



Article

# Host Tree Identity Influences Leaf Nutrient Relations of the Epiphyte *Dendrobium guamense* Ames.

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**Abstract:** Epiphytic orchid species grow in stressful conditions that include nutrient deficiency. A greater understanding of the factors that influence epiphytic orchid nutrition may benefit conservationists and horticulturists. Green and senesced leaf nutrient concentrations of the epiphytic *Dendrobium guamense* were quantified among six host tree species to more fully understand the role of contrasting host identity. Green leaves for *D. guamense* plants on species with vertical stems contained greater nitrogen, calcium, iron, manganese, zinc, and boron concentrations. Green leaves for *D. guamense* plants on species with horizontal stems contained greater phosphorus, magnesium, and copper concentrations. Senesced *D. guamense* leaves followed similar patterns among the host species for all elements except calcium, manganese, and boron. The stoichiometry among green leaf macronutrients indicated nitrogen was most limiting, and the limitation was more severe for plants on horizontal stems. Carbon:nitrogen of senesced leaves indicated *D. guamense* plants on vertical stems produced higher quality leaf litter, predicting slower decomposition of orchid leaf litter from plants growing on hosts with horizontal stems. The results may improve horticultural decisions to support conservation efforts and show that host identity is a variable that should be more fully studied to understand epiphyte biology.

**Keywords:** Guam; leaf litter quality; nutrient resorption efficiency; Orchidaceae

## 1. Introduction

Vascular epiphytes are plants that grow on plants, and many epiphyte species are prevalent in the horticulture trade. The abiotic constraints for growth of vascular epiphytes include water shortage and nutrient availability [1]. Adaptations to these abiotic stressors abound among epiphytes. For orchid epiphytes, the velamen radicum is one such adaptation [2]. This specialized orchid root is highly efficient in absorbing ephemeral arrivals of nutrients in solutions on the stem surfaces.

*Dendrobium guamense* Ames. is an epiphytic orchid with a small endemic range in the Mariana Island archipelago [3]. It is listed as threatened under the United States Endangered Species Act (ESA) [4]. The in-situ plants are conspicuous because of the mass of stems that radiate from a common base. The white flowers are borne in co-dominant pairs with concurrent anthesis on short peduncles at each stem node. The arborescent species that serve as host plants are abundant. Even invasive tree species such as *Leucaena leucocephala* (Lam.) de Wit and *Vitex parviflora* Juss. are prolific hosts for the endemic epiphyte (personal observations). These host trees vary greatly in canopy traits, some of which may directly influence the orchid's nutritional status.

Host stem orientation is a trait that is highly variable among *D. guamense* plants. The extent of abiotic stress may be influenced by the physical location of an epiphyte within the tree crowns [1]. For nutrient stress, if stemflow dominates the orchid's access of nutrients then host plants with ageotropic stem axial orientations may exhibit greater *D. guamense* leaf nutrient concentrations,

as vertical stems compared to horizontal stems may have greater stemflow volumes that the velamen radicum could intercept [5]. Contrarily, if a buildup of canopy detritus and development of a velamen radicum rhizosphere microbiome within that detritus dominates the orchid's access of nutrients, then host plants with plagiotropic stem axial orientations may produce greater *D. guamense* leaf nutrient concentrations. Therefore, I hypothesized that the concentrations of the *D. guamense* leaf mineral and metal elements which exhibit differences among the host plant species would separate into one group for the host species with near-vertical stem orientation and a second group for the host species with plagiotropic stem orientation. The results may inform management decisions to improve conservation efforts.

## 2. Materials and Methods

*Dendrobium guamense* plants used for leaf sampling were from a single habitat in Guam with karst soils (Clayey-skeletal, gibbsitic, nonacid, isohyperthermic Lithic Ustorthents) [6]. This orchid establishes on numerous host tree species in these soils. I reconnoitered the habitat in June and July 2015 to identify the list of host tree species for the orchid. The surveyed area was centered at 13.340854° N, 144.668138° E and spanned roughly 1.7 km latitudinally and 0.7 km longitudinally. Orchid plants on six trees of each of the following host species were selected for inclusion in this study:

- *Areca catechu* L. is one of two native palm species. This tree is the source of betel nut, a resource that is important to many cultures. The deviation from horizontal for the host stems of each sampled *D. guamense* was measured with a 50-cm bubble level and protractor. With this method, an orientation of 0° indicates horizontal and 90° indicates vertical. The stem angle above the horizontal ranged from 81° to 90°, with a mean of 85°.
- *Cycas micronesica* K.D. Hill is the only native gymnosperm in the Mariana Islands. Several recent specialist insect invasions have devastated the population of this cycad. As a result, the species was listed as endangered under the International Union for Conservation of Nature Red List [7] and as threatened under the ESA [4]. The stem angle above the horizontal ranged from 84° to 90°, with a mean of 86°.
- *Elaeocarpus joga* Merr. is endemic to the Mariana Islands. It is a striking horticultural specimen because of unique canopy architecture and blue fruits. This tree is not listed as threatened, although it has become less common in recent decades. The stem angle above the horizontal ranged from 2° to 18°, with a mean of 8°.
- *Guettarda speciosa* L. is a small Rubiaceae tree that has a very wide indigenous range in the Pacific Ocean. It is not very common on Guam but is not considered threatened. The stem angle above the horizontal ranged from 7° to 22°, with a mean of 18°.
- *Pandanus dubius* Spreng. is one of two native *Pandanus* species. The stem angle above the horizontal ranged from 70° to 84°, with a mean of 78°.
- *Pandanus tectorius* Parkinson ex Zucc. is the second native *Pandanus* species. It is much more prevalent than *P. dubius* throughout Guam. The stem angle above the horizontal ranged from 72° to 89°, with a mean of 81°.

The habitat also contained *D. guamense* plants growing on *Cocos nucifera* L., *Hibiscus tiliaceus* L., and *Leucaena leucocephala* trees, but I was unable to find enough trees for these host species to meet the requirement of six replications, so they were not included. The habitat contained other native epiphytic orchid species. However, the tree host range of these other orchids was not as broad as *D. guamense*, so they were also not included.

### 2.1. Field Methods

Leaf selection for each *D. guamense* plant followed established protocols for determining nutrient resorption during leaf senescence [8–10]. The host tree was considered a replication, so if more than one *D. guamense* plant occurred on an individual tree, appropriate leaves were obtained from all

plants. The youngest fully expanded leaf was collected to represent green leaves. Yellow and brown leaves attached to live stems were collected to represent senesced leaves. No detached senesced leaves were collected which ensured all samples were recently senesced. This minimized the potential for loss of nutrients by leaching after senescence. Green leaf collection was restricted to *D. guamense* stems that also contained a senesced leaf to justify intra-stem comparisons between the two leaf categories. The field sampling was conducted on several days in August 2015, and total dry weight ranged from 4 to 5 g for each sample.

## 2.2. Analytical Protocols

The leaf tissue was dried at 75 °C and milled to pass through a 20-mesh screen. Total carbon and nitrogen were determined by dry combustion (FLASH EA1112 CHN Analyzer, Thermo Fisher, Waltham, MA, USA) [11]. Samples were digested by a microwave system with nitric acid and peroxide, then phosphorus, potassium, calcium, magnesium, iron, manganese, zinc, and copper were quantified by inductively coupled plasma optical emission spectroscopy (Spectro Genesis; SPECTRO Analytical Instruments, Kleve, Germany) [12]. The stoichiometric variables were log-transformed prior to analysis. The remainder of the variables met parametric requirements. The data were subjected to a one-way ANOVA (SAS 9.3; SAS Institute, Cary, IN, USA), and means separation was by least significant difference.

## 3. Results

### 3.1. Macronutrients

Green leaf nitrogen, calcium, and magnesium concentrations were greater for *A. catechu*, *C. micronesica*, *P. dubius*, and *P. tectorius* than for *E. joga* and *G. speciosa* (Table 1). In contrast, green leaf phosphorus was greater in *E. joga* and *G. speciosa* than in the other host tree species. Carbon and potassium concentrations were not influenced by host tree species. Senesced leaf concentrations followed the pattern of green leaves for every element except calcium (Table 1). Senesced leaf calcium concentration was greater for *E. joga* and *G. speciosa* host trees than for *A. catechu*, *C. micronesica*, *P. dubius*, and *P. tectorius* host trees. Since calcium increased in concentration during the leaf aging process, the results indicate the up-accumulation was much greater for *D. guamense* plants growing on the tree species with horizontal stems.

**Table 1.** Concentration of carbon and macronutrients in green and senesced *Dendrobium guamense* leaves as influenced by host tree identity ( $n = 6$ ).

Element (mg g <sup>-1</sup> )	<i>Areca catechu</i>	<i>Cycas micronesica</i>	Green		<i>Pandanus dubius</i>	<i>Pandanus tectorius</i>	<i>p</i>
			<i>Elaeocarpus joga</i>	<i>Guettarda speciosa</i>			
Carbon	466.17	461.24	460.24	470.26	476.14	471.64	0.994
Nitrogen	7.02b <sup>1</sup>	7.68b	4.98a	4.41a	7.25b	7.18b	<0.001
Phosphorus	0.70a	0.73a	1.29b	1.13b	0.67a	0.65a	<0.001
Potassium	10.16	9.96	10.84	11.84	10.01	9.96	0.058
Calcium	9.12b	9.09b	7.19a	6.98a	8.57b	8.89b	<0.001
Magnesium	3.12a	3.06a	4.05b	3.89b	3.22a	3.25a	<0.001
			Senesced				
			Leaves				
Carbon	472.33	469.47	466.54	482.17	479.42	471.50	0.999
Nitrogen	4.30b	4.62b	2.99a	2.23a	4.88b	4.32b	<0.001
Phosphorus	0.48a	0.47a	0.81b	0.78b	0.46a	0.44a	<0.001
Potassium	4.46	4.45	4.30	4.94	4.25	4.24	0.170
Calcium	9.99a	10.43a	13.71b	12.12b	10.56a	10.59a	<0.001
Magnesium	2.88a	2.82a	4.06b	3.88b	2.82a	3.11a	<0.001

<sup>1</sup> Means followed by the same letter within each row are not different according to Least Significant Difference.

### 3.2. Micronutrients

Green leaf iron, manganese, zinc, and boron concentrations were greater for *A. catechu*, *C. micronesica*, *P. dubius*, and *P. tectorius* than for *E. joga* and *G. speciosa* (Table 2). Of these elements, zinc was the only one which differed between *E. joga* and *G. speciosa*, with *E. joga* trees containing *D. guamense* leaves with greater zinc than *G. speciosa* trees. Copper concentrations followed a contrasting pattern, with *E. joga* and *G. speciosa* host trees exhibiting greater copper than *A. catechu*, *C. micronesica*, *P. dubius*, and *P. tectorius* host trees. Senesced leaf concentrations followed the general pattern of green leaves for iron, zinc, and copper concentrations (Table 2). However, zinc concentration converged for *E. joga* and *G. speciosa* trees such that no difference occurred. The differences in manganese and boron that were evident in green leaves disappeared during leaf aging, as the senesced leaves did not exhibit differences among the host tree species.

**Table 2.** Concentration of micronutrients in green and senesced *D. guamense* leaves as influenced by host tree identity ( $n = 6$ ).

Element ( $\mu\text{g g}^{-1}$ )	<i>Areca catechu</i>	<i>Cycas micronesica</i>	<i>Elaeocarpus joga</i>	<i>Guettarda speciosa</i>	<i>Pandanus dubius</i>	<i>Pandanus tectorius</i>	<i>p</i>
Iron	56.17b <sup>1</sup>	61.68b	46.02a	44.18a	56.54b	59.92b	<0.001
Manganese	51.12b	53.64b	32.16a	35.51a	50.38b	48.76b	<0.001
Zinc	18.34c	19.22c	14.18b	11.64a	19.35c	20.49c	<0.001
Copper	3.21a	3.17a	4.81b	4.26b	2.95a	3.12a	<0.001
Boron	31.03b	30.41b	24.25a	23.79a	28.81b	28.66b	0.002
			Senesced	Leaves			
Iron	68.16b	69.72b	54.52a	51.37a	69.83b	72.36b	<0.001
Manganese	69.03	71.48	76.22	73.51	66.97	66.08	0.258
Zinc	15.05b	16.08b	12.44a	11.43a	16.17b	15.12b	<0.001
Copper	2.10a	2.48a	4.83b	4.03b	2.45a	2.44a	<0.001
Boron	25.74	27.24	24.12	23.01	25.73	25.13	0.230

<sup>1</sup> Means followed by the same letter within each row are not different according to Least Significant Difference.

### 3.3. Derived Leaf Traits

Stoichiometry among the pairs of leaf nutrients nitrogen, phosphorus, and potassium followed the same pattern for green and senesced leaves. Nitrogen:phosphorus, nitrogen:potassium, and potassium:phosphorus were always least for *D. guamense* plants on *E. joga* and *G. speciosa* host trees and greatest for *D. guamense* plants on *A. catechu*, *C. micronesica*, *P. dubius*, and *P. tectorius* host trees (Table 3). Within the two host species with horizontal stems or the four host species with vertical stems, the differences were either not significant or there was considerable overlap among the species. In contrast, the two horizontal host tree species never overlapped with any of the four vertical host tree species for any response variable. Senesced leaf carbon:nitrogen was least for the four host trees with vertical stems and greatest for the host trees with horizontal stems (Table 3). Senesced leaf carbon:phosphorus was least for the two host trees with horizontal stems, intermediate for the *A. catechu* and *C. micronesica* trees, and greatest for the *Pandanus* trees. Senesced leaf carbon:potassium exhibited few differences, with the *D. guamense* plants on *G. speciosa* having lower values than the other five host trees.

No differences occurred among the host tree species for nitrogen, phosphorus, or potassium resorption efficiency (Table 3). This result indicates the *D. guamense* plants with greater green leaf nutrient concentrations re-translocated greater absolute levels of each nutrient during leaf aging and senescence.

**Table 3.** Stoichiometry of nitrogen, phosphorus, and potassium concentrations in green and senesced *D. guamense* leaves and resorption efficiency of nitrogen, phosphorus, and potassium as influenced by host tree identity ( $n = 6$ ).

Trait	<i>Areca catechu</i>	<i>Cycas micronesica</i>	<i>Elaeocarpus joga</i>	<i>Guettarda speciosa</i>	<i>Pandanus dubius</i>	<i>Pandanus tectorius</i>	<i>p</i>
			Green	leaves			
Nitrogen:phosphorus	10.01b <sup>1</sup>	10.55b	3.86a	3.92a	10.84b	11.12b	<0.001 <sup>2</sup>
Nitrogen:potassium	0.69b	0.77b	0.46a	0.37a	0.72b	0.72b	<0.001
Potassium:phosphorus	14.49cd	13.67c	8.41a	10.51b	14.98d	15.43d	<0.001
			Senesced	leaves			
Nitrogen:phosphorus	8.7b	9.81b	3.71a	2.86a	10.82b	9.93b	<0.001
Nitrogen:potassium	0.96c	1.04c	0.69b	0.45a	1.17d	1.02c	<0.001
Potassium:phosphorus	9.30b	9.46b	5.35a	6.35a	9.20b	9.74b	<0.001
Carbon:nitrogen	109.87a	101.87a	156.87b	214.85c	100.93a	109.05a	<0.001
Carbon:phosphorus	985.34b	998.88b	582.26a	614.95a	1038.90c	1082.72d	<0.001
Carbon:potassium	105.96b	105.58b	108.93b	96.90a	112.93b	111.26b	0.014
Nitrogen resorption	38.74	38.96	39.89	49.67	31.34	39.74	0.068
Phosphorus resorption	31.52	34.29	37.40	30.96	30.86	32.29	0.450
Potassium resorption	56.13	54.58	60.26	58.36	57.59	57.43	0.173

<sup>1</sup> Means followed by the same letter within each row are not different according to Least Significant Difference.

<sup>2</sup> Analysis on log-transformed data for stoichiometric traits.

#### 4. Discussion

Ten mineral elements were determined in green *D. guamense* leaves, and only potassium was unaffected by host tree species. All of the nine elements that were influenced by host tree species behaved in a manner that the concentrations from the two host trees with horizontal stems separated from the concentrations from the four host trees with vertical stems. Therefore, the results confirm my hypothesis for every element that exhibited significant differences among the host trees.

There are no published orchid nutrition papers from the Mariana Islands to compare with my results. However, the relatively low concentrations of leaf macronutrients reported here were comparable to a range of epiphytic orchid species from Panama [13,14]. Stoichiometric variables indicated nitrogen was most limiting, yet nitrogen resorption efficiency was extremely low compared to phosphorus and potassium resorption efficiencies. These enigmatic relations were also shown for several epiphyte orchid species from Panama [14].

The highly stable potassium concentrations indicated *D. guamense* exerts greater control over leaf potassium homeostasis than any other essential element. The function of this element in maintaining leaf physiology and plant turgidity [15–17] may explain the need for this epiphyte species to control potassium concentrations within a narrow range to maintain desiccation tolerance. Moreover, nutrient budgets for the epiphytic orchid *Dimerandra emarginata* (G. Meyer) Hoehne revealed that fruit demands for potassium were substantial [13], and the potassium pool in vegetative organs may serve as a source to satisfy this need.

The manner in which epiphytic orchid plants influence habitat-level nutrient budgets and cycling cannot be fully understood without considering the bulk movement of elements from the living plant canopy to the soil. The transfer of nutrients that are sequestered in vegetation to the soil is mostly controlled by falling leaves and other plant parts. With time, the nutrients within these components of the litter layer are leached and released during decomposition. In addition to this phenomenon, precipitation causes the movement of nutrients from the vegetation to the soil as throughfall and stemflow [18–22]. Throughfall is the precipitation that is intercepted by a plant, then drips from the surfaces of components of the canopy. Stemflow is the intercepted rainfall that is transferred to the soil by running down the vertical stems. Throughfall and stemflow cause an increase in nutrient deposition in relation to precipitation because they contain nutrients that are leached from the vegetation canopy and nutrients that have accumulated on the surfaces of the canopy organs since the previous rainfall

event [23,24]. In contrast to the nutrients that are locked up in freshly fallen litter, these nutrients are available immediately. Throughfall and stemflow significantly contribute to soil nutrition in many ecosystems, and may be particularly important sources of input of severely limiting nutrients [25].

My results indicate nitrogen, calcium, iron, manganese, zinc, and boron may be more available to *D. guamense* plants via interception of stemflow solutions. In contrast, phosphorus, magnesium, and copper may be more available to *D. guamense* plants via development of a microbiome in the rhizosphere of canopy debris that accumulates on the top of plagiotropic branches. An attempt to separate the direct influences of host tree stem orientation from host tree species on *D. guamense* leaf nutrient traits is beyond the scope of this study. Furthermore, my dataset does not contain the continuum of host stem orientation from horizontal to vertical. The direct role of orientation could be directly tested with regression analysis by including a full range of stem orientations from horizontal to vertical within the same host tree species. Eudicot trees such as *E. joga* and *G. speciosa* would be required for this endeavor because the stems for the other host trees in this study are always ageotropic and therefore restricted to vertical orientation.

Several studies have focused on stemflow of palm species. A Brazilian forest dominated by the palm *Orbignya phalerata* Mart. exhibited stemflow that was 8% of incident rainfall, yet the total basal area of vegetation was only 0.3% of the plot area [26]. Comparisons of palms and eudicot tree species in Brazil showed palm trees to exhibit greater volumes and nutrient transfers in stemflow than the eudicot species [27,28]. The nutrient budgets of the palm *Rhopalostylis sapida* H. Wendl. & Drude were determined to show that stemflow contributed enough calcium, magnesium, and potassium to the base of the plant to meet all of the annual nutrient needs [29]. *Lodoicea maldivica* (J.F. Gmel.) Pers. was studied to show that the radiating leaves of a palm tree create an effective trap and funnel that intercepts aerosol particles including pollen, then transfers these particles to the soil as stemflow [30]. The canopy funnel trap phenomenon was further developed for the palm *Asterogyne martiana* Wendl. Ex Hemsl. by describing how copious amounts of alien litter were trapped then contributed nutrients to stemflow [31]. The electrical conductivity of stemflow varied almost three-fold among six palm species in a single location, illuminating the strong influence of canopy traits on stemflow chemistry [32].

The presence of epiphytes in a tree canopy can significantly reduce the volume of throughfall and stemflow [33,34] illuminating the ability of epiphytes to intercept rainfall, throughfall, and stemflow. The orchid velamen is highly efficient in capturing nutrients within solutions that are ephemerally available on stem surfaces. These nutrients can be absorbed in seconds whenever they become available on the stem surface [2]. Charged ions are retained after the rapid absorption while uncharged compounds are subsequently lost from the velamen. This form of nutrient absorption by *D. guamense* plants may be greater on vertical host stems because of the greater stemflow. Alternatively, the buildup of organic debris in microsites of tree canopies such as the top of large horizontal stems is referred to as “crown humus” or “canopy soil” and may occur in microsites in canopy trees where trapped organic matter becomes heavily decomposed [2]. Epiphyte roots that are able to access the nutrients that are released from this crown humus may have access to more available nutrients than epiphyte roots attached to vertical host stems. The rhizosphere of an epiphyte’s roots within this humus may be extensively colonized by microorganisms to create canopy hot-spots in which the interactions between organisms reach a very complex level. These contrasts of stemflow versus crown humus may explain my *D. guamense* results.

Stoichiometric relations among macronutrients have been used to estimate which elements are most limiting of plant health. The nitrogen:phosphorus mean of 7.35, the nitrogen:potassium mean of 0.55, and the potassium:phosphorus mean of 11.32 indicate that nutrient limitations for these *D. guamense* plants were greatest for nitrogen, least for potassium, and intermediate for phosphorus [35–39]. Moreover, nitrogen was more limiting in plants growing on horizontal host stems, and less so for plants growing on erect host stems. The inclusion of carbon in the relationships magnifies our ability to understand stoichiometry, as long-lived leaves tend to contain greater carbon construction costs which may dilute the mineral elements. The global C:N:P mean of 469:13:1 [40]

was not similar to the 679:11:1 for the *D. guamense* plants growing on erect host stems or the 387:4:1 for the *D. guamense* plants growing on horizontal host stems (calculated from Table 1). However, the difference of this metric for the *D. guamense* plants growing on erect host stems versus *D. guamense* plants growing on horizontal host stems provides more evidence of the direct influence of host plant on this orchid's leaf nutrition.

Nutrient resorption is a critical behavior among plants enabling a recycling of limiting elements prior to organ senescence [9,10]. This behavior affects plant processes by reducing the dependence on the rhizosphere to satisfy ongoing needs of mineral nutrition. Additionally, the behavior affects biogeochemical cycling by increasing elemental residence time within the plant body and reducing the minerals that are deposited into the litter layer when senescent organs are abscised from a plant. On average, plants withdraw 50% to 60% of leaf nitrogen and phosphorus prior to leaf abscission, and withdraw about 70% of the leaf potassium prior to leaf abscission [8,9,41–43]. Despite the relatively low green leaf concentrations, the *D. guamense* plants in this study were unable to adequately mobilize these three nutrients during leaf senescence, as the resorption efficiencies of nitrogen and phosphorus were less than 35% and that of potassium was only 50%.

Killingbeck [9] introduced the term resorption proficiency to describe the basal levels to which each nutrient was reduced in senesced leaves. Among woody perennials, a nitrogen resorption proficiency of  $<7 \text{ mg}\cdot\text{g}^{-1}$  and a phosphorus resorption proficiency of  $<0.5 \text{ mg}\cdot\text{g}^{-1}$  indicates complete withdrawal. Our resorption proficiencies did not approach these lower limits, confirming the conclusions from the resorption efficiencies that these orchid plants are not adept at recovering leaf nutrients during senescence.

Green leaf nutrient levels combine with resorption traits to mediate the litter quality upon leaf senescence. The relations among carbon and macronutrients exert profound influence in litter decomposition speed, with greater carbon:nitrogen indicating slower decomposition [44,45]. In this study, the influence of host tree on *D. guamense* leaf litter chemistry exerted robust changes in these traits. The species with vertical stems produced orchid litter with carbon:nitrogen averaging 105, while the *E. joga* trees produced orchid litter that was 1.5-fold greater and the *G. speciosa* trees produced orchid litter that was 2.0-fold greater than the species with vertical stems. Parasitic plants such as mistletoe have been shown to influence litterfall quality, quantity, seasonality, and spatial heterogeneity in a manner that alters habitat nutrient cycling [46,47]. In this light, my results indicate that host tree identity may exert a direct effect on how the epiphytic *D. guamense* influences nutrient turnover via litterfall. On average, the orchid plants growing on palms, cycads, and pandanus trees will contribute higher quality litterfall to the ecosystem than plants growing on *E. joga* or *G. speciosa* trees. These differences potentially improve habitat health by creating more spatial heterogeneity in soil nutrient status.

Conserving species-at-risk is often plagued with uncertainties and a failure to understand multi-species interactions. The conservation issues related to an epiphyte are magnified beyond direct threats to the plant in need of conservation. The requirement of a suitable host for an epiphyte indicates that threats to the range of host tree species also indirectly threatens an epiphyte species. The threats to *C. micronesica* illustrate this phenomenon. This tree was once the most abundant tree on Guam [48], illuminating the value in serving as potential host for *D. guamense*. However, an armored scale [49] and several other non-native specialist insects [50] have killed more than 90% of the tree population since 2003 [51]. This case study reveals the need to conserve host tree species as a component of a comprehensive conservation plan designed to conserve a threatened epiphyte species such as *D. guamense*.

More research is needed on tropical orchid species to fully understand the symbioses between the host plant and the orchid mycorrhizal fungi and other root endophytes [52]. My results illuminate two more research agendas that would benefit from the inclusion of more tropical orchid species. First, epiphytic orchid species with broad host tree range provide case studies to determine the influence of host tree on orchid nutritional status. My approach was to use two groups of host trees with highly

contrasting stem orientations, but there are other host tree traits that could also be studied in the context of orchid plant nutrition. For example, more direct measurements of stemflow and throughfall volume and chemistry among host trees would illuminate the differences among host trees. Second, much of the stemflow research has focused on canopy inputs to the solution that flows down the vertical stems. My results indicate epiphyte plant removal of nutrients from stemflow solutions should also be explicitly considered in the nutrient and water budgets during experimental design. In other words, what makes it into the solution at the canopy level may not be present when the stemflow enters the soil, especially in the case of epiphytic host tree species. The same may be said of throughfall, where mid-strata epiphytes may intercept and remove nutrients from throughfall that originates in the upper canopy. Collection of throughfall at more than one strata within the canopy would be able to quantify this function of canopy epiphytes in future throughfall research.

## 5. Conclusions

*Dendrobium guamense* plants in southern Guam contained greater green leaf nitrogen, calcium, iron, manganese, zinc, and boron on host trees with vertical stem orientation; and more phosphorus, magnesium, and copper on host trees with horizontal stems. Differences in leaf litter quality of senesced *D. guamense* leaves between the two host tree groups indicated a strong influence of the host tree on how *D. guamense* influences spatial heterogeneity within biogeochemical cycling. The results may improve horticultural decisions to support conservation efforts and add host stem orientation as a variable that should be more fully studied to understand epiphyte biology. The results provide ecological and physiological information about orchid species colonization that should be considered in horticultural management of these species. Conservation efforts on Guam include rescue and transplanting of ESA-listed orchid species and species introduction efforts occur for many orchid species globally. The transplanted or laboratory-grown plants are typically tied to the main stem or branch without consideration of how stem orientation affects nutrient processes. My results indicate the attachment of the *D. guamense* transplant to a vertical host stem may be more beneficial for plant nutrition than a horizontal host stem.

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## References

1. Zotz, G.; Hietz, P. The physiological ecology of vascular epiphytes: Current knowledge, open questions. *J. Exp. Bot.* **2001**, *52*, 2067–2078. [[CrossRef](#)] [[PubMed](#)]
2. Zotz, G.; Winkler, U. Aerial roots of epiphytic orchids: The velamen radicum and its role in water and nutrient uptake. *Oecologia* **2013**, *171*, 733–741. [[CrossRef](#)] [[PubMed](#)]
3. Raulerson, L.; Rinehart, A. *Ferns and Orchids of the Mariana Islands*; Raulerson and Rinehart: Mangilao, GU, USA, 1992; pp. 1–138. ISBN 9781878453099.
4. United States Fish and Wildlife Service (USFWS). Endangered and threatened wildlife and plants; endangered status for 16 species and threatened status for 7 species in Micronesia. *Fed. Regist.* **2015**, *80*, 59424–59497.
5. Corti, G.; Agnelli, A.; Cocco, S.; Cardelli, V.; Masse, J.; Courchesne, F. Soil affects throughfall and stemflow under Turkey oak (*Quercus cerris* L.). *Geoderma* **2019**, *333*, 43–56. [[CrossRef](#)]
6. Young, F.J. *Soil Survey of Territory of Guam*; U.S. Department of Agriculture Soil Conservation Service: Washington, DC, USA, 1988.
7. Marler, T.; Haynes, J.; Lindström, A. *Cycas micronesica*. *IUCN Red List Threat. Spec.* **2010**, e.T61316A12462113. [[CrossRef](#)]
8. Aerts, R. Nutrient resorption from senescing leaves of perennials: Are there general patterns? *J. Ecol.* **1996**, *84*, 597–608. [[CrossRef](#)]

9. Killingbeck, K.T. Nutrients in senesced leaves: Keys to the search for potential resorption and resorption proficiency. *Ecology* **1996**, *77*, 1716–1727. [[CrossRef](#)]
10. Killingbeck, K.T. Nutrient resorption. In *Plant Cell Death Processes*; Noodén, L.D., Ed.; Elsevier: Amsterdam, The Netherlands, 2004; pp. 215–226.
11. Dumas, J.B.A. Procédes de l'analyse organique. *Ann. Chim. Phys.* **1831**, *47*, 198–205.
12. Hou, X.; Jones, B.T. Inductively coupled plasma/optical emission spectrometry. In *Encyclopedia of Analytical Chemistry*; Meyers, R.A., Ed.; John Wiley & Sons: Chichester, UK, 2000; pp. 9468–9485.
13. Zotz, G. What are backshoots good for? Seasonal changes in mineral, carbohydrate and water content of different organs of the epiphytic orchid, *Dimerandra emarginata*. *Ann. Bot.* **1999**, *84*, 791–798. [[CrossRef](#)]
14. Zotz, G. The resorption of phosphorus is greater than that of nitrogen in senescing leaves of vascular epiphytes from lowland Panama. *J. Trop. Ecol.* **2004**, *20*, 693–696. [[CrossRef](#)]
15. Marschner, H. *Mineral Nutrition of Higher Plants*, 2nd ed.; Academic Press: San Diego, CA, USA, 1995.
16. Mengel, K.; Kirkby, E.A. *Principles of Plant Nutrition*, 5th ed.; Kluwer Academic Publishers: Dordrecht, The Netherlands, 2001.
17. Cakmak, I. The role of potassium in alleviating detrimental effects of abiotic stresses in plants. *J. Plant Nutr. Soil Sci.* **2005**, *168*, 521–530. [[CrossRef](#)]
18. Levia, D.F.; Frost, E.E. A review and evaluation of stemflow literature in the hydro-logic and biogeochemical cycles of forested and agricultural ecosystems. *J. Hydrol.* **2003**, *274*, 1–29. [[CrossRef](#)]
19. Ikawa, R. Literature review of stemflow generation and chemical characteristics in Japanese forests. *J. Jpn. Assoc. Hydrol. Sci.* **2007**, *37*, 187–200. [[CrossRef](#)]
20. Levia, D.F.; Germer, S. A review of stemflow generation dynamics and stemflow-environment interactions in forests and shrublands. *Rev. Geophys.* **2015**, *53*, 673–714. [[CrossRef](#)]
21. Van Stan, J.T.; Gordon, D.A. Mini-Review: Stemflow as a resource limitation to near-stem soils. *Front. Plant Sci.* **2018**, *9*, 248. [[CrossRef](#)] [[PubMed](#)]
22. Van Stan, J.T.; Stubbins, A. Tree-DOM: Dissolved organic matter in throughfall and stemflow. *Limnology Oceanogr. Lett.* **2018**, *3*, 199–214. [[CrossRef](#)]
23. Parker, G.G. Throughfall and stemflow in the forest nutrient cycle. *Adv. Ecol. Res.* **1983**, *13*, 58–135.
24. Chiwa, M.; Crossley, A.; Sheppard, L.J.; Sakugawa, H.; Cape, J.N. Throughfall chemistry and canopy interactions in a Sitka spruce plantation sprayed with six different simulated polluted mist treatments. *Environ. Pollut.* **2004**, *127*, 57–64. [[CrossRef](#)]
25. Fahey, T.J.; Yavitt, J.B.; Joyce, G. Precipitation and throughfall chemistry in *Pinus contorta* ssp. *latifolia* ecosystems, southeastern Wyoming. *Can. J. For. Res.* **1988**, *18*, 337–345.
26. Germer, S.; Werther, L.; Elsenbeer, H. Have we underestimated stemflow? Lessons from an open tropical rainforest. *J. Hydrol.* **2010**, *395*, 169–179. [[CrossRef](#)]
27. Schroth, G.; da Silva, L.F.; Wolf, M.A.; Teixeira, W.G.; Zech, W. Distribution of throughfall and stemflow in multi-strata agroforestry, perennial monoculture, fallow and primary forest in central Amazonia, Brazil. *Hydrol. Process.* **1999**, *13*, 1423–1436. [[CrossRef](#)]
28. Schroth, G.; Elias, M.E.A.; Uguen, K.; Seixas, R.; Zech, W. Nutrient fluxes in rainfall, throughfall and stemflow in tree-based land use systems and spontaneous tree vegetation of central Amazonia. *Agric. Ecosyst. Environ.* **2001**, *87*, 37–49. [[CrossRef](#)]
29. Enright, N.J. Stemflow as a nutrient source for nikau palm (*Rhopalostylis sapida*) in a New Zealand forest. *Austral Ecol.* **1987**, *12*, 17–24. [[CrossRef](#)]
30. Edwards, P.J.; Fleischer-Dogley, F.; Kaiser-Bunbury, C.N. The nutrient economy of *Lodoicea maldivica*, a monodominant palm producing the world's largest seed. *New Phytol.* **2015**, *206*, 990–999. [[CrossRef](#)] [[PubMed](#)]
31. Raich, J.W. Understory palms as nutrient traps: A hypothesis. *Brenesia* **1983**, *21*, 119–129.
32. Ghatge, S.D.; Sridhar, K.R. Rain-borne fungi in stemflow and throughfall of six tropical palm species. *Czech Mycol.* **2015**, *67*, 45–58.
33. Gay, T.E.; Van Stan, T.V.; Moore, L.D.; Lewis, E.S.; Reichard, J.S. Throughfall alterations by degree of *Tillandsia usneoides* cover in a Southeastern U.S. *Quercus virginiana* forest. *Can. J. For. Res.* **2015**. [[CrossRef](#)]
34. Howard, D.H.; Van Stan, J.T.; Whitetree, A.; Zhu, L.; Stubbins, A. Interstorm variability in the biolability of tree-derived dissolved organic matter (Tree-DOM) in throughfall and stemflow. *Forests* **2018**, *9*, 236. [[CrossRef](#)]

35. Koerselman, W.; Meuleman, A.F.M. The vegetation N:P ratio: A new tool to detect the nature of nutrient limitation. *J. Appl. Ecol.* **1996**, *33*, 1441–1450. [[CrossRef](#)]
36. Aerts, R.; Chapin, F.S. The mineral nutrition of wild plants revisited: A re-evaluation of processes and patterns. *Adv. Ecol. Res.* **2000**, *30*, 1–67.
37. Olde Venterink, H.; Wassen, M.J.; Verkroost, A.W.M.; de Ruiter, P.C. Species richness-productivity patterns differ between N-, P-, and K-limited wetlands. *Ecology* **2003**, *84*, 2191–2199. [[CrossRef](#)]
38. Tessier, J.T.; Raynal, D.J. Use of nitrogen to phosphorus ratios in plant tissue as an indicator of nutrient limitation and nitrogen saturation. *J. Appl. Ecol.* **2003**, *40*, 523–534. [[CrossRef](#)]
39. Güsewell, S. N:P ratios in terrestrial plants: Variation and functional significance. *New Phytol.* **2004**, *164*, 43–266. [[CrossRef](#)]
40. McGroddy, M.E.; Daufresne, T.; Hedin, L.O. Scaling of C:N:P stoichiometry in forests worldwide: Implications of terrestrial Redfield-type ratios. *Ecology* **2004**, *85*, 2390–2401. [[CrossRef](#)]
41. Yuan, Z.Y.; Chen, H.Y.H. Global trends in senesced-leaf nitrogen and phosphorus. *Glob. Ecol. Biogeogr.* **2009**, *18*, 532–542. [[CrossRef](#)]
42. Yuan, Z.Y.; Chen, H.Y.H. Global-scale patterns of nutrient resorption associated with latitude, temperature and precipitation. *Glob. Ecol. Biogeogr.* **2009**, *18*, 11–18. [[CrossRef](#)]
43. Vergutz, L.; Manzoni, S.; Porporato, A.; Novais, R.F.; Jackson, R.B. Global resorption efficiencies and concentrations of carbon and nutrients in leaves of terrestrial plants. *Ecol. Monogr.* **2012**, *82*, 205–220. [[CrossRef](#)]
44. Cornwell, W.K.; Cornelissen, J.H.C.; Amatangelo, K.; Dorrepaal, E.; Eviner, V.T.; Godoy, O.; Hobbie, S.E.; Hoorens, B.; Kurokawa, H.; Pérez-Harquindeguy, N.; et al. Plant species traits are the predominant control on litter decomposition rates within biomes worldwide. *Ecol. Lett.* **2008**, *11*, 1065–1071. [[CrossRef](#)] [[PubMed](#)]
45. Cornelissen, J.H.C.; Queded, H.M.; Gwynn-Jones, D.; Van Logtestijn, R.S.P.; De Beus, M.A.H.; Kondratchuk, A.; Callaghan, T.V.; Aerts, R. Leaf digestibility and litter decomposability are related in a wide range of subarctic plant species and types. *Funct. Ecol.* **2004**, *18*, 779–786. [[CrossRef](#)]
46. March, W.A.; Watson, D.M. Parasites boost productivity: Effects of mistletoe on litterfall dynamics in a temperate Australian forest. *Oecologia* **2007**, *154*, 339–347. [[CrossRef](#)] [[PubMed](#)]
47. March, W.A.; Watson, D.M. The contribution of mistletoes to nutrient returns: Evidence for a critical role in nutrient cycling. *Austral Ecol.* **2010**, *35*, 713–721. [[CrossRef](#)]
48. Donnegan, J.A.; Butler, S.L.; Grabowiecki, W.; Hiserote, B.A.; Limtiaco, D. *Guam's Forest Resources, 2002*; Resource Bulletin PNW-RB-243; U.S. Department of Agriculture, Forest Service, Pacific Northwest Research Station: Portland, OR, USA, 2004.
49. Marler, T.E. Cycad aulacaspis scale invades the Mariana Islands. *Mem. N. Y. Bot. Gard.* **2012**, *106*, 20–35.
50. Marler, T.E. Temporal variations in leaf miner, butterfly, and stem borer infestations of *Cycas micronesica* in relation to *Aulacaspis yasumatsui* incidence. *HortScience* **2013**, *48*, 1334–1338.
51. Marler, T.E.; Lawrence, J.H. Demography of *Cycas micronesica* on Guam following introduction of the armoured scale *Aulacaspis yasumatsui*. *J. Trop. Ecol.* **2012**, *28*, 233–242. [[CrossRef](#)]
52. McCormick, M.K.; Whigham, D.F.; Canchani-Viruet, A. Mycorrhizal fungi affect orchid distribution and population dynamics. *New Phytol.* **2018**, *219*, 1207–1215. [[CrossRef](#)] [[PubMed](#)]

