be detected, but increased "noise" from multiple pathway inhibitions creates difficulty in elucidating a specific mode of action for a given bioherbicide (Puig et al., 2018). This is a common observation with many bioherbicides. Only compounds that become more popular in the market, such as sarmantine and pelargonic acid, have had their physiological action evaluated across different potential MOAs (Dayan et al., 2015; López-González et al., 2024).

Most studies reviewed made a conscious effort to characterize the chemicals present in a compound through techniques such as high-performance liquid chromatography (HPLC), gas chromatography-mass spectrometry (GS-MS), and nuclear magnetic resonance (NMR) (Supplementary Table S1). The

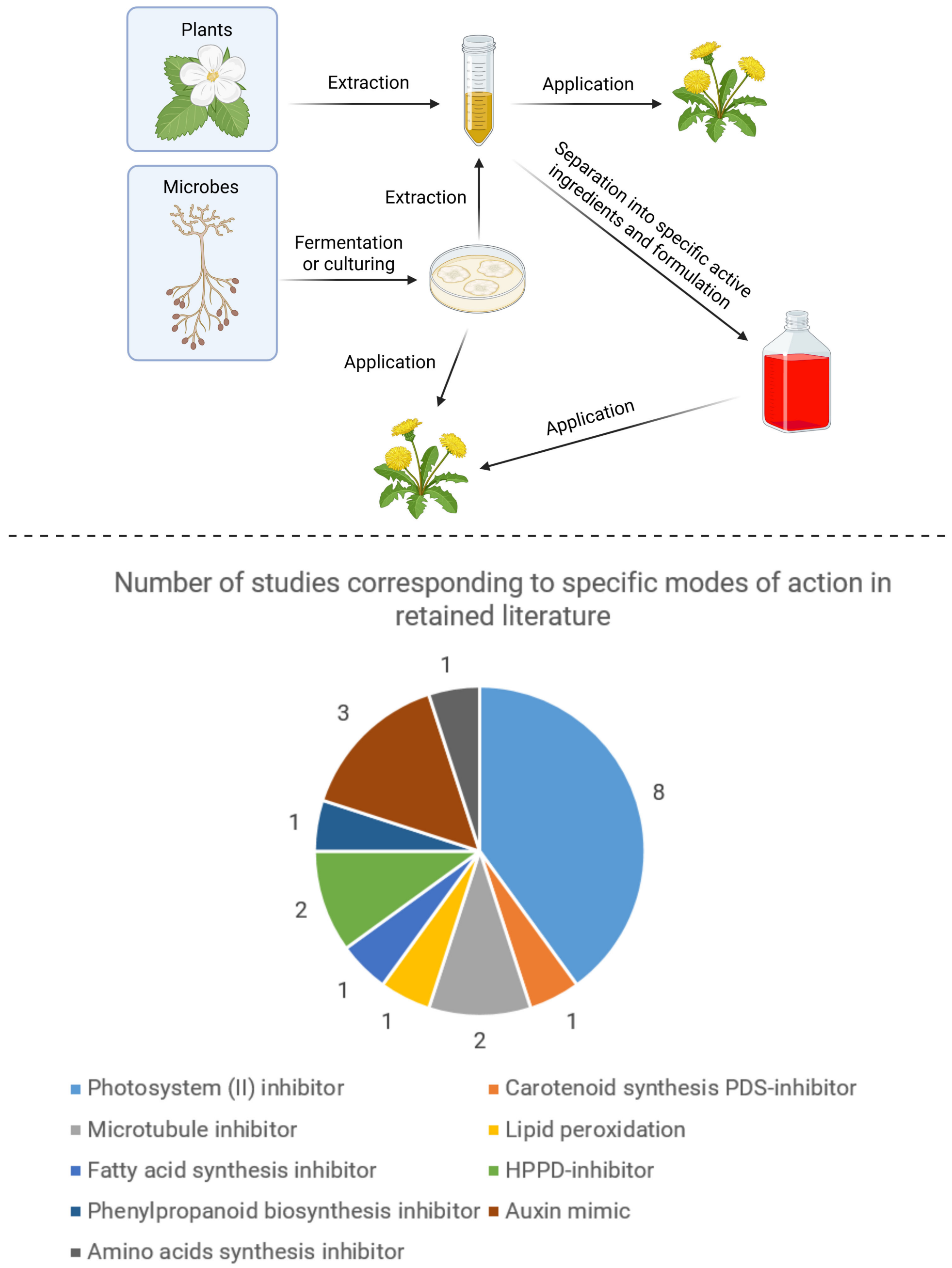

FIGURE 2
Schematic of bioherbicide synthesis and production from plant and microbial sources (top), and distribution of modes of action identified in the review on the physiological action of bioherbicides for weed control (bottom). Created in BioRender. Available online at: https://BioRender.com/ns703i0.

studies that were successful in elucidating a specific mode of action all performed chemical characterization or had done so in previous studies highlighting its importance (Table 1).

The large number of empirical studies screened in this review demonstrates the high efforts for finding new bioherbicides (Supplementary Table S1). While many other factors affect the market readiness of bioherbicides, screening various potential compounds early on that show significant phytotoxicity to the weeds is important to begin the process of discovery. Screening is time consuming, and companies interested in bioherbicides should invest in high-throughput methods for screening new compounds. Bioherbicides are also less stable than synthetic herbicides especially if the formulation contains live organisms, requiring specialized transportation and storage needs to maintain efficacy or increase shelf life, making it a challenge to study and work on (Duke et al., 2014).

Physiological action

Photosystem II inhibition

PS II inhibition was the most common MOA observed in the studies reviewed (Table 1). Patulin and citrinin are phytotoxins that are commonly synthesized in the fermentation of apple products by fungi such as *Penicillium, Aspergillus*, and *Byssochlamys*. Both Guo et al. (2021) and Yang et al. (2024) demonstrated that patulin and citrinin blocked electron transport from the primary to secondary plastoquinone acceptors (Q_A to Q_B) of PS II by binding to the electron acceptor site. Guo et al. (2021) and Yang et al. (2024) both utilized chlorophyll fluorescence, *in vitro* phytotoxicity assessments on plants, and *in-silico* modeling to determine binding to the D1 protein to histidine 252 and histidine 215 for patulin and citrinin, respectively.

The fungi *Trichoderma viride*, *Diaporthe phaseolorum, Penicillium simplicissimum* and *Trichoderma spirale* phytotoxins demonstrated inhibition of PS II (Xiao et al., 2020; Moura et al., 2020). These studies used indirect measurements to conclude that the PS II site was the site of action. Both studies measured electron transport efficiency, which allowed the effect on photosynthesis effect to be studied; however, no direct binding site was defined (Xiao et al., 2020; Moura et al., 2020). Xiao et al. (2020) also used chlorophyll fluorescence measurements.

Four essential oils derived from plants were also found to inhibit the PS II (Table 1). Pouresmaeil et al. (2020) and Alvarez-Rodríguez et al. (2023) both initially did a compound characterization to isolate the most abundant chemical and active ingredient in the essential oils. Anwar et al. (2023) and Dayan et al. (2015) had referenced previous studies of chemical characterization and were evaluating molecules much more widely known and used as bioherbicides, which were phenolic acids and sarmentine, respectively. All studies measured chlorophyll fluorescence and measures to estimate lipid or membrane oxidation induced electrolyte leakage measures and malondialdehyde and hydrogen peroxide concentrations (Pouresmaeil et al., 2020; Alvarez-Rodríguez et al., 2023; Anwar et al., 2023; Dayan et al., 2015). Only Anwar et al. (2023) and Dayan et al. (2015) followed with *in-silico* modeling techniques on the PS II docking protein, and Anwar et al. (2023) also measured the photosynthetic gene (psbA) expression after applications.

Microtubule disruption

Microtubule disruption is another MOA that was demonstrated in two studies (Table 1). Chaimovitsh et al. (2017) investigated the effect of monoterpenes which are naturally occurring allelopathic chemicals in plants on *Arabidopsis thaliana* (L.) seedlings. The compounds were applied to transgenic *A. thaliana* seedlings expressing the microtubule marker GFP-TUA6 and then stained with membrane marker FM4–64 before being evaluated by confocal microscopy. The results elucidated that the application of limonene, one of the plant derived essential oils evaluated, had a strong effect on disrupting microtubule formation and function (Chaimovitsh et al., 2017). The microscopy of limonene indicated the disappearance of the majority of microtubules and strongly suggest microtubule disruption as the MOA (Chaimovitsh et al., 2017).

The fungi derived phytotoxin, α,β-dehydrocurvularin, from the fungus *Curvularia eragrostidis* similarly elucidated the disruption of mitosis of root tip cells (Jiang et al., 2008). When α,β-dehydrocurvularin was applied at a concentration higher than 344 μM to garlic root tips, all mitotic cells were arrested in the late prophase stage, and became multinucleate cells (Jiang et al., 2008). Although there is no direct evidence showing microtubule disruption, it was hypothesized that microtubule function is disrupted during the late prophase stage, at which mitosis was halted. Additionally, α,β-dehydrocurvularin also decreased photosynthetic capacity by affecting chlorophyll A fluorescence, photophosphorylation, and Mg^{2+} ATPase activity at high concentrations in mature plants, possibly suggesting multiple MOAs (Jiang et al., 2008).

These two studies demonstrated an effort to learn if microtubule inhibition was significant enough to be the MOA. Many empirical studies screened in this review reported that germination was halted or reduced from field or greenhouse applications of bioherbicide; however, insufficient data was available to narrow down a pathway (Supplementary Table S1). The monoterpenes family was a common group of essential oils observed in many empirical studies reviewed like limonene evaluated by Chaimovitsh et al. (2017). However, even among the studies that demonstrated an MOA for an essential oil there was a diversity of MOAs concluded. Therefore, it will be worth exploring other molecules individually.

Carotenoid synthesis-inhibitors

In this review, Barickman et al. (2024) evaluated water soluble β-triketone extracted from Manuka oil (*Leptospermum scoparium*). While this study did not discover the MOA it is well documented by Dayan et al. (2007) that β-triketones target the hydroxyphenylpyruvate dioxygenase (HPPD) site on plants. The HPPD pathway is part of the carotenoid synthesis pathways that synthesize pigments for plants and therefore lead to bleaching injury or white like appearances on leaf tissue. The β-triketones from Manuka trees have been commercially synthesized and developed into successful herbicides used widely (Dayan et al., 2007). This is a great example of natural molecules being a pathway for synthetic herbicide discovery. Barickman et al. (2024) used the naturally derived β-triketone which showed success and similar results to other available bioherbicides. Manuka oil extract is a substance that is market ready with much more information and needs more applied research to expand its use.

Li et al. (2023) determined the essential oils from *Artemisa argyi* to be HPPD-inhibitors. The researchers used chemical characterization to determine that the oils were high in monoterpenes like including eucalyptol, thujone, β-caryophyllene, borneol, and camphor. The studies included phytotoxicity bioassays, estimation of electrolyte leakage, measuring reactive oxygen species and enzyme activity, and *in-silico* modeling techniques (Li et al., 2023). All molecules demonstrated an increase in reactive oxygen species and membrane disintegration;

however, modeling efforts showed all molecules binding with the HPPD enzyme which provides conclusive results for HPPD-inhibitors (Li et al., 2023).

The phytotoxins, macrocidins, are produced by *Phoma macrostoma* - a fungus. Macrocidins caused bleaching symptoms on leaf tissue after fungal infection (Hubbard et al., 2015). In this study, it was hypothesized that microcidins affected the phytoene desaturase (PDS) pathway in carotenoid synthesis because of the bleaching symptoms, and from previous work that demonstrated no inhibition on the HPPD enzyme; therefore, the PDS enzyme inhibition could answer the carotenoid buildup and reduction in impairment of the PS II observed from previous work (Hubbard et al., 2015). The study measured carotenoid and chlorophyll concentrations and compared them to diflufenican, a synthetic PDS-inhibiting herbicide widely known. PDS-inhibition was concluded from this study as the MOA of macrocidins (Hubbard et al., 2015).

Phenylpropanoid biosynthesis inhibition

Only one study demonstrated phenylpropanoid biosynthesis (PB) pathway inhibition in plants. The PB pathway is a precursor to lignin and flavonoids synthesis - which are important for plant growth at the seed stage and combat against environmental stresses (Tong et al., 2021). Tong et al. (2021) studied bruceine D, an essential oil derived from *Brucea javanica* (L.). The PB pathway was hypothesized to be inhibited because it is important for seed germination and the bioherbicides appeared to affect germination and early plant growth. The researchers measured lipid peroxidation, enzyme activity, lignin and flavonoid concentrations, while also measuring gene expression after an application from genes known to stimulate PB synthesis. (Tong et al., 2021). The results demonstrated that the PB pathway was disrupted and recorded a new pathway that may be affected by bioherbicides not previously mentioned (Tong et al., 2021).

Auxin mimics

Three studies demonstrated evidence of auxin mimic as the MOA. Pelargonic acid is a commonly known commercially available broad-spectrum bioherbicide, and the MOA was classified by the Herbicide Resistance Action Committee as an unknown multisite MOA (HRAC, 2024). López-González et al. (2024) make the case for pelargonic acid to be altering the auxin polar transport. The study used plant phytotoxicity studies, light and transmission electron microscopy to study movement in the plant roots, a pharmacological approach to evaluate auxin mimic MOA, assays with transgenic lines, and *in-silico* modeling to reach their conclusion (López-González et al., 2024). Pelargonic acid shared the same binding site as the natural auxin IAA and resulted in similar symptoms as auxin mimics in the roots (López-González et al., 2024). Pelargonic acid does have multiple MOAs, and this study concluded that one of the pathways is auxin mimic.

Bajsa-Hirschel et al. (2023) determined the Spliceostatin C, a phytotoxin from bacteria, behaves as an auxin mimic in *Arabidopsis thaliana* seedlings. The researchers used an approach with phytotoxicity bioassays, gravitropism assay for root tip re-orientation study, measured gene expression of known auxin genes, and finally *in-silico* modeling to reach their conclusion. All studies supported the MOA to be auxin mimic.

Out of 93 studies screened on the topic of biological control of weeds, five studies attempted to characterize the compounds and understand physiological action of the weed control (Supplementary Table S1); however, only one Radhakrishnan et al. (2016) was successful. Radhakrishnan et al. (2016) studied the MOA of *Enterobacter* sp. I-3 as it inoculates plants for weed control. The researchers inoculated seeds of weeds and measured abscisic acids, gibberellin concentrations, common natural auxins, along with amino acid concentrations while comparing them to known auxin herbicides applications. *Enterobacter* sp. I-3 demonstrated a disruption of plant hormones in the plant at the gibberellic acid metabolic pathway while also affecting the amino acid synthesis of the plant (Radhakrishnan et al., 2016).

Discussion

The majority of screened empirical studies did not identify a clear MOA but mainly discussed injury at the plant, tissue, or cellular level relating to the application of a bioherbicide. However, key injuries are described, which can be useful to develop hypothesis-driven research to determine the MOA (Supplementary Table S1). Electrolyte leakage and disruption of plasma membrane integrity were commonly observed as common symptoms (Ni et al., 2024; Kaur et al., 2011; Hasan et al., 2022). One example was with the commercially available bioherbicide Weedlock, which has a trademark active ingredient EGX-101™ derived from wild tomato plants (Hasan et al., 2022). Hasan et al., 2022 showed evidence of reactive oxygen species leading to membrane disintegration like protox inhibiting synthetic herbicides; however, many herbicides and even bioherbicides cause reactive oxygen species and further work should find clear evidence of the MOA. It was observed from the review that often multiple compounds cause injury in plants and weeds from the application of bioherbicides (Supplementary Table S1). However, the exact mechanism for injury is unknown and can largely be attributed to the composite nature of bioherbicides as they consist of multiple compounds that have different effects at the plant or cellular level. As such, it is pivotal that future research initially investigates the chemical composition of the herbicidal compounds.

Outlining chemical composition and testing the efficacy of compounds independently or in combination with others is an objective method to discover and develop bioherbicides. For example, Pardo-Muras et al. (2019) tested multiple combinations of volatile organic compounds present and showed that the verbenone and linalool pair were synergistic in herbicidal action, causing irreversible damage to *Digitaria sanguinalis* (L.) Scop. germination. The subsequent development of a bioherbicide can then be focused on optimization of the compounds or discovering additional herbicidal compounds with similar structures. However, chemical characterization alone cannot determine the MOA and additional studies are needed.

It is no surprise that PS II inhibition is the most observed MOA for bioherbicides since photosynthesis is the key physiochemical process for plants to grow and survive, and probably the most studied plant mechanism (Xiao et al., 2020). Sarmentine was the only molecule to be studied across different phytotoxicity routes, and it was determined that it had multiple MOAs, including lipid peroxidation in other areas and fatty acid synthesis inhibition, along with PS II inhibition (Dayan et al., 2015). The molecule sarmentine was studied across various mechanisms because it was thought to affect many areas based on previous studies and similarity to other compounds (Dayan et al., 2015). Given more available resources for bioherbicide research, it may be possible to observe multiple MOAs among other bioherbicide compounds. Essential oils are a mixture of various molecules and potentially target many plant processes (Dayan et al., 2015). The diverse MOA could be an appreciated trait to delay bioherbicide-resistant mechanisms from developing, but difficult to study.

Auxin mimics have been important herbicides and some of the first synthetic herbicides discovered and developed; therefore, it is not surprising that there are natural inhibitors created in nature from microbes that may infect plants, as is observed with many plant diseases that interfere with hormonal pathways of the plant (Radhakrishnan et al., 2016). The pelargonic acid compound was the only plant derived essential oil to affect auxin pathways in plants, and previously thought to affect cell membrane lipid peroxidation (López-González et al., 2024). The other bioherbicide derived from plant essential oils may have similar effects on the auxin pathways but have not been studied yet. The similarities of other essential oils to pelargonic acid in chemical structure or in injury symptoms could provide evidence to study this MOA pathway.

Pelargonic acid is a commonly used bioherbicide in crop production as a broad-spectrum weed control tool and is categorized in the Herbicide Resistance Action Committee (HRAC) as a compound with unknown MOA or multisite target action; however, López-González et al. (2024) demonstrated interference in auxin transport, invalidating the previous suggestions of lipid peroxidation as the MOA (López-González et al., 2024). The improved knowledge of the MOA may help advance field use by improving efficacy. For example, the synthetic auxin transport inhibitor, diflufenzopyr, has a synergistic effect on certain weed species when in mixture with other synthetic auxins (Enloe and Kniss, 2009). Similar applied research can be performed with pelargonic acid and other bioherbicides to determine potential synergistic effects when in mixtures that would be of value for practitioners.

The bioherbicides that demonstrated the MOA of microtubule inhibition and PB inhibition may only have activity during the germination stage or early weed seedling stages (Jiang et al., 2008; Chaimovitsh et al., 2017; Tong et al., 2021). Currently, the commercially available synthetic microtubule inhibitors will only have activity on germinating weed seeds or seedlings as pre-emergence or early post-emergence applications; however, when used appropriately and avoiding crop injury, they are effective weed management tools (Harker, 2013). If it is known that these bioherbicide compounds work on weed seed germination or seedlings only, then pre-emergence applications at the appropriate weed life cycle will improve control in the field.

The bioherbicide control agents resulted in diverse MOAs from the studies reviewed. No patterns were observed with the MOA and type of organism derived from or type of control agent (Table 1). The chemical structure and chemical group of the individual compounds may be more indicative of the physiological action in plants, and bioherbicides can be grouped into more comprehensible categories with future research.

The development and registration of a bioherbicide is contingent on the financial viability for a company to pursue. Screening bioherbicide on major agronomic crops (e.g. rice, wheat, soybean, corn, and cotton) establishes selectivity and showcases the potential for commercialization in important large-acreage cropping systems (Guo et al., 2021; Jiang et al., 2008). However, future research should also focus on bioherbicides in specialty crop systems. Especially for organic specialty crops, bioherbicides can be a new tool to alleviate labor costs on cultivation and hand weeding (Fennimore and Doohan, 2008). A new business paradigm for bioherbicide registrations or assistance from government agencies will be necessary to support efforts in specialty crops.

Bioherbicides can be an additional tool for weed management in crop production (Cordeau et al., 2016). Bioherbicides are often a "mixture" of multiple chemical compounds, making them difficult to screen for phytoselectivity and characterize their composition. Tested bioherbicides often cause multiple injuries, and the elucidation of MOA is challenging. Despite the difficulty in understanding how bioherbicides work, the trend of chemical characterization, hypothesis-driven research, gene expression, and molecular *in-silico* modeling observed in the retained studies help identify specific compounds of herbicidal activity and a MOA. From the studies reviewed, PS II inhibition, microtubule inhibition, carotenoid synthesis inhibition, cellular metabolism, and auxin mimic were identified as MOAs responsible for the injury by bioherbicides. However, it was not uncommon for bioherbicide compounds to demonstrate more than one MOA. More structured studies should uncover the chemical composition and physiological action of bioherbicides to help build a comprehensive and organized understanding of this group of herbicidal compounds.

Data availability statement

The original contributions presented in the study are included in the article/Supplementary Material. Further inquiries can be directed to the corresponding author.

Author contributions

ZZ: Conceptualization, Data curation, Formal Analysis, Investigation, Methodology, Validation, Writing – original draft, Writing – review & editing. AB: Conceptualization, Data curation,

Formal Analysis, Investigation, Methodology, Project administration, Validation, Writing – original draft, Writing – review & editing. KA: Conceptualization, Project administration, Resources, Visualization, Writing – original draft, Writing – review & editing.

Funding

The author(s) declare that no financial support was received for the research and/or publication of this article.

Conflict of interest

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Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fagro.2025.1633565/full#supplementary-material

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