

Efficacy of Aloe and Opuntia based composts on growth and phytochemical properties of *Salvia officinalis* and *Rosmarinus officinalis*



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Citation:

PRISA D., JAMAL A., 2025 - *Efficacy of Aloe and Opuntia based composts on growth and phytochemical properties of Salvia officinalis and Rosmarinus officinalis*. - Adv. Hort. Sci., 40(1): 29-46.

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Data Availability Statement:

All relevant data are within the paper and its Supporting Information files.

Competing Interests:

The authors declare no conflict of interests.

Received for publication 8 September 2025

Accepted for publication 2 December 2025

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Key words: Biostimulants, essential oil composition, organic cultivation, plant secondary metabolites, sustainable agriculture.

Abstract: The sustainable cultivation of medicinal and aromatic plants requires alternative substrates that reduce dependence on peat and synthetic fertilisers, while promoting acceptable plant growth and adequate phytochemical quality. This study evaluated the effects of five compost treatments on *Salvia officinalis* and *Rosmarinus officinalis* grown in greenhouses at CREA (Pescia, Italy). The treatments examined in this study include use of a peat-based control, commercial compost, monospecific composts derived from *Aloe arborescens*, *Aloe barbadensis* and *Opuntia ficus-indica*, plus a mixed Aloe-Opuntia compost. Growth parameters, photosynthetic performance, chlorophyll content, water and resource use efficiency, microbial biomass and secondary metabolites (phenols, flavonoids, essential oils) for both species were evaluated. For both species tested, the peat-based control supported consistently the best vegetative growth, highest phytochemical levels and was confirmed to be the best for physical support in greenhouse conditions. The mixed Aloe-Opuntia compost functioned similarly to the peat-based control, yet it showed a 5-8% reduction in vegetative growth, however, it still had a higher physiological/metabolic level than other compost based treatments. The results indicate that it is possible to use composts containing a combination of materials with complementary structural and biochemical properties to help overcome some of the limitations of single-species composts. On the other hand, composts made from *Aloe vera* alone exhibited the weakest agronomic and physiological performances likely due to having a higher electrical conductivity, high rapid decomposition rates, and having a poorly developed substrate structure even though it had a high total nutrient content. There was no difference in the safety of all composts; they caused no negative effects on plant survival, pest infestation, or disease occurrence. Such promising results were recorded particularly for mixed composts, but due to the fact that this was a greenhouse trial, there is a need for further replication to confirm findings. In conclusion, mixed plant composts could be a more sustainable solution, environmentally friendly for producing aromatic plants, and will reduce the reliance on the limited supply of peat.

1. Introduction

There has been growing interest in the development of sustainable growing conditions for plants that have been requested for use as sources of medicinal and aromatic oils due to the worldwide increase in demand for these products from both the food and beverage industries as well as from pharmaceutical and cosmetics companies (Lange, 2002; Pirani *et al.*, 2020; Chrysargyris *et al.*, 2022). Due to their high bioactive monoterpenes (1,8-cineole, camphor and α -pinene) content, *Salvia officinalis* and *Rosmarinus officinalis* are of great importance due to their use as essential oils in many applications and as important parts of many herbal preparations (El Euch *et al.*, 2019; Leporini *et al.*, 2020). These monoterpene substances have many health benefits, including antimicrobial, antioxidant and anti-inflammatory actions, and therefore are considered to be important components in the development of therapeutic products as well as in the preservation of natural food (Angioni *et al.*, 2004; Miguel, 2010; Shahina *et al.*, 2022). Cultivation practices that optimise both the quantity and quality of plants are crucial for sustaining the production and sales of MAPs such as *Salvia officinalis* and *Rosmarinus officinalis*, particularly when grown in controlled environments, such as greenhouses (Avasiloaiei *et al.*, 2025).

The advantages of using greenhouses as a method for growing crops are that they give producers a greater amount of control over their environment; however, the challenges of using alternative methods for managing substrates and providing nutrients to crops, as well as managing pests are many (Bu *et al.*, 2022; Chen *et al.*, 2025). In addition, there has been considerable debate about the negative effects on both people's health and the environment related to the increasing number of inputs into agricultural production, including pesticides and synthetic fertilizers (Dhankhar and Kumar, 2023; Jamal *et al.*, 2023). Due to this trend, many growers are beginning to make a shift away from the conventional style of agricultural production, which utilises an influx of synthetic materials, to a biodynamic approach to agricultural production (Brzozowski and Mazourek, 2018). Additionally, plant-based composts from renewable biomass are becoming increasingly common, as they provide both essential nutrients and bioactive compounds for promoting growth and increased resilience in plants (Salman *et al.*, 2023;

Ahmed *et al.*, 2025). Composts formulated from *Aloe barbadensis* (aloe vera), *Aloe arborescens*, and *Opuntia ficus-indica* (prickly pear cactus) have recently attracted attention as bio enhancing soil amendments (Semerel *et al.*, 2023; Bacchetta *et al.*, 2024). These species are well-known for their medicinal properties, which are derived from their diverse chemical profiles that are rich in polysaccharides, phenolic compounds, organic acids, and glycoproteins (Di Palma *et al.*, 2025). When incorporated into compost, these compounds may act as biostimulants, enhancing plant growth, improving tolerance to abiotic stress, and stimulating beneficial soil microbiota (Prisa and Gobbino, 2021).

Aloe species have been extensively studied for their antimicrobial, antifungal, and wound-healing effects in humans, and emerging evidence suggests that they may exert similar protective effects in soil-plant systems (Arsene *et al.*, 2022). *Aloe barbadensis*, in particular, contains aloin, emodin, and acemannan compounds that can stimulate plant defense pathways and promote systemic resistance to pathogens (Ahmad *et al.*, 2018). *Aloe arborescens*, though less commercially prominent, has shown superior antioxidant activity due to its higher phenolic content and greater diversity of bioactive metabolites (Maliehe *et al.*, 2023). Likewise, *Opuntia ficus-indica*, a drought-tolerant cactus species, has shown potential as a compost base due to its high mucilage content, nutrient accumulation capacity, and ability to enhance soil water retention and microbial activity (Procacci *et al.*, 2021).

Although plant-based composts are increasingly applied in open-field systems, their effects under greenhouse conditions remain underexplored, particularly for high-value aromatic crops (Oued Lhaj *et al.*, 2025). The confined root environment and controlled climate of greenhouses provide an ideal platform to test targeted compost formulations; however, their influence on growth performance, pest suppression, and secondary metabolite biosynthesis is not well understood (Zheng *et al.*, 2020). Furthermore, the bioactive properties of *Salvia officinalis* and *Rosmarinus officinalis* essential oils are strongly influenced by environmental and nutritional factors, including substrate composition (Rapposelli *et al.*, 2015). Previous studies on these two species have shown that organic amendments can modulate terpene biosynthesis pathways by altering both oil yield and chemical composition (Valiki and Ghanbari, 2015). Similarly, composts

enriched with phenolics or bioavailable micronutrients can enhance plant immunity and secondary metabolite accumulation in *S. officinalis* and *R. officinalis* (Naguib *et al.*, 2012; Montoya *et al.*, 2022).

However, most commercial composts used in organic horticulture are formulated from generic plant waste and lack the targeted bioactivity of the medicinal species. Composts made specifically from Aloe and Opuntia may provide added value by combining nutritional enrichment with biological protection (Procacci *et al.*, 2021). Such dual actions could be especially advantageous in greenhouse cultivation, where disease pressure and nutrient competition are intensified (Di Palma *et al.*, 2025). Integrating plant-based composts into MAP production also aligns with the principles of circular agriculture and zero-waste systems, particularly in the Mediterranean regions where Aloe and Opuntia are abundant and adapted to marginal soils (Zheng *et al.*, 2020).

Despite their potential, scientific data comparing Aloe and Opuntia based composts with commercial products under greenhouse conditions are scarce. Few studies have simultaneously evaluated the effects of compost on plant growth, pest incidence, and essential oil yield, and even fewer have examined its impact on the chemical composition of essential oils, which is a critical determinant of both market value and biological efficacy (Di Palma *et al.*, 2025).

Therefore, this study aimed to evaluate the agronomic performance, physiological responses, and secondary metabolite profiles of *Salvia officinalis* and *Rosmarinus officinalis* grown in greenhouse pot culture using composts derived from *Aloe barbadensis*, *Aloe arborescens*, and *Opuntia ficus-indica*, compared with commercial compost and an unfertilized control. It was hypothesized that plant-based composts formulated from bioactive species would enhance plant growth, physiological efficiency, and essential oil yield compared to the control and commercial composts, with mixed-species composts exhibiting superior effects over single-species formulations.

2. Materials and Methods

Experimental site and greenhouse conditions

The experiment was conducted in a controlled-

environment greenhouse at the Council for Agricultural Research and Economics - Research Centre for Vegetable and Ornamental Crops (CREA-OF), Pescia, Tuscany, Italy (43.8912° N, 10.6856° E; 35 m asl) from January to December 2024. The greenhouse was equipped with an automated climate control system regulating temperature, humidity, and ventilation. Daytime temperatures were maintained between 24-28°C and nighttime temperatures between 16-18°C. Relative humidity ranged from 60-75%. Supplementary LED lighting provided approximately 250 $\mu\text{mol m}^{-2} \text{s}^{-1}$ of photosynthetically active radiation (PAR) for 12 h day^{-1} , which lies within the optimal range reported for *Salvia officinalis* and *Rosmarinus officinalis* (200-600 $\mu\text{mol m}^{-2} \text{s}^{-1}$) to sustain healthy growth and essential oil production (Hussain *et al.*, 2010; Berkovich *et al.*, 2017; Papafotiou *et al.*, 2022).

Plant material and experimental design

Seedlings of *Salvia officinalis* L. and *Rosmarinus officinalis* L., propagated under identical nursery conditions, were transplanted at 60 days of age into 6 L plastic pots containing treatment-specific substrate mixtures. Each pot contained a single uniform seedling. The experimental design followed a completely randomized block (CRB) structure with six treatments with four replicates, with six plants per replicate and 24 plants per treatment. With a total of 144 plants per species. The six treatments were:

- T0 (Control): Peat-based substrate without compost (baseline).
- T1 (Commercial): Peat + 30% commercial green compost.
- T2 (*Aloe barbadensis*): Peat + 30% compost derived from *Aloe barbadensis*.
- T3 (*Aloe arborescens*): Peat + 30% compost derived from *Aloe arborescens*.
- T4 (Opuntia): Peat + 30% compost derived from *Opuntia ficus-indica*.
- T5 (Mixed): Peat + 10% compost from each of the three experimental composts (*Aloe barbadensis*, *Aloe arborescens*, *Opuntia ficus-indica*).

The experimental composts were produced through aerobic decomposition for 90 days using mature biomass from organically grown populations of *Aloe barbadensis*, *Aloe arborescens* and *Opuntia ficus-indica*. For the *Aloe* species, the compost

feedstock consisted mainly of senescent and pruned leaves, which are the main by-products generated during routine maintenance, while roots and stems were not included. For *Opuntia ficus-indica*, the composting material included pruned cladodes, collected after thinning and sanitary pruning; fruits, roots, and woody stems were excluded. All plant residues were shredded into 2-4 cm pieces prior to composting to ensure uniform decomposition. Prior to use, composts were characterized for pH, electrical conductivity, total nitrogen, organic matter content, and C:N ratio (Table 1). All plants were maintained under uniform irrigation, keeping the substrates at field capacity throughout the experimental period. No synthetic fertilizers or pesticides were applied during the trial.

Growth and morphological parameters

At 120 days after transplantation, various morphological traits were recorded for each plant. Growth and morphological parameters were monitored throughout the experiment. Plant height and leaf number were recorded biweekly, while final destructive measurements (leaf area, biomass, and root traits) were collected at 120 days after transplantation. Biweekly measurements allowed evaluation of growth trends over time, while final harvest data provided detailed information on biomass allocation and root development.

Plant height (cm) was measured from the substrate surface to the highest vegetative point using a flexible measuring tape, while the total

number of leaves was counted manually for each plant. Leaf area (cm²) was determined by collecting five fully expanded leaves per plant, scanning them, and analyzing the images using ImageJ software to calculate the mean surface area. For biomass assessment, plants were harvested, and roots were gently washed; the fresh biomass of both aboveground and belowground parts were recorded, followed by oven-drying at 65°C for 72 h to determine dry weight. Root length (cm) was measured as the longest root per plant using a digital caliper. Additionally, plant mortality (%) was recorded by counting the number of dead plants during the experimental period and calculating the mortality rate as a percentage of the total plants per treatment.

Photosynthetic and water-relations measurements

Photosynthetic parameters were measured biweekly, on clear days between 09:00 and 11:30 AM using a portable photosynthesis system (LI-6400XT, LI-COR Biosciences, Lincoln, NE, USA). Measurements were taken from the third fully expanded leaf from the shoot apex, avoiding shaded or damaged leaves. The recorded parameters included net photosynthetic rate (Pn, $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), stomatal conductance (Gs, $\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$), and transpiration rate (E, $\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$). Water use efficiency (WUE) was calculated as the ratio of photosynthesis to transpiration (Pn/E), indicating carbon gain per unit of water lost (Liu *et al.*, 2020; Mingyang *et al.*, 2022). All measurements were performed on three

Table 1 - Chemical and physical properties of composts used in the experiment

Parameter	Control (T0)	Commercial Compost (T1)	<i>Aloe barbadensis</i> (T2)	<i>Aloe arborescens</i> (T3)	<i>Opuntia ficus-indica</i> (T4)	Mixed compost (T5)
pH (H ₂ O 1:5)	6.4	6.8	6.6	6.9	6.7	6.7
Electrical conductivity (dS m ⁻¹)	0.9	1.6	1.8	1.7	1.7	1.7
Organic matter (%)	28.7	48.6	50.1	45.7	48.1	48.1
Total nitrogen (N, %)	0.7	1.5	1.4	1.3	1.4	1.4
C/N ratio	20.5	16.4	15.9	17.1	16.5	16.5
Available phosphorus (P ₂ O ₅ , mg kg ⁻¹)	510	910	950	890	920	920
Exchangeable potassium (K ₂ O, mg kg ⁻¹)	620	1120	1170	1090	1130	1130
Moisture content (%)	29.6	35.2	34.6	36.5	35.4	35.4
Bulk density (g cm ⁻³)	0.61	0.49	0.47	0.51	0.49	0.49
Microbial biomass (MBC, mg kg ⁻¹)	165	385	412	376	402	402

Values are means of three replicates. Composts were air-dried, sieved (≤ 10 mm), and analyzed according to standard soil and compost analysis protocols (EN 13039:2000 and ISO 14240-1 for MBC). Mixed compost (T5) was prepared by blending equal proportions (1:1:1) of T2, T3, and T4 by volume.

randomly selected plants per replicate, resulting in a total of 12 plants per treatment per species.

Chlorophyll content estimation

Relative chlorophyll content was measured biweekly using a SPAD 502 chlorophyll meter (Konica Minolta, Japan). Three SPAD readings were collected per plant from the same leaves used for gas-exchange measurements and averaged to obtain a representative chlorophyll index (Ling *et al.*, 2011; Yuan *et al.*, 2016).

Microbial biomass in substrate

At the end of the experiment, microbial biomass carbon (MBC) in the substrate was assessed using the chloroform fumigation-extraction method. Composite samples from each pot were extracted with 0.5 M K₂SO₄, and MBC was quantified via dichromate oxidation followed by spectrophotometric analysis. Data were expressed as mg MBC kg⁻¹ dry substrate (Vance *et al.*, 1987).

Leaf metabolite analysis

Leaf samples (~2 g fresh weight) were collected from each treatment, immediately flash-frozen in liquid nitrogen, and stored at -80°C until analysis. The samples were lyophilized and ground into a fine powder before extraction. Total phenolic content (TPC) was determined using the Folin-Ciocalteu method and expressed as mg gallic acid equivalents (GAE) g⁻¹ DW, while total flavonoid content (TFC) was quantified via the aluminum chloride colorimetric assay and expressed as mg quercetin equivalents (QE) g⁻¹ DW (Albayrak, 2013). Antioxidant capacity was evaluated using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay and expressed as IC₅₀ (µg mL⁻¹). For essential oil analysis, dried leaves were subjected to hydro distillation using a Clevenger apparatus, and the resulting oil was analyzed by GC-MS, with compounds such as 1,8-cineole, camphor, and α-pinene identified based on retention indices and comparison with NIST library spectra (Farhadi *et al.*, 2020).

Fertilization regime

To ensure a uniform nutrient baseline and prevent nutrient deficiency as a confounding factor, all treatments received a standardized organic fertilization schedule. A liquid organic fertilizer (NPK 5-2-5), derived from plant extracts and permitted under European organic farming regulations (Reg. EC

834/2007), was applied biweekly. Each plant received 50 mL of a diluted solution (1:100 v/v) to the root zone, starting 14 days after transplanting and continuing until day 90.

Micronutrient supplementation (Fe, Zn, Mn) was performed once at day 40 using an organic-certified foliar product based on seaweed extract and chelated trace elements. Applications were carried out in the early morning to minimize evapotranspiration and photodegradation.

Biological pest and disease control

An integrated pest management (IPM) strategy was implemented throughout the cultivation period, combining preventive monitoring and biological control. Weekly visual scouting and yellow sticky traps were used to monitor aphid (*Myzus persicae*) and whitefly (*Trialeurodes vaporariorum*) populations. When pest thresholds exceeded two adults per plant, biological interventions were initiated. *Beauveria bassiana* (strain ATCC 74040) was sprayed on foliage at 10-day intervals (1 × 10⁷ CFU mL⁻¹) during peak insect activity, while *Trichoderma harzianum* spores (10⁸ CFU g⁻¹) were applied to the root zone at 30 and 60 days after transplanting to prevent fungal diseases such as *Botrytis cinerea* and *Rhizoctonia solani*. All biocontrol products used were certified for organic cultivation and applied according to manufacturer guidelines, with no synthetic pesticides or systemic chemicals used during the experiment.

Statistical analysis

All quantitative data were first tested for normality using the Shapiro-Wilk test and for homogeneity of variances using Levene's test. When assumptions were met, differences among treatments were analyzed using one-way ANOVA, followed by Tukey's HSD post hoc test (α = 0.05) for mean separation. In cases where data did not meet parametric assumptions, the Kruskal-Wallis test was applied as a non-parametric alternative. Principal component analysis (PCA) and correlation plots were drawn to investigate the relationships among the studied variables using OriginPro Version 2024 (OriginLab Corporation, USA; www.originlab.com) software. Prior to PCA, the data were standardized using z-score normalization (mean-centered and scaled to unit variance) by enabling the "Standardize Variables" option in the PCA tool to ensure the equal contribution of all traits. PCA was performed using a

correlation matrix, and no rotation method was applied. Pearson’s correlation coefficients were calculated to construct correlation heatmaps, which were visualized with color-coded ellipses indicating the strength and direction of the relationships among variables.

Quality assurance and replication

All measurements were performed in biological triplicates or quadruplicates as appropriate. Analytical assays were conducted in duplicate, and instruments were calibrated regularly. Substrate samples and plant tissues were handled with sterile equipment to avoid cross-contamination.

3. Results

Salvia officinalis

Agronomic traits

Compost treatments significantly influenced the agronomic performance of *Salvia officinalis* (Table 2 and Fig. 1). The control substrate (T0) supported the most vigorous growth, producing the tallest plants (53.4 ± 1.2 cm), the greatest number of leaves (140.0 ± 1.0), the largest leaf area (580 ± 10.3 cm²), and the highest shoot biomass (43.0 ± 1.1 g) (Fig. 1).

The commercial compost (T1) performed comparably to the control, with non-significant reductions in plant height (-2.8%), leaf number (-1.8%), leaf area (-2.6%), and shoot biomass (-3.5%), indicating its suitability as an alternative growth medium. Similarly, the mixed Aloe and Opuntia compost (T5) produced only slight decreases in plant height (-5.6%), leaf number (-3.6%), and shoot biomass (-7.0%) relative to T0, suggesting its potential as an effective organic amendment (Fig. 1).

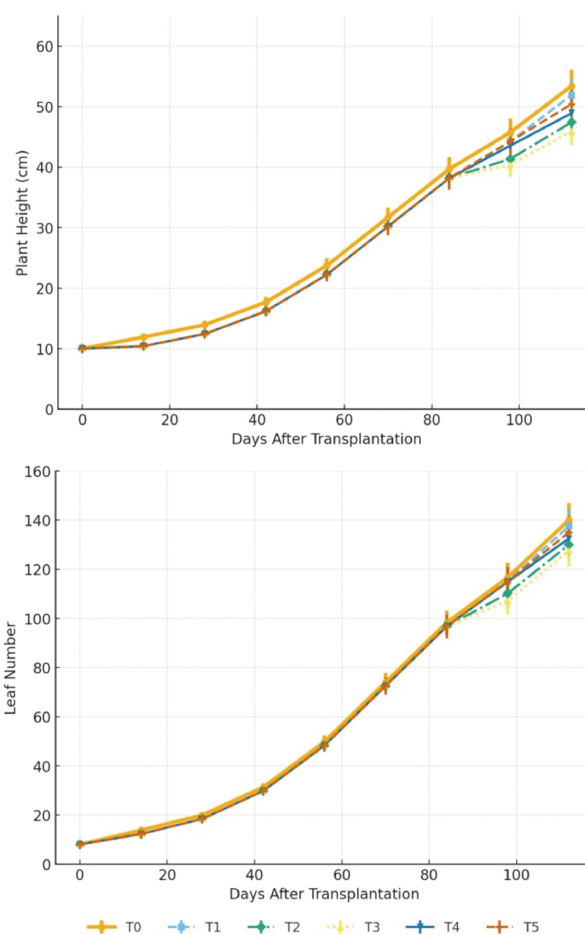


Fig. 1 - Biweekly growth (plant height and leaves number) dynamics of *Salvia officinalis* under different compost treatments.

In contrast, the Opuntia compost alone (T4) resulted in moderate reductions in plant height (-8.4%), leaf number (-5.4%), and shoot biomass (-10.7%) compared with T0. The Aloe-only treatments (T2 and T3) consistently underperformed. T2 plants exhibited an 11.2% reduction in height,

Table 2 - Agronomic parameters of *Salvia officinalis* under different compost treatments

Treatment	Plant height (cm)	Leaf number	Leaf area (cm ²)	Shoot biomass (g)	Root biomass (g)	Root length cm
T0	53.4 ± 1.2 a	140.0 ± 1.0 a	580 ± 10.3 a	43.0 ± 1.1 a	14.2±1.8 a	21.3±1.8 a
T1	51.9 ± 1.3 ab	137.5 ± 1.3 ab	565 ± 10.6 ab	41.5 ± 0.9 ab	13.5±1.6 bc	20.2±1.8 b
T2	47.4 ± 0.8 c	130.0 ± 1.2 c	520 ± 13.8 c	37.0 ± 0.9 c	11.8±1.3 e	18.3±1.6 d
T3	45.9 ± 1.1 c	127.5 ± 1.5 c	505 ± 12.1 c	35.5 ± 1.0 c	12.5±1.7 d	18.9±1.7 cd
T4	48.9 ± 1.1 bc	132.5 ± 1.1 bc	535 ± 10.5 bc	38.5 ± 0.9 bc	13.0±1.6 cd	19.4±1.9 c
T5	50.4 ± 1.0 b	135.0 ± 1.2 b	550 ± 11.7 b	40.0 ± 1.1 b	14.1±1.4 ab	20.2±1.5 b

Values are mean ± SD with statistical groupings indicated by letters (Tukey’s test, p < 0.05). Treatments: T0= Control; T1= Commercial; T2= *Aloe barbadensis*; T3= *Aloe arborescens*; T4= Opuntia; T5= Mixed.

7.1% fewer leaves, and 14.0% lower shoot biomass than the control, while T3 showed even greater reductions (14.1% shorter height, 8.9% fewer leaves, and 17.4% lower shoot biomass, 12.0% reduced root biomass, and 11.3% shorter roots). Although the Aloe-based composts (T2 and T3) contained higher levels of organic matter, available phosphorus, exchangeable potassium, and microbial biomass compared with the control (Table 1), they consistently resulted in the lowest plant height, leaf area, shoot biomass, and root biomass (Table 2). Both Aloe treatments exhibited significantly reduced vegetative growth compared with the control and with the commercial (T1), Opuntia (T4), and mixed compost treatments (T5) (Fig. 2). Despite the elevated nutrient and organic matter content of T2 and T3, the plants grown in these substrates showed reductions of 11-18% in plant height and 14-20% in shoot biomass relative to the control. Physiological measurements also showed decreased chlorophyll content, lower photosynthetic rates, and reduced stomatal conductance under Aloe-only treatments. These results indicate that the beneficial chemical characteristics of Aloe composts did not translate into improved plant performance in *Salvia officinalis* or *Rosmarinus officinalis* under the conditions of this experiment.

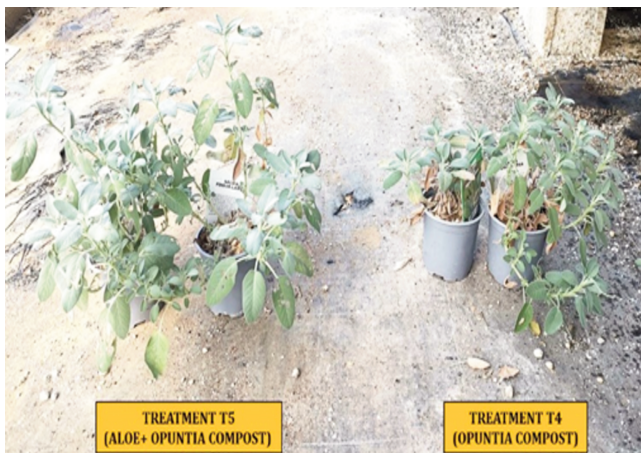


Fig. 2 - Effect of treatment T5 (Aloe + Opuntia compost) compared to T4 (Opuntia compost) on the height of *Salvia* plants.

These results demonstrate that while mixed or commercial composts can maintain growth comparable to the control substrate, single-source Aloe compost lacks the nutrient balance and structural benefits required for optimal plant development.

Physiological parameters

The physiological performance of *Salvia officinalis*, including chlorophyll content (SPAD), net photosynthetic rate (Pn), stomatal conductance (Gs), and water-use efficiency (WUE), varied significantly among treatments (Figs. 3, 4).

The highest SPAD index was observed in T0 (47), indicating maximum chlorophyll concentration. T1 (45) and T5 (44) exhibited only 4.3% and 6.4% lower SPAD values, re-spectively, compared with T0, whereas Aloe-only treatments (T2 and T3) recorded the lowest SPAD readings (38), representing a 19% reduction relative to the control (Fig. 3A).

A similar trend was found for photosynthetic activity. T0 achieved the highest Pn ($32 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), followed closely by T1 (30; -6.3%) and T5 (29; -9.4%). In contrast, T2 and T3 reduced photosynthetic rate to $24 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$, a 25% decline compared with the control (Fig. 3B), consistent with their reduced chlorophyll content and biomass production.

Stomatal conductance mirrored these patterns. T0

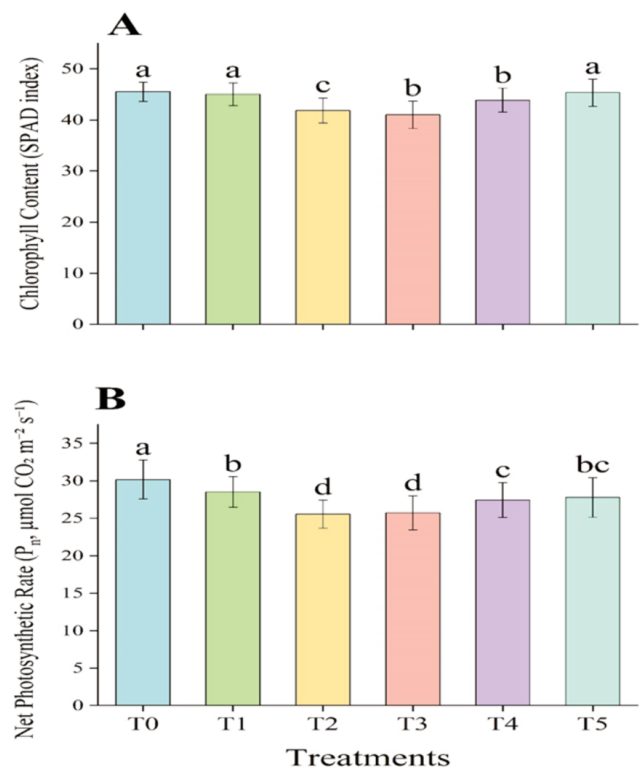


Fig. 3 - Chlorophyll content SPAD Index (A), and Photosynthetic Rate (Pn) (B) in *Salvia officinalis* under compost treatments. Note: Bars represent mean values ($n = 4$) with \pm SD. Letters above the bars denote statistically different groups (Tukey's HSD, $p < 0.05$). Treatments: T0= Control; T1= Commercial; T2= *Aloe barbadensis*; T3= *Aloe arborescens*; T4= Opuntia; T5= Mixed.

recorded the highest Gs ($0.55 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$), while T1 (0.50; -9.1%) and T5 (0.49; -10.9%) showed moderate reductions. T2 and T3 further decreased Gs to $0.42 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$, a 23.6% reduction relative to T0 (Fig. 4A), indicating restricted stomatal aperture and reduced transpiration capacity.

Interestingly, water-use efficiency (WUE) exhibited a slightly different response. T5 recorded the highest WUE (2.9), representing a 3.6% improvement over the control (2.8), despite a modestly lower photosynthetic rate. T1 maintained a similar WUE (2.7; -3.6%), whereas T2 and T3 showed the lowest efficiency (2.3), reflecting a 17.9% decline compared with T0 (Fig. 4B).

Collectively, these findings suggest that while commercial and mixed composts (T1 and T5) sustain gas exchange and chlorophyll synthesis near optimal levels, Aloe-only composts (T2 and T3) limit photosynthetic performance through reduced nutrient availability and stomatal activity. Notably, T5 enhanced WUE, suggesting potential advantages for water-limited cultivation systems.

Secondary metabolites and essential oils

Compost treatments significantly influenced secondary metabolite accumulation and essential oil production in *Salvia officinalis* (Table 3). The mixed Aloe-Opuntia compost (T5) markedly enhanced phenolic ($33.0 \pm 0.7 \text{ mg GAE g}^{-1}$) and flavonoid content ($18.0 \pm 0.6 \text{ mg QE g}^{-1}$), showing an increase of 10.0% and 11.1%, respectively, compared with the control (T0). T4 (Opuntia compost) also improved these metabolites relative to Aloe-only treatments but remained slightly lower than T5. In contrast, T2 and T3 (Aloe-only composts) resulted in the lowest phenolic (-18.2% for T3) and flavonoid contents (-27.8% for T3) relative to T0.

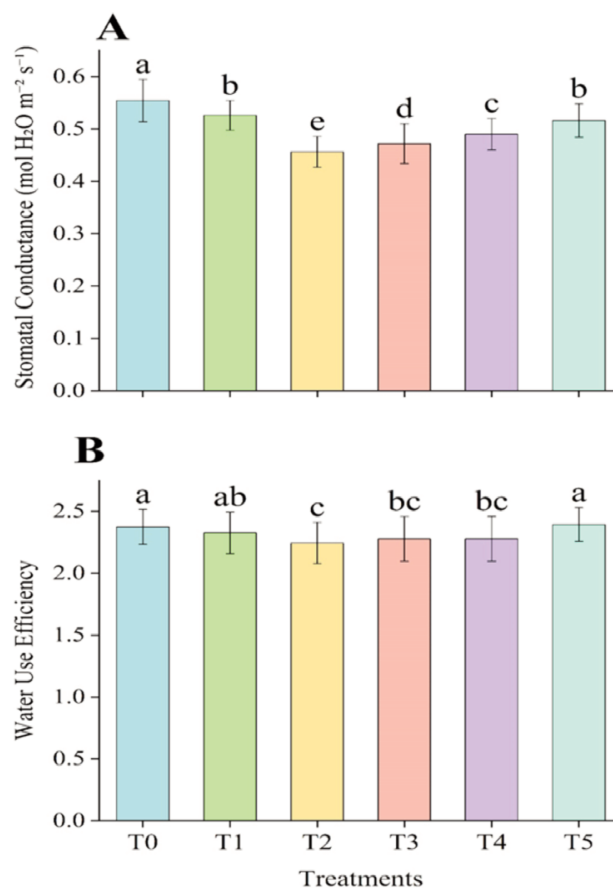


Fig. 4 - Stomatal conductance (A), and Water Use Efficiency (B) in *Salvia officinalis* under compost treatments. Note: Bars represent mean values (n = 4) with \pm SD. Letters above the bars denote statistically different groups (Tukey's HSD, $p < 0.05$). Treatments: T0= Control; T1= Commercial; T2= *Aloe barbadensis*; T3= *Aloe arborescens*; T4= Opuntia; T5= Mixed.

Antioxidant activity, assessed by IC_{50} values, showed that the control (T0) exhibited the strongest antioxidant capacity ($IC_{50} = 5.0 \pm 1.3 \mu\text{g mL}^{-1}$), consistent with its higher phenolic and flavonoid

Table 3 - Metabolite content in *Salvia officinalis* under different compost treatments

Treatment	Phenolic content (mg GAE g ⁻¹)	Flavonoids (mg QE g ⁻¹)	Antioxidant activity ($IC_{50} \mu\text{g mL}^{-1}$)	Essential oil Yield (%)	Microbial biomass ($\mu\text{g C/g soil}$)
T0	$33.0 \pm 0.7a$	$18 \pm 0.6a$	$50 \pm 1.3c$	$1.8 \pm 0.1 a$	$260.2 \pm 20.5a$
T1	$31.5 \pm 0.8a$	$17 \pm 0.6a$	$52 \pm 1.1c$	$1.6 \pm 0.1 ab$	$249.3 \pm 23.4b$
T2	$27.0 \pm 0.5d$	$14 \pm 0.4bc$	$58 \pm 1.0a$	$1.4 \pm 0.1 bc$	$238.1 \pm 21.2c$
T3	$25.5 \pm 0.8e$	$13 \pm 0.6c$	$60 \pm 1.3a$	$1.2 \pm 0.1 c$	$237.4 \pm 19.6c$
T4	$28.5 \pm 0.6c$	$15 \pm 0.7ab$	$56 \pm 1.0ab$	$1.4 \pm 0.1 bc$	$244.5 \pm 19.5bc$
T5	$30.0 \pm 0.7b$	$16 \pm 0.7a$	$54 \pm 1.1bc$	$1.6 \pm 0.1 ab$	$250.3 \pm 19.3b$

Values are mean \pm SD with statistical groupings indicated by letters (Tukey's test, $p < 0.05$). Treatments: T0= Control; T1= Commercial; T2= *Aloe barbadensis*; T3= *Aloe arborescens*; T4= Opuntia; T5= Mixed.

concentrations. Among compost treatments, T5 showed the best performance ($54 \pm 1.1 \mu\text{g mL}^{-1}$), followed by T4 ($56 \pm 1.0 \mu\text{g mL}^{-1}$). The Aloe-only treatments (T2 and T3) had the highest IC_{50} values ($58\text{-}60 \mu\text{g mL}^{-1}$), indicating the weakest antioxidant response. Thus, while mixed compost (T5) improved antioxidant capacity relative to other compost treatments, none surpassed the control substrate (T0).

Essential oil yield also followed this trend. T0 produced the highest oil content (1.8%), with T5 achieving a similar level (1.6%). T3 recorded the lowest yield (1.2%), representing a 33% reduction compared with T0.

Soil microbial biomass was highest in T0 and remained relatively high in T5, whereas Aloe-only composts (T2 and T3) showed an 8-9% reduction relative to the control.

Collectively, these results show that the mixed compost (T5) performed better than other compost treatments but did not exceed the control (T0) in antioxidant activity or essential oil yield.

Rosmarinus officinalis

Agronomic traits

Similar trends to *Salvia officinalis* were observed in *Rosmarinus officinalis* (Table 4, Fig. 5). The control treatment (T0) produced the most vigorous vegetative growth, yielding the tallest plants ($52.0 \pm 0.9 \text{ cm}$), the highest leaf number (138.0 ± 1.2), the largest leaf area ($570 \pm 11.4 \text{ cm}^2$), and the greatest shoot biomass ($42.0 \pm 1.0 \text{ g}$). Root traits also followed this pattern, with T0 showing the highest root biomass ($14.0 \pm 1.7 \text{ g}$) and longest roots ($20.6 \pm 1.8 \text{ cm}$).

Commercial compost (T1) performed comparably to the control, with only minor, non-significant

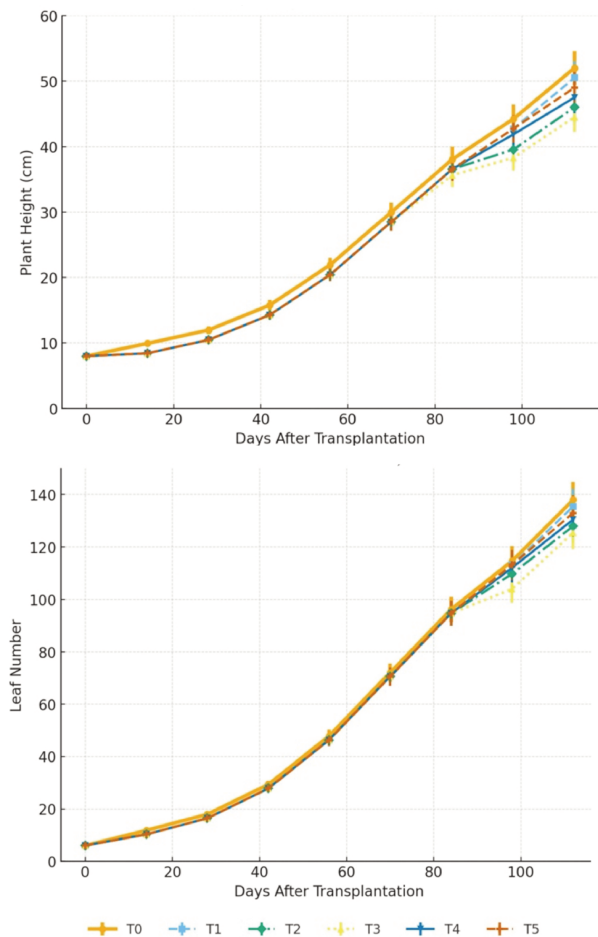


Fig. 5 - Biweekly growth dynamics of *Rosmarinus officinalis* under different compost treatments.

reductions in plant height (-2.9%), leaf number (-1.8%), leaf area (-2.6%), shoot biomass (-3.6%), root biomass (-4.3%), and root length (-3.9%). The mixed Aloe-Opuntia compost (T5) similarly supported substantial growth, showing 5.8% shorter plants, 3.6% fewer leaves, 5.3% smaller leaf area, and 7.1% lower shoot biomass, alongside moderate reductions

Table 4 - Agronomic parameters of *Rosmarinus officinalis*

Treatment	Plant height (cm)	Leaf number	Leaf area (cm^2)	Shoot biomass (g)	Root biomass (g)	Root length (cm)
T0	$52.0 \pm 0.9 \text{ a}$	$138.0 \pm 1.2 \text{ a}$	$570 \pm 11.4 \text{ a}$	$42.0 \pm 1.0 \text{ a}$	$14.0 \pm 1.7 \text{ a}$	$20.6 \pm 1.8 \text{ a}$
T1	$50.5 \pm 1.0 \text{ ab}$	$135.5 \pm 1.5 \text{ ab}$	$555 \pm 11.2 \text{ ab}$	$40.5 \pm 1.1 \text{ ab}$	$13.4 \pm 1.5 \text{ ab}$	$19.8 \pm 1.7 \text{ b}$
T2	$46.0 \pm 1.0 \text{ c}$	$128.0 \pm 1.4 \text{ c}$	$510 \pm 12.9 \text{ c}$	$36.0 \pm 1.3 \text{ c}$	$11.1 \pm 1.4 \text{ d}$	$17.6 \pm 1.4 \text{ d}$
T3	$44.5 \pm 1.2 \text{ c}$	$125.5 \pm 1.4 \text{ c}$	$495 \pm 12.8 \text{ c}$	$34.5 \pm 1.0 \text{ c}$	$11.3 \pm 1.5 \text{ d}$	$18.3 \pm 1.7 \text{ c}$
T4	$47.5 \pm 0.9 \text{ bc}$	$130.5 \pm 1.0 \text{ bc}$	$525 \pm 12.2 \text{ bc}$	$37.5 \pm 1.2 \text{ bc}$	$12.6 \pm 1.3 \text{ c}$	$19.4 \pm 1.6 \text{ b}$
T5	$49.0 \pm 1.2 \text{ b}$	$133.0 \pm 1.1 \text{ b}$	$540 \pm 11.2 \text{ b}$	$39.0 \pm 1.1 \text{ b}$	$12.8 \pm 1.8 \text{ bc}$	$19.7 \pm 1.6 \text{ b}$

Values are mean \pm SD with statistical groupings indicated by letters (Tukey's test, $p < 0.05$). Treatments: T0= Control; T1= Commercial; T2= *Aloe barbadensis*; T3= *Aloe arborescens*; T4= Opuntia; T5= Mixed.

in root biomass (-8.6%) and root length (-4.4%) compared with T0.

Moderate declines were recorded with Opuntia compost alone (T4), which resulted in 8.7% shorter plants (Fig. 6), 5.4% fewer leaves, 7.9% lower shoot biomass, 10.0% reduced root biomass, and 5.8% shorter roots relative to the control.



Fig. 6 - Effect of treatment T5 (Aloe + Opuntia compost) compared to T3 (Aloe compost) on the height of *Rosmarinus* plants.

In contrast, the Aloe-only treatments (T2 and T3) consistently produced the poorest growth. T2 reduced plant height by 11.5%, leaf number by 7.3%, leaf area by 10.5%, shoot biomass by 14.3%, root biomass by 20.7%, and root length by 14.6%. T3 showed even greater reductions, with 14.4% shorter plants, 9.0% fewer leaves, 13.2% smaller leaf area, 17.9% lower shoot biomass, 19.3% less root biomass, and 11.2% shorter roots compared with T0. These findings confirm that compost composition substantially affects both above- and below ground growth. Mixed or commercial composts provided a better nutrient balance and structural benefits, whereas Aloe-only composts were insufficient to support optimal root and shoot development.

Physiological parameters

Rosmarinus officinalis exhibited physiological responses similar to *Salvia officinalis* under different compost treatments (Figs. 7-8).

The SPAD index reached its maximum in control (T0, 47), indicating optimal chlorophyll content. T1 and T5 showed only 4-6% lower SPAD values, while Aloe-only treatments (T2 and T3) recorded the lowest readings (38), representing a 19% decline relative to T0 (Fig. 7A).

Net photosynthetic rate (Pn) peaked in T0 (31 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$). T1 and T5 maintained near-optimal photosynthetic rates with only 3-6% reductions, whereas T2 and T3 exhibited the lowest rates (24 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), corresponding to a 22.6% decline compared with the control (Fig. 7B).

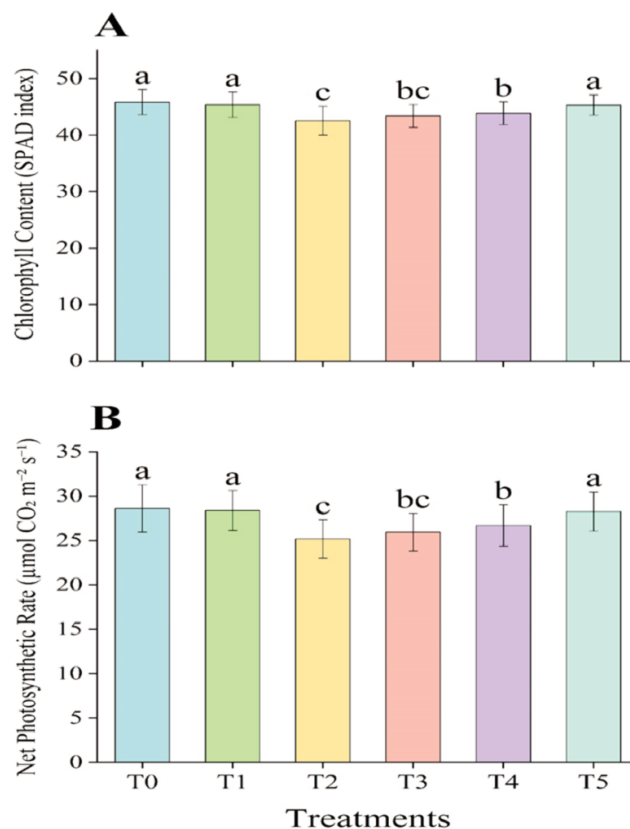


Fig. 7 - Chlorophyll content SPAD Index (A), and Photosynthetic Rate (B) in *Rosmarinus officinalis* under different compost treatments. Note: Bars represent mean values (n = 4) with \pm SD. Letters above the bars denote statistically different groups (Tukey's HSD, $p < 0.05$). Treatments: T0= Control; T1= Commercial; T2= *Aloe barbadensis*; T3= *Aloe arborescens*; T4= Opuntia; T5= Mixed.

Stomatal conductance (G_s) followed the same trend. T0 showed the highest conductance (0.55 $\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$), while T1 (0.50; -9%) and T5 (0.49; -11%) showed moderate reductions. T2 and T3 decreased G_s to 0.42 $\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$, a 23.6% reduction compared with T0 (Fig. 8A), suggesting limited stomatal aperture and reduced transpiration.

Interestingly, water-use efficiency (WUE) showed a slight advantage for the mixed compost. T5 achieved the highest WUE (2.9), a 3.6% improvement over T0 (2.8), while T1 maintained a similar efficiency

(2.7; -3.6%). T2 and T3 recorded the lowest WUE (2.3), a 17.9% decline relative to T0 (Fig. 8B).

These findings confirm that commercial (T1) and mixed composts (T5) sustain chlorophyll synthesis, gas exchange, and photosynthesis close to control levels, while Aloe-only composts significantly compromise physiological performance. Notably, the mixed compost (T5) enhanced WUE, indicating potential benefits under water-limited conditions.

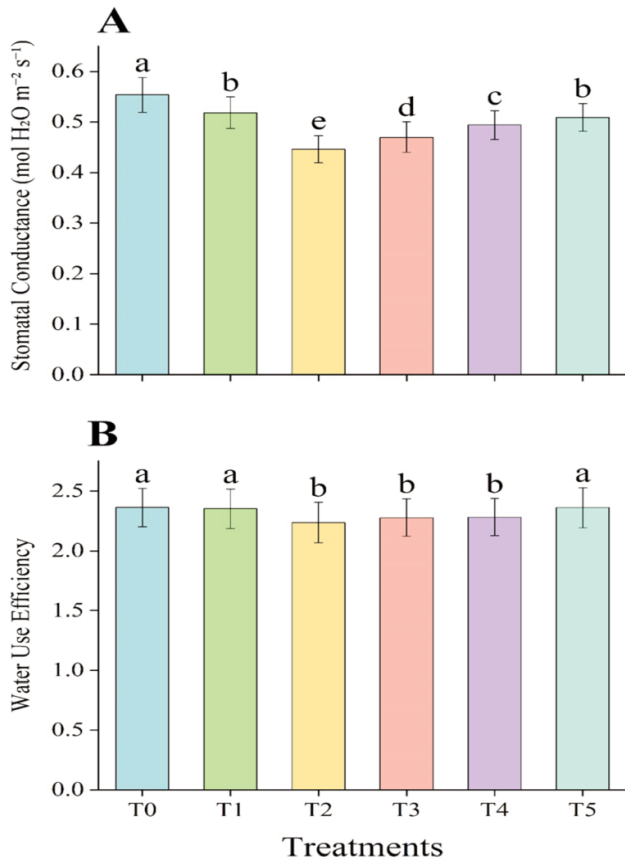


Fig. 8 - Stomatal conductance (A), and Water Use Efficiency (B) in *Rosmarinus officinalis* under different compost treatments. Note: Bars represent mean values ($n = 4$) with \pm SD. Letters above the bars denote statistically different groups (Tukey's HSD, $p < 0.05$). Treatments: T0= Control; T1= Commercial; T2= *Aloe barbadensis*; T3= *Aloe arborescens*; T4= *Opuntia*; T5= Mixed.

Metabolite content and essential oil yield

Compost treatments significantly affected secondary metabolite production and soil microbial biomass in *Rosmarinus officinalis* (Table 5). The control substrate (T0) yielded the highest phenolic content (32.0 ± 0.7 mg GAE g⁻¹) and flavonoid content (17.5 ± 0.6 mg QE g⁻¹).

The mixed Aloe-*Opuntia* compost (T5) performed slightly lower than the control, with -9.4% phenolics and -11.4% flavonoids, yet it was statistically superior to single-source composts. *Opuntia* compost alone (T4) followed a similar trend with -14.1% phenolic reduction compared with T0. In contrast, Aloe-only composts (T2 and T3) produced the lowest metabolite levels, reducing phenolic content by 18.8% (T2) and 23.4% (T3), and flavonoids by 22.9% and 28.6%, respectively, relative to the control.

Antioxidant activity, expressed as IC₅₀, showed the expected inverse trend, in which lower IC₅₀ values indicate stronger antioxidant capacity. The control treatment (T0) exhibited the highest antioxidant activity (48 ± 1.3 μ g mL⁻¹), followed by T1 (50 ± 1.4 μ g mL⁻¹). The mixed compost (T5) showed moderate antioxidant performance (52 ± 1.4 μ g mL⁻¹), whereas the Aloe-only treatments, particularly T3 (58 ± 1.0 μ g mL⁻¹), recorded the weakest antioxidant responses. Thus, T3 showed a 20.8% reduction in antioxidant efficiency relative to T0, confirming that Aloe-only composts were less effective in promoting antioxidant metabolism in rosemary.

Essential oil yield followed a similar pattern. T0 produced the highest yield ($1.7 \pm 0.1\%$), while T5 ($1.5 \pm 0.1\%$) and T4 ($1.4 \pm 0.1\%$) showed modest declines (11.8% and 17.6% reductions, respectively). The Aloe-only treatments resulted in the lowest oil content, with T3 yielding only $1.2 \pm 0.1\%$, a 29.4% reduction relative to the control.

Soil microbial biomass was highest under T0 (264.0 ± 20.7 μ g C g⁻¹) but remained relatively high under T5 (249.6 ± 19.2 μ g C g⁻¹; -5.5%). Aloe-only treatments significantly reduced microbial biomass, with T2 decreasing by 13.1% and T3 by 10.9% compared with the control (Table 5). No significant differences were observed in pest or disease incidence across treatments in either species. Preventive biological treatments (*Beauveria bassiana* and *Trichoderma harzianum*) maintained pest populations below damage thresholds throughout the trial. Thus, compost influence appeared limited to plant growth and metabolism rather than pest suppression. Collectively, these results indicate that mixed or *Opuntia*-based composts can partially sustain metabolite synthesis, antioxidant capacity, and essential oil production, whereas Aloe-only composts significantly compromise both plant biochemical quality and soil microbial activity.

Table 5 - Metabolite content in *Rosmarinus officinalis*

Treatment	Phenolic content (mg GAE g ⁻¹)	Flavonoids (mg QE g ⁻¹)	Antioxidant activity (IC ₅₀ µg mL ⁻¹)	Essential oil yield (%)	Microbial biomass (µg C/g soil)
T0	32.0 ± 0.7 a	17.5 ± 0.6 a	48 ± 1.3 c	1.7 ± 0.1 a	264.0±20.7 a
T1	30.5 ± 0.6 b	16.5 ± 0.7 a	50 ± 1.4 bc	1.6 ± 0.1 ab	252.1±22.5 b
T2	26.0 ± 0.7 e	13.5 ± 0.7 bc	56 ± 1.2 a	1.3 ± 0.1 bc	229.4±18.5 e
T3	24.5 ± 0.5 f	12.5 ± 0.7 bc	58 ± 1.0 a	1.2 ± 0.1 c	235.3±20.3 de
T4	27.5 ± 0.5 d	14.5 ± 0.8 ab	54 ± 1.2 ab	1.4 ± 0.1 bc	242.2±17.8 cd
T5	29.0 ± 0.7 c	15.5 ± 0.7 a	52 ± 1.4 ab	1.5 ± 0.1 ab	249.6±19.2 bc

Values are mean ± SD with statistical groupings indicated by letters (Tukey’s test, p < 0.05). Treatments: T0= Control; T1= Commercial; T2= *Aloe barbadensis*; T3= *Aloe arborescens*; T4= Opuntia; T5= Mixed.

Principal component and correlation analysis

The principal component analysis (PCA) of morpho-physiological and biochemical traits in *Salvia officinalis* and *Rosmarinus officinalis* under different compost treatments revealed distinct clustering patterns among treatments and species (Fig. 9A). The first two principal components (PCs) explained 95.1% of the total variance, with PC1 accounting for 91.2% and PC2 contributing 3.9%. PC1 was positively associated with water-use efficiency (WUE), chlorophyll content (Chl), root length (RL), evapotranspiration (ET), root biomass (RB), stomatal conductance (gs), and net photosynthetic rate (Pn). In contrast, plant height (PH), leaf area (LA), leaf number (LN), shoot biomass (SB), essential oil yield (EOY), total phenolic content (TPC), and total flavonoid content (TFC) were negatively loaded on PC1. PC2 was predominantly defined by antioxidant activity (AOA), indicating its distinct response relative to other traits.

Control treatments (T0) clustered toward the positive axis of PC1, indicating superior physiological performance, whereas T2 and T3 treated plants shifted toward the negative side, showing higher accumulation of secondary metabolites such as TPC, TFC, and EOY. Moreover, *Rosmarinus officinalis* tended to group closer to the positive PC1 axis, reflecting its relatively higher photosynthetic efficiency compared with *Salvia officinalis* under the same compost regimes.

Correlation analysis (Fig. 9B) further supported the PCA findings, showing strong positive correlations (r > 0.80, p < 0.001) among growth-related parameters such as (PH), LN, LA, SB, and RB, demonstrating their coordinated response to compost amendments. Photosynthetic parameters (Pn, gs, ET, and WUE) were also significantly

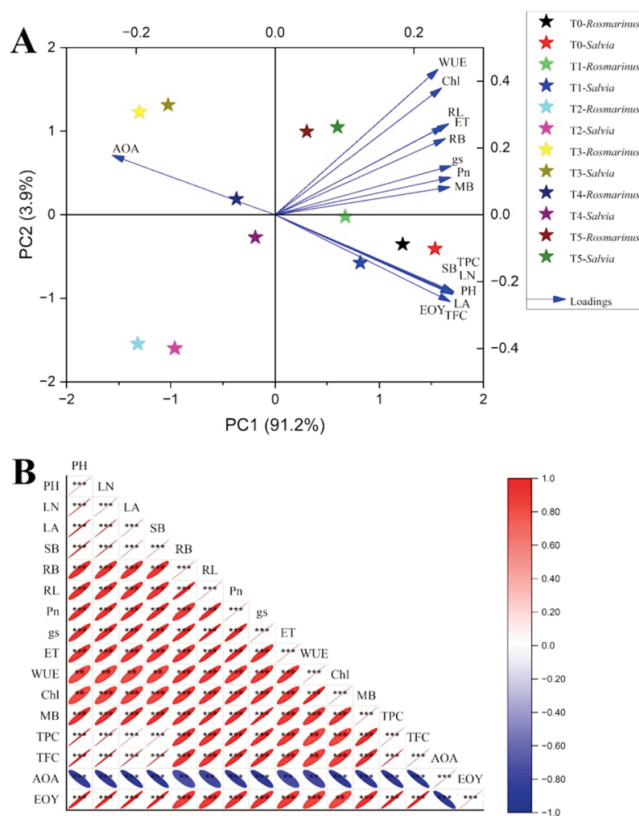


Fig. 9 - Principal component and correlation analysis of morpho-physiological and biochemical traits in *Salvia officinalis* and *Rosmarinus officinalis* under different compost treatments. (A) Principal component analysis (PCA) biplot showing the distribution of treatments and species along the first two principal components (PC1 = 91.2%, PC2 = 3.9%) with trait loadings indicated by blue arrows. (B) Pairwise correlation matrix of growth, physiological, and biochemical traits. The color scale indicates correlation coefficients (red = positive, blue = negative), with significance levels denoted as (*p ≤ 0.05, **p ≤ 0.01, ***p ≤ 0.001).

correlated with Chl and biomass accumulation (MB), highlighting the role of compost in enhancing photosynthetic efficiency and growth. Conversely, secondary metabolite traits (TPC, TFC), AOA, and EOY

were positively interrelated but showed significant negative correlations ($r < -0.60$, $p < 0.01$) with growth and photosynthetic parameters, suggesting a trade-off between vegetative growth and secondary metabolite production.

Notably, AOA exhibited the strongest negative correlation with WUE, Chl, and MB, indicating that antioxidant defenses were enhanced under conditions limiting photosynthetic performance. Furthermore, TPC and TFC displayed a positive relationship with EOY, emphasizing their role in essential oil biosynthesis under compost-induced stress conditions.

4. Discussion and Conclusions

This study highlights the influence of compost composition on the growth, physiological behaviour and secondary metabolism of *Salvia officinalis* and *Rosmarinus officinalis* grown under controlled conditions in a greenhouse. While the peat-based control substrate (T0) consistently produced the highest levels of growth and metabolites, the Aloe-Opuntia mixed compost (T5) showed considerable potential as a renewable and environmentally sustainable alternative, despite its slightly lower agronomic performance. The results provide important insights into how specific compost raw materials, particularly plant species with contrasting biochemical profiles, influence substrate quality, plant physiology and secondary metabolic pathways (Muscolo *et al.*, 2018; Veliu *et al.*, 2025). The superior performance of the control substrate (T0) is not unexpected, given the well-documented physical advantages of peat, including optimal aeration, drainage, high porosity and a stable supply of nutrients (Zhang *et al.*, 2025). These properties promote optimal root development, efficient water use and robust photosynthetic activity, which in turn lead to increased biomass production and secondary metabolite synthesis (Pan *et al.*, 2025). As peat-based substrates continue to be the industry standard for container-grown aromatic and medicinal crops, they provide a natural benchmark for evaluating the effectiveness of alternative amendments (Atzori *et al.*, 2021). However, environmental concerns related to peat extraction, including habitat destruction, carbon emissions and lack of renewability, necessitate the search for more sustainable substrate options (Räsänen *et al.*, 2023; Patel *et al.*, 2025).

In this context, the Aloe-Opuntia mixed compost (T5) emerged as the best performing organic soil amendment among those tested. Although it did not match T0 in terms of absolute growth, its performance remained relatively close, with reductions in vegetative traits generally ranging between 5 and 8%. Importantly, T5 consistently outperformed all other compost-based treatments in terms of agronomic, physiological, and biochemical parameters. This suggests that mixing aloe and opuntia biomass generates a substrate with more favourable physical and chemical characteristics than composts derived from a single raw material (Sortino *et al.*, 2024). The improved performance of T5 is likely attributable to the complementary nature of its components. Opuntia residues are rich in mucilaginous polysaccharides, known to increase water retention and improve the moisture buffering capacity of the substrate (Semerel *et al.*, 2023; Bacchetta *et al.*, 2024). This effect helps maintain stable hydration around the root zone, minimising water-related stress fluctuations commonly experienced in organic substrates and facilitating more controlled stomatal behaviour. Meanwhile, aloe residues contribute bioactive compounds such as polysaccharides, phenols, and glycoproteins that can function as plant biostimulants, improving nutrient uptake efficiency, osmotic regulation, and tolerance to minor environmental stressors (Prisa and Gobbino, 2021). These combined effects likely contributed to the greater physiological stability observed in T5, including higher chlorophyll content, improved stomatal conductance, and greater water use efficiency compared to other compost-based substrates (Read and Gregory, 1997). In contrast, aloe-only composts (T2 and T3) resulted in the weakest plant performance, despite their relatively high nutrient content. This highlights the importance of distinguishing between total nutrient concentration and actual agronomic suitability. Several factors may explain the limited effectiveness of aloe-based composts (Jaramillo *et al.*, 2025). First, both T2 and T3 showed the highest electrical conductivity values in the experiment (1.7-1.8 dS m⁻¹). It is well known that high salinity can limit water absorption by roots, reduce photosynthetic capacity and compromise stomatal function, reactions that are particularly evident in aromatic species native to Mediterranean regions (Tong *et al.*, 2025). Therefore, the high EC may have caused physiological stress that overshadowed the potential benefits of the available

nutrients (Atiyeh *et al.*, 2000).

Secondly, aloe biomass contains high levels of labile organic compounds, including mucilage, organic acids and simple sugars, which decompose rapidly and can temporarily create anaerobic microsites, organic acid accumulation and excessive microbial respiration (Maliehe *et al.*, 2023; Di Palma *et al.*, 2025). Such fluctuations can stress root systems, reduce oxygen availability, and create transient nutritional imbalances. This rapid decomposition dynamic is characteristic of monospecific composts derived from succulent plant material and has previously been associated with inconsistent nutrient release and reduced substrate stability (Gruber *et al.*, 2013).

Thirdly, aloe species contain biologically active metabolites such as aloin, haemodin and anthraquinones. Although composting reduces their concentration, residual levels may still affect root physiology. Some studies report inhibitory effects of secondary compounds derived from aloe on seedling root elongation and early plant development (Radha and Laxmipriya, 2014). This may partly explain the reduction in root biomass and shorter root length observed in treatments with aloe-only compost (Tawaraya *et al.*, 2007).

Finally, the physical structure of aloe tissue is characterised by low fibre content and high water content, which can result in compost with poor porosity, limited aeration and reduced structural stability. When used as the main component of the substrate, such composts can create compact or excessively moist environments, limiting root growth and compromising gas exchange (Dehouche *et al.*, 2020). The moderate performance of the Opuntia-only compost (T4) offers further insight into the influence of specific raw materials. Opuntia residues improved several metabolic characteristics and led to moderate but not optimal vegetative growth (Auteri *et al.*, 2025). This pattern suggests that, although Opuntia contributes to favourable moisture retention, microbial stimulation and nutrient cycling, its physical and chemical properties alone are not sufficient to match the agronomic efficiency of mixed compost (Matheri *et al.*, 2023). This reinforces the idea that substrate quality benefits from the biochemical and structural diversity of mixed raw materials, which improve nutrient release dynamics, microbial diversity, and physical stability (Naguib *et al.*, 2012). Secondary metabolism was strongly influenced by substrate type. For both species, T0

produced the highest levels of phenols, flavonoids, and essential oils, likely due to the overall higher vigour of the plants and carbon assimilation. T5, although slightly lower than the control, consistently ranked above the other compost treatments, indicating that mixed compost maintained a physiological environment favourable to secondary metabolite production (Montoya *et al.*, 2022). The performance of T5 suggests that mixed composts can support plant metabolic activity without inducing severe stress that would reduce secondary metabolite biosynthesis. In contrast, aloe-only composts showed the lowest accumulation of metabolites. Again, this may reflect the negative physiological effects of high EC and rapid decomposition dynamics, which impose sufficiently severe stress to limit carbon assimilation and reduce the metabolic resources available for secondary metabolite pathways (Biyada *et al.*, 2022; Sarwari *et al.*, 2024).

Importantly, none of the compost treatments adversely affected plant survival or increased susceptibility to pests or diseases. This suggests that all composts were sufficiently mature and free of phytotoxicity, supporting their potential use in organic greenhouse systems (Hadar and Papadopoulou, 2012). The absence of disease incidence is consistent with the well-known benefits of high-quality composts in suppressing soil-borne pathogens through competitive exclusion, microbial antagonism, and improved soil structure. Finally, the implications of these findings in terms of sustainability are significant (Pane *et al.*, 2019). Aloe and opuntia are drought-resistant species widely cultivated in arid and Mediterranean regions, producing significant by-products during pruning and processing (Liontakis and Tzouramani, 2016). The reuse of this biomass in compost is in line with the principles of circular agriculture, reducing waste and providing growers with renewable, low-input substrate alternatives. Although peat remains agronomically superior, mixed aloe and opuntia composts represent a promising step towards more sustainable greenhouse production of aromatic crops (Campos *et al.*, 2025).

Future research should aim to further characterise the microbial communities associated with each type of compost, as interactions between microbes and plants are increasingly recognised as determinants of nutrient availability, stress tolerance and secondary metabolite synthesis (Aguilar-Paredes *et al.*, 2023;

Gil-Martínez and Madejón, 2025). Field validation across multiple seasons and environments would further strengthen the practical relevance of these findings. Furthermore, technical-economic analyses would help determine the feasibility of large-scale compost production using aloe and opuntia residues (Pergola *et al.*, 2020; Gil-Martínez and Madejón, 2025).

The results of this research work confirmed that compost composition impacts the vegetative growth, physiological properties, and secondary metabolism of *Salvia officinalis* and *Rosmarinus officinalis* cultivated in a greenhouse environment. The control treatment containing peat as the growing substrate (T0) consistently provided greater growth and higher levels of phytochemical accumulation, indicating that it is a suitable growing medium for the conditions of this study. The mixed Aloe and Opuntia compost treatment (T5) produced less vegetative growth and phytochemical accumulation than the control treatment (T0) but had higher growth and phytochemical accumulation than the other compost treatments. This indicates that mixing plant residues can help lessen some of the disadvantages associated with using a compost consisting solely of one species of plant residue. While the differences seen between treatments should be approached cautiously, it is evident that more replicated trials in a variety of seasons/locations are needed to verify the clarity of the observed yield trends in this study, particularly for T5 moderate effectiveness and the lesser yielding ability of aloe-only composts. Although currently, peat (T0) is thought to be the best substrate, mixed plant-based composts such as T5 potentially present the opportunity for a more sustainable alternative. Their value lies not in the fact that they produce greater agronomic performance, but rather that they are renewable and consequently less harmful to the environment. When supported with further validation and additional levels of detailed evaluation, this research line may assist in the creation of environmentally sustainable composter for the farming of aromatherapy and medicinal crops.

Acknowledgements

The authors would like to express his heartfelt gratitude to colleagues and providers of plants and to all other sources for their cooperation and guidance

in writing this article.

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