



**ARTICLE**

# Evaluation of Commercial Potting Substrates for Reproducible Growth of *Arabidopsis thaliana* and *Nicotiana tabacum* under Laboratory Conditions

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**ABSTRACT:** The potting substrate is an important determinant of post-germination growth in *Arabidopsis thaliana* and *Nicotiana tabacum* under controlled laboratory conditions. We evaluated four commercially available soil substrates—Sta-Green potting mix plus fertilizer (SPM), Sta-Green flower & vegetable garden soil plus fertilizer (SGS), Miracle-Gro potting mix (MPM), and Miracle-Gro raised bed soil (MBS)—to assess their effects on seed germination and post-germination growth. Germination rates did not differ significantly among substrates for either species. In contrast, post-germination growth was strongly influenced by the substrate, with MPM consistently supporting greater biomass accumulation, stem elongation, and leaf production. Through integrated analysis of the manufacturer-reported nutrient composition and the substrate physical characteristics, including texture and moisture retention, we propose a mechanistic explanation for these growth differences. The superior performance of MPM likely reflects the combined effects of nutrient availability and favorable physical properties, including fine texture, aeration, and water-holding capacity, rather than nutrient content alone. Together, these results identify potting substrate choice as an important experimental variable affecting growth outcomes and reproducibility in laboratory-based plant research.

**KEYWORDS:** *Arabidopsis thaliana*; *Nicotiana tabacum*; potting substrates; germination rate; post-germination growth; experimental reproducibility

## 1 Introduction

The potting substrate is a critical component of plant-based research, influencing seed germination, vegetative growth, and biomass production. *Arabidopsis thaliana* and *Nicotiana tabacum* are widely used model plants in genetic, physiological, and molecular studies because of their well-characterized genomes, rapid life cycles, and established laboratory cultivation protocols [1,2]. *N. tabacum* is extensively used for gene transformation, to study plastid biology, and for developing and optimizing experimental approaches applicable to other plant species [3], whereas *Arabidopsis* is widely used as a model for studying gene function, plant development, and physiological and molecular responses to environmental cues [1,2]. Given the central role of these species in laboratory research, the use of standardized and reproducible growth substrates is essential for minimizing experimental variability and ensuring consistency across studies.

Although soil composition is known to influence post-germination development, its effect on seed germination remains debated [4,5]. Nutrient availability, moisture retention, aeration, and substrate structure can all significantly affect plant growth trajectories, yet relatively little systematic work has

examined how commercially available growth substrates influence model plant performance under controlled laboratory conditions. As a result, selection of the potting mix is often treated as a minor methodological detail, limiting standardization and reproducibility across research groups. In studies using *A. thaliana*, traits such as germination rate, total plant length, and dry biomass are commonly used to assess growth performance [2]. Similarly, *N. tabacum* growth is often evaluated using germination rate, stem length, leaf number, and biomass accumulation [3]. Germination rate is particularly important when seed availability is limited, as reliable establishment is essential for downstream experimentation [6]. Beyond germination, substrate-dependent differences in plant size, leaf production, and biomass can substantially influence experimental outcomes by determining the amount of tissue available for molecular, biochemical, and physiological analyses [7]. Therefore, identifying substrates that support consistent and vigorous growth is important not only for plant establishment but also for subsequent experiments.

Despite this importance, direct comparisons of commonly used commercial potting mixes or substrates in laboratory model systems remain limited. In this study, we systematically compared four widely available commercial mixes, differing in texture, nutrient composition, and intended application, to evaluate their effects on germination and post-germination growth of *A. thaliana* and *N. tabacum*. By assessing plant height, biomass accumulation, and leaf production and by integrating available information on the physical and chemical properties of the substrates, we aimed to provide mechanistically informed and actionable guidance for potting mix selection in laboratory-based plant research.

## 2 Materials and Methods

### 2.1 Substrate Selection and Composition

The four substrates used in this study were purchased from Lowe's, a widely available retailer in the United States of America. Sta-Green® Potting Mix Plus Fertilizer (SPM) and Sta-Green Flower & Vegetable Garden Soil Plus Fertilizer (SGS) are produced by Sta-Greens, New York, NY, USA, while Miracle-Gro Potting Mix (MPM), and Miracle-Gro Raised Bed Soil (MBS) are produced by The Scotts Miracle-Gro Company, Marysville, OH, USA. These substrates differ in nutrient content, organic matter composition, and physical structure. According to manufacturer's label, MPM contains a slow-release fertilizer blend with higher nitrogen and potassium levels and includes sphagnum peat moss and perlite, which are expected to enhance aeration and water retention. In contrast, MBS and SGS are formulated primarily for outdoor gardening and raised-bed use and have coarser textures, lower reported nutrient levels, and less uniform nutrient release. Soil properties are summarized in Table 1, representative substrate textures are shown in Fig. 1, and detailed compositional information is provided in Appendix A.

### 2.2 Experimental Design

Seeds of *A. thaliana* (ecotype Col-0) and *N. tabacum* (variety Honghua dajinyuan) were sown in pots as described previously [8] using each substrate. Three-inch square pots were filled with substrate to approximately 1 cm above the rim to allow for compaction after watering. The surface was gently leveled, and pots were placed in plastic flats. Each flat was initially watered with 1000 mL of distilled water to ensure uniform hydration. Flats were covered with clear plastic domes and placed in a growth chamber (Percival PR-106) at 22°C for 24 h before sowing. After sowing, the domes were placed for an additional 24 h and then removed. Thereafter, plants were maintained as described below (Section 2.3).

Seed germination and subsequent plant growth experiments were conducted to evaluate the effects of the different substrates under controlled laboratory conditions. For *A. thaliana*, five seeds were sown per pot, with ten pots per substrate type in each experimental run, for a total of 50 seeds per mix per

run. For *N. tabacum*, one seed was sown per pot, with five pots per substrate type in each experimental run. Germination was recorded when radicle emergence became visible. For germination analyses, each pot was treated as the replicate unit; thus, *A. thaliana* germination was evaluated across 10 replicates per experiment, and *N. tabacum* germination was evaluated across 5 replicates per experiment.

For post-germination growth analyses, each pot was considered an independent experimental unit. In *A. thaliana*, five seeds were initially sown per pot to ensure successful establishment, and seedlings were thinned to one plant per pot after germination to minimize within-pot competition and to standardize subsequent growth comparisons among substrates. In *N. tabacum*, one seedling was maintained per pot throughout the experiment.

Independent experiments were conducted to evaluate reproducibility. Germination experiments were repeated three times for *A. thaliana* and four times for *N. tabacum*. Growth trait experiments were conducted across three independent experiments. The resulting data were used for statistical analysis.

**Table 1:** Characteristics of four commercial soil substrates used for growing *A. thaliana* and *N. tabacum* under laboratory conditions.

Substrate	Nutrient Content (N-P-K, %)	Texture & Structure	Water Retention & Drainage
MPM (Miracle-Gro Potting Mix)	0.21-0.11-0.16	Fine, lightweight, high-quality perlite	Excellent aeration and moisture retention
SPM (Sta-Green Potting Mix)	0.10-0.08-0.06	Moderately fine, containing bark and peat moss	Good drainage with moderate moisture retention
SGS (Sta-Green Flower & Vegetable Soil)	0.05-0.04-0.03	Coarse, low organic matter	Reduced water retention, quicker drying
MBS (Miracle-Gro Raised Bed Soil)	0.09-0.08-0.09	Coarse, includes composts and peat	Moderate retention of water; it depends on the region

Note: The table summarizes the reported nutrient composition (nitrogen, phosphorus, and potassium; N-P-K), descriptive textural and structural properties, and water retention characteristics of each substrate.

### 2.3 Growing Conditions

*A. thaliana* plants were grown in a controlled growth chamber at 22°C under a 16-h light/8-h dark photoperiod with a light intensity of 100  $\mu\text{mol photons}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  PAR. After removal of the plastic dome, all flats were irrigated uniformly every 2–3 days, based on visual assessment of substrate moisture and plant condition, to maintain adequate moisture for germination and growth while avoiding waterlogging.

Germination rates were recorded 7 days after sowing. Plants were photographed 28 days after germination and after thinning to one plant per pot. Plants were harvested 75 days after sowing. At harvest, total plant length was measured by carefully uprooting plants, gently straightening the root system, and recording the distance from the root tip to the shoot apex. This measurement was used as an integrated indicator of whole-plant growth response to the different substrates [9]. Dry biomass was then determined after drying the samples at 44°C for 72 h.

*N. tabacum* plants were grown under similar watering, temperature and photoperiod conditions (22°C, 16-h light and 8-h dark) with a light intensity of 200  $\mu\text{mol photons}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  PAR. Germination was recorded 10 days after sowing. Plants were photographed 28 days after germination. At 34 days after sowing, seedlings were transplanted into larger pots (30 cm high  $\times$  10.5 cm diameter) with a new allotment of the same growth substrate and transferred from the growth chamber to a greenhouse receiving approximately 30% natural daylight through roof shading. Plants were harvested 60 days after sowing. Stem length, leaf number, and dry weights of stems and leaves were measured after drying samples at 70°C for 10 days.

Because *A. thaliana* and *N. tabacum* differ substantially in plant size, tissue mass, and growth habit, species-specific drying conditions were used to ensure complete dehydration without visible tissue scorching or deformation. We note, however, that these species-specific protocols limit direct comparison of absolute drying procedures between species and should be interpreted within the context of species-specific analyses.



**Figure 1:** Visual appearance and texture of the four commercial substrates used in this study. The images illustrate differences in color, particle size, and structural composition, corresponding to the physical characteristics summarized in Table 1.

## 2.4 Statistical Analysis

All statistical analyses were performed in RStudio version 2023.03.0. Germination data were analyzed using generalized linear models (GLMs) with substrate type, experimental run, and their interaction included as fixed effects. Germination was modeled as the number of germinated seeds relative to the total number of seeds sown within each experimental unit.

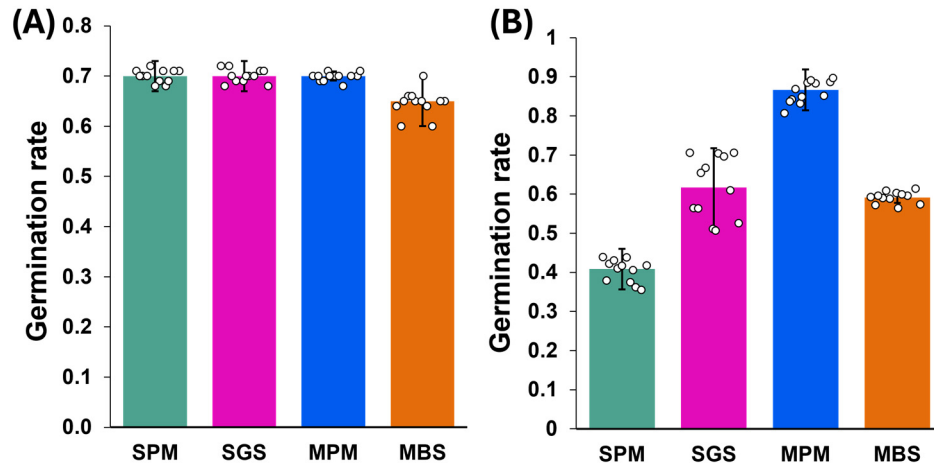
Plant growth traits, including biomass, total plant length or stem length, and leaf number, were analyzed using two-way analysis of variance (ANOVA), with substrate type and experimental run treated as fixed factors. When significant main effects were detected, pairwise comparisons among substrate types were conducted using Tukey's honestly significant difference (HSD) test. Because experimental run was included in the statistical models, variation among runs was accounted for during analysis.

Before ANOVA, model residuals were examined to assess the assumptions of parametric analysis. Normality and homogeneity of variance were evaluated with the Shapiro–Wilk and Levene's tests, respectively. Because independent experimental runs were conducted under the same controlled conditions and showed similar response patterns, data from repeated runs were pooled for presentation, while experimental run was retained in the statistical models.

## 3 Results

### 3.1 Germination Rate

There were no significant differences in germination rates among the tested substrates for either *N. tabacum* or *A. thaliana* according to the GLM analysis (Fig. 2). The interaction between substrate and experimental run was also not significant ( $p > 0.05$ ), indicating that germination responses were consistent across independent runs under the controlled conditions used in this study (Fig. 2). Furthermore, pairwise comparisons detected no significant differences among substrates for either species ( $p > 0.05$ ). These results suggested that, for the commercially formulated substrates tested here, the potting media did not measurably affect seed germination under these controlled conditions.



**Figure 2:** Germination rate by substrate type in (A) *N. tabacum* and (B) *A. thaliana*. For *N. tabacum*, one seed was sown per pot, with five pots per substrate in each experimental run ( $n = 5$  pots per substrate per run; 4 independent runs). For *A. thaliana*, five seeds were sown per pot, with ten pots per substrate in each experimental run ( $n = 10$  pots per substrate per run; 3 independent runs). Germination data were analyzed using generalized linear models (GLMs). No significant differences in germination rate were detected among the tested substrates ( $p > 0.05$ ). Bars represent group means  $\pm$  SD, and dots represent individual pot-level observations pooled across independent experimental runs.

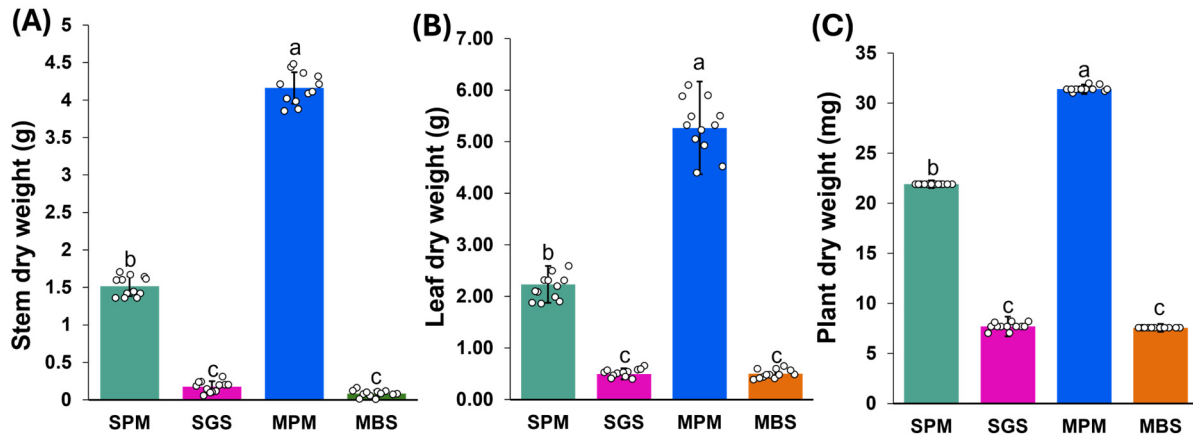
### 3.2 Biomass

Substrate type had a significant effect on dry biomass in both *N. tabacum* and *A. thaliana*. Among the substrates tested, the MPM substrate consistently produced the highest dry weights across all biomass-related measurements.

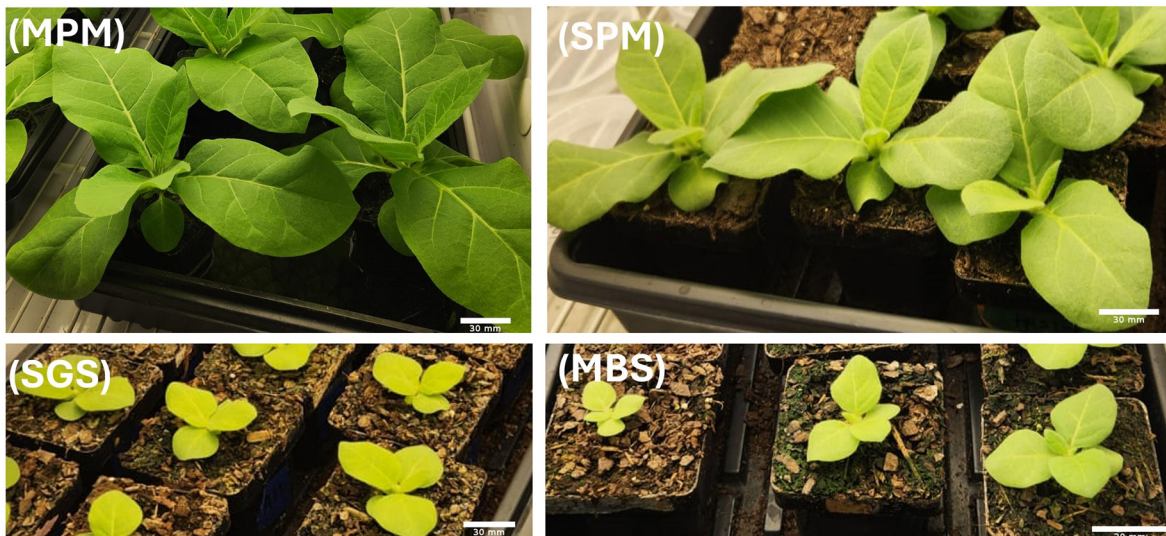
For *N. tabacum*, two-way ANOVA revealed a significant effect of substrate type on both leaf and stem dry weights ( $p < 0.0001$ ; Fig. 3A,B). Tukey's HSD test showed that MPM supported significantly greater biomass accumulation than all other substrates for both leaves and stems (Fig. 3A,B). In contrast, SGS and MBS produced significantly lower dry weights, with no significant difference between these two substrates (Fig. 3A,B). Neither experimental run nor the interaction between substrate and experimental run was significant, indicating that the effect of substrate on biomass was consistent across independent runs.

Similarly, for *A. thaliana*, substrate type had a significant effect on plant dry weight ( $p < 2e-16$ ; Fig. 3C). Plants grown in MPM had significantly greater dry weights than those grown in SPM, SGS, or MBS. Tukey's HSD test confirmed significant differences between MPM and all other substrates ( $p < 0.001$ ). In addition, SGS and MBS produced significantly lower dry weights than MPM and SPM ( $p < 0.0001$ ), whereas no significant difference was detected between SGS and MBS for either trait ( $p > 0.05$ ). As observed for *N. tabacum*, neither experimental run nor the interaction between substrate and experimental run was significant, indicating that the substrate effects were reproducible across experimental runs.

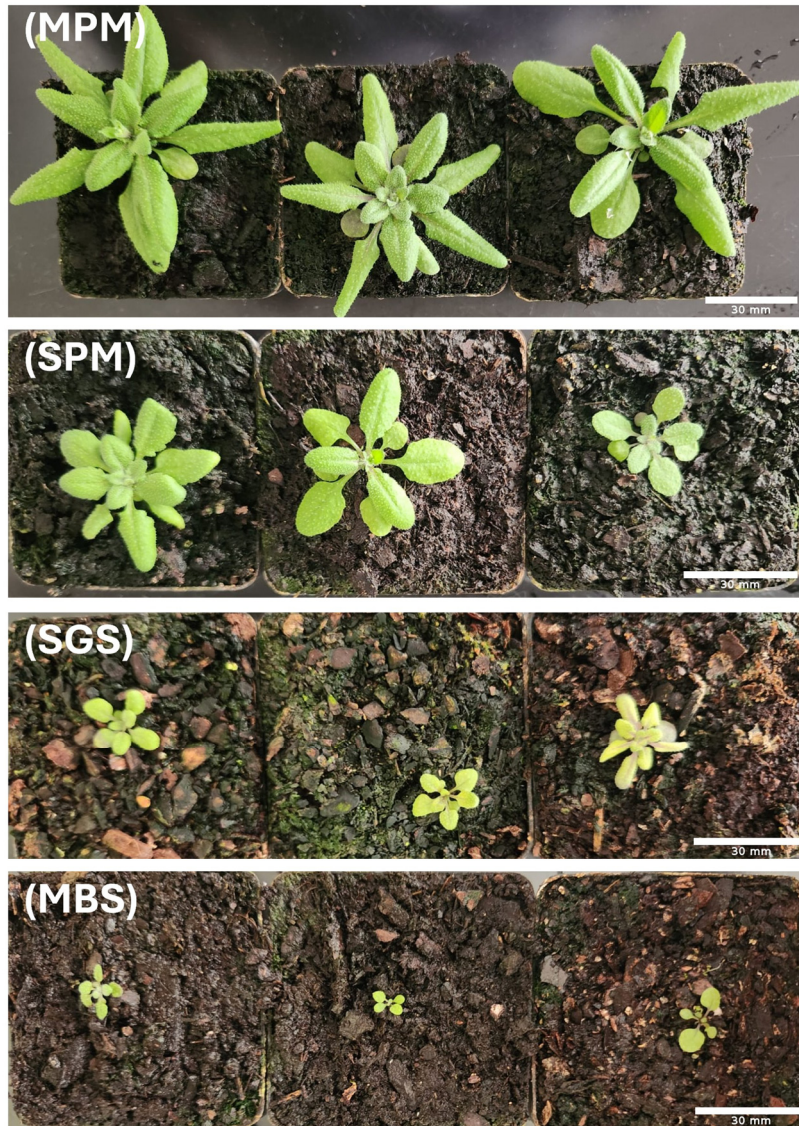
As shown in Figs. 4 and 5, both *A. thaliana* and *N. tabacum* also exhibited clear substrate-dependent morphological variation, including differences in overall plant size and leaf expansion. These visual observations are consistent with the quantitative biomass data and further support the conclusion that substrate type strongly influences post-germination growth in these two model plants.



**Figure 3:** Effect of substrate type on dry biomass in *N. tabacum* and *A. thaliana*. (A) Mean leaf dry weight and (B) mean stem dry weight of *N. tabacum* plants measured 60 days after sowing. (C) Mean whole-plant dry weight of *A. thaliana* plants measured 75 days after sowing. Bars represent group means, error bars indicate SD, and dots represent individual pot-level observations pooled from three independent runs, with five pots per substrate per run for *N. tabacum* and ten pots per substrate per run for *A. thaliana*. Data were analyzed using two-way ANOVA with substrate and experimental run as fixed factors, followed by Tukey's HSD test. Different lowercase letters indicate significant differences between the substrates for *N. tabacum* ( $p < 0.0001$ ) and *A. thaliana* ( $p < 0.001$ ); bars sharing the same letter are not significantly different ( $p > 0.05$ ).



**Figure 4:** Substrate type–dependent morphological variation in *N. tabacum* seedlings after 28 days of growth. Representative images show plants grown in four tested potting substrates (MPM, SPM, SGS, MBS as labeled in panels). Differences in overall plant size and leaf expansion are evident among treatments, indicating that substrate type influences early vegetative development. Scale bar = 30 mm.



**Figure 5:** Substrate-dependent morphological variation in *A. thaliana* seedlings after 28 days of growth. Representative images show plants grown in the four tested substrates. Differences in overall plant size and leaf expansion are evident among treatments, indicating that the substrate influences early vegetative development. Scale bar = 30 mm.

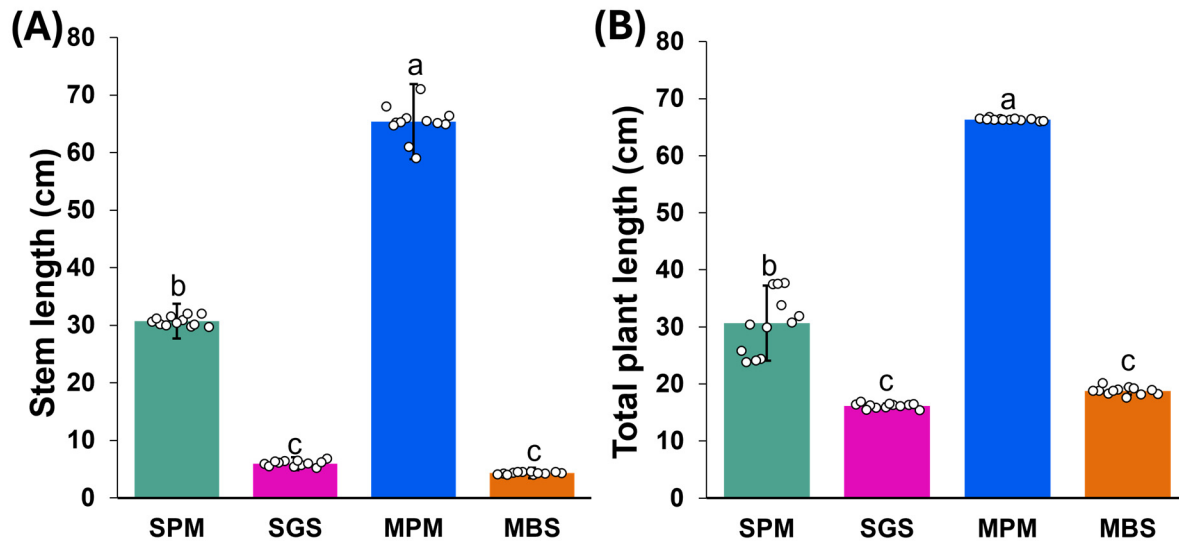
### 3.3 Plant Height

Substrate type had a significant effect on stem length in *N. tabacum* and total plant length in *A. thaliana*. Among the tested substrates, MPM consistently produced the greatest values for these growth traits.

For *N. tabacum*, two-way ANOVA showed a highly significant effect of substrate type on stem length ( $p < 2e-16$ ). Plants grown in MPM had significantly longer stems than those grown in SPM, SGS, and MBS (Fig. 6A). MBS produced the shortest stems and differed significantly from both SPM and MPM. No significant difference in stem length was detected between SGS and MBS ( $p > 0.05$ ).

Similarly, for *A. thaliana*, substrate type had a highly significant effect on total plant length ( $p < 2e-16$ ). Plants grown in MPM exhibited significantly greater total plant length than those grown in SPM, SGS, and MBS ( $p < 0.0001$ ; Fig. 6B). No significant difference was observed between SGS and MBS ( $p > 0.05$ ).

Overall, these results indicate that MPM was the most favorable substrate for promoting shoot elongation in both *N. tabacum* and overall plant length in *A. thaliana* under the conditions tested, whereas MBS and SGS generally supported lower growth.



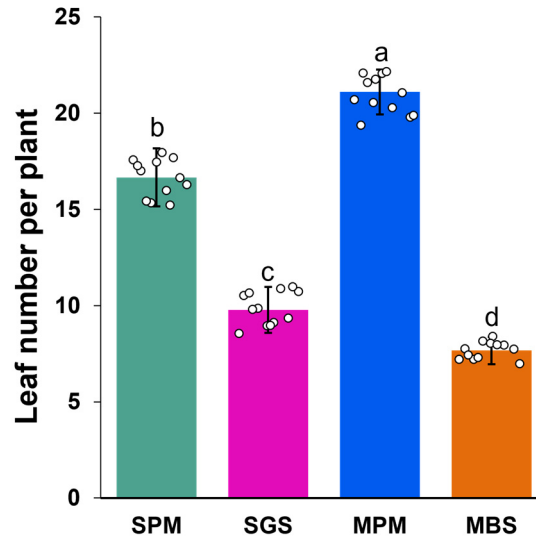
**Figure 6:** Effect of substrate type on stem length in *N. tabacum* and on total plant length in *A. thaliana*. (A) Mean stem length of *N. tabacum* plants measured 60 days after sowing. (B) Mean total plant length of *A. thaliana* plants measured 75 days after sowing. Total plant length was measured by gently removing each Arabidopsis plant from the media, straightening the root, and measuring the distance from the root tip to the apex of the main stem. Individual pot-level observations were pooled from three independent runs, with five pots per substrate per run for *N. tabacum* and ten pots per substrate per run for *A. thaliana*. Data were analyzed using two-way ANOVA with substrate and experimental run as fixed factors, followed by Tukey's HSD test. Different lowercase letters indicate statistically significant differences among the substrates for *N. tabacum* ( $p < 0.0001$ ) and *A. thaliana* ( $p < 0.0001$ ); bars sharing the same letter are not significantly different ( $p > 0.05$ ).

### 3.4 Leaf Number per Plant

Substrate type had a significant effect on leaf number in *N. tabacum* ( $p < 2e-16$ ), with MPM consistently producing the greatest leaf number per plant ( $p < 0.001$ ; Fig. 7). In contrast, MBS produced the fewest leaves and differed significantly from the other substrates. SGS produced fewer leaves than SPM but more than MBS, and these differences were statistically significant ( $p < 0.001$ ).

Experimental run had a smaller effect on leaf number than substrate type, and the interaction between substrate and experimental run was not significant. These results indicate that the effect of substrate on leaf production was consistent across independent runs.

Overall, MPM consistently supported greater dry biomass, plant length, and leaf production values under the conditions tested. Compared with SGS and MBS, the finer texture and greater water-holding capacity (Table 1) likely improved root-substrate contact and nutrient uptake. In addition, the higher reported nitrogen and potassium content of MPM may have contributed to enhanced overall growth. SPM showed intermediate performance, while SGS and MBS generally produced lower growth values.



**Figure 7:** Leaf number per plant of *N. tabacum* plants measured 60 days after sowing under different substrates. MPM produced the greatest leaf number per plant. Bars represent group means, error bars indicate SD, and dots represent individual pot-level observations pooled across three independent experimental runs, with five pots per substrate in each run. Data were analyzed using two-way ANOVA with substrate and experimental run as fixed factors, followed by Tukey's HSD test. Different lowercase letters indicate statistically significant differences among substrates ( $p < 0.001$ ); bars sharing the same letter are not significantly different ( $p > 0.05$ ).

#### 4 Discussion

The four potting substrates had no detectable effects on the germination of *N. tabacum* or *A. thaliana* under the controlled conditions used in this study. Pairwise comparisons likewise showed no significant differences in germination among the tested substrates. These findings are consistent with previous studies indicating that germination in *A. thaliana* is strongly influenced by genetic and environmental factors, including seasonal timing, photoperiod, and temperature [4]. Similarly, earlier work has shown that germination is governed primarily by environmental factors such as moisture and temperature, whereas substrate effects are often more evident during seedling establishment after germination [5].

The absence of substrate effects on *N. tabacum* germination further suggests that early seed establishment in this species is relatively robust under favorable environmental conditions. In contrast, substrate-dependent germination responses have been reported in studies comparing markedly different natural soils, such as sand and loess [10]. In the present study, however, all tested substrates were commercially formulated for plant cultivation and were therefore likely less divergent than natural soils in their basic growth-supporting properties. This likely explains the absence of significant differences in germination among substrates. Taken together, these results indicate that, under controlled laboratory conditions with adequate moisture and temperature, multiple commercially available substrates can support reliable germination of these two model species.

In contrast to germination, substrate type had a pronounced effect on post-germination growth in both *A. thaliana* and *N. tabacum*. Among the tested substrates, MPM consistently supported greater dry biomass accumulation, greater plant length, and, in *N. tabacum*, higher leaf number. This superior performance is likely associated with both the reported nutrient profile of MPM and its favorable physical characteristics. Compared with the other substrates, MPM contained higher reported levels of nitrogen, phosphorus, and potassium, exhibited a finer texture, contained perlite to enhance aeration and greater water-holding

capacity. These combined characteristics likely promoted more favorable root-zone conditions for water uptake, nutrient acquisition, and sustained plant growth.

Previous studies have similarly shown that peat-based substrates can support greater biomass accumulation in *Arabidopsis* than nutrient-poor or peat-free alternatives such as coir [11]. Nutrient availability, particularly nitrogen, is also known to influence biomass accumulation, shoot development, and leaf expansion [12–14]. Our results are consistent with these established relationships. However, the present findings also indicate that nutrient concentration alone is unlikely to explain the observed differences among substrates. Physical properties such as porosity, compaction, aeration, and moisture retention are also important determinants of root development and overall plant performance. Thus, the stronger performance of MPM likely reflects the combined effects of chemical and physical characteristics of the components rather than nutrient availability alone. While leaf number served as a practical indicator of growth in this study, future analyses incorporating leaf area measurements could provide additional resolution for substrate-dependent differences in canopy development and biomass accumulation.

The structural characteristics of the substrates also likely contributed substantially to the observed differences in plant growth. Fine-textured substrates with balanced porosity can promote more uniform water distribution and oxygen diffusion in the root zone, thereby supporting root development and improving nutrient uptake efficiency. By contrast, SGS and MBS contained coarser particles and more heterogeneous organic components, which may have reduced root–soil contact and limited nutrient accessibility in the small containers used in this study. Physical constraints such as compaction and reduced pore space are known to restrict root growth and influence plant performance [15–17]. These factors likely contributed to the lower biomass accumulation and reduced plant height observed in these substrates.

In addition to nutrient availability and physical structure, substrate salinity may also influence plant growth responses. Elevated electrical conductivity in growing media can impose osmotic stress, reduce water uptake, and limit root elongation and shoot development. *A. thaliana* is known to be sensitive to salt and osmotic stress, and increased salinity can negatively affect early growth and biomass accumulation [18]. Because electrical conductivity was not directly measured in the present study, the contribution of substrate salinity to the observed growth differences cannot be determined. Nevertheless, salinity remains an additional factor that may interact with substrate structure and nutrient availability to influence root-zone conditions and plant performance. Direct measurement of substrate electrical conductivity in future studies would help clarify this possibility.

Potassium availability may have further contributed to the observed growth differences, particularly in *N. tabacum*, where potassium uptake has been associated with improved growth and physiological performance [19–22]. In addition, the presence of slow-release fertilizer in MPM may have supported more sustained nutrient availability throughout the experimental period [23]. At the same time, the present study did not include nutrient-supplementation treatments or direct physicochemical measurements of the substrates. As a result, the relative contributions of nutrient supply, electrical conductivity, water dynamics, and physical structure could not be fully separated. This should be considered when interpreting the mechanistic basis of the observed differences.

Importantly, the comparatively lower performance of SGS and MBS in this study should not be interpreted as a general limitation of these substrates. Their coarser structure, slower nutrient release, and higher proportions of composted bark and peat may be advantageous for woody ornamentals, perennials, and longer term greenhouse experiments, where enhanced drainage, root aeration, and gradual mineralization support sustained growth and reduce the risk of waterlogging [24]. Although these properties were less suitable for the small potted systems used here, they may be beneficial under other

cultivation conditions. Recognizing these context-dependent strengths broadens the practical relevance of the present findings.

Several limitations should also be acknowledged. First, detailed physicochemical properties were not determined experimentally for the substrates, including electrical conductivity and direct measurements of nutrient availability. Second, the study focused on growth performance under a specific laboratory setup and therefore does not capture all possible substrate-by-environment interactions. Third, although the selected growth traits were appropriate for comparing substrate performance in routine cultivation, additional parameters such as leaf area, root architecture, and reproductive output could provide a more comprehensive assessment of plant responses. In addition, because *A. thaliana* plants were harvested 75 days after sowing, the resulting measurements should be interpreted as overall post-germination growth rather than strictly early vegetative growth. Future studies incorporating direct substrate characterization, earlier stage-specific sampling, and expanded phenotypic measurements will help refine the mechanistic interpretation of substrate-dependent growth differences.

Overall, our results show that while substrate type does not strongly influence germination under controlled conditions, it plays an important role in post-germination growth. By considering nutrient availability together with physical structure and the potential contribution of salinity, this study identifies substrate selection as an important and often underreported variable in laboratory plant research. These findings underscore the value of careful substrate choice for improving growth consistency, plant health, and experimental reproducibility in controlled-environment studies using model plant systems.

## 5 Conclusions

The choice of potting substrate significantly influences post-germination growth of *A. thaliana* and *N. tabacum* under controlled laboratory conditions. Among the substrates tested, the commercially available Miracle-Gro potting mix (MPM) consistently supported the strongest overall growth performance, likely because of the combined effects of nutrient availability and favorable physical structure. These findings identify substrate choice as an important experimental variable affecting growth outcomes and reproducibility in laboratory-based plant research.

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**Author Contributions:** Ethan Brister: formal analysis; investigation; writing—review and editing. Ramtin Vamenani: conceptualization; formal analysis; investigation; methodology; writing—original draft. Ling Li: conceptualization; data curation; funding acquisition; investigation; methodology; project administration; resources; supervision; writing—review and editing. All authors reviewed and approved the final version of the manuscript.

**Availability of Data and Materials:** The authors confirm that the data supporting the findings of this study are available within the article.

**Ethics Approval:** Not applicable.

**Conflicts of Interest:** The authors declare no conflicts of interest.

## Appendix A

Sta-Green Potting Mix Plus Fertilizer (SPM) is formulated for a wide range of flowering plants and vegetables. It contains a blend of organic materials, including composted bark and peat moss, which provide good drainage and aeration while retaining moisture. SPM is enriched with a slow-release fertilizer containing nitrogen (0.10%), phosphate (0.08%), and potassium (0.06%), designed to support initial plant growth. Regionally formulated, it also includes ingredients such as sphagnum peat moss, horticultural perlite, and dolomitic limestone, which adjusts soil pH. The polymer-coated fertilizer provides a steady release of nutrients, ensuring sustained availability over time.

Sta-Green Flower & Vegetable Garden Soil (SGS) shares similar characteristics with SPM but differs slightly in its physical composition. With a lower nutrient content—nitrogen (0.05%), phosphate (0.04%), and potassium (0.03%)—SGS may provide less sustained nutrient availability compared to SPM. However, it also features a polymer-coated slow-release fertilizer to maintain nutrient levels, although at the lower concentrations. SGS is designed to cater to a variety of garden plants but may offer less nutritional support for high-demand plants.

Miracle-Gro Potting Mix (MPM) is a commercially popular soil for container plants. It features a lighter and airier structure, likely due to the inclusion of perlite, which enhances aeration and prevents soil compaction. MPM is enriched with Miracle-Gro plant food, designed to feed plants for up to six months. The nutrient profile is significantly higher than SPM and SGS, with 0.21% nitrogen, 0.11% phosphate, and 0.16% potassium. A portion of these nutrients is coated for slow release, providing a long-term nutrient supply. The improved drainage and high nutrient availability make MPM a favorable option for promoting plant growth in various conditions.

Miracle-Gro Raised Bed Soil (MBS) is optimized for use in raised beds, with a focus on moisture retention and proper drainage. It is formulated with a combination of peat, processed forest products, sphagnum peat moss, and organic materials such as poultry litter, bone meal, and earthworm castings. MBS provides moderate levels of nitrogen (0.09%), phosphate (0.08%), and potassium (0.09%), with slow-release nitrogen accounting for 0.054% of the total nitrogen. The product's formulation varies by region but consistently aims to maintain adequate soil moisture and provide long-term nutrient availability, which makes it suitable for raised bed applications but potentially less effective in container environments.

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