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Preliminary Observation: Prolonged Glyphosate Application Effects on Soil Microbial Biomass Carbon in Coffee Cultivation Systems

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Abstract

Coffee Arabica, native to Ethiopia and accounting for 60–70% of global production, thrives on healthy soil, which supports its high-quality beans. Soil characteristics influence plant health and bean quality, while coffee cultivation helps protect soil from erosion, retain moisture, and support microbial activity. Sustainable practices improve soil health by enhancing organic matter, microorganisms, and nutrient mobility. Effective weed management boosts productivity by improving soil conditions and reducing competition, while proper control mitigates pest risks and enhances market value. From this preliminary observation, prolonged herbicide use harms soil health, disrupts microbial communities, and reduces fertility. Balanced weed management strategies are essential for sustainable coffee cultivation. Soil samples were tested for pH, with glyphosate-treated plots showing lower pH values (5.20 at 0-20 cm and 5.22 at 20-40 cm) compared to untreated plots (5.86 and 5.58). Prolonged glyphosate use reduced organic matter, causing soil acidification, decreased microbial biomass, and slower herbicide degradation, which increases persistence and complicates weed management. The data shows that herbicide use negatively affects soil microbial biomass carbon and pH, with broader implications for soil health and ecosystem sustainability. Reduced microbial biomass and pH indicate impaired microbial activity, nutrient cycling, and soil stability, all essential for agricultural productivity. These results highlight the need for sustainable agricultural practices to protect soil health. However, the studies limited scope calls for more research on herbicide impacts and strategies for maintaining soil health while managing weeds.

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Introduction

Coffee is one of the most popular beverages globally, enjoyed by millions for its rich flavor and stimulating effects due to various beneficial stimulants found it (McCook, 2024). Coffee Arabica (*Coffea Arabica*) cultivation and trade have a long history, dating back to the 15th century, with roots in the high lands of Ethiopia and highly regarded coffee species in the World. Native to Ethiopia, Coffee Arabica accounts around 60-70% of global coffee production and it is known for its high quality, smooth flavor, bright acidity, and complex aroma, making it favorite beverages among coffee enthusiasts. The interaction between coffee plants and soil is crucial for the successful cultivation of high-quality coffee (AdJR et al., 2023). Soil provides the essential nutrients, water, and support needed for coffee plants to grow and produce beans with desirable flavors.

Coffee plants, particularly the widely cultivated *Coffea arabica* and *Coffea canephora*, thrive in specific soil conditions, and the characteristics of the soil play a significant role in the health, productivity, and quality of coffee beans (Getachew et al., 2023). Coffee in its turn protects the soil from erosion, supplying it moisture for the appropriate functioning of soil and its components (soil organic matter, living microbes, gases and water) for their movement and release exudates which soil microorganisms feed from it (Nurcholis et al., 2024).

(Kobusinge et al., 2023) also found that, coffee serves to safeguard the soil against erosion, providing essential moisture that facilitates the optimal functioning of soil and its constituents (including soil organic matter, living microorganisms, minerals, gases and water), thereby enabling their mobility and the secretion of exudates that serve as nourishment for soil microbes. Furthermore, the cultivation of coffee possesses the potential to augment soil health and quality resilience through certain practices (Nurcholis et al., 2024).

Weed management exerts a substantial impact on the cultivation of coffee, affecting both its productivity and quality. The implementation of weed control measures can increase coffee yields through the enhancement of soil conditions and the mitigation of resource competition. Weeds possess the potential to serve as reservoirs for pests and pathogens; diminishing the quality of coffee. The implementation of effective control measures mitigates these threats; enhancing the market value of the coffee produced (Alcântara et al., 2015). Different weed management systems modify soil composition and moisture retention, thereby influencing the overall health of coffee plants (Junior et al., 2011). Holistic weed management methodologies, which encompasses non-chemical techniques, are crucial for the sustainable cultivation of coffee (Wortley, 2022). Bio-control agents play a significant role in promoting soil health through the regulation of nematodes populations and the augmentation of microbial community diversity (Saikai et al., 2023).

Although effective weed control can promote coffee agriculture, an overreliance on herbicides results in enduring soil health complications and the development of resistance among weed species; underscoring the necessity for a balanced strategy in weed management. Application of herbicides exerts profound effects on soil health by altering microbial communities and biochemical processes that are critical for nutrient cycling. The presence of herbicide residues implies a negative correlation with microbial biomass and enzyme activities, which are imperative for sustaining soil fertility (Liu et al., 2024; Bhardwaj et al., 2024). The prolonged presence of glyphosate in the soil system is associated with enduring ecological ramifications, which adversely impact crop productivity and overall soil ecosystem and health (Badani et al., 2023). Residual herbicide remains in the soil, resulting in persistent detrimental effects on microbial communities and soil health; the resistance of glyphosate to degradation presents significant risks to soil biodiversity (Badani et al., 2023). Studies revealed that glyphosate can increase soil compaction indirectly by affecting earthworms and soil fauna; which help in improving soil aeration, water movement and aggregation of soil. Reduced biological activity can diminish the natural processes that reduce compaction. Glyphosate indirectly affect soil organic matter content which leads to changes in the soil’s structure; which influence water infiltration rates and retention, including aggregation; due to reduced microbial activity or organic matter can result in reduced porosity, leading to lower infiltration rates and increased runoff, potentially affecting erosion.

The observation of the effect of glyphosate on soil microbial carbon (MBC) is an important area of research in environmental science and agriculture, as well as precise mechanism by which glyphosate affects biomass, including its interaction with microbial enzymes, nutrient cycling and soil matrix, remain underexplored. While glyphosate is widely used as herbicide, its long-term impact on soil health, particularly, microbial biomass is not fully understood. In addition to this hypothesis, even though the glyphosate was applied in order to control the prevalence of weed, contributing in boosting coffee production and productivity, its impact on soil microbial biomass carbon was not considered for those long years. The objective of this observation was conducted on a farm plot where glyphosate has been sprayed for long period of time starting from 2008; for the last 16m years; up to the date of soil sample collection for observation.

Materials and Methods

Study area description

The study area was situated at the Jimma Agricultural

Research Center, which is geographically positioned at 07°40'140" latitude and 036°46'943" longitude.

The site is located of 1770 meters above sea level agro-ecological zone. This region is characterized by its conducive climatic and soil condition, which are favorable for agricultural research and coffee cultivation. The center serves as a prominent hub for agricultural studies, focusing on enhancing productivity and sustainability in farming practices. Its strategic location also ensures diverse agro-ecological conditions, making it ideal for wide range of agricultural experiments and innovations.

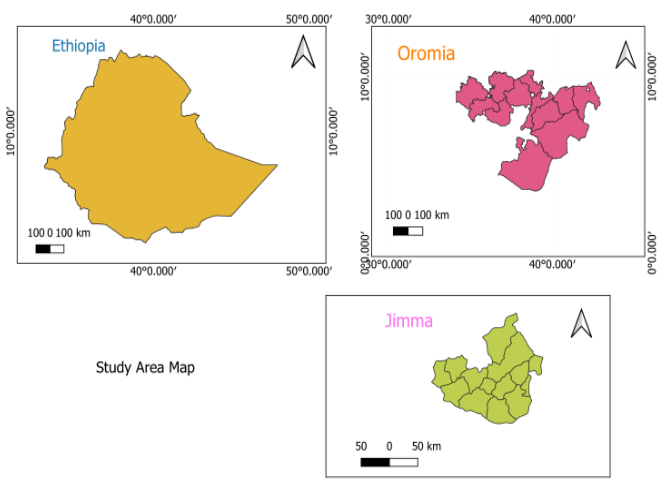

Figure.1 Map of the study area

Soil Sample Collection and Preparation

Soil samples were collected from coffee herbicide testing field farm on which glyphosate has been applied for long period of time and adjacent coffee farm where glyphosate was not sprayed before. The plot size of the area on which the research has been conducted was 42 m * 20 m; where the coffee was planted 2m*2m. Soil samples were collected randomly at two depths; 0cm – 20cm and 20cm – 40cm. Twenty four soil samples were sampled in order to quantify the long term effect of glyphosate application on microbial biomass carbon across different depths.

Fresh soil samples were collected and sieved using 2mm sieve in order to remove large particles, roots and stones. Ten gram of soil samples collected from 0cm-20cm and 20cm-40cm were weighed in to a beaker in triplicate for both fumigated and non-fumigated soils. The beakers were placed containing 25 mL of 99% alcohol free chloroform and without the chloroform were incubated for 24 hours at room temperature under sealed desiccator in dark room. This mechanism induces the destruction of microbial cellular structures, thereby liberating intracellular constituent into the soil matrix. After overnight incubation, both samples, treated and non-treated with chloroform containing beaker were removed from the desiccator allowing the chloroform vapor to escape. The samples were left open in a fume hood for few minutes in order to ensure the complete removal of chloroform. Both chloroform fumigated and non-fumigate samples were weighed and their weight were recorded using the following formulas.

MBC = C_f-C_{uf}

$$MBC\left(\frac{mg}{kg}\right) = (Cf - Cuf) \div kc$$

Cf- carbon concentration in the fumigated soil sample
Cuf- carbon concentration in un-fumigated soil sample
Kc- efficiency factor (0.45)

Application of glyphosate

From the beginning of 2008 different rate of glyphosate with different trade name has been applied in order to control weeds and increase coffee productivity.

Each year, only one herbicide which constitute glyphosate active ingredient sprayed to the specified plot of coffee testing site. The purpose of using this herbicide with different trade names was to examine their efficacy in control of weed infestation. The herbicides imported from different countries by traders/importers, after communicating with ministry of agriculture and getting permission for their trial sent to Jimma Agricultural Research Center, department of protection; tested for their effectiveness in suppressing different weed types and their result were reported for the respective department at the end of the year after completion of treatment.

Reagents and Materials used

Microbial biomass carbon (MBC) is a measure of the amount of carbon in the microbial biomass of soil. The chloroform fumigation extraction /CFE/ method is widely used to determine MBC in soil because it is effective and relatively straightforward.

Statistical Analysis

In order to quantify and identify if there is a significant difference among each treatment along the depth and between the glyphosate treated and non-treated one R-software was used at 95% significance level

Results and Discussions

Table.1 Soil sample results obtained from the sample across depth and treatment

Soil depth (cm)	MBC (mg/kg)	pH (1:2.5)
0-20		
Treated	63.56[c]	5.20
Non-treated	117.70[a]	5.86
20-40		
Sprayed	75.36[c]	5.22
Non-sprayed	93.60[b]	5.58

Soil Sample collection and soil pH

The collected soil samples were divided in to two in order to analyze for different purposes. Half of it was air dried, crushed and sieved by < 2mm, prepared for further analysis. The pH of the soil was analyzed by 1:2.5 (soil: water) ratio after shaking it for thirty minutes on an orbital shaker. The soil samples collected from the herbicide trial site and with the adjacent; on which the herbicide were not applied; tested for their pH level. Since the area has high rain fall throughout the year and soil acidity is another problem of the area, the pH of the samples were in the range of acidic. The glyphosate sprayed plot has 5.20 and 5.22 across the depth, 0-20cm and 20-40cm respectively. The adjacent coffee site trial had a high pH relative to the herbicide sprayed plots. The pH of the non-treated plot was 5.86 for the 0-20cm depth and 5.58 for the soil collected from the depth of 20-40cm respectively. The above result indicated that due to the removal of organic matter because of the glyphosate sprayed for the long period of years, soil fertility enhancer like soil organic matter were reduced and resulted in soil pH decline. Non-treated and non-sprayed soils consistently showed higher pH values, closer to neutral. This indicated that glyphosate application for longer time acidified the soil, which could indirectly affect microbial populations and activity. The microbial biomass data of the soil result indicated that a clear negative impact of the herbicide application and suggested that potential link to soil pH changes. The treated plot of soil pH value is significantly lower compared to the controls over which the glyphosate was not sprayed. Some herbicides can directly acidify the soil, while others might indirectly contribute to acidification by altering microbial processes that influence soil pH resulting in soil fertility decline. Herbicides can reduce soil pH, especially in treated areas compared to untreated ones, as demonstrated in orchards where herbicides treated strips showed significantly lower pH level than grass-covered alleys as indicated (Haynes, 1981). A lower pH of soil can increase herbicide persistence by slowing the degradation process of these chemicals, leading to their prolonged activity in the soil. Acidic conditions often reduce the effectiveness of microbial activity and chemical reactions that breakdown herbicides, allowing them to retain in the soil for extended periods. This increased persistence can complicate weed management strategies, as it results in residual herbicides affecting subsequent crops or causing unintended harm to non-targeted plants (Raeder et al., 2015). Soil pH was identified as a key determinant in shaping the distribution and composition of microbial communities. Its influence extends beyond microbial carbon, as it plays a pivotal role in regulating other critical soil properties, such as the availability and balance of other nutrients. By altering the soil's chemical environment, pH impacts nutrient solubility, cation exchange capacity, and the overall interactions between soil particles and microbial populations, thereby driving both biological and chemical processes within the soil ecosystem (Wang et al., 2022).

Soil Microbial Biomass Carbon

Soil microbial biomass carbon is one of the indicators of soil health which is very crucial in agriculture to enhance productivity. In both 0-20 and 20-40 cm depths, the glyphosate applied or sprayed soils exhibit significantly lower MBC compared to the non-treated /non-sprayed controls. This decrease in MBC indicated the decline in the living microbial biomass within the soil, which is very crucial for various soil functions like nutrient cycling, organic matter decomposition and soil aggregation. A significant decrease in in microbial diversity was observed at the surface (0-20cm) depth as compared to the deeper depth (20-40cm), where the soil is more exposed for the applied glyphosate, highlighting the adverse effects in declining pH levels on the complexity and richness of microbial community. The decrease in microbial biomass tends to be more pronounced in soils with low organic matter, as higher organic matter levels can help buffer against some of the negative impacts (Amritha & Devi, 2017). The MBC differences were observed in pronounced quantity between the glyphosate applied plot and non-applied one at the surface level potentially because, the deeper soils were less exposed to the herbicide applied. This reduction suggests that acidification created unfavorable conditions for many microbial species, potentially disrupting ecological functions, nutrient cycling and overall soil health (Wang et al., 2022). Glyphosate itself could be directly toxic to soil microorganisms, leading to their death and a subsequent a decrease in MBC. The application of herbicides can suppress soil respiration and dehydrogenase activity, which are critical indicators of microbial health (Rouhi-Kelarlou et al., 2024) (Douibi et al., 2024). Non-sprayed coffee plots, at both depths exhibited a higher MBC than sprayed coffee plot, though the difference was less dramatic than at the surface layer. This suggests that herbicide application affected microbial communities more strongly in the upper layers of soil, possibly due to reduced penetration at greater depths. Herbicides have been found to reduce microbial biomass carbon, with research highlighting a significant decline shortly after their application (Amritha & Devi, 2017).

In conclusion, the data presented provides compelling evidence of the detrimental effects of herbicide application on both soil microbial biomass carbon and soil pH, emphasizing the broader implications for soil health and ecosystem sustainability. The observed reduction in soil microbial biomass carbon is indicative of a significant decline in the abundance and metabolic activity of soil microorganisms, which play a pivotal role in maintaining essential ecological processes. These microorganisms are critical for nutrient cycling, organic matter decomposition, and the formation and stabilization of soil structure, all of which are fundamental to sustaining agricultural productivity and ecosystem balance.

In parallel, the observed decrease in soil pH reflects an increase in soil acidity, which can exacerbate the stress on microbial communities by creating unfavorable living conditions. Acidic soils can limit the availability of essential nutrients, inhibit the growth of beneficial microorganisms, and shift the composition of microbial communities toward less desirable species. This, in turn, can impair the soil's overall functionality, including its capacity to support healthy plant growth and resist environmental stressors.

These findings underscore the urgent need to recognize and address the ecological consequences of herbicide usage in agricultural systems. This observation highlighted the importance of adopting sustainable and integrated agricultural practices that minimize negative impacts on soil health. By prioritizing soil health, it is possible to achieve more sustainable agricultural systems that balance productivity with long-term environmental stewardship.

This analysis relied on a limited dataset and provides only a preliminary understanding of the interactions between herbicide use, soil microbial communities, and overall soil health. The dynamics involved are highly complex and influenced by various factors, including herbicide type, application rate, soil properties, climatic conditions, and the inherent resilience of microbial communities. Further investigation is essential to explore these variables in greater depth, assess long-term effects, and identify potential mitigation strategies to preserve soil health while managing weed populations. Comprehensive studies across diverse ecosystems and under varying agricultural practices are needed to draw more robust conclusions and inform sustainable land management practices.

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