The impact of glyphosate herbicide doses with different time interval and soil type On some soil physiochemical properties

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Abstract

Glyphosate herbicide widely used in agricultural and nonagricultural places in Iraq/Kurdistan governorate. This herbicide has a great environmental impacts on human health and soil physiochemical properties. The objectives of this study is to evaluate the effects of glyphosate herbicides in different doses on some important soil physio-chemical properties. The soil type (soil texture) significantly (p≤ 0.005) influenced the physio-chemical properties. The application of different glyphosate doses significantly influenced the soil's physio-chemical properties after eight weeks. The p**hosphorus (P) availability** in the **overdose**, **recommended dose**, and **under dose** treatments significantly increased soil P levels (29.04–32.84 ppm) compared to the **control** (5.75 ppm). **Ammonium (NH_4^+) and nitrate (NO_3^-), NH_4^+** peaked at w**eek 2 (203.9 ppm)** before decreasing at w**eeks 4 and 8 (147.5–159.7 ppm)**, possibly due to initial glyphosate-induced inhibition of nitrifires followed by recovery. **OM% declined over time** in all treatments, with the sharpest decrease in **overdose (loam: 1.05% to 0.358%)** and **recommended dose (clay: 1.698% to 1.113%),**the decline in OM suggests potential negative effects on soil carbon cycling, possibly due to shifts in microbial community structure. All glyphosate treatments resulted in a slight decrease in pH but significant (7.682 –7.22), possibly due to organic acid release during glyphosate degradation. **(EC)** indicate that the **overdose** treatment had the highest EC (1291 μS), followed by the **recommended dose** (1142 μS) and **under dose** (1053 μS), compared to the **control** (439 μS).In c**onclusion**, the results demonstrate that **soil type and glyphosate dosage significantly influence soil physicochemical properties.** Glyphosate application, particularly at **overdose levels**, alters soil chemistry by **increasing P, NO_3^- , and EC while reducing OM%**.

Keywords: glyphosate, Herbicide, Soil Pollution, Overdose, Roundup, Soil type

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Introduction

Glyphosate, also known as *N-(phosphonomethyl*) glycine, is a widely used broad-spectrum organophosphorus herbicide. **[8,24]**. It is a common herbicide used in home gardens, urban areas, and agricultural to combat annual and perennial weeds. **[37]**. Its application has significantly expanded in recent years as a result of genetically modified crops and weed control in fallows. **[1]**. Glyphosate is highly adsorbed to soil particles as it enters the soil. **[25]**One of the key elements influencing the herbicide's behavior and fate seems to be its adsorption to clay particles. **[6].** It is a necessary part of post-emergent and non-selective herbicides that fight woody plants, grasses, and annual broadleaved weeds. **[5].** The phosphonic hydrogen in this molecule tends to detach and join the amine group, making it a zwitterion. Glyphosate inhibits EPSPS (5-enolpyruvylshikimate-3-phosphate synthase), which delays the production of necessary secondary metabolites and proteins and inhibits the critical energy pathways in plants and soil microorganisms. **[7]**. According to a study, glyphosate changes the microbial diversity and soil texture by increasing the number of phytopathogenic fungi and decreasing microbial richness. **[11].** Because of its possible interaction with soil nutrients and impacts on microorganisms directly involved in residue decomposition and nutrient cycling, long-term glyphosate use may have both direct and indirect effects on soil chemistry**[29]**. Herbicide management had an impact on the concentrations of accessible P, Fe, K, NO3-N, and SO4-S, but it had no discernible influence on the pH, SOM, exchangeable Ca, Mg, Mn, or Zn of the soil. Herbicide management had an impact on the concentration of exchangeable K, which might be obscured by crop removal. However, the natural higher K levels and crop removal outweigh the quantity of K given to the soil as a result of glyphosate treatment. Herbicide management may have an indirect impact on these nutrients in the soil, even if NO3-N buildup is a temporary occurrence.

[31]. The chemistry of the soil may be impacted by the additives used with herbicides [35]. It has been clearly known that the amount of absorbed glyphosate strongly depends on pH. There is an agreement that glyphosate adsorption reduces with rising pH **[27].** On the other hand, reports regarding the impact of an acidic pH are not entirely consistent [38]. The most significant variables influencing glyphosate adsorption on so are pH, clay content, and iron oxides, according to numerous studies [6]. A common component of many fertilizer blends, phosphorous is an essential nutrient that supports plant growth. However, if too much phosphorous finds its way into the area's watershed, it can disrupt wetlands, streams, and lakes, resulting in toxic algae blooms (such as the extreme cyanobacteria algae blooms in Lake Erie last year) and dissolved oxygen depletion, which can Occasionally cause fish and other aquatic life to die off in large numbers. **[13]**. Furthermore, like phosphorus (P) sorption, glyphosate sorption to the soil occurs via legend exchange through the phosphonate group. For the same sorption sites in the soil exchange complex, glyphosate thus faces competition from P. **[9].** Long-term glyphosate use may have an impact on P cycling in soils due to this glyphosate characteristic. Glyphosate that has already been sorbed may desorb and remobilize as a result of elevated P levels in the soil [10].Glyphosate sorption in soil is influenced by a number of factors, such as clay concentration, SOM, and Fe/Al

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oxides. Glyphosate sorption is also expected to be influenced by factors including pH, which affects glyphosate speciation, and inorganic P, which competes for sorption sites in Fe/Al-oxides. [4]. Thus, the current study's goals are to assess how glyphosate herbicides at varying dosages affect key soil chemical characteristics such as PH, EC, OM content, available N, and available P following glyphosate application at various intervals.

Material and Methods

Study Area: experimental location description

In the September of 2024, the glyphosate application experiment was performed at the college of agricultural engineering science in the University of Duhok, Kurdistan region, Iraq. The college of agriculture in duhok has a latitude of approximately 36°51'32.39"N and a longitude of approximately 42°52'58.95"E, with an altitude of around 473 meters above sea level.(38)pots were prepared under the green house in the college of agriculture engineering science, under the same environmental condition.as showed in the bellow figure.

Sampling collection and pots experiment

In this study, two different types of soils with different physical and chemical characteristics used, Clay soil collected from the A-horizon of cultivated soil in the college of agricultural engineering science by the way of randomize methods to represent all of the field, with a reported history of GLP non application .the second soil type was loamy soil, the commercial soil that used in the gardens and nurseries .The characteristics of the soils are given in Table 1. Both soils were collected and sieved by (2mm) sieve, after sieving the soil, the particle size were analyzed to estimate the soil texture, Applying glyphosate to them and to compare between them. the soils puts into plastic planting pots with a surface diameter of (25) cm, a height of (30) cm and 2 kg soil for each pots, the bottom drainage holes of the pots were opened to drain excessive water and prevent water logging. Two plant types cultivated in pots, Cynodon dactylon, commonly known as a (Bermuda grass) is a perennial grass species widely recognized for its durability and adaptability.Were harvested in the college gardens.and the second plant type was avena sativa L, Anatolia type that is native to or cultivated in the Anatolian region, primarily Turkey.

Weed control

Weed control type used for this study were commercial herbicides (Round up) PILARSATO A **soluble liquid (SL) herbicide** containing **approximately 480 g/L glyphosate** (48% SL) acid equivalent — a non-selective, systemic compound used worldwide for broad leaf and grass weed control. the herbicide were applied on the plants as overdose 30 ml /L ,Recommended dose 15ml /L and under dose 7.5 ml /L.

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Determination of soil physiochemical properties

Soil PH was analyzed in a 1:5(v/v) ratio, the extracts were measured by pH meter and glass electrode according to verma and kalamdhad (2014).Soil EC was analyzed in a 1:5(v/v),the soil EC of the extracts were measured by EC meter and glass electrode according to **[33]**The soil organic matter was estimated by the reduction of potassium dichromate (K2Cr2O7) by OC compounds and subsequent determination of the unreduced dichromate by oxidation-reduction titration with ferrous ammonium sulfate. This method is referred to as the Walkley-Black method **[34].** The Kjeldahl methods were used to determine the levels of available NO3-N and NH4-N. after the air dried soil samples were extracted with 2M KCL (1:5),(W/V) when 25 ml of 2M KCL were added to 5g of air dried soil, then subjected to steam distillation and titration methods **[26]**The available phosphorous was estimated by The sodium bicarbonate ($NaHCO_3$) procedure of Olsen et al(1954).

Statistical Analysis.

Data analysis was done by using the Analysis of Variance method (ANOVA) with the General Linear Methods (GLM) procedure to compare between various glyphosate herbicide doses significant effects ($P < 0.005$) in clayey and loamy textured soils on some soil physio-chemical properties over 4 time intervals , using the Minitab software package 19. The Tuckey test was used to determine whether there were significant differences between treatment means.

Results and Discussion

3.1. Results

The application of different glyphosate doses significantly($p \leq 0.001$) influenced the soil's physio-chemical properties after eight weeks (Table 2).The effects of glyphosate doses on some soil physio-chemical properties as shown in the table 2, the p**hosphorus (P) availability** in the **overdose**, **recommended dose**, and **under dose** treatments significantly increased soil P levels (29.04–32.84 ppm) compared to the **control** (5.75 ppm), indicating that glyphosate application enhances P availability, possibly due to glyphosate's ability to chelate metal ions, releasing bound P**[2]**.also because phosphorus and glyphosate share the adsorption mechanisms and sites in soils. If glyphosate outcompetes phosphorus for sorption sites in soils (glyphosate-based-herbicides) (GBH) usage increase the amount of phosphorus available to plants **[12]**. Our findings are consistent with those reported by Grenier et al. (2022), who observed that,A significant increase in PO4 3– content was recorded for the control between T28 and T112, with values going from 605.8 to 1,027.5 μgPg–1. The increase was significant between T0 and T112 for the GBH treatment, with PO4 3– going from 652.3 to 990.5 μgPg–1. **[32]**.was done his research on Dissipation and effect of glyphosate during composting of organic wastes. **Regarding organic matter (OM %),** the **control** had the highest OM (2.17%), while all glyphosate-treated soils showed significantly lower OM (1.23–1.30%). This suggests that glyphosate may accelerate microbial decomposition of organic matter or inhibit microbial activity involved in OM stabilization **[19]**. Similar to that (Liu, H., 2022) said, "Spraying

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glyphosate had an adverse effect to decrease organic carbon, probably leading to reducing soil nutrition, fertility and contributing to the increase of CO_2 in the atmosphere". A**mmonium (NH_4^+) and nitrate (NO_3^-)** shows that **control** had the highest NH_4^+ (206.2 ppm), whereas glyphosate treatments reduced NH_4^+ levels (117.9–132.4 ppm), possibly due to altered microbial nitrification processes **[39]**. Similar to the results of Grenier et al. (2022), "we found that The NH3/NH4 + content did not differ between treatments but decreased significantly over the course of the experiment". Because no leachate was collected during the experiment, this result suggests that ammonium was primarily used by the microorganisms as a source of N and was oxidized in the nitrification process, although loss through volatilization of NH3 is also possible. [32].Conversely, NO_3^- was highest in the **overdose** treatment (644.4 ppm) compared to the **control** (171.6 ppm), suggesting that glyphosate may stimulate nitrification or inhibit DE-nitrification process **[22].**similar to Grenier et al. (2022) results who found that, A significant increase in NO2 —NO3 – content was measured between T7 and T28 for the control and GBH treatments, from 30.1 to 147.2 µgNg–1 and from 5.52 to 136.4 µgNg–1, respectively .The increase in content between T28 and T112 was significant for all three treatments, with values increasing from 147.2 to 1,007.1 µgNg–1 for the control, from 118. to 877.1 µg N g–1 for AG glyphosate, and from 136.4 to 783.4 µg N g–1 for GBH. [32].**While the electrical conductivity (EC)** indicate that the **overdose** treatment had the ($p \leq 0.005$) highest EC (1291 µS), followed by the **recommended dose** (1142 µS) and **under dose** (1053 µS), compared to the **control** (439 µS) which is the lowest value observed. This increase in EC may be due to the release of ions from glyphosate degradation or altered soil microbial activity **[30].**Because it introduces and promotes the **accumulation of soluble ions** in the soil. Table 1 declare that the s**oil pH** in the **control** had the highest pH (7.75), while all glyphosate treatments resulted in a slight decrease but significant ($p \leq 0.005$) (7.51–7.62), and in the first week was (6.883) possibly due to organic acid release during glyphosate degradation. **[23]**. There is fully agreement between our findings and those of **[20]**, particularly regarding that the soil pH in all groups was basically neutral to weakly acidic with values from 6.67 to 7.90. Overall, the effect of glyphosate application on soil pH exhibited less significant in soil with transgenic glyphosate-resistant soybeans cultivation history than that of recipient soybeans. At the initial stage of glyphosate spraying for 7 days, soils were more acidic than control. The possible reason for this phenomenon is that a decrease of pH could urge the sorption of glyphosate to minerals and benefit glyphosate degradation **[20,27]**.his research was on (Effects Of Glyphosate Application on Soil Ecological Health ..) but it was neutralized and slightly increased after some days, this is due to soil PH buffering capacity .That property resist change in PH.

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Table 1:-characteristics of both soils that were sprayed with glyphosate soluble powder.

Soil type	pH	EC (Us)	NO_3 (ppm)	NH_4 (ppm)	OM%	Soil Texture			P(ppm)
						Sand%	Silt%	Clay%	
Clay soil	7.700	536	329.2	118.2	2.600	5.00	32.75	62.25	7.800
Loamy soil	7.800	342	14.01	294.2	1.740	56.60	32.95	10.45	3.700

Table 2-: The effects of glyphosate doses on some soil physio-chemical properties.

Doses	P(ppm)	OM %	NH_4(ppm)	NO_3(ppm)	EC (Us)	PH
Control	5.75^b	2.170^a	206.2^a	171.6^c	439^c	7.750^a
Overdose	32.84^a	1.228^b	132.4^b	644.4^a	1291^a	7.509^c
Recom. dose	29.56^a	1.295^b	122.4^b	504.8^b	1142^b	7.506^c
Under dose	29.04^a	1.249^b	117.9^b	399.3^b	1053^b	7.618^b
p-value	0.000	0.000	0.000	0.000	0.000	0.000

The values that do not share a letter are significantly different. According to Duncan multiple ranges test at significant level of 5%.

The soil type (soil texture) significantly influenced the physio-chemical properties, as shown in the table 3. Regarding p**hosphorus (P) availability**, the l**oam** soil had significantly higher P (25.62 ppm) compared to **clay** (22.97 ppm) (p≤ 0.005). This is due to better P mobility in loam soils, which have a balanced texture allowing for greater nutrient diffusion [3].The differences in P availability between **loam** and **clay** soils align with previous studies showing that medium-textured soils (loam) often exhibit better P mobility due to reduced fixation compared to fine-textured soils (clay) [3].**While, organic matter (OM %)has a fully significant difference (p-value<0.001).which in Clay** soil retained significantly higher OM (1.85%) than **loam** (1.12%), likely due to clay's higher cation exchange capacity (CEC) and ability to stabilize organic compounds[15].The higher OM in **clay** supports the well-documented role

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of clay minerals in protecting organic matter from microbial decomposition[15]. **Ammonium (NH_4^+) and Nitrate (NO_3^-) also have a fully significant difference (p-value<0.001) with soil type.**NH_4^+ was higher in **loamy soil** (173.8 ppm) than in **clay soil** (115.6 ppm), possibly due to faster mineralization rates in well-aerated loam soils [17]. **NO_3^-** was higher in **clay** (488.8 ppm) than in **loam** (371.3 ppm), suggesting that clay's reduced oxygen conditions may slow denitrification, allowing NO_3^- accumulation [16].The contrasting trends in NH_4^+ and NO_3^- between soil types suggest that **loam** promotes faster N mineralization, while **clay** may favor NO_3^- retention due to restricted microbial activity under lower oxygen conditions [16]. **Electrical conductivity (EC)**, **Clay** had significantly higher EC (1116 µS) than **loam** (846 µS) **(p-value<0.001).**likely due to its greater surface area and ion retention capacity **[3]**.The higher EC in **clay** is consistent with its greater capacity to retain soluble ions **[3]**. S**oil pH** shows no significant difference between **clay soil** (7.61) and **loamy soil** (7.58) **(p-value=0.185), so the (p-value>0.005).** Indicating that soil texture had minimal effect on pH under these conditions. And it's driven by chemical properties like (base saturation, organic matter, and inputs), not the soil texture. These findings highlight the importance of considering soil type in nutrient management strategies, as texture significantly influences nutrient availability and microbial processes.

Table 3:- The effects of soil type on some physiochemical properties.

Soil type	P(ppm)	OM %	NH_4(ppm)	NO_3(ppm)	EC (Us)	PH
Clay	22.97 [b]	1.847 [a]	115.6 [b]	488.8[a]	1116.3 [a]	7.612 [a]
Loam	25.62[a]	1.123[b]	173.8[a]	371.3[b]	846.2[b]	7.579[a]
p-value	0.014	0.000	0.000	0.000	0.000	0.185

The values that do not share a letter are significantly different. According to Duncan multiple ranges test at significant level of 5%.

The duration of glyphosate exposure (1, 2, 4, and 8 weeks) significantly influenced soil physiochemical properties (Table 3).The effect on **phosphorus (P) availability,** P levels were lowest at w**eek 1 (8.36 ppm)** but sharply increased (p≤ 0.5)by **Week 2 (29.59 ppm)** and remained high (28.92–30.32 ppm) through w**eeks 4 and 8**.This suggests that glyphosate degradation or microbial activity releases P over time, consistent with findings that phosphonate herbicides enhance P solubility (Gimsing et al., 2004). The influence of time in o**rganic matter (OM %),**OM declined progressively from w**eek 1 (1.76%)** to w**eek 8 (1.21%)**, indicating accelerated decomposition or reduced microbial biomass due to prolonged glyphosate exposure[19]. **Ammonium**

(NH_4^+) and nitrate (NO_3^-),NH_4^+ peaked at w**eek 2 (203.9 ppm)** before decreasing at w**eeks 4 and 8 (147.5–159.7 ppm)**, possibly due to initial glyphosate-induced inhibition of nitrifiers followed by recovery [28].**NO_3^-** increased steadily, doubling from w**eek 1 (197.0 ppm)** to w**eeks 4 and 8 (~590 ppm)**, suggesting delayed nitrification stimulation [19].**Electrical conductivity** EC rose sharply by w**eek 4(1289 µS)**, likely from glyphosate breakdown products [30].then declined slightly by w**eek 8 (947 µS)**, possibly due to leaching or microbial

Assimilation. The pH dipped at week 2 (7.53) and week 8 (7.50), potentially from organic acid release during glyphosate degradation [7**].**but rebounded at week 4 (7.67), possibly due to buffering from clay or carbonate minerals.

The temporal trends reveal that glyphosate's impact on soil is dynamic, and can be categorized in three stages .**Early stage (1–2 weeks)**, Rapid P release and NH_4^+ accumulation suggest initial glyphosate breakdown and temporary suppression of nitrification.**Mid stage (4 weeks),** peak EC and NO_3^- levels indicate advanced degradation and nitrification recovery. **Late stage (8 weeks).**These findings align with studies showing glyphosate's time-dependent effects on microbial activity and nutrient cycling **[28,30]**. The persistent NO_3^- rise raises concerns about potential leaching risks in agro ecosystems.

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Table 4:- The effects of time on some physiochemical properties.

Time week	P(ppm)	OM %	NH_4(ppm)	NO_3(ppm)	EC (Us)	**PH**
1	8.36[b]	1.760[a]	67.8[c]	197.0[c]	787.3[c]	7.682[a]
2	29.59[a]	1.594[b]	203.9[a]	342.8[b]	901.7[b]	7.526[b]
4	28.92[a]	1.374[c]	147.5[b]	590.2[a]	1289.3[a]	7.673[a]
8	30.32[a]	1.214[d]	159.7[b]	590.1[a]	946.7[b]	7.503[b]
p-value	0.000	0.000	0.000	0.000	0.000	0.000

The values that do not share a letter are significantly different. According to Duncan multiple ranges test at significant level of 5%.

Also, the study examined the effects of different glyphosate doses (control, under dose, recommended dose, and overdose) on the physicochemical properties of **clay and loam soils** over four time intervals (1, 2, 4, and 8 weeks) as shown in table 3 & 4. **Phosphorus (P) availability in overdose treatments** led to a significant ($p \leq 0.005$) and ($p \leq 0.001$) increase in P concentration, particularly in **loam soil (up to 46.22 ppm at weeks 4 and 8)**, compared to clay soil (34.48 ppm at week 8).The **recommended dose** showed a moderate increase in P, while the **under dose** had minimal effects. **Control treatments** maintained stable P levels (3.70–7.80 ppm), indicating no glyphosate-induced P release.

OM% declined over time in all treatments, with the sharpest decrease in **overdose (loam: 1.05% to 0.358%)** and **recommended dose (clay: 1.698% to 1.113%)**. The decrease of total organic matter over time is rational as it decomposed by microbial communities exhibited in the soil or it suggests that glyphosate application may accelerate microbial decomposition of organic matter **[7]**. **The dynamic of NH_4^+ levels fluctuated**, with **higher initial values in control loam soil (294.2 ppm)**, likely due to natural mineralization. **NO_3^- accumulation was highest in overdose treatments**, particularly in **clay soil (1068.3 ppm at week 4)**, possibly due to glyphosate-induced nitrification **[21]**.The **recommended dose** resulted in moderate NO_3^- increases, while the **under dose** had variable effects.

EC spiked in overdose treatments, reaching **1910.3 µS in clay at week 4**, indicating potential salt accumulation from glyphosate degradation products.

PH remained near-neutral (7.2–7.8), but **slightly decreased in overdose loam soil (7.212 at week 2)**, possibly due to acidic glyphosate metabolites **[21]**

Discussion 4.1.

Our findings indicate that glyphosate application, even at recommended doses, significantly alters soil nutrient dynamics. The increase in P availability aligns with studies showing that glyphosate acts as a phosphonate, releasing fixed P **[14]**.Glyphosate is known to compete with P for binding sites on the soil exchange complex **[14]**, and this can affect P availability in the soil. It is plausible that the presence of glyphosate caused increased retention of P on the soil exchange sites and explains greater P concentration observed in the glyphosate only treated plots compared to other herbicide treatments. High P contents in the soil after glyphosate application, may be due to the release of P from soil minerals as the herbicide release acids to the soil addition to the high P content in its chemical composition and the limited shape and size of pots that prevent mobility of P to far distance. However, **loamy soil exhibited higher P mobility**, possibly due to lower adsorption compared to clay **[36]**.The reduction in NH_4^+ alongside increased NO_3^- suggests that glyphosate may promote nitrification, consistent with findings by **[39]**However, the decline in OM suggests potential negative effects on soil carbon cycling, possibly due to shifts in microbial community structure **[19]**The elevated EC in glyphosate-treated soils indicates increased salinity, possibly from glyphosate breakdown products **[30].**The elevated EC in glyphosate-treated soils indicates increased salinity, possibly from glyphosate breakdown products **[30].**The slight pH reduction could be due to organic acids produced during microbial degradation of glyphosate **[7].**These changes highlight the need for careful glyphosate management, as repeated applications may lead to long-term soil health degradation, affecting nutrient cycling and microbial balance.**Clay soil retained higher OM% and NO_3^-**, likely due to its greater cation exchange capacity

(CEC) and nutrient-holding capacity **[3].Overdosing glyphosate increased EC and NO_3^-**, which could lead to **soil salinization and nutrient imbalances**, as observed in previous studies **[18].**

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5. Conclusions

Depending on the results of this study revealed the following conclusion:

- In c**onclusion**, the results demonstrate that **soil type and glyphosate dosage significantly influence soil physicochemical properties.**
- Glyphosate application, particularly at **overdose levels**, alters soil chemistry by **increasing P, NO_3^-, and EC while reducing OM%**.
- Declining OM and EC suggest long-term microbial community shifts or nutrient leaching.
- **Clay soil mitigates some effects due to its buffering capacity**, whereas **loam soil is more susceptible to nutrient leaching and pH shifts**.

These findings highlight the importance of **proper glyphosate dosing to maintain soil health.**

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