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VERMICOMPOST EFFECTS ON SOIL CHEMISTRY AND BIOLOGY: CORRELATIONS WITH BASIL'S (*Ocimum basilicum* L.) TOTAL PHENOLIC CONTENT AND PHENOLOGICAL TRAITS

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Abstract: This study investigates the effects of vermicompost on the chemical and biological properties of soils, their nutrient content, and the effects on the growth and phenolic content of basil (*Ocimum basilicum* L.). Using a controlled experimental setup, we tested five dosages of vermicompost (0%, 4%, 12%, 20%, and 24%, w/w) to evaluate their influence on soil biological activity by measuring basal respiration (CO_2-C), microbial biomass C (MBC-C), and dehydrogenase activity (DHA) as well as basil's growth parameters and total phenolic content (TPC). The results show that vermicompost addition to soil enhanced soil microbial activity in direct proportion to the dose of vermicompost. The application of lower dosages of vermicompost (4% and 12%) significantly enhanced both fresh and dry weights. However, higher dosages (20% and 24%) were associated with reduced growth metrics. Notably, the highest vermicompost concentration (24%) led to a substantial increase in total phenolic content (TPC) in basil leaves, correlating with decreased growth metrics. The values for CO_2-C, MBC-C, and DHA were determined as 0.135, 20.756, and 12.806, respectively, at the highest solid vermicompost application dose of 24%. Fresh and dry weight were determined at 12% vermicompost application, and plant height and leaf length were also determined at 12% vermicompost application. The TPC showed a remarkable increase at the 24% application dose. This response indicates a defense mechanism of the plant against oxidative stress caused by excess nutrients or salinity from the vermicompost. A multiple regression analysis following a correlation analysis also revealed an inversely proportional relationship between phosphorus content in the soil and total phenolic content in basil leaves. Our findings illustrate that while moderate vermicompost dosages optimize plant growth and health, higher concentrations can strategically enhance phenolic content due to nutrient overload or salt-induced stress. These results offer critical insights for tailoring organic amendment applications to balance plant growth and biochemical properties in agricultural practices.

Keywords: *Ocimum basilicum* L., Phenolic content, Soil biology, Soil chemical parameters, Soil microbial activity, Vermicompost

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1. Introduction

Vermicompost, derived from the breakdown of organic material through earthworm activity, has been increasingly recognized for its potential to enhance soil quality and plant growth. Research indicates that vermicompost enriches soil with nutrients, improves its structure, and enhances microbial activity, all of which contribute to better plant health and productivity (Edwards and Burrows, 1988; Atiyeh et al., 2000; Kizilkaya et al., 2012). Studies have shown that plants grown in vermicomposted soil exhibit increased growth rates, yield, and resistance to pests and diseases compared to those grown in non-vermicomposted soils (Arancon et al., 2005; Bachman and Metzger, 2008). Moreover, vermicompost has been found to influence the synthesis of plant secondary metabolites, including phenolic compounds, which play a crucial role in plant defense mechanisms (Szczech, 1999). Given the global push towards sustainable agriculture, understanding the specific effects of vermicompost on crop species such as basil (*Ocimum basilicum* L.), known for its economic and medicinal value, is particularly relevant.

Additionally, the interactions between compost, vermicompost, and earthworms have been found to significantly influence plant growth and yield, emphasizing the importance of integrating these natural processes into greenhouse and field settings (Doan et al., 2013). The positive effects of vermicompost on nutrient dynamics and the reduction of heavy metals in soil further illustrate its role in creating healthier, more productive soil environments (Kannadasan et al., 2013). Earthworms, often described as "ecosystem engineers", contribute to these improvements by enhancing soil structure, nutrient cycling, and plant growth, which are essential for robust agricultural systems (Wang et al., 2021). Moreover, the research by Samaranayake and Wijekoon (2011) highlights the ability of specific earthworm species to boost soil fertility and enhance the growth of crops such as maize, showcasing the practical applications of vermicompost in agriculture. Vermicomposts are commonly referred to as biological fertilizers due to their high microbial content. These organic fertilizers are known to promote soil microbial biodiversity by introducing a diverse range of beneficial microbes such as Arbuscular Mycorrhizal (AM) fungi, Azotobacter, Agrobacterium, and Rhizobacteria into the soil (Domínguez et al., 2014; Broz et al., 2016). Vermicomposts contain much larger populations of bacteria (5.7×10^7), fungi (22.7×10^4), and actinomycetes (17.7×10^6) compared to those in conventional thermophilic composts (Edwards et al., 2010). This superiority of vermicompost is due to its very high microbial activity. Additionally, vermicompost has a higher concentration of beneficial microbial populations, readily available plant nutrients, and plant growth regulators compared to conventional compost (Türkay, 2023). This study seeks to build upon these findings by specifically investigating the impact of varying vermicompost concentrations on basil, aiming to optimize plant health and soil quality while mitigating potential negative effects associated with excessive organic amendments.

In recent years, there has been a growing interest in studying the effects of vermicompost on plant growth and the phenolic content of various plants, including basil. Vermicompost, which is organic fertilizer produced from earthworms, has been found to significantly impact the growth, chemical composition, and oil yield of plants like basil. Studies have shown that vermicompost applications at different concentrations can lead to increased fresh and dry weight, leaf area, essential oil compounds, and nutrient levels in plants like basil (Türkay and Öztürk, 2019; Massoud et al., 2022; Türkay and Öztürk, 2023; Türkay et al., 2024).

The phenolic content of plants, including basil, has been a subject of interest due to its antioxidant properties. Basil extracts have been reported to possess a higher total phenolic acid content and greater antioxidant activity, making them valuable for potential pharmacological effects (Mintas et al., 2021). Furthermore, studies have highlighted the rich source of phenolic compounds in basil leaves, including various phenolic acids, flavonol-glycosides, and anthocyanins (Zlotek et al., 2016). These compounds contribute to the antioxidant capacity of basil and are essential for its health benefits (Türkay et al., 2024).

Research has also explored the antioxidant capacity of basil extracts, comparing different extraction methods and studying the antioxidant properties of various parts of the plant. Studies have investigated the antioxidant properties of different extracts of *Ocimum basilicum* and *Origanum vulgare*, emphasizing the importance of these plants as potential sources of antioxidants (Kaurinovic et al., 2011). Additionally, investigations into the effects of different drying methods on the proximate composition and antioxidant activities of *Ocimum basilicum* leaves have provided insights into preserving the bioactive compounds in basil (Mahirah et al., 2018).

Moreover, the influence of environmental factors on the phenolic composition of basil plants has been studied. Factors such as nitrogen starvation, different soil types, water stress, and pre-harvest UV-B supplementation have been found to affect the phenolic content of basil leaves, highlighting the importance of environmental conditions in determining the phytochemical profile of plants like *Ocimum basilicum* (Luna et al., 2015; dos Santos Nascimiento et al., 2020; Prinsi et al., 2020). Additionally, the use of humates from vermicompost to mitigate the effects of salinity on basil growth further underscores the potential of organic amendments in enhancing plant resilience (Reyes-Pérez et al., 2017).

Furthermore, the potential allelochemical effects of basil on weed control in other crops have been investigated, shedding light on the broader ecological implications of basil cultivation (Kamel et al., 2021). Additionally, studies have explored the biofortification of basil leaves with selenium to enhance their quality and shelf life, demonstrating innovative approaches to improving the nutritional value of basil plants (Puccinelli et al., 2020). The role of mycorrhizal fungi and microalgae in modulating the antioxidant capacity of basil plants further emphasizes the intricate interactions between plants and their symbiotic partners in influencing phenolic content (Hristozkova et al., 2018).

In conclusion, the research on vermicompost applications, phenolic content, and antioxidant properties of basil and other plants provides valuable insights into the factors influencing plant growth, chemical composition, and bioactive compound levels. Understanding the effects of vermicompost, environmental conditions, and extraction methods on plant phenolics is crucial for maximizing the nutritional and medicinal potential of plants like basil. These studies contribute to the broader knowledge of plant science and offer practical implications for sustainable agriculture and herbal medicine.

The primary objective of this study is to investigate the impact of different concentrations of vermicompost on the growth characteristics and total phenolic content of basil (*Ocimum basilicum* L.) due to the high microbial activity content of vermicompost. Specifically, the research aims to delineate how varying vermicompost dosages influence plant height, leaf length, and fresh and dry biomass, thereby providing insights into the optimal vermicompost concentration for enhancing plant growth and health. Additionally, the study seeks to explore the relationship between changes in soil microbial activity caused by applied vermicompost dosages and the production of phenolic compounds, which are vital for plant defense mechanisms. By analyzing these relationships, the study aims to provide practical guidelines for the application of vermicompost in agriculture, particularly in enhancing soil quality and plant health while minimizing potential adverse effects related to nutrient overload or salt stress. This comprehensive analysis is intended to aid farmers and agricultural practitioners in making informed decisions regarding the use of organic amendments like vermicompost in sustainable farming practices.

2. Materials and Methods

Before the experiment, the soil was air-dried to standardize moisture content and sieved through a 2 mm sieve to remove debris and ensure a uniform texture. This was crucial for accurate vermicompost integration. The soil underwent a detailed characterization to establish its baseline chemical properties (Table 1), including pH, electrical conductivity (EC), organic matter content, and essential nutrients like nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), iron (Fe), copper (Cu), zinc (Zn), and manganese (Mn). These analyses were conducted using standard soil testing techniques, such as pH and EC measurements in a soil-water suspension, organic matter by the modified Walkley-Black method (Nelson and Sommers, 1996), and nutrient quantification through spectrometric or titrimetric methods. This initial characterization provided a crucial understanding of the soil's nutrient status and fertility, important for assessing the impacts of vermicompost on soil health and plant growth during the experiment.

The vermicompost used in this study was produced in-house at the Kırşehir Ahi Evran University, Faculty of Agriculture, using 100% barnyard manure and *Eisenia fetida* earthworms, which are effective composters. The earthworms were housed in plastic containers filled with manure under controlled conditions suitable for vermicomposting. After completion, the vermicompost was passed through a 2-mm sieve to ensure consistency and remove larger uncomposted particles, crucial for maintaining quality. To preserve its microbiological properties, the vermicompost was refrigerated at +4°C for a day before use, maintaining microbial integrity vital for its effectiveness as a soil amendment. Chemical and biological analyses of the vermicompost included pH, electrical conductivity (EC), organic matter content, and concentrations of nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), and trace elements like iron (Fe), copper (Cu), zinc (Zn), and manganese (Mn). These analyses ensured the vermicompost's quality met the experimental needs, establishing a baseline for assessing its impact on soil health and plant growth in the study.

For the production of basil seedlings, the experiment began with the germination of basil seeds, which were bought from a local agricultural products supplier, in a controlled environment. The seeds were sown in 50 cc pots, three seeds per pot, across 10 replicates, ensuring an adequate number of samples for the study. At the end of the second week of germination, an evaluation was conducted to select the healthiest seedlings for continuation in the experiment. In each pot, one healthy individual was selected based on its overall health and developmental progress compared to its peers, and the remaining seedlings were carefully removed.

2.1. Experimental Design

The experiment assessed the impacts of vermicompost on basil through a structured treatment setup under controlled conditions, aiming for high scientific standards and replicability. Five vermicompost doses were prepared: 0% (control), 4%, 12%, 20%, and 24%, representing the proportion mixed with base soil to explore a spectrum of effects. The results of each treatment was obtained from the samples which taken from three pots in each group, totaling 15 experimental pots. The experiment took place in a plant growth cabinet, enabling precise control over temperature, humidity, and light. The temperature was consistently held at 25 °C, optimal for basil growth, humidity was consistently held at 55-60% and lighting (16h light: 8h dark photoperiods) was adjusted to promote full photosynthesis without causing photobleaching or heat stress.

Before planting, thorough mixing of soil and vermicompost for each treatment ensured uniformity, crucial for eliminating growth variability due to nutrient distribution differences. Soil moisture was closely monitored to maintain field capacity by weighing the pots daily and adjusting water levels accordingly.

Table 1. Analyses and methods used to determine the chemical properties of the soil used in the experiment (Kacar, 1994).

Analysis	Method
Soil Texture (% sand, silt, clay sized particles)	Hydrometer method
Soil reaction (pH)	Measured with a pH meter in a 1:1 (w/v) soil:distilled water mixture
Electrical Conductivity (EC, dS m^{-1})	Measured with an EC meter in a 1:1 (w/v) soil:distilled water mixture
Organic Matter, %	Modified Walkley-Black method
Total N, %	Kjeldahl method
Available P, mg.kg^{-1}	Olsen method
Exchangeable K, Ca, Mg, me100 g^{-1}	Ammonium acetate extraction method
Available Fe, Cu, Zn, Mn, mg·kg^{-1}	DTPA extraction method

At the end of the experiment, critical growth parameters such as plant height, leaf length, and biomass (both fresh and dry weights) were measured for each individual in application groups. Additionally, the phenolic content in the basil leaves was analyzed by the Folin-Ciocalteu method to determine the effects of the vermicompost treatments.

2.2. Data Collection Methods

2.2.1. Description of plant growth measurements

In the study, detailed phenological and growth metrics were recorded to evaluate vermicompost's influence on basil. Plant height was measured from the stem base to the apex of the tallest shoot using a ruler, with measurements noted in millimeters. Similarly, leaf length was assessed from the base to the tip of the longest leaf on each plant, also in millimeters. At the end of the 10-week growth period, the fresh weight of each plant was promptly recorded post-harvest to prevent dehydration. For dry weight assessments, plants were placed in a drying area away from direct sunlight and kept at room temperature until thoroughly dried, after which dry weights were noted to evaluate biomass accumulation across different vermicompost treatments. Additionally, 15 grams of the top leaves from each plant were collected, dried as described, and analyzed for total phenolic content (TPC) using the Folin-Ciocalteu method. This spectrophotometric technique measures phenolic compounds, offering insights into the plants' antioxidant potential.

2.2.2. Soil and vermicompost sampling methods

Soil samples were collected at two critical times: before vermicompost application to establish a baseline, and after the 10-week growth period to evaluate treatment effects. The representative soil samples, which taken from the pots, were split into two portions; one was refrigerated at +4 °C to preserve biological properties for microbial analysis (Table 2), and the other was air-dried, sieved, and analyzed for chemical properties. The samples underwent detailed analysis for pH, electrical conductivity, nutrient content and microbial activity using standard laboratory methods, ensuring thorough evaluation of vermicompost's effects on soil health and plant growth.

This detailed approach to sampling and analysis ensures that the data collected will provide a comprehensive insight into how vermicompost affects soil properties over the course of the experiment, thereby facilitating a better understanding of the interactions between soil amendments and plant responses.

2.2.3. Total phenolic content analysis method

To evaluate the impact of vermicompost on the total phenolic content (TPC) of basil, we employed the Folin-Ciocalteu method, a standard for quantifying phenolic compounds in plant tissues and assessing antioxidant capacity. At the conclusion of the 10-week growing period, top leaves from each basil plant were uniformly harvested, with approximately 15 grams of leaf material collected from similar positions on each plant to ensure consistency. These leaves were then dried in the shade at room temperature to preserve the phenolic compounds and maintain consistent dry weight. The dried leaves were weighed, and a specific amount was used for extraction with an 80% methanol solution, chosen for its effectiveness in extracting phenolic substances. The extraction involved macerating the leaves in methanol under controlled conditions to ensure thorough extraction of phenolic compounds. The Folin-Ciocalteu reagent, prepared following standard laboratory protocols, reacts with the phenolic compounds to form a blue complex, the intensity of which was measured spectrophotometrically at 765 nm.

Table 2. The microbiological analyses used to determine the biological properties of soil

Analysis	Protocol
Basal Respiration (CO_2-C)	50 g of soil is moistened with distilled water until it reaches 55% of its maximum water holding capacity and placed into 1-liter Isermeyer jars. 25 mL of 0.05 M NaOH is added to the alkaline tube of the jar, and the jars are incubated at 25°C for 3 days. The CO_2 released by microbial respiration is trapped by the alkali, and the remaining OH- is titrated with standardized HCl in the presence of phenolphthalein indicator. The result is expressed as µg CO_2-C g^{-1} dry soil.
Microbial Biomass Carbon (MBC)	50 g of soil is moistened with distilled water until it reaches 55% of its maximum water holding capacity, then 200 mg of glucose is added and placed into 1-liter Isermeyer jars. The amount of CO_2 released from the soil is determined hourly as described in section 3.5.1. The maximum respiration at the end of 4 hours is calculated using the equation 40.04 µg CO_2 g^{-1} + 3.75 and the result is expressed as µg CO_2-C g^{-1} dry soil.
Dehydrogenase Activity (DHA)	After adding 30 mg glucose, 1 mL of 3% 2,3,5-triphenyltetrazolium chloride (TTC) substrate solution, and 2.5 mL of distilled water to a 6 g soil sample, the mixture is incubated at 37 °C for 24 hours. At the end of the incubation, the released 1,3,5-triphenylformazan (TPF) is determined spectrophotometrically at a wavelength of 485 nm, and the result is expressed as µg TPF g^{-1} dry soil.

Absorbance readings are directly proportional to the phenolic content in the samples. A calibration curve using known concentrations of gallic acid enabled the accurate calculation of the TPC, expressed as mg of gallic acid equivalents (GAE) per gram of dry weight.

2.3. Data Analysis

To evaluate the effects of varying vermicompost concentrations on basil growth parameters and TPC, we performed an Analysis of Variance (ANOVA) followed by Tukey post-hoc test (Genç and Soysal, 2018). This combination identifies significant differences between groups, denoted by distinct letters, indicating statistically significant results at $P<0.05$. Additionally, Principal component analysis (PCA) was employed to reduce data dimensionality and highlight the main variables affecting plant and soil traits (Kurnaz et al., 2022). This method uncovered patterns in how vermicompost dosages influence growth and soil characteristics. Pearson correlation coefficients were calculated to explore the relationships between growth parameters, soil properties, and total phenolic content (TPC), providing insights into the linear interactions within the dataset. Multiple regression analysis (Kurnaz et al., 2021) was used to examine the impact of changes in soil parameters, notably organic matter and phosphorus content, on TPC. This analysis provided a comprehensive understanding of the interactions between soil and phenolic compounds. All statistical tests were conducted with a 95% confidence interval, ensuring the robustness of our findings. The statistical analyses were conducted using the provided calculators on the Social Science Statistics (2024).

3. Results and Discussion

3.1. Characteristics of Soil and Vermicompost in Basil Cultivation

Tables 3 and Table 4 provide detailed chemical analyses of the soil and vermicompost used in our experiments, crucial for understanding the initial conditions affecting basil growth. Table 3 focuses on the soil used as a growing medium, documenting key parameters like pH, electrical conductivity, organic matter content, and essential nutrients such as nitrogen, phosphorus, and potassium. These parameters are vital for assessing the soil's fertility and its ability to support healthy plant growth. The data serves as a baseline, reflecting the soil's condition prior to any experimental treatments, and acts as a control for evaluating the impacts of subsequent vermicompost additions.

Table 4 describes the chemical properties of the vermicompost utilized in the experiments, providing insights into its quality and effectiveness as a soil amendment. This table outlines various characteristics of the vermicompost, including its nutrient content and biological properties, which are important for judging its capacity to enhance plant health and growth. The information is instrumental in assessing how vermicompost influences not only the phenological and biochemical traits of the basil plants but also the overall biological quality of the soil after its application.

Together, both tables are essential for establishing a scientific framework for the experimental treatments. The soil evaluated in our study has a slightly alkaline pH of 7.32 which is suitable for many plants, including basil, which thrive in a pH range of 6.0 to 7.5 (Couto, 2018; Neina, 2019). The electrical conductivity (EC) of 529.5 µS cm^{-1} indicates a moderate salt content, which should not significantly affect basil's water uptake (Rao et al., 2020). However, the low organic matter content of 1.151% suggests limited nutrient reserves and microbial activity, which are important for robust plant growth and soil structure (Tisdall and Oades, 1982; Chang et al., 2014). Essential nutrients like nitrogen, phosphorus, and potassium are present but in modest amounts, indicating basic soil fertility sufficient for basic plant needs but likely requiring supplementation for optimal growth.

Table 3. Chemical properties of the soil used in the experiment

Chemical properties	Values
pH	7.32
EC, µS cm^{-1}	529.5
Organic matter, %	1.151
Total N, %	0.100
Available P, ppm	1.681
Exchangeable K, me100 g^{-1}	0.427
Exchangeable Ca, me100 g^{-1}	33.711
Exchangeable Mg, me 100 g^{-1}	3.189
Available Fe, mg.kg^{-1}	3.41
Available Cu, mg.kg^{-1}	1.01
Available Zn, mg.kg^{-1}	0.32
Available Mn, mg.kg^{-1}	3.20

Table 4. Chemical properties of vermicompost used in the preparation of plant growing medium in the experimental setup

Chemical properties	Values
pH	7.10
EC, dS m^{-1}	12.8
Organic matter, %	41.6
Total N,%	2.08
C/N	11.59
NO_3-N mg.g^{-1}	713.05
NH_4-N mg.g^{-1}	518.2
Total P, %	0.81
Total K, %	1.72
Total Ca, %	1.64
Total Mg, %	0.73
Fe, mg.kg^{-1}	1146.90
Cu, mg.kg^{-1}	9.95
Zn, mg.kg^{-1}	25.47
Mn, mg.kg^{-1}	137.92

The vermicompost used in our study is slightly more acidic with a pH of 7.10, falling within a beneficial range for basil cultivation. This acidity level is advantageous as vermicompost contains a combination of macro and micro-nutrients that positively impact plant nutrition, growth, photosynthesis, and chlorophyll content, all of which are essential for basil cultivation (Moustafa et al., 2022). It has a high EC of 12.8 dS m^{-1}, suggesting significant salt content that could cause salinity issues if over-applied. The vermicompost is rich in organic matter (41.6%), enhancing soil structure, moisture retention, microbial life, and plant health. It contains high levels of essential nutrients and is abundant in micronutrients like iron, copper, zinc, and manganese, crucial for plant enzymatic processes and disease resistance.

Using vermicompost can significantly enhance soil nutrient levels and improve soil structure, benefiting water retention, aeration, and microbial activity. This helps in nutrient cycling and increases nutrient uptake by plants. Studies have shown that vermicompost applications lead to improvements in soil microbial biomass, soil porosity, water holding capacity, nutrient content, and plant growth (Sim and-Wu, 2010; Tejada et al., 2010; Pramanik et al., 2010; Lim et al., 2014; Pereira et al., 2014; Akhzari et al., 2015; Yadav and Garg, 2015; Rekha et al., 2018; Jahanbakhshi and Kheiralipour, 2019; Rivier et al., 2022). However, the high EC of vermicompost requires careful management to avoid salinity stress. Proper application rates and methods are crucial to maximize benefits and prevent adverse effects in the soil-plant system, ensuring enhanced soil fertility and support for plant growth.

3.2. The Effects of Vermicompost Applications on Soil Properties and Microbial Activity

The addition of vermicompost to soil at increasing doses (0%, 4%, 12%, 20%, and 24%) significantly enhances various chemical and biological properties of the soil (Table 5). As vermicompost doses increase, soil microbial activity, represented by CO_2-C and DHA, and microbial biomass, indicated by MBC-C, show marked improvements. Specifically, CO_2-C rises from 0.054 µg.g^{-1} at control (0%) to 0.135 µg.g^{-1} at 24%, and MBC-C increases from 7.759 µg.g^{-1} to 20.756 µg.g^{-1} over the same range. Additionally, dehydrogenase activity (DHA) increases from 2.416 µg TPF g^{-1} at 0% to 12.806 µg TPF g^{-1} at 24% (Table 5). These changes reflect enhanced soil microbial health and activity due to the addition of organic material from vermicompost.

Moreover, vermicompost application leads to significant improvements in soil fertility indicators, including organic matter (OM), total nitrogen (N Total), and phosphorus (P). OM content rises dramatically from 1.151% at 0% to 15.262% at 24%, while total nitrogen and phosphorus increase from 0.100% to 0.537% and 1.681 ppm to 13.876 ppm, respectively. Although soil pH decreases with higher vermicompost doses, from 7.315 to 7.212, electrical conductivity (EC), which measures soluble salt content, increases from 0.529 dS m^{-1} to 0.981 dS m^{-1}. These changes highlight the substantial benefits of vermicompost in enriching soil nutrients and enhancing soil structure, making it an effective amendment for sustainable soil management.

Overall, the addition of vermicompost at increasing doses positively affects the chemical properties of the soil, enhancing microbial activity, organic matter content, and nutrient availability. However, the increase in electrical conductivity indicates a potential risk of salinity stress at higher vermicompost doses.

3.3. Observations on Basil Growth and TPC under Different Vermicompost Treatments

Figure 1 and Figure 2 offer insights into the influence of varying doses of vermicompost on the phenological traits and total phenolic content (TPC) of basil, respectively.

Figure 1 reveals that applying vermicompost at concentrations of 4% and 12% significantly increased the fresh weights of basil to 21.78 grams and 22.93 grams, respectively, compared to 9.72 grams in the control group. However, fresh weight declined at a concentration of 20% and decreased further at 24%, suggesting that lower doses are more effective for biomass accumulation. The dry weight peaked at a dosage of 4% (1.83 grams) and decreased slightly at higher dosages but remained above control levels, indicating that moderate vermicompost levels effectively support biomass retention. Plant height was highest at 373.30 mm with 4% vermicompost, compared to 314.90 mm in the control, and decreased with higher dosages, though all treated plants were taller than the control. The maximum leaf length (108.40 mm) was observed at 12% dosage, with significant lengths also noted at 4% and 24% dosages, suggesting that mid-range vermicompost dosages might optimize leaf development.

Table 5. Chemical and biological properties of the soils of vermicompost (VC) application groups

VC Dose	CO_2-C µg.g^{-1}	MBC-C µg.g^{-1}	DHA µg TPF g^{-1}	pH	EC Ds m^{-1}	OM %	Total N %	P ppm
0%	0.054^{e}	7.759^{e}	2.416^{e}	7.315^{a}	0.529^{e}	1.151^{e}	0.100^{e}	1.681^{e}
4%	0.067^{d}	11.423^{d}	10.456^{d}	7.290^{b}	0.612^{d}	3.574^{d}	0.227^{d}	2.487^{d}
12%	0.096^{c}	15.035^{c}	12.007^{c}	7.264^{c}	0.830^{c}	7.987^{c}	0.360^{c}	6.889^{c}
20%	0.124^{b}	18.943^{b}	12.556^{b}	7.237^{d}	0.934^{b}	12.288^{b}	0.477^{b}	11.316^{b}
24%	0.135^{a}	20.756^{a}	12.806^{a}	7.212^{e}	0.981^{a}	15.262^{a}	0.537^{a}	13.876^{a}

The data are reported as means of 3 replicates. Means followed by the different letters are statistically significant at $P<0.05$.

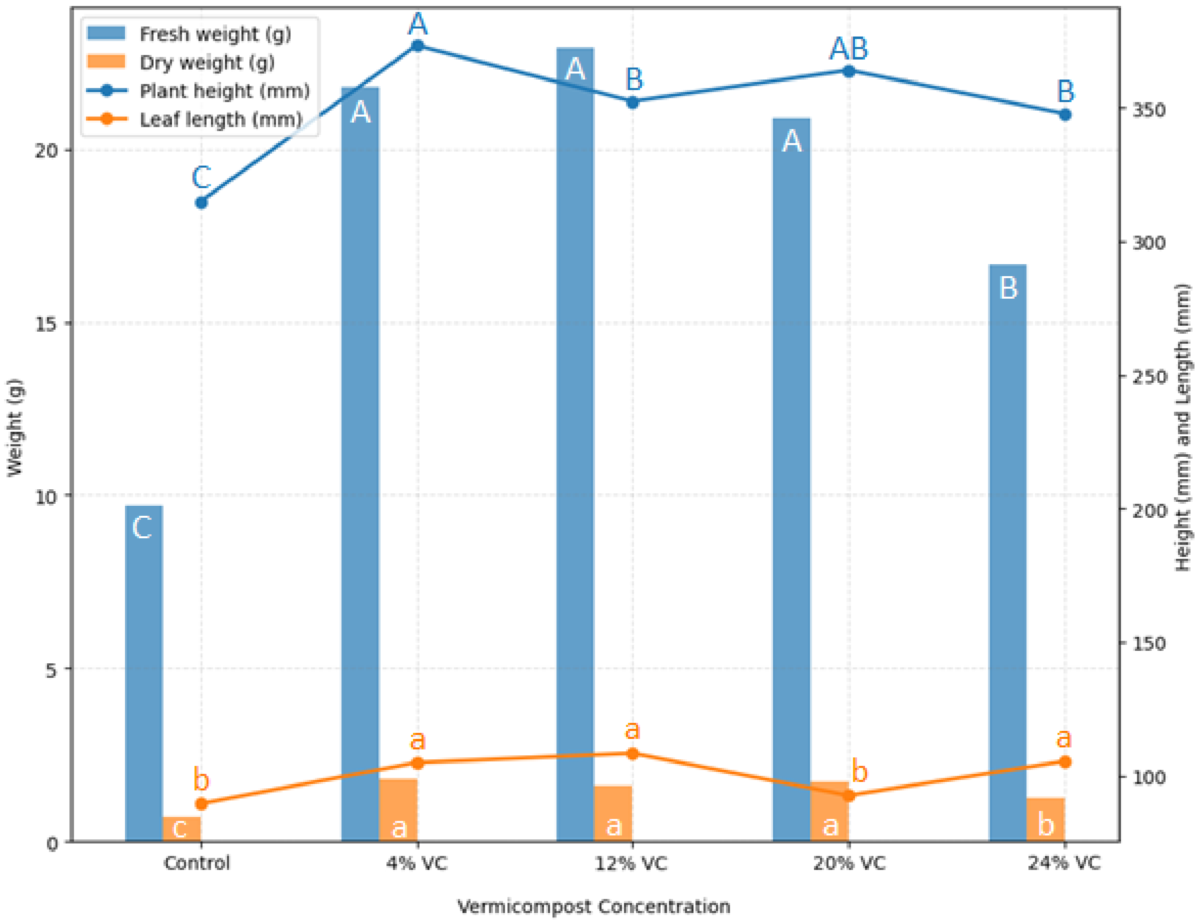


Figure 1. Effects of varying vermicompost doses on the phenological traits of basil.

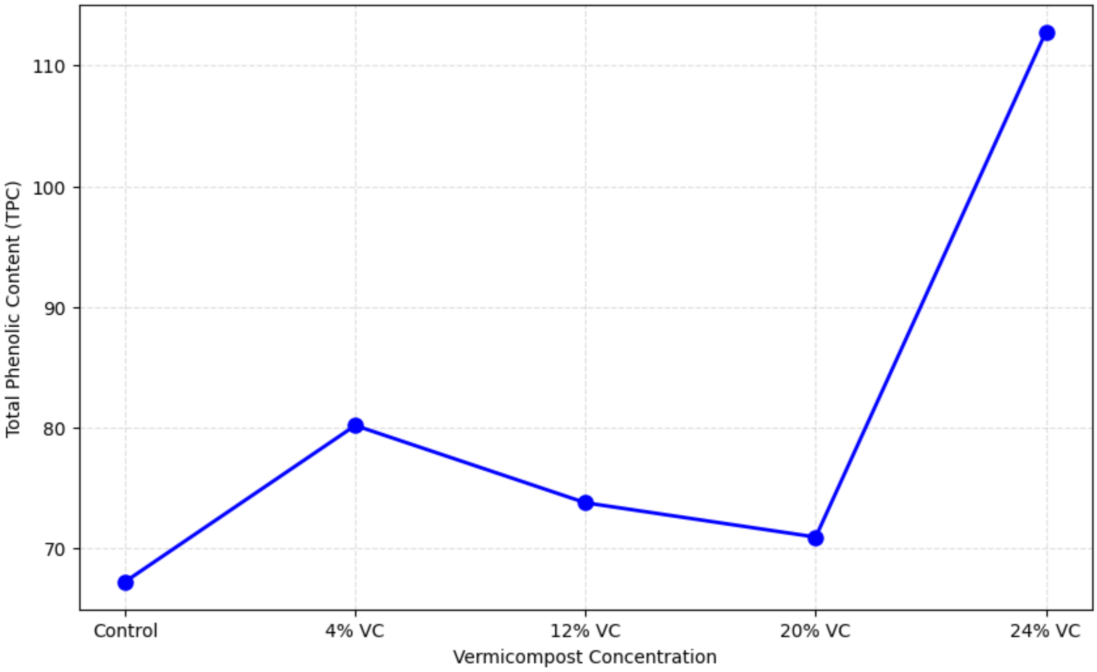


Figure 2. Impact of different vermicompost dosages on the total phenolic content (TPC) of basil.

TPC significantly increased with vermicompost application, with the control group having the lowest value at 67.25 mg GAE/g (Figure 2). TPC rose to 80.19 mg GAE/g at a 4% dosage but slightly decreased at 12% and 20%. The highest phenolic content (112.77 mg GAE/g) was observed at the 24% dosage, indicating a potential dose-response relationship where phenolic content might increase substantially at higher vermicompost concentrations.

The results suggest a nonlinear relationship between vermicompost dosage and both phenological traits and TPC. Moderate vermicompost applications (around 4% to

12%) generally enhanced growth parameters more effectively than no vermicompost. Very high dosages (24%), however, seemed to induce stress, reflected in reduced growth parameters but increased phenolic content, possibly as a stress response mechanism in basil. This increase in phenolics at high vermicompost levels could be due to enhanced secondary metabolite production, which plants often increase under stress conditions (Akula and Ravishankar, 2011; Kumar and Sharma, 2018; Tikoria et al., 2022; Sharma et al., 2023; Türkay and Öztürk, 2023; Türkay et al., 2024).

These findings suggest that while moderate vermicompost applications can enhance growth and overall biomass, higher concentrations might be used strategically to boost phenolic content, potentially enhancing the medicinal and nutritional value of basil. Thus, the choice of vermicompost dosage could be tailored depending on the desired outcome, whether it is maximizing growth or enhancing specific phytochemicals.

Overall, the experiment underscores how organic amendments like vermicompost can influence plant growth and secondary metabolite production, particularly phenolic compounds. This aligns with existing research suggesting that vermicompost not only improves soil fertility and plant growth but also enhances the production of plant secondary metabolites due to increased nutrient availability and improved soil health (Atiyeh et al., 2000; Nunes et al., 2018; Nurhidayati et al., 2018).

This study demonstrates that lower vermicompost dosages (4% and 12%) enhance fresh and dry weights of plants, suggesting improved nutrient uptake and soil structure which boost root growth and nutrient absorption, consistent with findings from Gill et al. (2018). Conversely, higher dosages (20% and 24%) may cause nutrient overload or salt stress, potentially impeding plant growth despite the rich nutrient content of vermicompost. Optimal growth in plant height and leaf length at mid-range dosages confirms that moderate levels provide a balanced nutrient mix, avoiding physiological stress. A study by Ibrahim et al. (2022) found that higher rates of vermicompost, especially when combined with high concentrations of ascorbic acid, resulted in optimal growth and yield of tomato plants grown in saline soil, suggesting that very high or low dosages could potentially lead to nutrient overload or salt stress. Pérez-Gómez et al. (2017) reported that higher dosages of vermicompost could lead to salt stress, impacting potato plants' growth negatively, whereas mid-range dosages minimized these stress effects and were optimal for growth. Adamipour et al. (2016) observed that mid-range doses of vermicompost produced the highest growth values for *Festuca arundinacea* Schreb. under salinity stress conditions, suggesting that higher doses may cause detrimental effects due to nutrient overload or exacerbation of stress conditions.

A notable increase in total phenolic content (TPC) at the highest vermicompost dosage (24%) correlates with reduced growth metrics, indicating a stress-induced enhancement in phenolic synthesis. Plants likely increase phenolics as a defense mechanism against oxidative stress caused by high nutrient levels or salinity from the vermicompost. Phenolics are crucial in plant defense against abiotic stresses such as nutrient and salt stress, which is consistent with the increased phenolic content observed. Aliyar et al. (2021) discuss the effect of vermicompost application on the growth and water relationships of quinoa plants under salinity stress conditions, suggesting that higher vermicompost concentrations lead to an increase in phenolic content as a response to oxidative stress. Beykkhormizi et al. (2016) noted that the application of vermicompost under salinity stress conditions led to significant changes in phenolic content in bean plants, pointing to a stress response mechanism involving phenolic synthesis due to high nutrient levels or salinity.

The results highlight a nonlinear relationship between vermicompost dosage and the resulting phenological and biochemical traits, suggesting that vermicompost is beneficial up to a threshold; beyond this, it may induce stress that compromises growth while boosting secondary metabolite production. This duality offers strategic opportunities for agricultural applications and enhancing phytochemical properties. For crops focusing on growth and yield, maintaining moderate vermicompost applications (around 4% to 12%) is ideal for optimal plant growth and health. For medicinal and aromatic plants, where secondary metabolites like phenolics are valued, higher vermicompost dosages can be utilized to enhance the concentration of bioactive compounds, increasing their therapeutic and nutritional value.

In conclusion, vermicompost is a valuable soil amendment for improving plant growth and soil properties, but its application must be carefully managed to prevent adverse effects at higher concentrations. Tailoring application rates to meet specific crop needs and desired outcomes allows growers to maximize the benefits of vermicompost, supporting sustainable agricultural practices and enhancing the nutritional and medicinal value of crops like basil.

3.4. Statistical Insights: Understanding the Data through PCA, Correlation Coefficients, and Multiple Regression Analysis

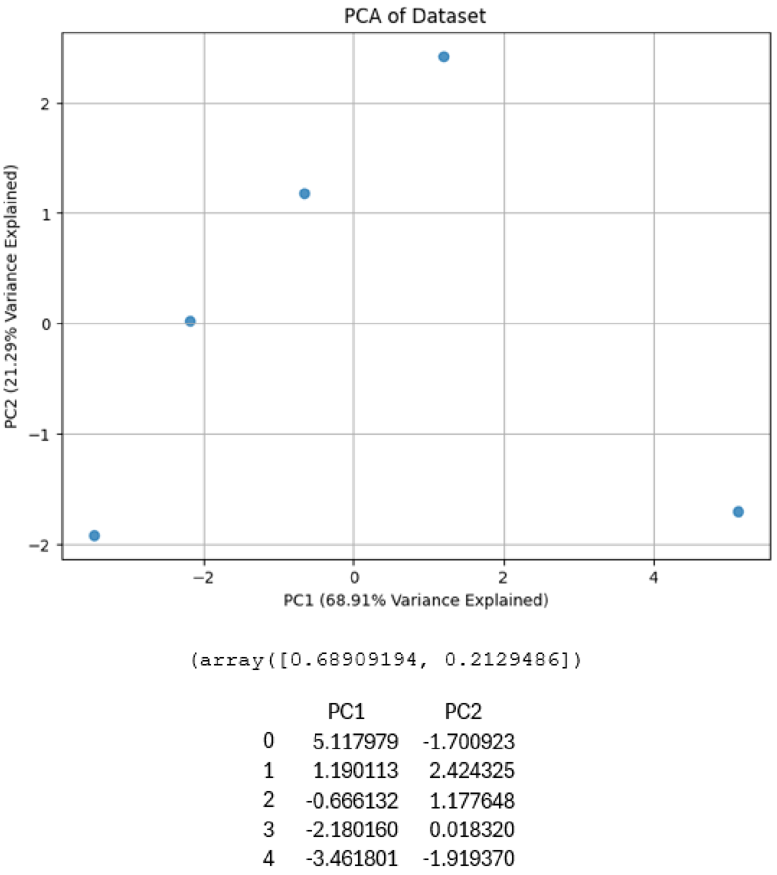
In our study, principal component analysis (PCA) was applied to investigate the relationships and underlying patterns in the data collected from various vermicompost (VC) dosing levels and their impact on plant growth parameters and soil properties (Figure 3). This multivariate statistical technique was utilized to reduce the dimensionality of the dataset, comprising measurements such as plant height, leaf length, fresh and dry weight of plants, total phenolic content (TPC), microbial biomass carbon (MBC-C), dehydrogenase activity (DHA), basal respiration (CO_2-C), pH, electrical conductivity (EC), organic matter (OM) content, total N, and P content.

According to the results, PCA deciphers the patterns in how VC affects plant growth and soil properties, identifying two principal components that explain 90.20% of the data variance. The first principal component (PC1) explained 68.91% of the variance, highlighting its correlation with key growth parameters and nutrient dynamics, showcasing VC's significant impact on nutrient enhancement in plants and soil. The second principal component (PC2), accounting for 21.29% of the variance, focused on more specific soil chemical properties and microbial interactions, underscoring subtle effects on soil health.

The PCA scatter plot illustrated the distribution and relationship of different VC dosing groups, revealing how similar or unique their impacts are, which helps in identifying optimal VC dosages. This analysis proves invaluable in simplifying complex multivariate data, allowing for a concentrated examination of essential factors affecting plant and soil health. It guides further research and practical applications by improving understanding of VC's broad and nuanced impacts, thus enhancing agricultural outcomes. PC1 primarily indicates that VC boosts plant growth through nutrient availability, while PC2 suggests VC's secondary effects modify soil chemistry and microbial activity, crucial for nutrient cycling. This streamlined approach helps in optimizing VC dosing, enhancing plant productivity, soil health, agricultural sustainability, and crop quality.

The Pearson correlation coefficient analysis in our study aimed to determine the relationships between various plant growth parameters, soil properties, and biochemical indicators (Table 6).

The outcomes of Pearson correlation coefficient analysis highlight significant relationships between plant growth parameters, soil properties, and vermicompost dosages, providing valuable insights for enhancing agricultural practices and sustainability. The analysis reveals strong positive correlations between plant height, leaf length, fresh and dry weights, indicating that plants with greater height and leaf length generally exhibit higher biomass. This correlation extends to the total phenolic content (TPC), which correlates positively with plant biomass, CO_2-C, microbial biomass carbon (MBC-C), and total nitrogen (Total N, %), suggesting that healthier, more robust plants tend to accumulate more phenolics, beneficial for plant defense mechanisms. Moreover, soil biochemical properties such as CO_2-C and MBC-C, indicators of microbial activity, show positive correlations with each other and dehydrogenase activity (DHA), indicating interconnected microbial processes. These markers, however, negatively correlate with soil pH, implying that lower pH levels could enhance microbial and enzyme activities, beneficial for basal respiration and microbial biomass.



```
(array([0.68909194, 0.2129486])
```

```
         PC1       PC2
0   5.117979 -1.700923
1   1.190113  2.424325
2  -0.666132  1.177648
3  -2.180160  0.018320
4  -3.461801 -1.919370
```

Figure 3. Principal component analysis (PCA) of the dataset showing the distribution of different vermicompost dosages and their impact on basil.

Table 6. Correlation matrix showing the relationships among various plant growth parameters, soil properties, and biochemical indicators for basil

	PH	LL	FW	DW	TPC	CO_2-C	MBC-C	DHA	pH	EC	OM	Total N	P	K
PH			☑	☑				☑						
LL			☑		✓			✓						
FW	☑	☑		☑				☑						
DW	☑		☑					☑						
TPC		✓				✓	✓			✓	☑	✓	☑	✓
CO_2-C					✓		☑	☑	⊗	☑	☑	☑	☑	☑
MBC-C					✓	☑		☑	⊗	☑	☑	☑	☑	☑
DHA	☑	✓	☑	☑		☑	☑		⊗	☑	☑	☑	☑	☑
pH						⊗	⊗	⊗		⊗	⊗	⊗	⊗	⊗
EC					✓	☑	☑	☑	⊗		☑	☑	☑	☑
OM					☑	☑	☑	☑	⊗	☑		☑	☑	☑
Total N					✓	☑	☑	☑	⊗	☑	☑		☑	☑
P					☑	☑	☑	☑	⊗	☑	☑	☑		☑
K					✓	☑	☑	☑	⊗	☑	☑	☑	☑	

✓= positive correlation ($P<0.05$), ☑= strong positive correlation ($P<0.01$), ⊗= strong negative correlation ($P<0.01$), *n*= 15, PH= plant height, LL= leaf length, FW= fresh weight, DW= dry weight.

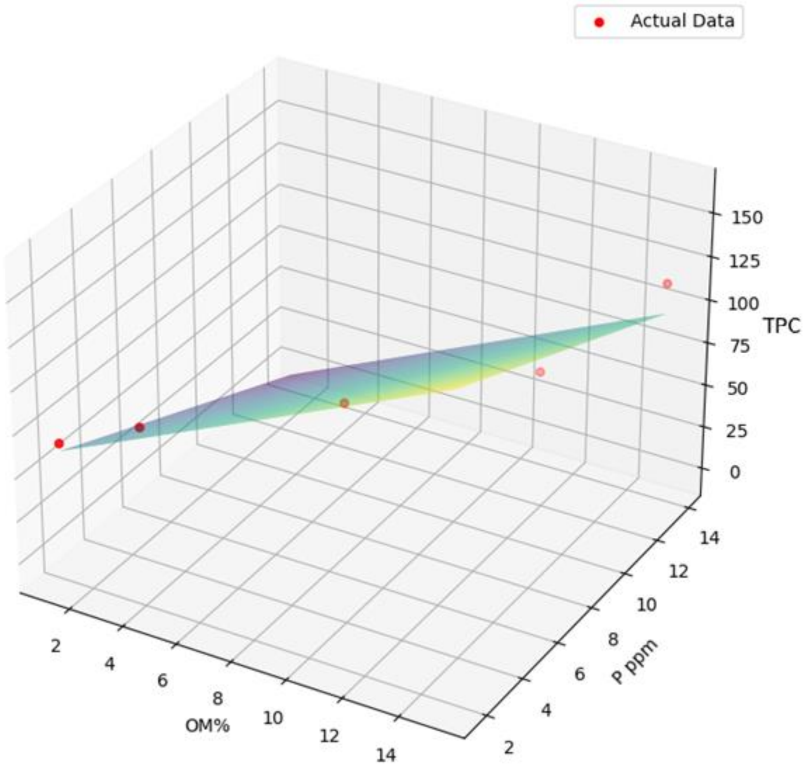


Figure 4. 3D plot of the regression model illustrating the relationship between soil organic matter, (OM) content (%), phosphorus (P) content (ppm), and total phenolic content (TPC) in basil plants.

The analysis also demonstrates strong negative correlations between soil pH and microbial activities, underscoring the critical role of pH in enhancing microbial efficiency and nutrient cycling. Optimally managing soil pH can significantly improve nutrient availability and uptake by plants. In addition, strong positive correlations between organic matter content and key soil nutrients like total N content and phosphorus (P) highlight the importance of organic matter as a nutrient reservoir, improving soil structure and fertility. The relationships between TPC and nutrients such as total N and phosphorus suggest that nutrient management influences phenolic content, important for plant health and disease resistance. Balanced fertilization strategies can enhance microbial activity, supporting overall soil health and plant growth.

Multiple regression analysis was also carried out to explore the impact of soil properties, specifically organic matter (OM) and phosphorus (P) contents, on the total phenolic content (TPC) of basil plants because of the result as a strong correlation according to Pearson correlation analysis. We utilized multiple regression analysis to model the relationship between these variables, providing a quantitative assessment of how changes in soil composition influence TPC. The model was constructed using data gathered from various vermicompost treatments applied to the soil, which were expected to modify the soil's OM (%) and P content (ppm) (Figure 4).

The derived multiple regression equation given in Equation 1:

$$y = 7.09356 \times OM - 5.53497 \times P + 63.99723 \qquad (1)$$

where $\hat{y}$ represents the predicted TPC. The model indicates that each unit increase in OM%, holding P constant, is associated with an increase of 7.09356 in TPC. Conversely, each unit increase in P, with OM% held constant, is associated with a decrease of 5.53497 in TPC. Our research results emphasize the need for careful management of P levels in soil to avoid inhibiting phenolic production, particularly in phosphorus-rich environments. This finding is consistent with the observations of Malusà et al. (2006), who reported that phosphate deficiency increased soluble phenolic content in bean roots, suggesting that high P levels have the opposite effect. Similarly, Asami et al. (2003) highlighted the importance of managing soil phosphorus to prevent a reduction in phenolic synthesis. Further supporting our findings, Phares et al. (2020) observed that higher P availability for plant uptake was associated with reduced total flavonoid content, which includes phenolic compounds. This suggests a disruption in phenolic synthesis due to elevated soil pH resulting from increased P levels. Zargoosh et al. (2019) also noted a correlation between soil P availability and total phenol content, indicating that increased soil phosphorus could play a key role in reducing phenol levels in specific environmental contexts. Additionally, Pang et al. (2022) found that phosphorus addition reduced lignin and vanillyl-type phenols, indicative of disrupted phenolic synthesis in plants. Zhang et al. (2023) further reported that high soil phosphorus inhibited the accumulation of polyphenols in tea plants, aligning with our conclusions about the negative impacts of excessive P on phenolic content in plants. This study also highlights the complex interactions influenced by agricultural practices and environmental conditions. High P levels have been consistently shown to correlate negatively with phenolic content in various plants, suggesting that phosphorus might suppress the synthesis of defense-related secondary metabolites.

4. Conclusion

Further research is essential to understand the intricate relationships between different soil nutrients and secondary metabolite production in plants. This includes exploring interactions between phosphorus and other micronutrients, as well as the effects of varying soil pH and microbial activity on phenolic synthesis. By leveraging these insights, agricultural practitioners can refine their fertilization strategies to improve crop yield and quality, focusing on the beneficial phenolic compounds that contribute to the health benefits of many plants and herbs like basil. This analysis underscores the complexity of plant-soil interactions and highlights the importance of understanding these relationships for optimizing agricultural inputs, improving crop quality, and enhancing environmental sustainability. Future studies will be pivotal in deepening our understanding of these dynamics, leading to more informed and sustainable agricultural practices.

Author Contributions

The percentage of the author(s) contributions is presented below. The author reviewed and approved the final version of the manuscript.

	F.Ş.H.T.
C	100
D	100
S	100
DCP	100
DAI	100
L	100
W	100
CR	100
SR	100
PM	100
FA	100

C=Concept, D= design, S= supervision, DCP= data collection and/or processing, DAI= data analysis and/or interpretation, L= literature search, W= writing, CR= critical review, SR= submission and revision, PM= project management, FA= funding acquisition.

Conflict of Interest

The author declared that there is no conflict of interest.

Ethical Consideration

Ethics committee approval was not required for this study because of there was no study on animals or humans.

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