

ISSN Print: 2617-4693 ISSN Online: 2617-4707 IJABR 2024; SP-8(7): 554-560 www.biochemjournal.com Received: 23-05-2024 Accepted: 25-06-2024

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A review on effect of herbicides on soil biology and its functions

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DOI: https://doi.org/10.33545/26174693.2024.v8.i7Sg.1575

Abstract

There is a growing awareness among farmers about the importance of soil for sustaining crop production and providing beneficial ecosystem services. Over the last 2 decades, global herbicide use has increased as farmers have shifted to more sustainable conservation tillage practices and have adopted herbicide-tolerant crop cultivars. While the effects of increased herbicide use on soil biology are under scrutiny, there is a lack of a comprehensive review on this subject. Within this chapter, we detail the chemistry and application of major herbicide categories, and assess soil functions pertinent to crop production. Subsequently, we gather and analyse evidence regarding the impacts of herbicides on soil biota and activity. Overall, the majority of studies indicate that the effects of herbicide application on soil function are predominantly minor and/or temporary. Nonetheless, there are specific cases where findings consistently indicate effects that may substantially change soil function. These instances comprise disturbances to earthworm ecology in soils subjected to glyphosate and atrazine; hindrance of soil nitrogen cycling (encompassing biological nitrogen fixation, mineralization, and nitrification) by sulfonylurea herbicides in alkaline or low organic matter soils; and localized rises in disease due to the use of diverse herbicides. Challenges in extending these findings to broadacre farming encompass the absence of a uniform framework for evaluating herbicide risk to soil biology, the significance of herbicide impact magnitude compared to other soil management practices like tillage or crop rotation, the intricate nature of herbicide formulations and combinations, and the scarcity of long-term field studies.

Keywords: Herbicides, mineralization, nitrification earthworm

Introduction

Soil, a vibrant ecosystem, profoundly influences food production, environmental sustainability, and global equilibrium (Escudey et al., 2019; Andrews et al., 2004; Snapp et al., 2010) [31, 2, 74]. Soil quality, defined as its ability to function within ecosystem boundaries to sustain biological productivity, uphold environmental integrity, and foster plant and animal well-being, hinges on its biological activity. Soil health indicators such as microbial biomass, enzyme activity, earthworm population, and organic carbon content serve as reliable measures (Pandey and Palni, 2010; Mann et al., 2019; Rottler et al., 2019; Moebius et al., 2016) [63, 52, 70, 58]. These bioindicators, responsive to changes in land management practices and environmental conditions, reflect soil biology and health accurately (He and Sikora, 2017; Anderson et al., 2008; Arshad and Coen, 1992; Bending et al., 2004) [39, 1, 3, 11]. They also promptly respond to environmental changes and adequately reflect biological changes induced by pollution and contamination (Singh et al., 2011; Bunemann et al., 2018; Bhowmik et al., 2019) [72, 19, 14]. Their shifts directly impact carbon and nutrient cycling (Bardgett and Putten, 2014; Bhaduri et al., 2018; Bhaduri et al., 2015) [78, 12, 13]. Soil enzymes, crucial for biochemical soil processes, regulate organic matter production and nutrient cycling (Burns, 2013; Caldwell et al., 1999; Farrell et al., 2020; Ciarkowska et al., 2014) [20, ^{21, 32, 22}]. Earthworms contribute to carbon and nitrogen recycling through organic residue decomposition (Edwards and Bohlen, 1996; Tiwari et al., 1989; Parle, 1963) [29, 76]. In contemporary agriculture, herbicides represent a cornerstone for effective weed control and enhanced productivity (Duke, 2012; Gerwick, 2010; Zimdahl, 2004) [27]. While modern herbicides boast high biological activity and selectivity, their inappropriate or continuous use can yield adverse environmental consequences (Singh, 2018; Kraehmer, 2012) [73, 46].

The effects of herbicides on soil ecology hinge on factors such as active substance type, application rates, soil oxidation-reduction potential, and physicochemical properties. Assessing changes induced by herbicides involves analyzing microbial responses, enzymatic activities, and earthworm populations (Meghvansi and Varma, 2010) [55]. Understanding the impact of herbicides on soil health indicators is pivotal for fostering sustainable crop production practices.

History of herbicides development

For the first time in 1880 herbicidal action of some compounds example NaCl, other inorganic salts of K and Mg was highlighted. They were non selective and could hardly be used in crops. As a result, salts of CuSO₄, FeSO₄ and ammonium sulfamate were put into use since late 1890's. In 1919, sodium chlorate was first widely used as soil sterilant for controlling perennial weeds. It also used as total weed killer in roadside, non-crop area but it poses fire hazard and risky to handle (Duke and Powles, 2008) [28]. Later cyanates, thiocyanates and borates were non selective in nature. Hence first discovery in the field of selective weed control as contact herbicide was introduction of DNOC (4,6- dinitro-O-cresol) is member of dinitrophenol in 1933. Because of narrow spectrum activity, crop phytotoxicity it could not become popular in world (Hayes and Laws, 1991) [38]. In 1940-1941 Zimmerman and Hitchcock synthesised 2,4-D in USA. It is the first selective organic herbicide in world. Because of wide spread, longterm use in crops and less risky so gained popularity (Lebaron et al., 2008) [50]. Later different herbicides were developed and classified based on chemical nature.

Soil Biology

Soil biology investigates the microbial and faunal activity, as well as the ecology, within soil (Nannipieri and Eldor, 2009; Coleman et al., 2004; Lavelle et al., 2004) [62, 23, 48]. The term "soil biota" encompasses organisms spending a significant portion of their life cycle within soil or at its interface with litter (Bardgett and Putten, 2014; Bardgett, 2005; Avis, *et al.*, 2008) [7, 8, 4]. These organisms range from earthworms, nematodes, and protozoa to fungi, bacteria, and various arthropods, along with some reptiles (like snakes) and burrowing mammals such as gophers, moles, and prairie dogs. Soil biology profoundly influences numerous soil characteristics (Pankhurst et al., 1997; Baker et al., 2006; Bardgett, et al., 2005) [64, 6, 7]. The decomposition of organic matter by soil organisms significantly impacts soil fertility, plant growth, soil structure, and carbon storage (Wardle et al., 2004; Birkhofer, et al., 2008) [80, 15]. Soil hosts a substantial proportion of the world's biodiversity (Wall et al., 2012; Bronick and Lal, 2005) [79, 18]. The intricate connections between soil organisms and soil functions are pivotal (Wall *et al.*, 2012; Fierer and Jackson, 2006) [79, 33]. Given the complexity of the soil "food web," any assessment of soil function must consider interactions with the living communities therein (Bardgett and Putten, 2014; Gyaneshwar et al., 2002) [8, 35]. Soil organisms decompose organic matter, releasing nutrients for plant and organism uptake (Nannipieri and Eldor, 2009; Hardarson and Atkins, 2003) [62, 36]. Nutrients stored in soil organisms' bodies help prevent nutrient loss via leaching (Vries and Bardgett, 2016) [78]. Microbial exudates contribute to soil structure maintenance, while earthworms are essential

bioturbation (Rillig and Mummey, 2006) ^[69]. In balanced soil, plants thrive in an active, stable environment (White, 2013) ^[81]. While soil mineral content and structure are vital for plant well-being, it's the life within soil that drives its cycles and fertility (Montgomery, 2007) ^[61]. Without soil organisms' activities, organic materials would accumulate, littering the soil surface, and plants would lack essential nutrients (Brady and Weil, 2008) ^[17]. The soil biota classification:

- **Megafauna:** size range-20 mm upward, e.g. moles, rabbits, and rodents.
- Macrofauna: size range 2 to 20 mm, e.g. woodlice, earthworms, beetles, centipedes, slugs, sn ails, ants, and harvestmen.
- **Mesofauna:** size range 100 micrometres to 2 mm, e.g. tardigrades, mites and springtails.
- Microfauna and Microflora: size range 1 to 100 micrometres, e.g. yeasts, bacteria (commonly actinobacteria), fungi, protozoa, roundworms, and rotifers.

Within these soil organisms, bacteria and fungi play pivotal roles in maintaining soil health (Sylvia *et al.*, 2015) ^[75]. They function as decomposers, breaking down organic materials into detritus and other breakdown products (Sylvia *et al.*, 2015; Jackson, *et al.*, 2007) ^[75, 42]. Soil detritivores such as earthworms consume detritus and aid in its decomposition (Edwards and Bohlen, 1996) ^[29]. Ants (macrofauna) contribute similarly to decomposition while providing mechanical agitation through their movements (Lavelle *et al.*, 2016) ^[49]. Additionally, rodents and woodeaters enhance soil absorbency (Lavelle *et al.*, 2016; Johnson *et al.*, 2007) ^[49].

Functions of soil biology

Microorganisms play a vital role in decomposing organic matter through enzymatic breakdown of polymers into monomers (Nannipieri and Eldor, 2009; Jones, et al., 2004) [62, 44]. This turnover of organic matter releases nutrients essential for crop growth, while the balance between turnover and stabilization determines the loss of carbon from the system, primarily as dissolved organic matter or gaseous molecules such as CO2 and CH4 (Lehmann and Kleber, 2015; Kibblewhite et al., 2008) [51]. Consequently, organic matter turnover significantly influences climate regulation (Lehmann and Kleber, 2015; Paul, 2007) [51, 66]. They also play role in nitrogen fixation, mineralization, nitrification and denitrification (Singh et al., 2011) [72]. example: Nitrifying bacteria such as Nitrosomonas, Nitrobacter, Nitrococcus etc. Denitrifying bacteria such as Achromobacter, Pseudomonas etc. They also help in phosphorus solubilization (Richardson, 2001) [68] example: Bacillus, Pseudomonas and phosphorus mobilization example: VAM. Some soil organisms cause plant disease and some suppress disease (Mendes et al., 2013) [56].

Effect of herbicides on soil bacteria

Mohanty *et al.* (2004) ^[59] reported that population of methanogenic bacteria was significantly inhibited at higher concentrations of 50 and 100 mg g⁻¹ soil amendment of butachlor. This is mainly due to application of butachlor inhibits methane monooxygenase enzyme which inhibits the conversion of methane to methanol thereby affecting the metabolism of methanotrophic bacteria. Latha and Gopal

(2010) [47] found that population of bacteria was found to be significantly influenced by the type of herbicides, concentrations and the days after application of herbicides. Among the herbicides, butachlor application significantly reduced the population of total heterotrophic bacteria (18.25 CFU g-1 soil) compared to the herbicides pyrazosulfuron ethyl (33.46 CFU g⁻¹ soil) and pretilachlor (32.45 CFU g⁻¹ soil). The bacterial population at 7 and 30 days after herbicide application was significantly higher than the population at 15 days after herbicide application. The bacterial population with herbicides applied at 1 times field rate (35.92 CFU g⁻¹ soil) and 2 times field rate (33.71 CFU g⁻¹ soil) was significantly higher compared to 5 (31.66 CFU g⁻¹ soil), 10 (24.01 CFU g⁻¹ soil) and 100 (19.00 CFU g⁻¹ soil) times of the recommended rates of 2,4-DEE application. This is due to application of butachlor can penetrate the cell easily and disturb the bacterial metabolism and often cause death of bacteria. Baxter and Cummings (2008) compared microcosms containing 10 and 50 mg kg⁻¹ bromoxynil showed that the abundance of the four microbial taxa was significantly different at days 7 and 35. This is due to bromoxynil inhibit photosynthesis by binding to D₁ protein of photosystem II in thylakoid membrane. It blocks electron transport and CO₂ fixation as a result blocking of electron transport in photosystem II promotes formation of highly reactive oxygen species causing lipid and protein membrane destruction that results in membrane leakage allowing cells and cell organelles to dry and rapidly disintegrate. Mariusz and Zofi (2007) [53] concluded that response of nitrifying bacteria to herbicide is strongly correlated with the dosages of the linuron used. The highest rates of herbicide negatively affected the viable counts of nitrifying bacteria during whole experimental period. In turn, denitrifying bacteria were less sensitive than nitrifying bacteria.

Effect of herbicides on soil fungi

Dubey et al. (2018) [26] observed significant effects of various herbicide treatments on fungal counts 90 days after crop sowing. Notably, weed-free and weedy check treatments exhibited lower fungal counts compared to herbicidal treatments, indicating a more favourable environment for microorganisms in the latter. The herbicidal treatments fostered higher fungal populations, likely due to improved soil conditions and increased root exudation, providing a carbon source for microbial growth. Enhanced soil biological properties in well-aerated aerobic soil conditions of direct-seeded rice suggest improvements in nutrient status and physical soil conditions, facilitating microbial growth. It can be inferred that microbial populations began to recover after herbicide application eradicated weeds, enriching the soil with nutrients. Furthermore, the degradation of herbicides may serve as a carbon source for microbial growth. Contradictory to above findings, Rathod (2020) [67] observed that weed free (two hand weeding) treatment recorded significantly higher fungal count at 7, 15, 21, 30 and 45 DAS (9.59, 10.02, 10.13, 9.93 and 9.70×10^4 cfu g⁻¹ soil, respectively) as compared to initial value (8.6 x 10⁴ cfu g⁻¹ soil). Significantly lower value of fungal count (5.81 x 10⁴ cfu g⁻¹ soil) was recorded in the treatment of pendimethalin @ 1000 g ha⁻¹ (PE) followed by 2,4 D ethyl ester @ 1000 g ha⁻¹ (PoE) at 45 DAS. The soil fungal count was considerably decreased with advanced period of field experimentation due to application of pre and post emergence herbicides. This reduction due to applied herbicides affect fungi physiologically by changing their biosynthetic mechanism, supress cell division as a consequence of disturbing nucleic acid metabolism and protein synthesis finally causes death of the fungi. Vivek (2010) [67] revealed that there was an increase in fungal population from 0 to 50 DAS in hand weeded and weedy check plots. Recommended herbicide application inhibited the fungal growth from 0 to 50 DAS. This is due to application of fenoxaprop-p-ethyl as post emergent inhibit synthesis of acetyl CoA carboxyl (ACC) enzyme required for biosynthesis of fatty acid by carboxylation of acetyl CoA to produce malonyl CoA. In which inhibition of ACC results in fatty acid depletion leading to rapid cell death due to membrane dysfunction.

Effect of herbicides on soil actinomycetes

According to Dubey et al. (2018) [26] counts of actinomycetes were significantly affected by different herbicides treatments at 90 days after sowing of the crop. Among different herbicides treatments, there were significantly lower counts of actinomycetes were found in the weed free and weedy check. Significantly higher microbial populations in the herbicidal treatments are due to microorganisms are able to degrade herbicides and utilize them as a source of biogenic elements for their own physiological processes. According to Santric et al. nicosulfuron showed significant effects on actinomycetes growth and development in soil. For the two highest concentrations tested (10X and 50X the recommended field application rate of 0.3 mg kg-1 soil) a decrease in actinomycetes counts was observed after the application of nicosulfuron. The number of these microorganisms then decreased, and the reduction was significant (between 13 and 31% compared to the control) from the 3rd to 30th day after treatment. This reduction was greater for the higher herbicide concentrations. This is due to as nicosulfuron belongs to sulfonyl urea group inhibit acetohydroxyacid synthase also known as acetolactate synthase. This enzyme catalyses the formation of 2-acetolactate and 2-aceto-2hydroxybutyrate as first step in biosynthesis of branched chain amino acids isoleucine, leucine and valine. Actinomycetes die in response to inhibition of branched chain amino acid synthesis. The same authors also worked on the effect of glyphosate on the abundance of actinomycetes (10⁴ g⁻¹dry soil). The effect of glyphosate (10X and 50X recommended field application rate of 32.6 mg kg⁻¹ soil) showed a decreasing trend from the 3rd to 30th day after treatment and their number was reduced between 19 and 45%, compared to the control. The reduction in actinomycetes count is due to as glyphosate mainly inhibits synthesis of EPSP enzyme i.e., 5-enolpyruvyl shikimate-3phosphate synthase is a key enzyme of shikimate pathway required for synthesis of three essential amino acids i.e., phenyl alanine, tyrosine and tryptophan.

Effect of herbicides on soil ciliate

According to Bonnet *et al.* (2007) [16] found that there is relatively higher population growth of ciliate *Tetrahymena pyriformis* in control as compared to alachlor treated plots at different concentrations. This is due to alachlor inhibits cell division which causes blockage of DNA replication followed by a cell cycle arrest. As a result, population growth decreases.

Effect of herbicides on soil earthworm

Mohapatra (2014) [60] reported that the maximum percent loss of biomass of Eudrilus eugineae over the incubation period of 20 days was observed in high (0.6 ml 100 ml⁻¹ of distilled water) herbicide dose and the minimum in low (0.2 ml 100 ml⁻¹ of distilled water) herbicide dose of dalapon. Eudrilus eugineae treated with herbicide dalapon at different concentrations indicate that with increase in concentration of herbicide in soil there is considerable damage to muscle fibres which might affect the ecological function of each species of earthworm. According to Hemlata (2015) [40] observed effect of herbicides on the survival rate of earthworms and it was found number of earthworms decreased as the concentration increased. The percent decrease in population of earthworms in pinoxaden @ 3.0 mg kg⁻¹ soil was 44.59% whereas in sulfuron @ 3.0 mg kg-1 soil it was 40.26%. This is due to toxicity of herbicides it leads to cessation of their biological activity which resulted in their mortality. High tolerance of earthworms to chemicals is due to detoxification by metallothionine protein present in the posterior part of their alimentary canal. The survival rate of an organism depends upon the exposure of the organism to the stress, its duration and dose. According to Xiao et al. (2005) [82] concluded that in control earthworms had the highest mean cocoon production of 2.52 cocoons per worm after eight weeks of incubation. At the concentrations of 5, 10, 20 and 80 mg kg ¹ of acetochlor. This is due to earthworms exposed to long term effects of acetochlor results in increased mortality and occurrence of defects in reproductive cells that affect reproductive potential. Usually, cocoons are produced as a secretion of slime tube (pre capsule) from clitellum. As earthworm is a hermaphrodite, so sperm, eggs and symbionts are deposited in pre capsule. As direct inhibition of acetochlor on pre capsule cause defects in reproductive cells.

Effect of herbicides on soil termite

Ejomah *et al.* (2020) ^[30] found significant variations in the running speed of termites, with those treated with water (control treatment) exhibiting the highest speed (3.56 cm s⁻¹) compared to termites treated with different concentrations (1.563, 3.125, and 6.25 ml) of 2,4-D. The decreased mobility observed in worker termites may result from direct intoxication, leading to a knock-down effect, trembling, and rotating. This reduced mobility in worker termites could potentially hinder their ability to transport food to the colony and carry out various ecological functions. Additionally, decreased mobility in worker termites may render them more susceptible to predation and adverse environmental conditions.

Effect of herbicides on soil nematode

According to Ibrahim *et al.* (2020) ^[41] reported that decrease in population of both bacterivore and total number of nematodes after treatment of Tribenuron-methyl herbicide compared to before treatment. This is due to Tribenuron-methyl belongs to sulfonyl group inhibit synthesis of branched chain amino acids such as leucine, valine and isoleucine which is required for protein synthesis.

Effect of herbicides on soil biological functions

Mohanty et al. (2004) [59] observed that the content of readily mineralizable carbon (RMC) was significantly

elevated in soils treated with butachlor compared to the control, with the highest levels recorded at higher concentrations of butachlor amendment. RMC serves as an indicator of a soil's methanogenic potential, with its levels increasing during soil incubation but declining in later stages. The higher RMC content in butachlor-amended soils indicates that the herbicide application did not impede the decomposition of organic matter or the accumulation of RMC. According to Medo et al. (2021) [54] they concluded that microbial biomass carbon was affected by length of incubation as the values gradually declined to the top in the 3rd day toward the minimum in the 112th day. The effect of treatments was significant only when high doses of dimethachlor were applied. This is due to microorganisms use herbicide molecules as source of organic carbon, nitrogen or phosphorus. Hardeep (2014) [37] observed that right from the zero day of incubation, application of herbicide (clodinafop propargyl) decreased the NH⁴⁺ -N₂ content in soil even in the presence of nitrogen upto 56 days of incubation. On the 56th day of incubation, application of herbicide in the presence nitrogen @ 100 and 200 mg kg-1 decreased the NH⁴⁺ -N₂ content in soil from 34.45 to 25.62 mg kg⁻¹ and from 42.96 to 37.65 mg kg⁻¹, respectively over nitrogen alone. The use of herbicide suppressed the process of ammonification. This is due to mode of action of clodinafop propargyl is inhibition of acetyl-CoAcarboxylase enzyme which results in depletion of fatty acids leading to death of the cells of nitrogen fixing bacteria as a result reduces ammonia content in the soil. Min et al. (2001) found that the temporary inhibition of denitrification by butachlor occurred immediately within the first 3 days postapplication. This inhibition was succeeded by stimulation during the period from the 4th to the 16th day, followed by a secondary inhibition after the 16th day. The study also revealed that higher concentrations of butachlor resulted in more pronounced inhibition of denitrification during both the initial and secondary inhibition periods. These effects stem from the impact of butachlor application on diverse microorganism populations, enzymatic activities, and microbial transformation processes in paddy rice soil. The concentration of butachlor applied emerges as a crucial influencing populations of various factor the microorganisms and enzyme activities in paddy soil, independent of butachlor's inherent characteristics. Moreover, anaerobic microorganisms such as sulphatereducing bacteria have been shown to efficiently degrade butachlor in paddy soil. Additionally, the soil's nature and the application method also influence the behavior of butachlor in paddy rice soil. Das et al. (2003) [25] found that the oxyfluorfen treatment exhibited a higher concentration of available phosphorus compared to the oxadiazon treatment. This difference can be attributed to increased solubilization of insoluble phosphates by phosphatesolubilizing microorganisms, as well as a higher content of organic acids present in the root exudates of the growing plants, leading to greater release of available phosphorus in the rhizosphere soil of rice. According to Das and Dev (2014) [24] observed that as compared to control soil, maximum stimulation of total P was recorded in soil when fenoxaprop was applied along with pendimethalin (15.4%), followed by paraquat with fenoxaprop (14.3%) pendimethalin (14.1%). The enhanced microbial biomass accentuated more amount of P due to greater utilization of herbicides and the degraded products in soil. The greater

solubilization of insoluble phosphates by the increased microbial biomass due to application of herbicides manifested greater accumulation of available P in soil. Incidentally, the availability of soluble P was positively correlated with microbial biomass P indicating that the greater activities of the phosphate-solubilizing microorganisms released greater amount of available P in soil.

Effect of herbicides on soil enzymes

According to Baboo et al. (2013) [5], the activity of amylase in soil treated with butachlor, pyrazosulfuron, and glyphosate displayed an increasing trend from the 7th to the 21st day, followed by a decrease on the 28th day after treatment. This pattern was statistically significant across different herbicides and days post-treatment, attributed to changes induced by the applied herbicides in starchdegrading enzymes. Invertase activity in butachlor-treated soil increased steadily from the 7th to the 28th day, while paraquat-treated soil exhibited a consistent increase until the 14th day and then gradually declined by the 21st day, mirroring the trend observed in glyphosate-treated soil. The variation in soil invertase activity was linked to the herbicides causing the death of microorganisms responsible for producing and secreting invertase enzyme. Urease activity in butachlor, pyrazosulfuron, paraquat, and glyphosate-treated soil increased from the 7th to the 28th day, albeit lower compared to other enzyme activities. Dehydrogenase activity showed an increasing trend across all herbicide-treated soils from the 7th to the 28th day, with glyphosate-treated soil exhibiting higher activity on the 28th day. The variation in soil dehydrogenase activity was attributed to an increase in microbial community composition capable of utilizing herbicides as a carbon source.

Conclusion

In conclusion, the impact of herbicides on soil biology and its functions is significant and multifaceted. While these chemicals are effective in managing weed populations and enhancing agricultural productivity, their indiscriminate use can lead to adverse effects on soil microbial communities, nutrient cycling, and overall soil health. The disturbance of microbial populations can disrupt vital processes such as decomposition, nitrogen fixation, and organic matter turnover, ultimately compromising soil fertility and ecosystem resilience. Moreover, the long-term implications of herbicide residues on soil organisms and their interactions remain a subject of concern, highlighting the need for sustainable agricultural practices and alternative weed management strategies. Integrating approaches such as crop rotation, cover cropping, and reduced tillage can mitigate the negative impacts of herbicides while promoting soil biodiversity and ecosystem stability. In light of the intricate relationship between herbicides, soil biology, and ecosystem functions, continued research and monitoring efforts are crucial for understanding the full extent of their effects and developing informed management practices that prioritize agricultural productivity and environmental sustainability. By adopting holistic approaches that consider the complex dynamics of soil ecosystems, we can strive towards a balance where weed control coexists harmoniously with soil health and biodiversity conservation.

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