

## URBAN VEGETATION DECLINE UNDER *BRACHYCHITON DISCOLOR* F. MUELL. TREE

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### Abstract

*Brachychiton discolor* F.Muell. (Malvaceae) is a deciduous ornamental tree native to Australia, which has gained ecological significance in urban ecosystems worldwide. This study evaluates the impact of leaf litter from *B. discolor* canopies on ground cover and plant diversity in urban environments. Areas under *B. discolor* canopies exhibited significant reductions in the cover of *Cynodon dactylon* (L.) Pers., *Euphorbia hirta* L., *Malva parviflora* L., *Medicago polymorpha* L., *Melilotus indicus* (L.) All., *Poa annua* L., and *Sisymbrium irio* L., along with Shannon-Wiener diversity (H') and evenness indices (E) declined. Moreover, leaf litter from *B. discolor* increased soil organic matter by 18.6% and enhanced the concentrations of certain nutrients, including potassium (16.24%) and copper (11.07%), in the affected urban soils. Greenhouse experiments demonstrated that leaf residues of *B. discolor* significantly inhibited *C. dactylon* germination (by 37.1%) and reduced the establishment of *C. dactylon* (by 54.6%) and *M. parviflora* (by 58.64%). This tree much reduced the light intensity under the tree canopy. High-performance liquid chromatography analysis identified key phenolic and flavonoid allelochemicals, including ellagic acid, pyrogallol, and benzoic acid among phenolics and kaempferol among flavonoids. These results suggested that reduced vegetation cover and diversity under *B. discolor* are primarily driven by its leaf litter and shading effects. The allelopathic impact of its chemical compounds appeared more pronounced than its mechanical effects. Consequently, *B. discolor* significantly alters urban vegetation cover and diversity, highlighting the need for conservation strategies to mitigate its impact and protect urban ecosystems.

**Key words:** Allelopathy, Weeds, HPLC, *Brachychiton discolor*, Urban ecology.

### Introduction

The leaf litter of different trees can be found in many terrestrial habitats, particularly in sites where deciduous trees exist. In these locations, leaf residues from trees may spread and accumulate during the leaf abscission period. The litter of these trees was a factor influencing cover, composition, diversity and dynamics of the associated weed vegetation (Facelli & Pickett, 1991; Hassan *et al.*, 2021). It inversely affects seed germination, seedling establishment and plant recruitment in different habitats (Xiong & Nilsson, 1999). Tree litter in urban environments is a persistent event that occurs continuously and may cause problems for urban green spaces. The studies regarding the potential impacts of tree litter have been increasing worldwide, predominantly in the grasslands and forests (Loydi *et al.*, 2013; Bai *et al.*, 2023). Nevertheless, a few studies were also conducted to explore the ecological role of tree litter on the green cover in urban environments.

Allelopathy is a common ecological phenomenon concerning the potential release of some metabolic compounds called allelochemicals, which influence germination, establishment and development of the associated plants (Rice, 1984). These allelochemicals could be secreted from plants to the immediate environment from their roots or leached from the different plant residues, including stems and leaves (Djurđević *et al.*, 2004, 2011; Hassan *et al.*, 2014a and b). Tree species can also exhibit allelopathic stress against understory vegetation through their leaf litter via the allelopathy process (Lorenzo *et al.*, 2011; Hassan, 2018). Another effect of litter that should be considered is its mechanical effect. The litter of trees, particularly those of broad-

leaved ones, represents a mechanical block preventing the possible extension of plant shoot systems of the emerging seedlings (Bosy & Reader, 1995). Therefore, the overall effect of litter may depend primarily upon litter quantity in the affected sites.

Under field conditions, plant residues affect soil characteristics, including the physical and chemical structures that principally affect germination and growth of the coexisting species (Batish *et al.*, 2002; Hassan *et al.*, 2014b). Tree litter may alter the soil conditions to change the cover and diversity of coexisting weeds (Krishna & Muhan, 2017). On the other hand, tree canopy may create a shading condition causing reduction in light availability that certainly affects emergence, establishment and growth performance of the understory species.

*Brachychiton* are tree species native to eastern Australia (Forster *et al.*, 2022). Recently, these trees have been widely introduced for ornamental purposes to hot, dry regions such as the Mediterranean, South Africa, the United States and the Kingdom of Saudi Arabia (Rathie, 2014; Yassin *et al.*, 2021; Bieler *et al.*, 2014). *Brachychiton discolor* F.Muell. (Malvaceae) is a typical rainforest tree species. Leaves of this tree showed potent anti-allergic and anti-inflammatory activities as they contain different biologically active compounds (Thabet *et al.*, 2018). Besides, many phenolic ingredients were characterised in the leaves of such trees (Ragheb *et al.*, 2021). However, the allelopathic potential and ecology of such tree have not been investigated yet. This tree's leaf litter may persist on the understorey vegetation in the urban gardens, revealing some adverse effects in the urban green areas. In this study, these issues are undertaken.

In our initial field study, the understory vegetation of *Brachychiton discolor* exhibited distinct gaps, forming a mosaic pattern of vegetation interspersed with bare sites often covered by the leaf litter. This study investigates how the leaf litter from *B. discolor* influence understory vegetation, including potential allelopathic or mechanical effects and modifications to soil properties that may reduce plant cover and diversity. This study aims to evaluate the impact of *Brachychiton discolor* on urban vegetation through three main hypotheses:

1. Soil modification: *B. discolor* alters soil properties through litter deposition. To test this, soil and vegetation characteristics were compared between plots under the tree canopy and outside its influence.
2. Allelopathic and mechanical effects: The leaf litter of *B. discolor* negatively affects the emergence, establishment, and growth of understory species through both chemical and mechanical mechanisms. A greenhouse experiment was conducted using two target understory species to assess the mulching effects of litter at varying doses. The mechanical impact was simulated using inert materials, while potential allelochemicals, including phenolics and flavonoids, were identified via high-performance liquid chromatography (HPLC).
3. Shading effects: The tree canopy reduces light availability, potentially suppressing understory vegetation. Light intensity was measured under and outside tree canopies to quantify this effect.

## Material and Methods

**Field study:** The area under study was well-illustrated by Hassan & Hassan (2019). This study was conducted during the 2022-2023 growing seasons. To investigate the impact of introduced *Brachychiton discolor* trees on understory vegetation, 40 plots of five m<sup>2</sup> each were randomly selected. These plots included vegetation under the tree canopy and in adjacent areas unaffected by the trees. Twenty plots affected by litter under the tree canopy were designated as treatments, while the remaining plots served as controls. The studied *B. discolor* trees were approximately 20 years old, 20 to 30 meters tall, with canopies spanning about 5 meters in width. Plot selection was made during mid-spring and late summer to capture seasonal vegetation variations and assess the influence of the trees on both winter and summer plant species.

**Vegetation characteristics:** Following vegetation characteristics were assessed in each plot.

1. Species richness (S) was determined by enumerating the species monitored in each plot.
2. The cover value of each plant species and the total vegetative cover of all plants found were measured, with plant cover estimated as the area (m<sup>2</sup>) on the soil employed by every species. The relative cover for a given plant was determined by the equation: relative cover = (cover of species i / cover of all species) × 100. Bare areas, or areas that were not covered by vegetation, were also recorded.

3. Additional diversity indices, namely: Shannon-Wiener (H'), Evenness (E), and Simpson's (D) indices for diversity, were useful to evaluate the floristic diversity at each location (Pielou, 1975). Besides, the incident sunlight was monitored by measuring the incident illumination intensity onto the soil via a digital lux meter (Victor 1010A, China). Species identification and scientific naming followed Boulos (1999, 2000, 2002, 2005).

**Soil analysis:** Representative soil samples from each site were collected from 0-20 cm depth and combined to form a composite sample for each plot. These samples were air-dried, sieved through a 2 mm sieve to remove plant residues, and stored in plastic bags for potential analyses. The soil parameters measured were measured as follows:

1. Soil pH and EC were determined in a soil-water extract (1:5 w/v). Soil pH was measured using a digital pH meter (AD 3000), while soil EC was determined employing a conductivity meter (Jenway 3305).
2. Soil moisture content was measured by subtracting the weight of the oven-dried soil from the weight of the freshly collected soil, and thereafter dividing by the weight of the dry soil. Soil water content was expressed as a percentage (%).
3. Available soil N, P, K, and Zn were measured using atomic absorption spectrometry following Allen's (1989) methods.
4. Soil organic carbon (OC) was investigated via the Walkley and Black (1934) method.

## Greenhouse investigation

**Litter and soil collection:** Leaf litter from *B. discolor* was collected at the end of winter 2023, a period characterised by abundant leaf residues and tree stress. The litter was sliced into small portions and stored in a dark, cold chamber until needed. Concurrently, soil samples were collected from *Brachychiton*-free sites, specifically from the upper 0-20 cm soil surface. This soil was air-dried, sieved through a 2 mm sieve, and placed in plastic black pots (13 cm in diameter × 18 cm in depth).

**Target understory-tested species:** Two native understory species, *Cynodon dactylon* Pers. (Poaceae) and *Malva parviflora* L. (Malvaceae), were selected for their ecological relevance to the study area (Hassan & Hassan, 2019). These species represent two different families and classifications (monocotyledons and dicotyledons), thus expecting to cover a wide range of responses in the plant world. Both weeds exhibit lower cover under the tree canopy than in *Brachychiton*-free areas.

**Effect of tree litter:** Twenty seeds of each target species were equally scattered at a depth of 0.2 cm in each pot, and chopped dry leaves of *B. discolor* evenly spread on the soil surface in treated pots at 4.5 g and 10 g, reflecting the typical litter quantities in the field. Litter-free pots were marked as control.

A second set of pots was prepared with small pieces of lightweight plastic bars corresponding to 4.5 g and 10 g to evaluate the mechanical effect of the litter. Untreated pots served as controls.

**Residual toxicity in soil:** To investigate the potential toxicity of the soils under the tree canopies, adequate soil samples were collected from the sites which were under the tree canopies and used as substrate for germination and growth of the above-mentioned target species, *Cynodon dactylon* and *Malva parviflora*. The soils just outside the tree canopies, i.e. neither affected by tree litter nor superficial roots of the tree, were used as control. This method could be common to study the soil under the species affecting vegetation (AL-Huqail *et al.*, 2025). Both treatment and control labelled soils were inserted in 500 ml plastic rounded pots and maintained the same field conditions. The seeds of the target species were placed at 0.3 cm depth, and irrigation was performed regularly by a drop-wise process when needed.

**Measurements:** The experiments were conducted under natural environmental conditions using a fully randomised design, with 5 replicates per treatment. The study lasted four weeks, during which the number of emerging individuals was recorded daily. Once emergence ceased, the total number of plants in each pot was counted and expressed as the percentage of seeds sown that successfully emerged. Establishment percentage was calculated as the proportion of individuals that completed the emergence process. Additionally, root depth, shoot height, and biomass were measured for each pot.

#### HPLC analysis for measurement of free phenolics and flavonoids in *B. discolor* leaf litter

**(a) Extraction:** Free phenolic and flavonoid compounds were extracted using the Lin *et al.*, (1996) procedure.

**(b) HPLC system:** The analysis was achieved by HPLC (Agilent 1100) and the Agilent ChemStation software. The HPLC system was composed of two LC- pumps and a UV-DIODE ARRAY detector adjusted at 280 nm to detect the existing phenolics and flavonoids. Compounds were separated in a Kromasil C18 column of dimensions 125 mm × 4.60 mm and 5 µm particle size. The mobile phase in the HPLC system is composed of two solvents 0.1% methanol: phosphoric acid (50: 50 v/v) in an isocratic mode for possible separation of phenolics with a continuous flow rate adjusted to 1.0 ml min<sup>-1</sup>. Different standards of phenolics (Sigma Aldrich, USA) were used to identify the compounds in the plant extracts. Concentrations of these compounds were calculated using the area under peaks from the standards.

**Statistical analyses:** Primarily, the data gathered were first examined regarding their normality and homogeneity via Kolmogorov–Smirnov and Levene's tests, respectively. The parametric independent T-test analysed the normal and homogeneous results of the field trial and the test due to residual toxicity in soil. The non-parametric Mann-Witney U test was performed when both data types were abnormal.

The data representing the effect of litter, as the dose applied in both testes, in the greenhouse that showed normality and homoscedasticity were examined by the parametric test (i.e. the one-way ANOVA) followed by the parametric Duncan post-hoc test for pairwise comparison of means. When the data were not normal, the Kruskal-Wallis H test was applied. Dunn's test was accomplished for a potential non-parametric pairwise comparison of means. To discriminate between the effects of dry leaf litter and the mechanical effect only represented by the plastic pieces, the independent-sample T-test was applied to compare each pair of the same amount (i.e., 4.5 vs 4.5 and 10 vs 10 g per pot, respectively). A two-way ANOVA was also carried out to check the interactive effect of the litter amount applied and the type of litter on the measured criteria. SPSS software package version 20.0 (IBM Corporation, USA) was used to conduct these analyses at  $p < 0.05$  and  $p < 0.01$ .

## Results

**Fieldwork:** Nine species from six families were identified in the vegetation associated with *Brachychiton discolor* (Table 1). Only *Cynodon dactylon* was perennial among these, while the remaining species were annuals. Most species in the understory vegetation, except for *Coronopus didymus* and *Eragrostis pilosa*, exhibited lower cover values under the tree canopy compared to open areas (Table 1). The total plant cover beneath the canopy was significantly reduced, with a corresponding increase on bare ground ( $p < 0.01$ ) (Table 2). Additionally, plant diversity, as indicated by Shannon-Wiener and Simpson indices, was significantly lower under the canopy than in open areas ( $p < 0.05$ ) (Table 2).

The reduction in understory vegetation was accompanied by a significant decrease in the intensity of sunlight beneath the tree canopy ( $p < 0.01$ ) (Table 2). Soil analysis revealed notable differences between litter-affected and litter-free soils (Table 3).

## Greenhouse trial

**Effect of litter:** The effects of *Brachychiton discolor* leaf litter on the emergence, establishment, and growth of the tested species were influenced primarily by both the type of litter (leaf litter or plastic fragments) and application rate (Table 4). At the lower application rate, *B. discolor* leaf litter significantly reduced the biomass of *Malva parviflora*, although other variables, such as emergence and establishment, remained unaffected. This reduction in biomass suggests that the leaf litter may have inhibited the overall growth of *M. parviflora* through both chemical and physical mechanisms, possibly involving allelopathic compounds released from the litter or physical shading effects. In contrast, plastic fragments applied at the same rate did not show any significant effects on either species, suggesting that the observed suppression was due to the chemical properties of the leaf litter rather than merely the mechanical presence of material on the soil surface. Notably, leaf litter had a more pronounced inhibitory effect on root growth of *M. parviflora* compared to plastic fragments, pointing to the potential role of allelopathic compounds such as phenolics and flavonoids, which are commonly found in the litter of many plant species and can interfere with root development (Table 4).

At the higher application rate, *B. discolor* leaf litter caused a significant reduction in the emergence of *Cynodon dactylon* and the establishment of both species. The higher dose likely led to more intense chemical interactions in the soil, possibly through the accumulation of organic acids or other allelochemicals that inhibited seed germination and seedling growth. Additionally, root depth and total biomass of *C. dactylon* and *M. parviflora* were markedly inhibited, suggesting that at higher litter doses, both chemical and mechanical effects (such as altered soil aeration or moisture retention) might have contributed in the reduction of growth. Shoot growth was more sensitive in *C. dactylon*, showing a noticeable reduction, while *M. parviflora* shoot growth was unaffected by this higher dose, indicating a species-specific response to the litter and its compounds. This differential sensitivity could be related to the varying ability of the species to tolerate or adapt to allelopathic stress or to their different root architecture and growth patterns.

Regarding mechanical effects, plastic fragments had minimal impact, causing only slight reductions in the root length of *M. parviflora* and biomass of *C. dactylon* (Table 4). These findings reinforce the idea that the primary inhibitory effects of the litter were

chemical, as the plastic fragments lacked the bioactive compounds present in the leaf litter.

Soils under the influence of *B. discolor* exhibited higher levels of total nutrients, as reflected by increased soil electrical conductivity (EC) ( $p < 0.01$ ), organic carbon (OC), and available potassium and copper ( $p < 0.05$ ).

Overall, the inhibitory effects of *B. discolor* leaf litter on emergence, establishment, and growth performance were significantly greater than those of plastic fragments, except for the shoot extension of *M. parviflora*, which was not affected by either treatment (Table 4). This suggested that chemical allelopathic interactions were the primary mechanism for the suppression of vegetation rather than mechanical effects from litter accumulation.

These findings were further supported by two-way ANOVA (Table 5), which revealed significant interactive effects between litter type and application rate. For *C. dactylon*, both factors significantly influenced all measured parameters, emphasising the combined influence of chemical and physical factors on plant growth. For *M. parviflora*, the amount of litter, its type, and their interaction were significant for establishment, root growth, and total biomass (Table 5), suggesting that the litter's quantity and chemical composition play a crucial role in influencing plant performance.

**Table 1. Mean covering area (m<sup>2</sup>) of the plant species detected in the plots with and without *Brachychiton discolor*.**

Species	Family	Outside tree canopy	Under tree canopy
<i>Coronopus didymus</i> (L.) Sm.	Brassicaceae	8.64	5.0
<i>Cynodon dactylon</i> (L.) Pers	Poaceae	42.12	9.76****
<i>Eragrostis pilosa</i> L.	Poaceae	18.0	9.0
<i>Euphorbia hirta</i> L.	Euphorbiaceae	11.15	8.00*
<i>Malva parviflora</i> L.	Malvaceae	12.24	6.00*
<i>Medicago polymorpha</i> L.	Fabaceae	14.4	6.50*
<i>Melilotus indicus</i> (L.) All.	Fabaceae	13.20	7.00*
<i>Poa annua</i> L.	Poaceae	12.64	6.0*
<i>Sisymbrium irio</i> L.	Brassicaceae	13.5	5.50**

\* Significant differences at  $p \leq 0.05$ ; \*\* Significant differences at  $p \leq 0.01$ ; \*\*\*\* Significant differences at  $p \leq 0.0001$ .

**Table 2. Total plant cover (%) and diversity indices (mean  $\pm$  SE) in the plots under and outside the tree canopy.**

Parameter	Outside tree canopy	Under tree canopy
Total plant cover (%)	72.06 $\pm$ 4.62	26.18** $\pm$ 3.31
Bare area (%)	27.94 $\pm$ 4.62	73.82** $\pm$ 3.31
Light intensity on soil (Lux)	114825.0 $\pm$ 3328.76	10255** $\pm$ 1234.0
Species richness (S)	2.82 $\pm$ 0.29	2.53 $\pm$ 0.24
Shannon-Wiener index (H')	0.88 $\pm$ 0.11	0.75* $\pm$ 0.11
Evenness index (E)	0.83 $\pm$ 0.77	0.74 $\pm$ 0.09
Simpson's index (D)	2.52 $\pm$ 0.26	2.19* $\pm$ 0.20

\* Significant differences at  $p < 0.05$ ; \*\* Significant differences at  $p < 0.01$

**Table 3. Soil properties (Mean  $\pm$  SE) in the plots under and outside the tree canopy.**

Soil parameter	Outside tree canopy	Under tree canopy	
pH	8.83 $\pm$ 0.034	8.79 $\pm$ 0.024	
Electrical conductivity ( $\mu\text{S cm}^{-1}$ )	449.17 $\pm$ 3.55	470.50 ** $\pm$ 6.30	
Soil moisture content (%)	8.25 $\pm$ 2.09	5.04 $\pm$ 1.72	
Organic carbon (%)	2.193 $\pm$ 1.24	2.60 * $\pm$ 1.47	
<b>Available nutrients</b>			
<b>Available</b>	N ( $\text{mg g}^{-1}$ soil)	17.46 $\pm$ 0.57	17.11 $\pm$ 0.62
	P ( $\text{mg g}^{-1}$ soil)	27.76 $\pm$ 1.18	29.13 $\pm$ 2.76
	K ( $\text{mg g}^{-1}$ soil)	65.70 $\pm$ 3.37	76.37* $\pm$ 2.87
	Zn ( $\mu\text{g g}^{-1}$ soil)	187.04 $\pm$ 2.51	187.5 $\pm$ 3.70
	Cu ( $\mu\text{g g}^{-1}$ soil)	3.07 $\pm$ 0.08	3.41* $\pm$ 0.13

\* Significant differences at  $p < 0.05$ ; \*\* Significant differences at  $p < 0.01$

**Table 4. Effects of leaf litter and mechanical effect of *Brachychiton discolor* leaf litter on emergence, establishment, root length, shoot height and biomass (mean  $\pm$  SE) of some understory species.**

Target species	Control	Litter amount (g pot <sup>-1</sup> )	
		4.5	10
<b>Emergence (%)</b>			
<i>Cynodon dactylon</i>	97.0 $\pm$ 2.0 (97.0 $\pm$ 2.0)	91.0 $\pm$ 2.45 (94.0 $\pm$ 2.92)	61.0 <sup>****</sup> $\pm$ 4.0 (83.0 $\pm$ 6.25) <sup>SS</sup>
<i>Malva parviflora</i>	82.0 $\pm$ 3.74 (82.0 $\pm$ 3.74)	78.0 $\pm$ 3.74 (80.0 $\pm$ 3.16)	66.0 $\pm$ 7.48 (80.0 $\pm$ 3.16) <sup>SS</sup>
<b>Establishment (%)</b>			
<i>Cynodon dactylon</i>	97.0 $\pm$ 2.0 (97.0 $\pm$ 2.0)	91.0 $\pm$ 2.45 (94.0 $\pm$ 2.92)	46.0 <sup>****</sup> $\pm$ 3.67 (83.0 $\pm$ 6.25) <sup>S</sup>
<i>Malva parviflora</i>	82.0 $\pm$ 3.74 (82.0 $\pm$ 3.74)58.6 % 58.64	78.0 $\pm$ 3.74 (80.0 $\pm$ 3.16)	34.0 <sup>****</sup> $\pm$ 2.45 (80.0 $\pm$ 3.16) <sup>SS</sup>
<b>Root length (cm)</b>			
<i>Cynodon dactylon</i>	2.82 $\pm$ 0.11 (2.82 $\pm$ 0.11)	2.72 $\pm$ 0.15 (2.72 $\pm$ 0.15)	1.28 <sup>***</sup> $\pm$ 0.14 (2.48 $\pm$ 0.09) <sup>SS</sup>
<i>Malva parviflora</i>	5.56 $\pm$ 0.42 (5.56 $\pm$ 0.42)	4.64 $\pm$ 0.22 (5.94 $\pm$ 0.27) <sup>S</sup>	2.0 <sup>****</sup> $\pm$ 0.14 (3.88 <sup>*</sup> $\pm$ 0.35) <sup>S</sup>
<b>Shoot height (cm)</b>			
<i>Cynodon dactylon</i>	2.14 $\pm$ 0.10 (2.14 $\pm$ 0.10)	2.16 $\pm$ 0.12 (2.16 $\pm$ 0.12)	1.06 <sup>**</sup> $\pm$ 0.07 (2.16 $\pm$ 0.09) <sup>SS</sup>
<i>Malva parviflora</i>	5.94 $\pm$ 0.36 (5.94 $\pm$ 0.36)	6.18 $\pm$ 0.37 (5.30 $\pm$ 0.21)	5.92 $\pm$ 0.12 (5.48 $\pm$ 0.18)
<b>Biomass (mg pot<sup>-1</sup>)</b>			
<i>Cynodon dactylon</i>	25.2 $\pm$ 1.85 (25.2 $\pm$ 1.85)	23.0 $\pm$ 1.41 (24.6 $\pm$ 1.72)	11.6 <sup>****</sup> $\pm$ 1.21 (19.2 <sup>*</sup> $\pm$ 0.86) <sup>S</sup>
<i>Malva parviflora</i>	191.40 $\pm$ 9.02 (196.20 $\pm$ 12.21)	155.0 <sup>*</sup> $\pm$ 8.66 (198.0 $\pm$ 12.80)	45.0 <sup>****</sup> $\pm$ 4.47 (172.0 $\pm$ 8.60) <sup>SS</sup>

† Values outside the parentheses are the effects due to leaf litter, whereas those found inside them are the effects due to mechanical effect of plastic pieces.

\* Significant differences from control at  $p \leq 0.05$  according to Duncan's test

\*\* Significant differences from control at  $p \leq 0.01$  according to Duncan's test

\*\*\* Significant differences from control at  $p \leq 0.001$  according to Duncan's test

\*\*\*\* Significant differences from control at  $p \leq 0.0001$  according to Duncan's test

<sup>S</sup> Significant difference between the effect of litter mulching and that of the corresponding litter amount amended with soil at  $p < 0.05$  according to independent-Samples T-test

<sup>SS</sup> Significant difference between the effect of litter mulching and that of the corresponding litter amount amended with soil at  $p < 0.01$  according to independent-Samples T-test

**Table 5. F values of the two-way ANOVA for the effect of the litter amount, type of litter applied and their interactive effects on the measured criteria in the tested species.**

Parameter	Litter amount	df	Litter type	df	Litter amount $\times$ litter type	df	Error
<i>Cynodon dactylon</i>							
Emergence (%)	49.61 <sup>***</sup>	2	21.33 <sup>***</sup>	1	16.89 <sup>***</sup>	2	24
Establishment (%)	27.50 <sup>***</sup>	2	8.10 <sup>**</sup>	1	5.51 <sup>*</sup>	2	24
Root length (cm)	34.24 <sup>***</sup>	2	15.42 <sup>***</sup>	1	15.42 <sup>***</sup>	2	24
Shoot height (cm)	18.36 <sup>***</sup>	2	19.03 <sup>***</sup>	1	19.03 <sup>***</sup>	2	24
Biomass (mg)	24.01 <sup>***</sup>	2	6.03 <sup>*</sup>	1	3.43 <sup>*</sup>	2	24
<i>Malva parviflora</i>							
Emergence (%)	2.14	2	2.17	1	1.46	2	24
Establishment (%)	32.88 <sup>***</sup>	2	33.88 <sup>***</sup>	1	29.82 <sup>***</sup>	2	24
Root length (cm)	40.14 <sup>***</sup>	2	16.29 <sup>***</sup>	1	4.48 <sup>*</sup>	2	24
Shoot height (cm)	0.41	2	3.61	1	1.20	2	24
Biomass (mg)	24.01 <sup>***</sup>	2	6.03 <sup>*</sup>	1	3.43 <sup>*</sup>	2	24

\* Significant results at  $p < 0.05$ ; \*\* Significant results at  $p < 0.01$ ; \*\*\* Significant results at  $p < 0.001$

**Residual toxicity in soil:** Soils beneath the *Brachychiton discolor* canopy exhibited a significant reduction in the emergence and biomass of *Cynodon dactylon*. The impact was also evident in the suppressed root growth observed in both tested species. These results suggested that the soil under the tree canopy, potentially influenced by accumulated leaf litter and other factors, creates an inhospitable environment for plant establishment and development. The decreased emergence and biomass, along with reduced root growth, indicate a residual toxicity effect of the soil, which may be related to the chemical composition of the litter and its interaction with soil properties (Fig. 1). This residual effect emphasises the potential role of *B. discolor* in altering soil conditions and negatively impacting the growth of understory vegetation.

**HPLC analysis:** High-performance liquid chromatography (HPLC) analysis of *Brachychiton discolor* leaf litter revealed the presence of several free phenolic acids and flavonoids, indicating a complex chemical composition with potential allelopathic properties (Table 6). Eight phenolic acids and five flavonoids were identified in the leaf litter.

Among the phenolic acids, ellagic acid was the most abundant, followed by pyrogallol, benzoic acid, and syringic acid, suggesting that these compounds may contribute to the inhibitory effects observed in the vegetation. Regarding flavonoids, kaempferol was the most predominant, and it is known for its biological activity and potential role in plant interactions. These findings highlight the chemical diversity within *B. discolor* leaf litter, which may play a significant role in suppressing understory vegetation through both allelopathic and mechanical effects (Table 6).

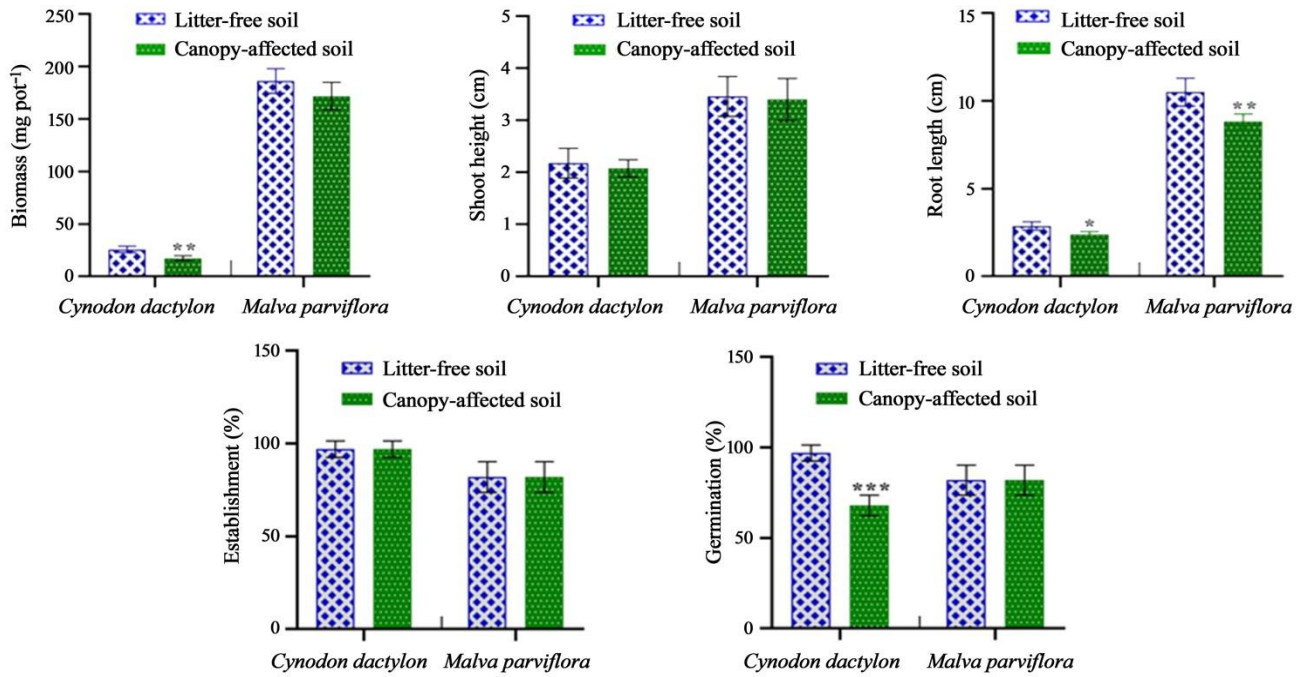


Fig. 1. Germination (%), establishment (%) and the measured growth criteria of the target species when grown in the soil under the tree canopy (canopy-affected soil) and soil outside tree canopy.

**Table 6. Contents (Mean  $\pm$  SD) of different phenolic acids and flavonoids measured in the leaf litter of *B. discolor*.**

Compounds	Concentration $\mu\text{g g}^{-1}$ litter
<b>Phenolic acids</b>	
Benzoic acid	14.60 $\pm$ 1.14
Chlorogenic acid	7.58 $\pm$ 2.73
Ellagic acid	19.84 $\pm$ 5.96
Ferulic acid	6.18 $\pm$ 1.23
Pyrogallol	16.29 $\pm$ 6.10
Salicylic acid	3.25 $\pm$ 1.04
Syringic acid	11.39 $\pm$ 4.86
Trans-cinnamic acid	6.41 $\pm$ 0.65
<b>Flavonoids</b>	
Catechin	11.45 $\pm$ 0.46
Kaempferol	20.10 $\pm$ 6.80
Luteolin	8.76 $\pm$ 3.33
Myricetin	7.04 $\pm$ 1.16
Naringenin	9.18 $\pm$ 2.92

## Discussion

**Vegetation cover and soil characteristics:** This study demonstrated that native vegetation cover and diversity indices were significantly lower under the canopy of *Brachychiton discolor*. This suppression can be attributed to several factors, including the influence of tree litter. Soil analysis revealed that litter-affected soils under the canopy exhibited higher electrical conductivity (EC), likely due to the accumulation of ions from decomposing leaf litter. This finding aligns with Ouyang *et al.* (1998), who reported that increased soil nutrients, such as nitrogen (N) and potassium (K), can significantly elevate EC. In this case, the elevated concentrations of K and copper (Cu) detected under the canopy are consistent with the presence of these elements in the leaf litter of *B. discolor*.

Although K toxicity is rare due to plants' regulatory mechanisms for potassium ion uptake (White *et al.*, 2021), the role of Cu warrants further investigation; while Cu concentrations were higher under the canopy, they remained below phytotoxic thresholds (Gharbi *et al.*, 2005). Therefore, it is unlikely that the observed suppression of vegetation cover and diversity is solely due to the measured soil characteristics. Instead, the interaction between nutrient enrichment, allelochemical release, and shading likely contributes to reducing diversity and cover under the canopy.

**Role of leaf litter:** The accumulation of leaf litter on the soil surface can create unique microenvironmental conditions that affect seed germination and growth. Previous studies have shown that litter modifies light availability and soil temperature, negatively impacting seedling establishment (Hassan *et al.*, 2024; Zhang *et al.*, 2021). In this study, the canopy and litter of *B. discolor* restricted sunlight penetration, likely reducing the cover and diversity of coexisting species. Limited light availability is a critical factor regulating seed germination and plant growth, which typically peaks under full sunlight (Broncano *et al.*, 1998; Chen *et al.*, 2023).

Phytochemical analysis of *B. discolor* leaf litter revealed the presence of phenolic acids and flavonoid compounds, such as ellagic acid and kaempferol, known for their phytotoxic properties (Pardo-Muras *et al.*, 2020a; Mousavi *et al.*, 2021). These allelochemicals are released during litter decomposition and have been shown to inhibit germination and growth of other species (Al Harun *et al.*, 2015; Chaves *et al.*, 2015; Zhang *et al.*, 2015; Hassan, 2018). Additionally, studies have indicated that volatile compounds in *B. discolor* may inhibit microbial activity and probably seedling growth (Thabet *et al.*, 2020). Releasing these compounds at different times of the year could contribute to the reduced floristic cover and diversity under the canopy.

**Species-specific responses in the greenhouse experiment:**

Greenhouse experiments confirmed that the effects of *B. discolor* litter were species-specific and concentration-dependent. At low concentrations, litter had no impact on the emergence, establishment, or growth of the tested species. However, germination, establishment, and growth were significantly inhibited at high concentrations, with more pronounced effects on *Cynodon dactylon* than *Malva parviflora*. These results align with previous studies showing species-specific responses to allelopathic effects of leaf litter (Ahmed *et al.*, 2008). Such variations can be attributed to differences in the physiological traits and seedling morphology of the species.

Although grasses like *Cynodon* species are generally less susceptible to the effects of litter (Barritt & Facelli, 2001), the results showed that *Cynodon* species was more sensitive than *Malva* to both the chemical effects of the litter and the canopy-affected soil. This suggests that the inhibitory effects of *B. discolor* on *Cynodon* may involve not only allelopathy but also competition for resources and root exudates from the tree, as observed in other tree species (Fernandez *et al.*, 2021).

**Mechanical vs. chemical effects:** Using plastic fragments to simulate the mechanical effect of litter demonstrated that their inhibitory effects on germination and growth were minor compared to natural leaf litter (Ruprecht *et al.*, 2010; Pardo-Muras *et al.*, 2020b). This supports the idea that the observed suppression in the field was primarily due to chemical effects (allelopathy) rather than physical barriers. These results are consistent with prior studies showing that litter affect neighbouring species through multiple pathways, including physical suppression, nutrient alteration, and chemical inhibition (Elgersma *et al.*, 2012; Asplund *et al.*, 2018). Overall, the results indicate that allelopathic compounds in *B. discolor* litter play a dominant role in inhibiting the target species' germination, establishment, and growth. This reinforces the need to consider chemical and physical effects in future studies on plant-plant interactions in urban and natural environments.

**Conclusion**

This study demonstrated that *Brachychiton discolor* leaf litter negatively impacts the cover and diversity of urban vegetation, with the intensity of the effect depending on the amount of litter. The inhibitory effects are likely due to both the litter's chemical (allelopathic) and mechanical properties, as well as phytotoxins released into the soil or through root exudates. Despite the enrichment of organic matter and nutrients in litter-affected soils, the decline in plant cover and diversity cannot be solely attributed to changes in soil conditions. These findings underline the potential threat of *B. discolor* litter to urban vegetation and highlight the importance of further research to confirm these results. Promoting native species in urban landscapes is crucial for preserving biodiversity and enhancing quality of life.

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