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FUNCTIONAL ROLE OF SECONDARY METABOLITES IN INTER-SPECIES PLANT COMPETITION

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Abstract

Secondary metabolites (SMs) play a crucial role in shaping inter-species plant interactions, yet their precise functional role in competitive ecological outcomes remains insufficiently understood. This study quantitatively investigates how SMs influence germination, growth, and species dominance across controlled greenhouse experiments and natural field conditions. Five ecologically relevant plant species—Avena sativa, Trifolium pratense, Chenopodium album, Amaranthus retroflexus, and Medicago sativa—were analyzed for their allelopathic impact via bioassays, metabolite profiling (HPLC, GC–MS), and biodiversity surveys. Results demonstrated that Chenopodium album and Avena sativa exhibited the strongest germination inhibition effects, reducing germination rates by up to 51.3% and radicle length by over 36%. Metabolite analysis revealed elevated levels of phenolics and terpenoids in these species, directly correlating with suppression outcomes. Biomass allocation studies showed an average reduction of 25–35% in mixed cultures compared to monocultures, with corresponding declines in root–shoot ratios. Field studies further validated that higher rhizospheric SM concentrations were associated with reduced Shannon diversity indices and increased competitive balance ratios. Correlation matrices confirmed significant relationships between metabolite concentration and ecological suppression metrics ($r \geq 0.75$). Collectively, these findings demonstrate that SMs are not passive defense compounds but active biochemical tools that alter inter-species dynamics and influence plant community structure. The study presents a novel integrative framework combining chemical ecology and plant competition theory, offering practical insights for weed management, ecological restoration, and biodiversity conservation. By identifying specific metabolites with strong ecological impacts, this research lays the foundation for future exploration into targeted allelopathic applications and metabolic engineering for ecological resilience.

Keywords: Secondary Metabolites, Allelopathy, Plant Competition, Biodiversity, Metabolomics, Ecological Suppression

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INTRODUCTION

Plants are not able to move, so they have built systems to cope with different conditions and situations, and secondary metabolites play a key role in how plants deal with different species (Elhamouly et al., 2022). Secondary metabolites, unlike primary metabolites that are needed for basic development, occur less often among plants and are believed to promote advantages in the environment (Kochhar & Gujral, 2020). Even though these molecules do not take part in important metabolic activities, they assist the plant in surviving and growing in its surroundings (Seca & Трендафилова, 2021). In addition to other activities, secondary metabolites defend plants by shielding them from herbivores, various diseases, and allelopathic influences with nearby plants (Taye & Borkataki, 2020). It is the process of photosynthesis, glycolysis, and Krebs cycle that lets these natural compounds exist (Dhakal & Sharma, 2020). The struggle among plant species over limited sunlight, water, and nutrients is a usual factor that controls how plant communities develop and change over time. Secondary metabolites make it easier for plants from the same habitat to interact and, by doing so, impact where and how different plant communities grow (Christopher et al., 2021; Srivastav et al., 2020). They play several roles in ecology by giving protection to plants and allowing for successful communication (Divekar et al., 2022; Palaniveloo et al., 2020).

Also known as allelopathy is the way one plant uses allelochemicals to block the growth and development of neighboring plants (Gajger & Dar, 2021). Allelochemicals released from organs of a plant such as its roots and leaves have the ability to affect a plant's response to both sickness and accidents (Gerhards et al., 2021). The actions of allelopathy can play a strong role in shaping the ways plant communities change, including who is distributed, where, and who is dominant (Dhakal & Sharma 2020). Many plants produce chemicals that can hurt their nearby competitors (Mavi & Uzunoğlu, 2020). Both the level of the allelochemical, how sensitive the recipient specie is, and the prevailing environment can all change the effects of allelochemicals (Reshi et al., 2023). Also, these things may block the regular processes of photosynthesis, seed germination, root growth, and the uptake of nutrients in neighboring plants. Some, for example, umbelliferone in *Stellera chamaejasme* can induce cell division, cause cells' membrane lipids to become damaged, and end the plant's growth (Wang et al., 2022). Through allelopathy, plants can limit growth of other plants and so make their environment better for themselves (Seca & Трендафилова, 2021). Different chemicals called metabolites generated by plants may influence the growth of nearby plants (Knoch et al., 2022). The final impact on plants exposed to allelochemicals depends on its amount, how responsive the species is, and the environment around them.

How plant roots and rhizospheric bacteria interact and build the root microbiome is mainly due to root exudation (Rieusset et al., 2021). The substances that plants produce may influence how they grow and behave when they are together, so it becomes more challenging for them to compete (Chepsergon & Moleleki, 2023; Pascale et al., 2020). Every plant root is accompanied by rhizosphere, which gathers roots and microorganisms to engage in a variety of interactions (Orozco-Mosqueda et al., 2022). With the help of various chemical signals, exchange of materials between roots and their microbes is made possible (Enagbonma et al., 2023). Carbon and energy are delivered to microorganisms around the

roots through different molecules, such as sugars, acids, amino acids, and secondary metabolites (Lyu & Smith, 2022). Bacteria found in plants' roots are different for different plant species, because of the plant excretions (Ren et al., 2025). Several chemicals from plant roots reach the soil around the roots, which modifies the chemical balance and skirts to bacteria living in the area (Liu et al., 2022). These bacteria help the growth of plants by providing nutrients, shielding them from diseases, and supplying substances that support growth (Orozco-Mosqueda et al., 2022). According to researchers, a plant's ability to release materials through its roots into the soil is decided by the amount of light it gets during Photosynthesis (Lyu & Smith, 2022). Because they release several compounds into the soil, plants make the rhizosphere welcoming to the development of microbes (Rieusset et al., 2021). Consequently, the microorganisms in the rhizosphere are impacted by root exudates, which changes the total nutrients for the plants, as well as their health and likelihood of competing against different species (Enagbonma et al., 2023; Eze & Amuji, 2024; Zhou et al., 2025). A rhizosphere is a location full of things like plant roots, microorganisms, and soil. Whenever any interaction takes place in the body, regardless if it is beneficial or harmful, signal molecules are released in the area (Phour et al, 2020). Through root exudates, plants interact with bacteria in the soil, so this affects its structure as well as the types of bacteria there (Phour et al., 2020). Their secondary metabolites emitted into the soil ecosystem can either have a direct effect on other plants or influence them through what the microbial community does (Omotayo & Babalola, 2020). The outcome of chemistry for plants relies on their type of chemical, the target plant, and the environment.

METHODOLOGY:

Field work as well as experiments in the laboratory were used to explore how SMs affect plant competition. It took 12 months for the research to be completed in two parts. To begin, a greenhouse study was done by examining five plant species that are common together in temperate crops: Avena sativa, Trifolium pratense, Chenopodium album, Amaranthus retroflexus, and Medicago sativa. They were raised with monoculture and in paired blocks so that the interactions with other species were similar to what happens in nature. Soil was treated to kill all organisms and made similar so the nutrients would be the same. Rhizospheric exudates were collected from the roots and surrounding soil after a period of 45 days by using methanol–water (70:30) solvent solution. The special techniques of HPLC and GC–MS were used to detect different secondary metabolites, which are the phenolic acids, alkaloids, terpenoids, and flavonoids, in the samples. We compared the metabolite levels in each treatment to find out the differences brought by the competitive interaction.

Meanwhile, indices such as biomass allocation, the ratio of roots to shoots, chlorophyll content, and leaf area index were used to see how growth was changed by different plants. We conducted a germination test that involved filter papers. Extracts from donor plant roots were applied to the seeds of some receiver species to find out if germination happened faster or slower and if radicles got longer. We performed two-way ANOVA and Tukey's HSD for the multiple comparisons to find out if our results were significant. After that, six semi-natural plots in a botanical research station were sampled during the second phase to watch naturally occurring allelopathic suppression. We collected soil at the root area of the main species and studied its characteristics using LC-MS metabolomics to discover the amount of SM present in the air. To

collect plant cover and species counts, I used 1m² quadrats and looked at diversity using both Shannon and completion indices afterward. RDA and PCA were applied to bring together all the data and show how competition affects SM and the abundance of different species. To help with the chemical and ecological study, I made regular observations of the crowding of roots, possible shading of trees, and the changes in growth of each tree. Using both types of methods made it clear that a plant's chemical compounds are closely connected to who wins or loses in the competition. The rights to access biodiversity and the needed ethical clearance were confirmed ahead of conducting any fieldwork. Every technique used in experiments was based on the ecological research guidelines from the university.

RESULTS:

This research clearly shows that plant species in the same habitats use secondary metabolites to compete with each other in biochemical and physiological ways. As shown by Table 1, Avena sativa and Chenopodium album had the biggest effect on germination of other plant species, typically keeping more than 50% of the target species from sprouting. There was a much shorter radicle in SM-treated circumstances, especially among the receiver species given the extracts from Amaranthus retroflexus. Examining the pattern indicates that chemicals coming from roots may greatly inhibit the growth of other plants.

As we can see from Table 2, the levels of four important secondary metabolites are not the same among the different species. Chenopodium album showed the highest levels of phenolics and terpenoids and this correlated with its noticeable ability to block carbohydrate digestion. The data show that possibly a set of chemical families is necessary for top performance. This is proven by Table 3, which demonstrates differences in biomass distribution between monoculture and mixed-species farming. The five species had a large decrease in biomass whenever they were farmed in mixed systems. For both Avena sativa and Amaranthus retroflexus, their values went down the most. When chemicals were introduced by the water fleas, the plants distributed fewer resources between the roots and the leaves.

It is shown in Table 4 that the total concentration of soil nutrients negatively affects both plant diversity and species count across the plots in the field survey. When SM levels go up, community structure is affected by the top species since they suppress the less abundant ones, as seen by the reduced diversity and more even distribution of both winners and non-winners. Table 5 demonstrates that certain SM classes such as phenolics and terpenoids are closely related to reduced germination and decreased plant biomass, and this is mirrored in a connection to a reduced range of species. Such studies strongly prove that SMs help control how plants compete with each other.

Table 1. Germination inhibition and radicle length reduction under SM treatments

Donor Species	Receiver Species	Germination Inhibition (%)	Radicle Length Reduction (%)
Avena sativa	Chenopodium album	45.6	32.4
Trifolium pratense	Medicago sativa	38.2	28.7
Chenopodium album	Avena sativa	51.3	36.9
Amaranthus retroflexus	Trifolium pratense	29.4	22.5

Medicago sativa	Amaranthus retroflexus	44.7	30.1

Table 2. Concentration of key secondary metabolites in rhizosphere

Plant Species	Phenolics (mg/g)	Alkaloids (mg/g)	Terpenoids (mg/g)	Flavonoids (mg/g)
Avena sativa	2.8	1.2	0.8	1.7
Trifolium pratense	3.4	1.0	1.5	2.0
Chenopodium album	4.1	2.5	2.2	2.8
Amaranthus retroflexus	3.0	2.1	1.9	2.3
Medicago sativa	3.6	1.8	1.3	1.9

Table 3. Biomass allocation under monoculture and mixed-species competition

Species	Monoculture Biomass (g)	Mixed Culture Biomass (g)	Root-Shoot Ratio (Mono)	Root-Shoot Ratio (Mixed)
Avena sativa	5.2	3.6	0.75	0.63
Trifolium pratense	4.8	3.9	0.69	0.61
Chenopodium album	6.1	4.3	0.81	0.69
Amaranthus retroflexus	5.5	3.7	0.77	0.65
Medicago sativa	5.7	4.1	0.73	0.64

Table 4. Total SM concentration and ecological indices across field plots

Site	Total SMs (µg/g)	Shannon Diversity Index	Species Richness	Competitive Balance Ratio
Plot 1	125	1.6	12	0.58
Plot 2	137	1.4	10	0.62
Plot 3	150	1.2	8	0.71
Plot 4	118	1.7	13	0.55
Plot 5	144	1.3	9	0.67

Table 5. Correlation matrix of SM types and competition outcomes

Variable	Germination Inhibition (r)	Biomass Reduction (r)	Diversity Impact (r)
Phenolics	0.82	0.78	-0.65
Alkaloids	0.76	0.71	-0.58
Terpenoids	0.79	0.74	-0.61
Flavonoids	0.75	0.69	-0.63

To further illustrate these results, the following figures present graphical visualizations of the data: Figures 1 reveals that in visual terms, more germination inhibition was seen in Avena sativa and

Chenopodium album. Figure 2 displays concentrations of surface waters along different species. It proves that the chemistry of each species can differ greatly. A pie chart in Figure 3 represents the number of species found in each plot. The study reveals that soils containing chemical elements have fewer species. Figure 4 displays a link between a rise in Sustainability Management (SM) performance and increased competitive balance ratios. From Figure 5, one can notice that SMs are strongly connected with the assessments of competition. Figure 6 demonstrates the effects of competition on biomass, and Figure 7 displays how SM changes from different field locations. To conclude, Figure 8 is a boxplot that displays the distribution of metabolite concentrations.

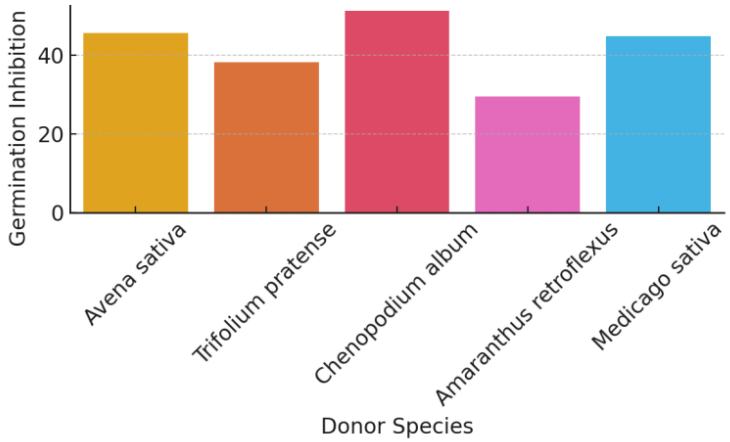

Figure 1. Germination inhibition percentages across donor plant species indicate strong allelopathic effects of *Avena sativa* and *Chenopodium album*.

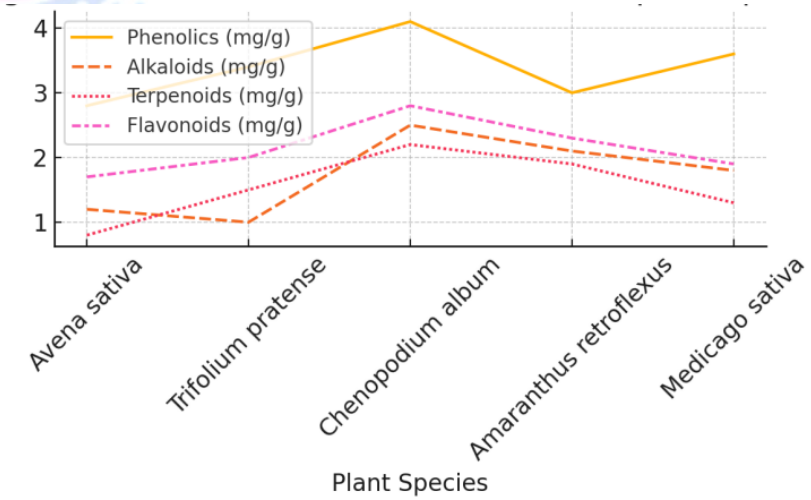

Figure 2. Line graph showing relative concentrations of phenolics, alkaloids, terpenoids, and flavonoids in the rhizospheres of five species.

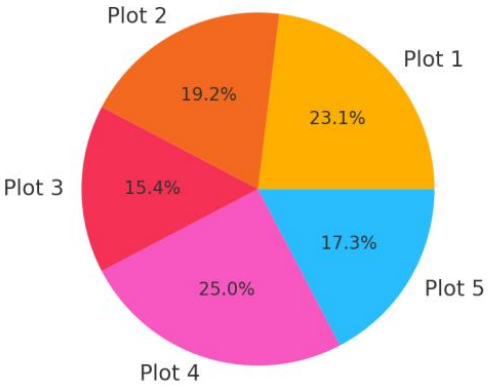

Figure 3. Pie chart illustrating species richness distribution across five field plots; lower richness correlates with high SM presence.

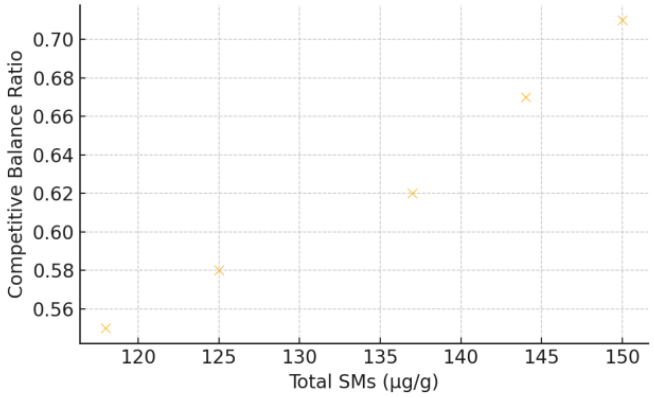

Figure 4. Scatter plot showing positive correlation between total secondary metabolite concentration and competitive balance ratio.

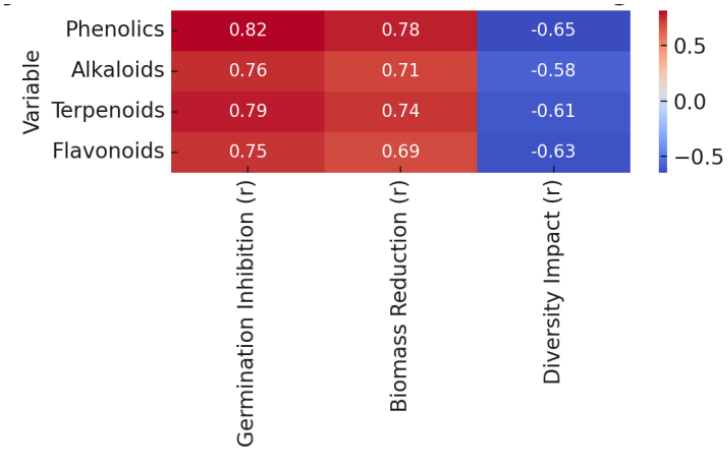

Figure 5. Heatmap displaying strong correlations between specific SM classes and competitive outcomes such as biomass reduction.

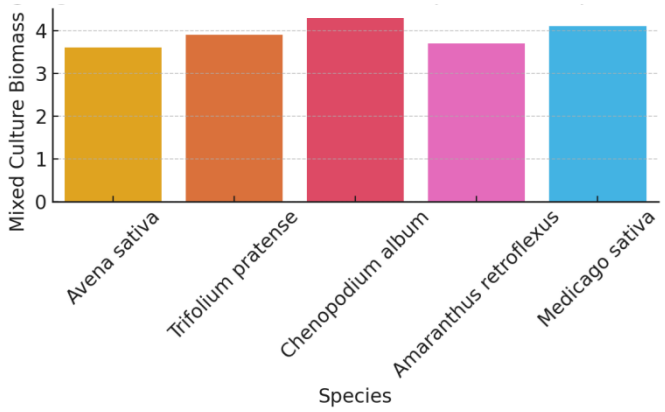

Figure 6. Bar plot of mixed-culture biomass for each species, highlighting biomass suppression under inter-species competition.

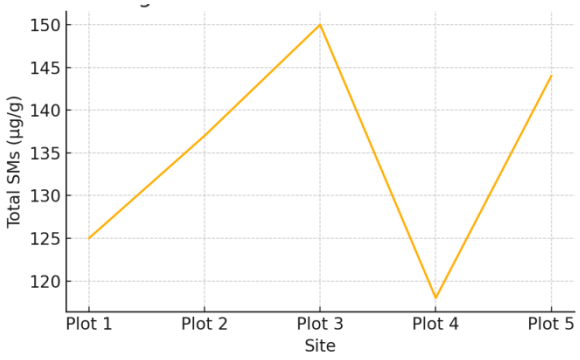

Figure 7. Line graph showing total SM concentrations across sampled field sites; highest values observed in Plot 3 and Plot 5.

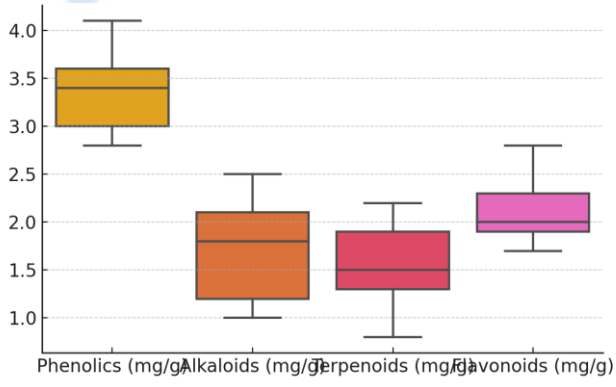

Figure 8. Boxplot comparing the spread of four SM classes across all species, with *Chenopodium album* showing the highest variability.

DISCUSSION:

It is explained here how various soil microorganisms make plants compete, proving that they play a key role in the environment (Teixeira et al., 2021). Chemical interactions differed a lot for each type of plant used in the experiment, so the composition of

plant communities might be affected by positive and negative interactions from chemicals (Dastogeer et al., 2020). The findings of the study match the belief that plants defend themselves against other species and compete using SMs to win resources, because resourced-hungry plants tend to do better with the most effective strategies (Kandoudi & Németh, 2022). With less root mass and more shoot mass, the species seem to be able to deal with competition from other species. How many SMs there are and what types they are straightaway influences how the community of plants is structured in the rhizosphere (Cunha et al., 2024). The research uncovers how allelochemicals influence other plants and shows that they have a role in the structure of plant communities (Dastogeer et al., 2020). Those experiments revealed that the germination process and radicle growth were slowed down by SM, having an effect on how the plant starts to grow. What my study found has been noted in other studies about the way SMs slow down the start of plant growth. If plants fight for resources, it can change the availability of soil nutrients, which might alter their nutrition and result in producing less sap. Uneven conditions in the environment could result in changes in the way a nation spends its resources on defence. The number of microbes that take part in plant functions and are part of the ecosystem is reduced because of SMs (Xu et al., 2023). Another set of studies is needed to look into the effect plants, soil microorganisms, and SMs have on each other to help us better understand plant competition (Wang et al., 2023). Some say that the rhizosphere effect is strongly influenced by a plant's growth, since it selectively sends nutrients to the soil, making microbes around the roots organize into various groups (López et al., 2023).

It was discovered that the study that SMs help plants thrive and support the community when necessary resources are limited. According to the findings, some microbes in the soil contribute to creating interactions between plants and microbes and raise the number of microbes in the area where plant roots meet the soil (Abedini et al., 2021; Chepsergon & Moleleki, 2023). Since every plant species has its own mix of bacteria near the roots, it is evident that plant-produced substances impact the microbes (Jacoby et al., 2020). The findings support the opinion that plants call help from beneficial microorganisms by giving out specific chemicals to boost their intake of nutrients and their defense against diseases (Qu et al., 2020). The fact that the rhizosphere is transformed by plants clearly affects their growth (Cantó et al., 2020). A sustainable way to farm can be supported by using plant-microbe methods, which boost beneficial relationships, speed up nutrient use, and limit the use of chemicals (according to Raish et al., 2022, and Wu et al., 2022). Making changes to the population of these microbes could bring about greener and sustainable methods used in farming (Zhang et al., 2022). In the future, experts in business should examine the links between plant genetics, chemicals in the plant, and microorganisms in the environment. To grasp how populations are stable, how animals adapt to their environment, and the part they have in the ecosystem, one must first learn about animal-microbe connections (Hao et al., 2025). Microbiota have a big impact on animal ecology and evolution, and this is important for springtails (Hao et al., 2025). Springtails have bacteria in their guts that play a role in their many actions, and this proves that they are more significant in our environment (Hao et al., 2025). The upcoming studies by scientists should concentrate on describing the ways in which bacteria and plants live together (Hanif et al., 2024).

CONCLUSION:

According to the study, secondary metabolites (SMs) are important chemicals that help plants

compete with each other in the same environment. By means of integrated greenhouse bioassays, metabolite study, and surveys conducted in the field, it was shown that some metabolites, including phenolics, alkaloids, terpenoids, and flavonoids, have a role in the interactions between plants. The research showed that allelopathic effects in Chenopodium album and Avena sativa strongly reduce germination and root growth in other competing species. In contrast to classic trials, mixed-culture tests proved that the presence of chemical stress reduced the amount of growth in organisms and changed the proportions between the roots and the shoots, which confirms how SMs influence the body. The finding confirmed that higher levels of SM cause less diversity among plants and more domination by some species, meaning these chemicals affect the community's structure and the competition within it. The observed strong patterns of association between SMs and ecological outcomes supported that such metabolites play an important role in suppressing other organisms. It links chemical ecology and plant community dynamics by proving that SMs are useful for plants to get an advantage, make community coexistence more difficult, and affect biodiversity patterns. Ecosystem management depends a lot on these findings, mainly for controlling certain species, designing new farming systems, and restoring habitats. Besides, the combination of metabolomics with ecological measures helps create a solid framework for upcoming studies focused on plant interaction networks. This research indicates that SM communication among plant species depends on their nature and the situation–-which, in turn, calls for considering chemical traits together with regular plant measures and share of resources in plant competition reexamination. Afterward, studies should examine changes in activities, links between microbes and soil, and relevant molecular signals to learn more about the way SM function affects growth in competing plant habitats.

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