

Hemiparasitic features and antioxidant activity of *Dendrophthoe pentandra* and *Scurrula atropurpurea* on *Diospyros rhodocalyx*

NONTHIWAT TAESUK¹, SIRITHIDA BOONPEN¹, KULLASATREE MANEE¹, PASAKORN BUNCHALEE¹, TEERAPORN KATISART¹, VACHIRAPORN PIKULTHONG², SUTHIRA MANEECHAI^{1,*}

¹Department of Biology, Faculty of Science, Mahasarakham University, Kham Riang, Kantharawichai District, Maha Sarakham 44150, Thailand. Tel./fax.: +66-2160-1143, *email: suthira.m@msu.ac.th

²Division of Biology, Department of Science, Faculty of Science and Technology, Suan Sunandha Rajabhat University, U-Thong nok Rd., Dusit, Bangkok 10300, Thailand

Manuscript received: 25 February 2025. Revision accepted: 16 May 2025.

Abstract. Taesuk N, Boonpen S, Manee K, Bunchalee P, Katisart T, Pikulthong V, Maneechai S. 2025. Hemiparasitic features and antioxidant activity of *Dendrophthoe pentandra* and *Scurrula atropurpurea* on *Diospyros rhodocalyx*. *Biodiversitas* 26: 2531-2539. Mistletoe, a hemiparasitic plant, exhibits broad ecological adaptability, establishing parasitic relationships with a wide range of host species across diverse habitats and environmental conditions. *Dendrophthoe pentandra* and *Scurrula atropurpurea*, belonging to the family Loranthaceae, represent prominent hemiparasitic species with ecological and phytochemical significance. This study investigated the characteristics and phytochemical composition of mistletoe species growing on shared host trees. The phenolic and flavonoid contents, as well as the antioxidant activities (DPPH and ABTS assays) of these two hemiparasitic mistletoe species were examined in relation to their common host plant, *Diospyros rhodocalyx*. Comparative distribution analysis revealed that *D. pentandra* was more widely distributed across host trees than *S. atropurpurea*. Morphological assessments of reproductive structures indicated that *D. pentandra* produced larger and more vividly colored flowers and fruits compared to *S. atropurpurea*. The phytochemical profiles, total phenolic and flavonoid contents, and antioxidant activities of the leaf and stem extracts were investigated using different extraction solvents. Crude extracts were prepared by Soxhlet extraction with dichloromethane, ethanol, and ethyl acetate. The ethanol extract of *D. pentandra* stems provided the highest total phenolic content as determined by the Folin-Ciocalteu method. Antioxidant activities were assessed using the DPPH and ABTS assays, with the ethyl acetate extract of the stem from *S. atropurpurea* showing the highest antioxidant activity at IC₅₀ values of 0.282±0.005 and 0.483±0.041 µg/mL, respectively. These findings provide insights into the ecological interactions between flower and fruit characteristics, chemical profiles, and potential bioactivities of mistletoe species.

Keywords: Antioxidant activity, *Dendrophthoe pentandra*, *Diospyros rhodocalyx*, Mistletoe-hemiparasitic plants, *Scurrula atropurpurea*

INTRODUCTION

Dendrophthoe pentandra (L.) Miq. and *Scurrula atropurpurea* (Blume) Danser are hemiparasitic plants belonging to the Loranthaceae family, commonly known as mistletoes. Hemiparasites partially rely on host plants for water, nutrients, and minerals, but are capable of photosynthesis to generate a portion of their energy. Both these species attach to the branches or trunks of host plants using specialized structures called haustoria, which are adaptable to various host types, allowing hemiparasites to establish themselves on various species (Teixeira-Costa and David 2021). *D. pentandra* and *S. atropurpurea* exhibit key hemiparasitic traits, including photosynthesis, specialized haustoria for nutrient uptake, and host adaptability. Their tubular, brightly colored flowers attract pollinators, while fleshy, bird-dispersed fruits aid propagation. Despite their parasitic nature, they support biodiversity by providing food and habitat for birds and insects, contributing to ecosystem stability and seed dispersal (Těšitel et al. 2021).

D. pentandra is known for its bright flowers and broad host range, often infesting fruit trees and ornamental species. *S. atropurpurea* has dark purple flowers and infects various plant species, though it may exhibit some

host preference. Understanding their parasitic behavior helps manage the impact of these two mistletoe species on economically and ecologically significant host plants while recognizing their roles in the broader ecosystem. Bird-mediated seed dispersal is a cornerstone in the establishment and proliferation of mistletoes. Kuramana et al. (2020) described the hemiparasitic features of the mistletoe *Dendrophthoe falcata* (L.fil.) Blume, which has vibrant tubular flowers that attract avian pollinators and sticky, berry-like fruits that aid in seed dispersal. These adaptations enhance host attachment, reproduction, and ecological interactions; thereby, ensuring plant survival. Mucho et al. (2022) comprehensively reviewed host defenses, including mechanical and chemical barriers. These findings complement the ecological observations by revealing the long-term stress imposed on host plants, including reduced water and nutrient availability. Biochemical analyses have indicated that the bioactivity of mistletoe indicated that mistletoe is closely tied to its host. Phytochemical profiling of *D. pentandra* revealed a diverse array of secondary metabolites, including alkaloids, flavonoids, and terpenoids, with significant antioxidant, anticancer, and antidiabetic properties. This connection was also explored by Awang et al. (2023), who demonstrated that host species influence

the concentration and efficacy of bioactive compounds. Morphological adaptations observed in *D. pentandra* provide a tangible link between host-derived biochemical variations and ecological significance, offering a multidimensional understanding of host-parasite relationships. Taxonomic studies, such as the phenetic analysis of *D. pentandra*, underscore the high morphological variability among populations associated with different host species. Variations in leaf thickness, stomatal density, and reproductive structures suggest adaptive responses to host-specific microenvironments (Adiansyah et al. 2023).

Previous studies on *S. atropurpurea* and *D. pentandra* have highlighted their host-dependent phytochemical profiles and bioactivities, which are driven by flower and fruit characteristics, as well as biochemical interactions with various host plants. Both mistletoes exhibit key bioactive compounds, including quercetin, catechin, tannins, and flavonoids, which are associated with antioxidant, anticancer, and antihypertensive properties, though the concentration and efficacy of these metabolites vary depending on the host (Endharti et al. 2018; Yismairai et al. 2019; Bunchalee et al. 2022). Mistletoe has been traditionally used in herbal medicine because its bioactive compounds exhibit antioxidant, anti-inflammatory, and anticancer properties. Several studies (Kasmiyati and Kristiani 2022; Kong et al. 2023) have highlighted the pharmacological potential of mistletoes by identifying bioactive compounds with antioxidative and anticancer properties, thereby positioning mistletoes as promising candidates for drug development. Despite these advancements, significant gaps remain in integrating ecological and phytochemical findings to provide a cohesive understanding of mistletoe

biology. The exploration of mistletoe-host relationships in diverse ecosystems can offer insights into their evolutionary strategies and potential applications in sustainable agriculture and pharmacology.

This study investigated the phytochemical composition and antioxidant activities of two hemiparasites growing on the same host tree, *Diospyros rhodocalyx*. The objective was to determine how a shared host affects its secondary metabolite profiles and antioxidant potential, and to provide insights into its ecological interactions and pharmacological relevance.

MATERIALS AND METHODS

Study area

The localities for investigating a type of mistletoe host plant were conducted at Mahasarakham University (MSU) in Thailand across three locations. The first site was the Faculty of Science and Technology, located in Kham Riang Subdistrict on the MSU main campus (16°14'51.13"N, 103°15'3.48" E). The second site was the President Building, also situated in Kham Riang Subdistrict within the main campus (16°14'39.14" N, 103°14'56.53" E). The third site included the Faculty of Medicine and the MSU city campus, both part of Mahasarakham University's former campus (16°12'0.03"N, 103°17'4.66"E) (Figure 1). All maps were created using ArcGIS Pro version 2.9.6 (Esri 2021). The host trees of mistletoes were located using a Geographic Information System (GPS).

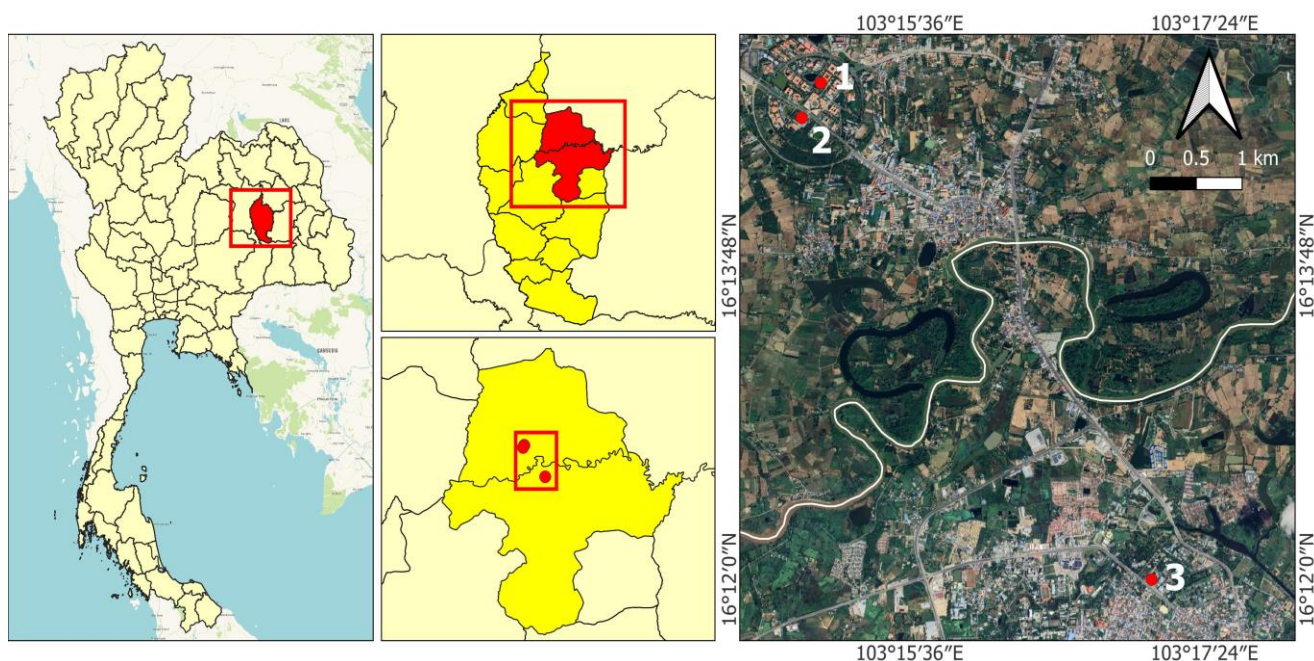


Figure 1. Location of the study area: 1. Faculty of Science and the Faculty of Technology area; 2. President Building area; and 3. Faculty of Medicine and the MSU city campus, Mahasarakham, Thailand

Plant collection, morphological study, and preparation for phytochemical analysis

Samples of *D. pentandra* and *S. atropurpurea* were collected from *D. rhodocalyx* in its natural forest habitat between March 2022 and April 2024. The plants were identified, and voucher specimens were deposited in the herbarium of the Department of Biology, Faculty of Science, Mahasarakham University.

Flower and fruit characteristics

The flower and fruit characteristics, including shape and color, were recorded, and their sizes were measured individually using a vernier caliper.

Extract preparation

Samples of *D. pentandra* and *S. atropurpurea* were cleaned, air-dried, and ground into a fine powder for further analysis. 5 g of the powdered stem and leaf material were extracted using 400 mL of dichloromethane, ethyl acetate, and ethanol in a Soxhlet apparatus for 6 hours. This was followed by evaporation in a water bath at 60°C. The resulting extracts were stored at -4°C for future use.

Phytochemical analysis

Total phenolic content

The Total Phenolic Content (TPC) was measured using the Folin-Ciocalteu method and expressed as mg gallic acid equivalent per gram (mg GAE/g). The TPC was determined using a modified Folin-Ciocalteu colorimetric method as described by Amin et al. (2006). Gallic acid, at concentrations ranging from 12.5 to 100 mg/mL, was used as the standard. The plant extracts were dissolved in methanol, and 0.5 mL of each extract (1 mg/mL) was combined with 2.5 mL of Folin-Ciocalteu reagent and left to react at room temperature for 5 minutes. Then, 2 mL of Na₂CO₃ solution was added, thoroughly mixed, and the volume was adjusted to 5 mL with distilled water. The mixture was then incubated at room temperature for 2 hours. The absorbance was recorded at 760 nm using a UV-Visible Spectrophotometer (Thermo Scientific GENESYS 10S Series) manufactured by Thermo Fisher Scientific, USA. The TPC was determined using the gallic acid standard curve and expressed as mg of Gallic Acid Equivalent (GAE) per gram of extract.

Total flavonoid content

The Total Flavonoid Content (TFC) was determined using the aluminum chloride colorimetric assay and expressed as mg quercetin equivalent per gram (mg QE/g). The TFC was assessed using a modified version of the method described by Suriyaprom et al. (2021). Quercetin concentrations, ranging from 15.625 to 250 µg/mL, served as the standard. For each sample (500 µL at 1 mg/mL), 1,500 µL of 95% ethanol was added and incubated at room temperature for 6 minutes. Next, 100 µL of 10% aluminum chloride was added, and the mixture was allowed to stand for 5 minutes before 100 µL of 1 M potassium acetate was introduced. The volume was adjusted to 5,000 µL with distilled water, and the mixture was kept in the dark at room temperature for 30 minutes. The absorbance was

measured at 415 nm in triplicate. The TFC was calculated against the quercetin standard curve and expressed as mg of quercetin per gram of extract.

DPPH radical scavenging assay

The 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay was performed to evaluate the free radical scavenging ability, as described by Likhitwitayawuid et al. (2006). First, 100 µM DPPH solutions were prepared in methanol. Solutions of varying concentrations (1000, 500, 250, 125, and 62.5 µg/mL) were made for each crude extract and ascorbic acid, with plant extracts also prepared in methanol at a concentration of 1 mg/mL. Then, 180 µL of the DPPH solution was combined in a microplate with 20 µL of the extract solution. The mixtures were incubated in the dark at room temperature for 20 minutes, and the absorbance was measured at 517 nm using a microplate reader. The absorbances of the extract samples (As) were compared to methanol (blank, Ac), with ascorbic acid used as the standard. The percentage of radical scavenging activity was calculated using the following equation:

$$\text{DPPH radical scavenging activity (\%)} = \frac{Ac - As}{Ac} \times 100\%$$

ABTS radical scavenging assay

The ABTS assay was conducted following a modified version of the method used by Suriyaprom et al. (2021). The ABTS^{•+} cation radical was produced by reacting 7 mM ABTS in water with 2.45 mM potassium persulfate. The mixture was left in the dark at room temperature for 12 to 16 hours before use. The resulting ABTS solution was diluted with ethanol to achieve an absorbance of 0.700±0.02 at 734 nm. Solutions at varying concentrations (1000, 500, 250, 125, and 62.5 µg/mL) were prepared for each crude extract and ascorbic acid. Triplicate measurements were taken, using methanol as the blank and ascorbic acid as the standard. The percentage inhibition was calculated using the following equation:

$$\text{Percentage radical scavenging (\%)} = \frac{Ac - As}{Ac} \times 100\%$$

Statistical analysis

All statistical analyses were performed using R software version 4.1.2 (R Core Team 2021). One-way ANOVA followed by Tukey's post hoc test was used to assess differences in the phytochemical profiles of the samples. A *p*-value<0.05 was considered statistically significant.

RESULTS AND DISCUSSION

Flower and fruit characteristics

The inflorescence of *D. pentandra* is racemose, with obclavate flowers, there are developing from light green or green buds into bright yellow or orange when mature, and are covered with brownish appress stellate hairs. The pedicel is 10.0-15.0 mm long, bearing 5-7 florets arranged oppositely or suboppositely decussate. The peduncle is 2.5-4.0 mm long. The light greenish ovate bract, 2.0-2.5 by 2.0-

2.5 mm, is attached distally to the rachilla. The five sepals are cup-shaped, light green, connate, approximately 1.0 mm in depth and 3.0-3.5 mm in diameter. The five petals are also connate, with free and reflexed apex lobes when mature, 18-22 mm in length and 4.0-6.0 mm in width, expanding up to 12 mm when the flower blooms, the petal lobes 6.0-8.0 mm long. The five yellow stamens are adnate to the middle of the petals, with filiform-like filaments, 4.5-5.0 mm long. The filaments are attached to the base of the anthers. The linear ditheous anthers are 4.5-5.0 mm long with longitudinal apertures. The pistil consists of an inferior ovary that is ovate, green, 2.0-2.5 by 2.0-2.5 mm, with the ovule attached to the base of the ovary. The style is filiform, 18-22 mm long, terminating in a capitate stigma. The fruits are berry, ovate, 10-14 by 5-7 mm, yellow to reddish-orange when ripe, glabrous to glabrous. The exocarp, mesocarp, and endocarp are prominent, while the integuments (testa and tegment) develop into the viscin layer. The seed is oblong, 4.0-4.5 by 2.0-2.5 mm, with smooth seed coats and covered with a viscin layer (mucilage pectins) (Figures A, C, E and G).

The inflorescences of *S. atropurpurea* are racemose, slender, obclavate flowers and curved, developing from light brown in buds into light gray when mature, and covered with brownish, verticillate hairs. The pedicel 4.0-10.0 mm long, bearing 4-5 florets arranged oppositely decussate; the peduncle 4.0-8.0 mm long. The ovate bract, approximately 1.0 by 1.0 mm, is attached distally to the rachilla. The four sepals are very small and scale-like. The four petals are connate, with free and reflexed apex lobes when mature, 20-30 mm in length and 1.5-2.0 mm in width, expanding up to 10 mm when the flower blooms, the petal lobes 6.0-8.0 mm long. The four dark red stamens are adnate to the subapex of the petals, with filiform-like filaments, 3.0-3.5 mm long. The filament is attached to the base of the anthers. The linear ditheous anthers are 1.8-2.0 mm long with longitudinal apertures. The pistil consists of an inferior ovary that is obovate, light brown, 3.0-3.2 by 2.0-2.2 mm, with the ovule attached to the base of the ovary. The style is filiform, 25-28 mm long, terminating in a capitate stigma. The fruits are berry, obovate, 5.0-7.0 by 2.5-3.0 mm, light brown when ripe, with stellate hairs. The exocarp and mesocarp are prominent, while the endocarp develops into the viscin layer. The seed is oblanceolate, 6.0-6.5 by 1.0-1.4 mm, and the seed coat is present (Figures 2 B, D, F, and H)

The flowers, fruits, and seeds of *D. pentandra* are more showy and larger than those of *S. atropurpurea* when mature (Figure 2 and Table 1). These characteristics contribute to the more successful reproduction of *D. pentandra*, as pollinators can more easily detect its flowers compared to those of *S. atropurpurea*. Additionally, the fruits are clearly visible to birds, which are the primary animal aiding in seed dispersal (Figure 2 and Table 1). This observation corresponds with the findings of Adiansyah et al. (2023). Their morphological assessments of *D. pentandra* reveal significant variability across different host species, with adaptations observed in leaf thickness, stomatal density, and reproductive structures. *D. pentandra* produces larger and more vividly colored flowers and fruits compared to *S.*

atropurpurea, which likely enhances its attractiveness to pollinators and seed dispersers. Devkota and Kunwar (2006) also highlighted the complexity of mistletoe-avian interactions, emphasizing that while bright fruit colors crucial role in attracting birds. Additional traits, such as fruit availability, optimal fruit size, seed stickiness, and adaptive fruit displays, further influence seed dispersal. Collectively, these studies highlight the ecological and morphological adaptability of mistletoes, which is shaped by host-specific microenvironments and interactions with dispersal agents. In addition, the size and position of branches on the tree crown affect to propagation of mistletoe; if their less than 5 cm in diameter can found more clumps of them (Ma et al. 2020). Generally, mistletoe seed germination is influenced by light and temperature. Luo and Zhang (2013) tested the effects of these factors on *D. pentandra* seeds; their results revealed that light conditions and an optimal temperature of 20°C are most effective for seed germination. In addition, the factor that significantly contributes to the successful spread of mistletoe is birds. The germination percentage of seeds increases when the fruits are digested by birds compared to when the pericarp is manually peeled. Moreover, the viscin layer, a sticky mucilaginous tissue, is a critical factor that helps seeds adhere to host surfaces. Subsequently, the primary root penetrates the vascular tissue to absorb water and minerals from the host plants (Gedalovich et al. 1988). Although the seeds of *D. pentandra* and *S. atropurpurea* possess mucilage tissue, its origin differs between the two species. In *D. pentandra*, this mucilage tissue is derived from the ovule integuments. In comparison, in *S. atropurpurea*, it is transformed from the endocarp (Figures G and H). These findings suggest that the origin of the viscin layer can develop from various tissues in the pistils. The viscin mucilage major component of *Phoradendron californicum* and *Arceuthobium americanum* (Viscaceae), is composed of pectin sugar (xylose and arabinose), proteins (glycine and histidine). Meanwhile, *Phthirusa pyrifolia* (Loranthaceae) mucilage contented higher neutral sugars and proteins (Gedalovich-Shedletzky et al. 1989).

Sjakoer et al. (2021) evaluated the bioactivity and safety of phytochemicals derived from *S. atropurpurea* and *D. pentandra* as potential antihypertensive agents using an in silico approach. They identified key bioactive compounds, including quercetin, kaempferol, and casticin, which demonstrated significant ACE inhibitory activity, as well as other beneficial cardiovascular effects. These findings support the traditional use of these mistletoe species in the management of hypertension and highlight their potential for developing novel, effective, and low-toxicity therapeutic agents.

Distribution of host trees

The twenty-six host tree species, 23 genera and 14 families across three study areas were parasitized by the mistletoes. Mostly, twenty-two host species were infected by *D. pentandra*, while three host was occupied by both *D. pentandra* and *S. atropurpurea*, and only one host species was parasitized by *S. atropurpurea* (Table 2). These findings indicate that *D. pentandra* is more successfully

distributed than *S. atropurpurea* in the same environment. It means that the characteristics of flowers and fruits are keystones for dispersal. The Fabaceae family had the highest number of host species, with 10 species recorded.

Among these, seven were hemi-parasitized by *D. pentandra*, two by both mistletoe species, and only one infected by *S. atropurpurea*. Additionally, two host species from Annonaceae and one species from other families were occupies.

Table 1. Flower and fruit characteristics of *D. pentandra* and *S. atropurpurea*

Characteristics		<i>D. pentandra</i>)n = 10(<i>S. atropurpurea</i>)n = 10(
Flower characteristics	Length of corolla tube (mm)	18-22 12 mm 16.7-19.6	20-30
	Width of corolla tube (mm)	4.0-6.0 (-12 when flower blooms)	1.5-2.0 (-10 when flower blooms)
	Shape of corolla tube	Obclavate	Tubular and curved
	Color of corolla tube	Bright yellow or orange	Light gray
Fruit characteristics	Length of fruit (mm)	10-14	5.0-7.0
	Width of fruit (mm)	5-7	2.5-3.0
	Shape of fruit	Ovate	Obovate
	Color of fruit	Reddish-orange	Light brown

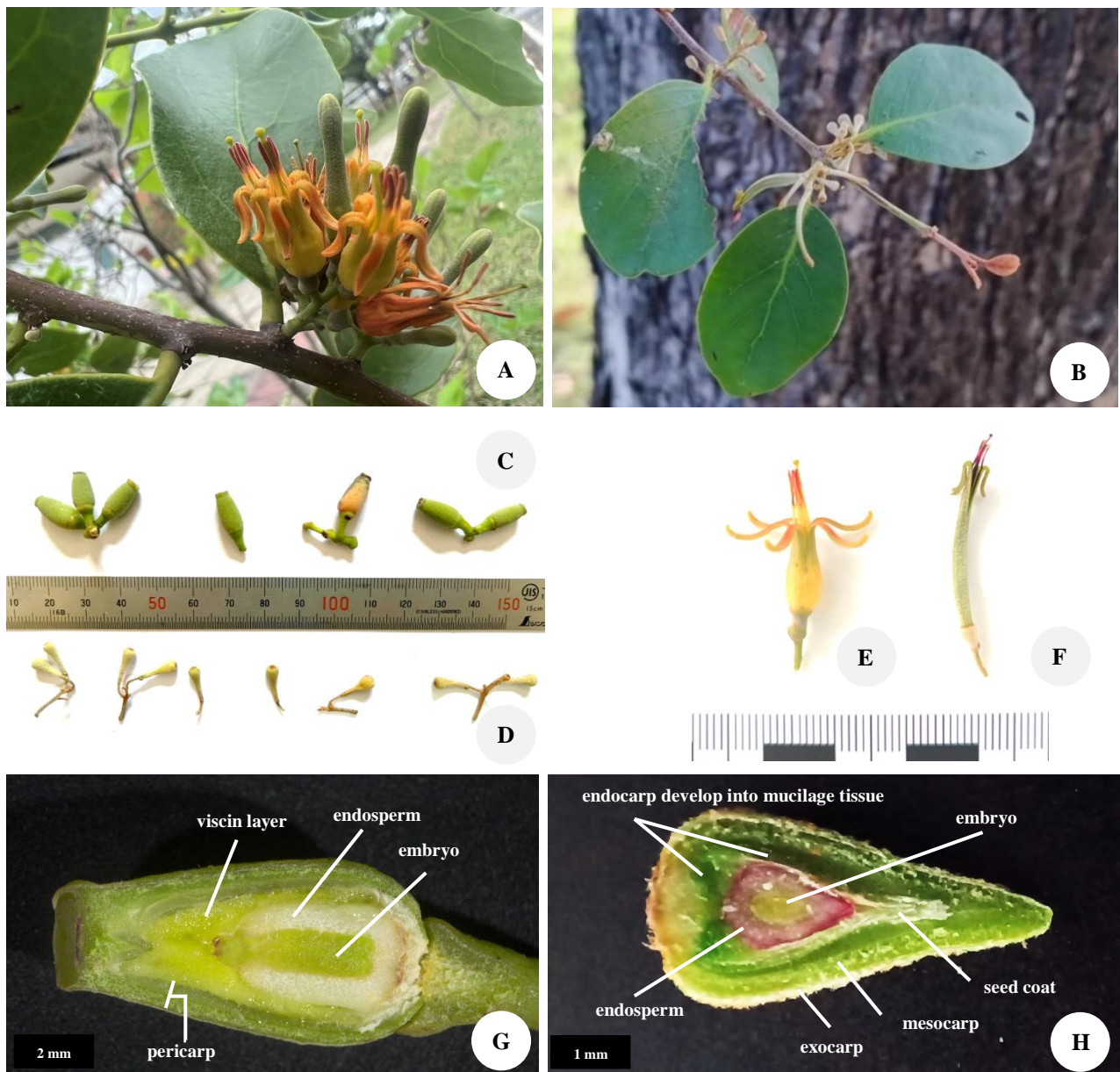


Figure 2. Flower and fruit of *D. pentandra* (A, C, E, G) and *S. atropurpurea* (B, D, F, H). Endocarp develops to mucilage tissue

Table 2. List of host trees of *D. pentandra* and *S. atropurpurea*

Mistletoe	Host plant	Host family	Study area
A	<i>Mangifera indica</i> L.	Anacardiaceae	1
A	<i>Huberantha cerasoides</i> Chaowasku	Anonaceae	2
A	<i>Uvaria siamensis</i> L.L.Zhou, Y.C.F.Su & R.M.K.Saunders	Anonaceae	1
A	<i>Terminalia ivorensis</i> A.Chev.	Combretaceae	1, 2, 3
A	<i>Anthoshorea roxburghii</i> P.S.Ashton & J.Heck.	Dipterocarpaceae	1, 2
A, B	<i>Diospyros rhodocalyx</i> Kurz	Ebenaceae	1, 2
A	<i>Butea monosperma</i> Kuntze	Fabaceae	1
A	<i>Cassia bakeriana</i> Craib	Fabaceae	1
A	<i>Cassia fistula</i> L.	Fabaceae	1
B	<i>Erythrophleum succirubrum</i> Gagnep.	Fabaceae	1, 2
A	<i>Peltophorum pterocarpum</i> K.Heyne	Fabaceae	1, 2, 3
A, B	<i>Pterocarpus indicus</i> Willd.	Fabaceae	1, 2, 3
A, B	<i>Samanea saman</i> Merr.	Fabaceae	1, 2
A	<i>Senna garrettiana</i> H.S.Irwin & Barneby	Fabaceae	1
A	<i>Senna siamea</i> H.S.Irwin & Barneby	Fabaceae	1
A	<i>Senna surattensis</i> H.S.Irwin & Barneby	Fabaceae	1
A	<i>Tectona grandis</i> L.f.	Lamiaceae	2
A	<i>Lagerstroemia floribunda</i> Jack	Lythraceae	1
A	<i>Lagerstroemia macrocarpa</i> Kurz	Lythraceae	1, 2, 3
A	<i>Bombax ceiba</i> L.	Malvaceae	1
A	<i>Swietenia macrophylla</i> King	Meliaceae	1
A	<i>Morus alba</i> L.	Moraceae	1
A	<i>Muntingia calabura</i> L.	Muntingiaceae	1
A	<i>Melaleuca viminalis</i> (Gaertn.) Byrnes	Myrtaceae	2
A	<i>Callistemon viminalis</i> (Sol. ex Gaertn.) G.Don	Myrtaceae	2
A	<i>Syzygium cumini</i> Skeels	Myrtaceae	2
A	<i>Schoutenia glomerata</i> King	TiLiaceae	1

Note: A: *D. pentandra*; B: *S. atropurpurea*. Study areas: Faculty of Science and Technology, located in Kham Riang Subdistrict on the MSU main campus; President Building, located in Kham Rieng Subdistrict on the MSU main campus; Faculty of Medicine and the MSU city campus, located in MSU city campus

Mistletoe-host specificity varied between the two species. *D. pentandra* exhibited a broad host range, parasitizing over 26 tree species, highlighting its ecological versatility and adaptability to diverse host chemistries. By contrast, *S. atropurpurea* was found on only four host species, suggesting a more specialized parasitic strategy.

Some host tree species can be found across study areas including *Lagerstroemia macrocarpa*, *Peltophorum pterocarpum*, *Pterocarpus indicus* and *Terminalia ivorensis*, exhibited a wide distribution and present in all the three study areas. The species *Anthoshorea roxburghii*, *Diospyros rhodocalyx*, *Erythrophleum succirubrum* and *Samanea saman* were found in both the study area 1 and 2. Anyway, some host trees were limited to a single study area, *Bombax ceiba*, *Butea monosperma*, *Cassia bakeriana*, *Cassia fistula*, *Lagerstroemia floribunda*, *Mangifera indica*, *Morus alba*, *Muntingia calabura*, *Schoutenia glomerata*, *Senna garrettiana*, *Senna siamea*, *Senna surattensis*, *Swietenia macrophylla* and *Uvaria siamensis* were found in the study area 1, while *Huberantha cerasoides*, *Melaleuca viminalis* and *Syzygium cumini* were found only in the study area 2.

Study areas 1 and 2 exhibited a high diversity of host tree species that were occupied by mistletoes, with multiple species shared between them, including *Shorea talura* and *D. rhodocalyx*. Study area 3 had fewer host tree species, primarily consisting of widespread trees such as *P. indicus*,

P. pterocarpum, and *T. ivorensis*. The distribution patterns of the mistletoes are shown in Figure 3.

Phytochemical composition and antioxidant activity

The phytochemical analysis revealed significant variations in TFC and antioxidant activities (DPPH and ABTS assays) among the host tree (*D. rhodocalyx*) and the two hemiparasitic plants (*D. pentandra* and *S. atropurpurea*) across different plant parts (stem and leaf) and extraction solvents (Table 3). The ethanol extracts generally had the highest bioactive compound contents and antioxidant activities, followed by ethyl acetate, while dichloromethane extracts were the least effective. *Scurrula atropurpurea* exhibited a higher Total Phenolic Content (TPC) and antioxidant activity compared to *D. pentandra* and *D. rhodocalyx*. This finding concurred with Rahmawati et al. (2014), who evaluated the antioxidant potential of five mistletoe species, including *Scurrula*, *Dendrophthoe*, and *Macrosolen*; they reported that *Scurrula* exhibited the highest antioxidant activity compared to *Dendrophthoe*. The HPLC analysis of mistletoe extracts identified key phenolic compounds, including gallic acid, catechin, epicatechin, rutin, and quercetin-4-glucoside.

Total phenolic content

Among the ethanol extracts, *D. pentandra* exhibited the highest TPC in the stems (414.26±4.06 mg GAE/g) and leaves (289.57±4.46 mg GAE/g), significantly exceeding

the values observed in *S. atropurpurea* and the host tree *D. rhodocalyx*. The lowest TPC was found in the ethanol stem extract of the host tree *D. rhodocalyx* (93.42±4.55 mg GAE/g). For ethyl acetate extracts, *S. atropurpurea* had the highest TPC in the stem (379.65±3.00 mg GAE/g), while

D. pentandra exhibited the highest TPC in the leaves (211.94±3.11 mg GAE/g). The dichloromethane extracts generally had lower TPC values across all species, with *D. pentandra* showing the highest leaf TPC (108.51±3.63 mg GAE/g).

Table 3. Total Phenolic Content (TPC), Total Flavonoid Content (TFC), and antioxidant activity (%) of the plants were extracted

Solvent	Part	Bioactive compounds	Species		
			<i>Diospyros rhodocalyx</i>	<i>Dendrophthoe pentandra</i>	<i>Scurrula atropurpurea</i>
Ethanol	Stem	TPC) mg GAE/g(93.42±4.55 ^c	414.26±4.06 ^a	375.88±2.17 ^b
		TFC) mg QE/g(9.44±1.67 ^b	12.96±1.53 ^b	19.57±1.35 ^a
		DPPH)%(37.91±4.57 ^b	69.15±4.08 ^a	66.74±0.60 ^a
		ABTS)%(31.88±4.37 ^b	66.18±2.44 ^a	35.12±4.62 ^b
	Leaf	TPC) mg GAE/g(104.80±2.38 ^c	289.57±4.46 ^a	148.91±3.47 ^b
		TFC) mg QE/g(40.11±2.72 ^c	193.21±2.60 ^a	64.04±0.60 ^b
Ethyl acetate	Stem	TPC) mg GAE/g(90.58±1.61 ^b	89.84±2.74 ^b	379.65±3.00 ^a
		TFC) mg QE/g(77.87±2.26 ^a	80.75±3.22 ^a	39.38±2.12 ^b
		DPPH)%(36.07±1.32 ^b	26.71±0.37 ^c	68.43±1.07 ^a
		ABTS)%(27.26±2.60 ^b	29.21±1.67 ^b	50.66±1.72 ^a
	Leaf	TPC) mg GAE/g(85.31±3.06 ^c	211.94±3.11 ^a	130.00±2.99 ^b
		TFC) mg QE/g(116.74±1.44 ^c	182.63±1.44 ^a	136.89±0.51 ^b
Dichloromethane	Stem	TPC) mg GAE/g(61.73±3.17 ^b	77.90±3.43 ^a	67.23±2.41 ^b
		TFC) mg QE/g(106.01±0.84 ^a	81.76±1.61 ^b	59.14±2.82 ^c
		DPPH)%(33.94±2.45 ^a	8.96±0.63 ^b	14.61±3.32 ^b
		ABTS)%(24.00±1.39 ^a	14.50±1.41 ^b	5.96±0.93 ^c
	Leaf	TPC) mg GAE/g(82.58±1.22 ^b	108.51±3.63 ^a	56.91±4.99 ^c
		TFC) mg QE/g(137.23±1.66 ^b	152.99±3.49 ^a	142.90±1.96 ^b
		DPPH)%(7.14±1.03 ^b	11.98±1.31 ^a	6.69±1.01 ^b
		ABTS)%(18.57±1.16 ^a	10.33±0.86 ^c	14.00±0.80 ^b

Note: Results are presented as mean±SD of duplicate measurements. Mean values in the same row with different superscript letters are significantly different (p<0.05)

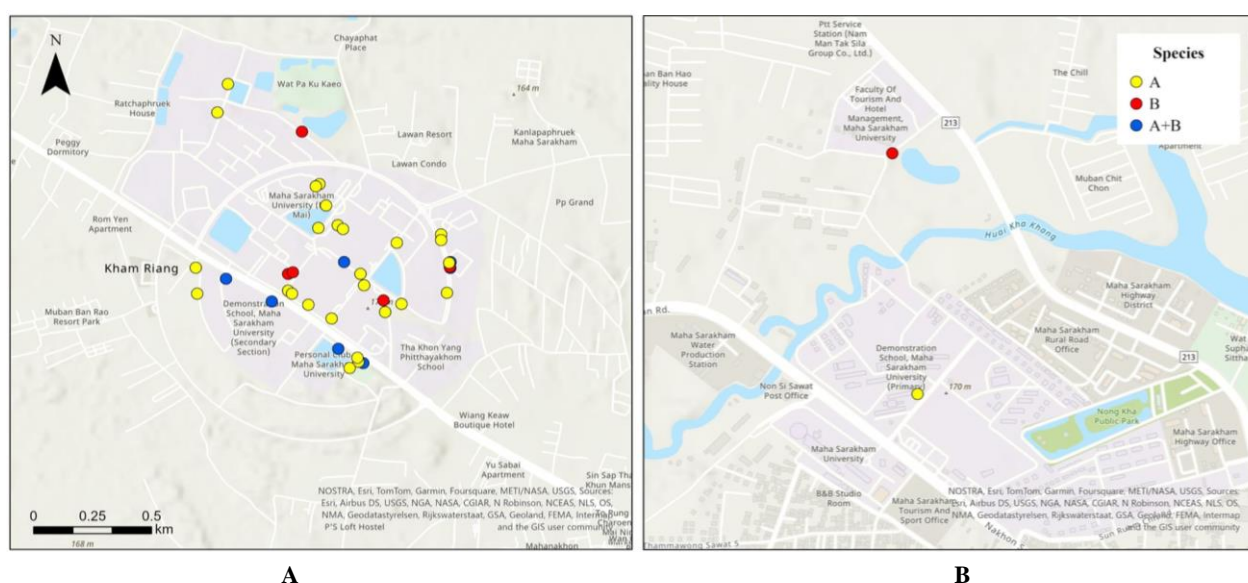


Figure 3. Distribution of hemiparasitic plant species and host trees in: A. The MSU main campus (study areas 1 and 2); and B. The MSU city campus (study area 3). Species are represented by colored circles, where yellow indicates *D. pentandra*, red represents *S. atropurpurea*, and blue denotes the presence of *D. pentandra* and *S. atropurpurea*

Total flavonoid content

D. pentandra leaf extracts had the highest TFC across all solvents, particularly in ethanol (193.21±2.60 mg QE/g), which was significantly higher than that of *S. atropurpurea* (64.04±0.60 mg QE/g) and *D. rhodocalyx* (40.11±2.72 mg QE/g). Ethyl acetate extraction yielded high flavonoid content in *D. pentandra* leaves (182.63±1.44 mg QE/g). The host tree *D. rhodocalyx* exhibited the highest TFC in dichloromethane stem extracts (106.01±0.84 mg QE/g), whereas *D. pentandra* and *S. atropurpurea* had lower values.

Antioxidant activity (DPPH and ABTS)

The antioxidant capacities varied depending on the extraction solvent. In the DPPH assay, ethanol extracts of *D. pentandra* showed the highest scavenging activity (69.15±4.08% in stems and 60.35±1.04% in leaves), while the host tree *D. rhodocalyx* had the lowest values (37.91±4.57% in stems and 31.92±2.32% in leaves). Ethyl acetate extraction gave the highest antioxidant activity in *S. atropurpurea* (68.43±1.07% in stems). Dichloromethane extracts displayed significantly lower DPPH activity across all species, with the highest activity found in *D. rhodocalyx* stems (33.94±2.45%).

In the ABTS assay, ethanol extracts of *D. pentandra* also demonstrated the highest antioxidant activity (66.18±2.44% in stems and 48.84±0.82% in leaves), while *S. atropurpurea* had moderate activity. Ethyl acetate extracts of *S. atropurpurea* stems also showed high ABTS scavenging activity (50.66±1.72%). Dichloromethane extracts yielded the lowest ABTS values, particularly in *S. atropurpurea* stems (5.96±0.93%).

Host influence on phytochemical profiles and bioactivity

The results indicated that *D. pentandra* exhibited a broader host range, possibly contributing to the greater variability observed in its secondary metabolite composition (Yee et al. 2017; Yismairai et al. 2019; Kristiningrum et al. 2020). By contrast, *S. atropurpurea*, which parasitizes fewer host species, may exhibit a more stable phytochemical profile, with consistent levels of bioactive compounds (Mustarichie et al. 2017; Yuniwati et al. 2018; Aditiyarini et al. 2022). Despite sharing *D. rhodocalyx* as a common host, the two mistletoes displayed distinct patterns of phenolic and flavonoid accumulation, suggesting that host-specific interactions influence their metabolic pathways differently. Interestingly, ethanol extracts of *S. atropurpurea* demonstrated superior antioxidant activity compared to *D. pentandra*, highlighting differences in their phytochemical compositions. The high antioxidant efficacy of *S. atropurpurea* may be attributed to its ability to synthesize potent bioactive compounds despite being restricted to a more limited range of host species (Sjakoer et al. 2021). These results further underscore the role of host-parasite interactions in shaping secondary metabolite production and bioactivity. The substantial accumulation of phenolics and flavonoids in *D. pentandra* and *S. atropurpurea* compared to their host tree suggests that hemiparasitic plants may possess adaptive advantages that enhance their survival and ecological success. The strong antioxidant potential of these mistletoes supports their traditional use in

herbal medicine and indicates their potential as a source of natural antioxidants for pharmacological applications.

In conclusion, this research underscores the importance of host-parasite dynamics in shaping phytochemical diversity and antioxidant capacity in hemiparasitic plants. Our findings suggest that *D. rhodocalyx* plays a significant role in influencing the secondary metabolite profiles of *D. pentandra* and *S. atropurpurea*. The two mistletoes exhibit distinct phytochemical responses despite sharing the same host. The observed differences in antioxidant activity further emphasize the potential pharmacological applications of these mistletoes, particularly *S. atropurpurea*, which demonstrated the highest bioactivity in ethanol extracts. Future studies should focus on long-term monitoring of mistletoe-host interactions, investigating how seasonal variations, environmental factors, and co-evolutionary pressures influence phytochemical production. Further exploration of the pharmacological properties of these bioactive compounds could provide new insights into their potential use in medicine and nutraceutical applications. These findings contribute to a broader understanding of the ecological and medicinal significance of hemiparasitic plants, paving the way for further research into their biochemical interactions with host trees and their potential role in natural product development.

ACKNOWLEDGEMENTS

This research project was financially supported by Mahasarakham University, Thailand.

REFERENCES

- Adiansyah NP, Islami PD, Purnomo P. 2023. Variation and phenetic relationships of *Dendrophthoe pentandra* (L.) Miq. from various host trees based on morphological characters. *Biogenesis: Jurnal Ilmiah Biologi* 11 (1): 48-58. DOI: 10.24252/bio.v11i1.34540.
- Aditiyarini D, Restiani R, Evieyana. 2022. Profiling secondary metabolites and antioxidant activity of tea mistletoe leaves (*Scurrula atropurpurea* (Bl.) Danser) in Nglinggo, Kulon Progo, Yogyakarta. *Biogenesis: Jurnal Ilmiah Biologi* 10 (2): 196-205. DOI: 10.24252/bio.v10i2.31258.
- Amin I, Norazaidah Y, Hainida KIE. 2006. Antioxidant activity and phenolic content of raw and blanched *Amaranthus* species. *Food Chem* 94 (1): 47-52. DOI: 10.1016/j.foodchem.2004.10.048.
- Awang MA, Daud NNNM, Ismail NIM, Abdullah FI, Benjamin MAZ. 2023. A review of *Dendrophthoe pentandra* (Mistletoe): Phytomorphology, extraction techniques, phytochemicals, and biological activities. *Processes* 11 (8): 2348. DOI: 10.3390/pr11082348.
- Bunchalee P, Monthakantirat O, Luecha P, Srichai T, Maneechai S. 2022. GC-MS analysis and antioxidant potential of the stem and leaf extracts of *Dendrophthoe pentandra* (L.) Miq. hemiparasite on *Shorea roxburghii* G. Don and *Cassia fistula* L. *Trop J Nat Prod Res* 6 (10): 1638-1643. DOI: 10.26538/tjnp/v6i10.13.
- Devkota MP, Kunwar RM. 2006. Pollination and dispersal of three *Scurrula* species (Loranthaceae) in Godawari area of Kathmandu Valley, Nepal. *Indian J Bot Res* 2 (2): 115-128.
- Endharti AT, Wahyuningtyas TE, Hardini, Handono K, Widjajanto E, Permana S. 2018. *Dendrophthoe pentandra* leaves extract promotes apoptotic effects of doxorubicin in human breast cancer cell via modulation of intracellular calcium and survivin. *J Appl Pharm Sci* 8 (8): 39-43. DOI: 10.7324/japs.2018.8806.
- Esri Inc. 2021. ArcGIS Pro (Version 3.0). Environmental Systems Research Institute, Redlands, California.

- Gedalovich E, Kuijt J, Carpita NC. 1988. Chemical composition of viscin, an adhesive involved in dispersal of the parasite *Phoradendron californicum* (Viscaceae). *Physiol Mol Plant Pathol* 32 (1): 61-76. DOI: 10.1016/S0885-5765(88)80006-7.
- Gedalovich-Shedletzky E, Delmer DP, Kuijt J. 1989. Chemical composition of viscin mucilage from three Mistletoe species-A comparison. *Ann Bot* 64 (3): 249-252. DOI: 10.1093/oxfordjournals.aob.a087838.
- Kasmiyati S, Kristiani EBE. 2022. The potential of *Dendrophthoe pentandra* (L.) Miq and *Scurrula ferruginea* stem from *Syzygium aqueum* as source of natural antioxidant. *Biosaintifika: J Biol Biol Educ* 14 (3): 348-355. DOI: 10.15294/biosaintifika.v14i3.39250.
- Kong D, Wang L, Niu Y, Cheng L, Sang B, Wang D, Tian J, Zhao W, Liu X, Chen Y, Wang F, Zhou H, Jia R. 2023. Traditional uses, phytochemistry, pharmacology, toxicity, and applications of *Dendrophthoe falcata* and *Dendrophthoe pentandra*: Advancing parasitic plants to the frontier of phytomedicine. *Front Pharmacol* 14: 1096379. DOI: 10.3389/fphar.2023.1096379.
- Kristiningrum N, Ridlo M, Pratoko DK. 2020. Phytochemical screening and determination of total phenolic content of *Dendrophthoe pentandra* L. leaves ethanolic extract on mango host. *Ann Trop Med Public Health* 23 (3A): 98-107. DOI: 10.36295/asro.2020.2334.
- Kuramana S, Gandipilli G, Ratna Kumar PK. 2020. Studies on biology, seed dispersal and host interaction of *Dendrophthoe falcata* (L.f.) Etting. - A stem parasite of *Mangifera indica* L. (Mango). *World J Adv Res Rev* 8 (3): 97-103. DOI: 10.30574/wjarr.2020.8.3.0465.
- Likhitwitayawuid K, Sornsute A, Sritularak B, Ploypradith P. 2006. Chemical transformations of ox resveratrol (trans-2,4,3,5-tetrahydroxystilbene) into a potent tyrosinase inhibitor and a strong cytotoxic agent. *Bioorg Med Chem Lett* 16 (21): 5650-5653. DOI: 10.1016/j.bmcl.2006.08.018.
- Luo Y-H, Zhang L. 2013. Germination characteristics of *Dendrophthoe pentandra* seeds. *Plant Divers Resour* 35 (1): 73-80. DOI: 10.7677/ynzwjy201312064. [Chinese]
- Ma R, Miao N, Zhang H, Tao W, Mao K, Moermond TC. 2020. Generalist mistletoes and their hosts and potential hosts in an urban area in southwest China. *Urban For Urban Green* 53: 126717. DOI: 10.1016/j.ufug.2020.126717.
- Muche M, Muasya AM, Tsegay BA. 2022. Biology and resource acquisition of mistletoes, and the defense responses of host plants. *Ecol Proc* 11: 24. DOI: 10.1186/s13717-021-00355-9.
- Mustarichie R, Runadi D, Ramdhani D. 2017. The antioxidant activity and phytochemical screening of ethanol extract, fractions of water, ethyl acetate, and n-hexane from mistletoe tea (*Scurrula atropurpurea* Bl. Dans). *Asian J Pharm Clin Res* 10 (2): 343-347. DOI: 10.22159/ajpcr.2017.v10i2.15724.
- R Core Team. 2021. R: A language and environment for statistical computing. R Foundation for Statistical Computing. <https://www.r-project.org/>.
- Rahmawati SI, Ishimaru K, Hou D-X, Hayashi N. 2014. Antioxidant activity and phenolic content of mistletoe extracts following high-temperature batch extraction. *Food Sci Technol Res* 20 (2): 201-206. DOI: 10.3136/fstr.20.201.
- Sjakoer NAA, Mubarakati NJ, Taufiq A. 2021. Investigation of excellent ACE inhibitor agents from *Scurrula atropurpurea* and *Dendrophthoe pentandra* for anti-hypertension. *Chiang Mai Univ J Nat Sci* 20 (3): e2021068. DOI: 10.12982/cmujns.2021.068.
- Suriyaprom S, Kaewkod T, Promputtha I, Desvaux M, Tragoolpua Y. 2021. Evaluation of antioxidant and antibacterial activities of white mulberry (*Morus alba* L.) fruit extracts. *Plants* 10 (12): 2736. DOI: 10.3390/plants10122736.
- Teixeira-Costa L, Davis CC. 2021. Life history, diversity, and distribution in parasitic flowering plants. *Plant Physiol* 187 (1): 32-51. DOI: 10.1093/plphys/kiab279.
- Těšitel J, Li A-R, Knotková K, McLellan R, Bandaranayake PCG, Watson DM. 2021. The bright side of parasitic plants: What are they good for?. *Plant Physiol* 185 (4): 1309-1324. DOI: 10.1093/plphys/kiab069.
- Yee LS, Mohd Fauzi NF, Najihah NN, Daud NM, Sulain MD. 2017. Study of *Dendrophthoe pentandra* ethyl acetate extract as potential anticancer candidate on safety and toxicity aspects. *J Anal Pharm Res* 6 (1): 00167. DOI: 10.15406/japlr.2017.06.00167.
- Yismairai E, Hemelda NM, Yasman, Handayani W. 2019. Antioxidant activity of extract of mistletoe, *Dendrophthoe pentandra* (L.) Miq., lived in three different host plants, collected from Kampus UI, Depok. *AIP Conf Proc* 2168: 020100. DOI: 10.1063/1.5132527.
- Yuniwati C, Ramli N, Purwita E, Yusnaini Y, Nurdahlia N, Miko A, Liana I, Andriani A, Maharani M. 2018. Molecular docking for active compounds of *Scurrula atropurpurea* as anti-inflammatory candidate in endometriosis. *Acta Inform Med* 26 (4): 254-257. DOI: 10.5455/aim.2018.26.254-257.
- Zhang H, Florentine S, Tennakoon KU. 2022. The Angiosperm stem hemiparasitic genus *Cassytha* (Lauraceae) and its host interactions: A review. *Front Plant Sci* 13: 864110. DOI: 10.3389/fpls.2022.864110.