Chilades pandava Damage among 85 *Cycas* Species in a Common Garden Setting

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Abstract. The extent of *Chilades pandava* Horsfield herbivory among 85 Cycadaceae species was determined by three evaluators in a common garden setting in Thailand to identify patterns that may improve horticultural and conservation management practices. The significant differences in herbivory damage from this invasive lepidopteran pest ranged 8.7-fold among the species. Phylogenetic sections of this monogeneric cycad family did not correspond to the relative differences among the species, and country of nativity was also not informative for this purpose. We suggest the *Cycas* L. species that share native habitat with this butterfly or the closely related *Theclinesthes onycha* Hewitson are among the least damaged taxa when they are comingled with other *Cycas* species in a common landscape. Grouping the most damaged *Cycas* species together in a managed landscape may reduce costs associated with plant protection. The inclusion of non-native *Cycas* plants in gardens nearby native *Cycas* habitats carries the potential of disrupting the delicate specialist relationship that native butterfly populations have with host *Cycas* species.

Invasive arthropod herbivores comprise one of the greatest threats to cycad conservation both in situ and ex situ. The lepidopteran *Chilades pandava* (Plains Cupid Butterfly, Fig. 1B) is a specialist insect native to southern Asia that invaded Guam in recent years (Moore et al., 2005), and it is among the invasive species that are collectively threatening the endemic *Cycas micronesica* K.D. Hill (Marler and Lawrence, 2012). This insect depends on a sustained close relationship with native *Cycas* within its indigenous range. It causes extensive damage to *Cycas* populations within habitats in its invasive range like Guam. These factors engender a horticultural conflict between the need to protect *Cycas* species from damage within garden settings and within the butterfly's invasive range and the arguable need to conserve its dependence on *Cycas* plants in situ.

An extensive ex situ germplasm collection of Guam's *C. micronesica* was established at

Nong Nooch Tropical Botanical Garden in Thailand as part of our efforts to conserve this endangered cycad. In this setting where the butterfly is native, herbivory is highly heterogeneous among comingled *Cycas* species. Some species like *Cycas revoluta* Thunb. (Fig. 1C) and *C. micronesica* (Fig. 1E) exhibit extensive, unsightly damage, whereas other species exhibit minimal damage (Figs. 1D and 1F). Management decisions for this situation would benefit from understanding how the butterfly relates to its native host *Cycas* species in natural habitats and how it relates as a pest to novel *Cycas* host species in horticultural settings.

Most of the 107 described *Cycas* species (Osborne et al., 2012) are being conserved in the ex situ cycad germplasm collections in Nong Nooch Tropical Botanical Garden. Pesticide protection of susceptible *Cycas* taxa resulting from this pest represents one of the greatest costs for success of these conservation efforts. We used this germplasm collection growing in a common garden setting to determine relative butterfly herbivory at the species level. Our objective was to then mine these data for linkages between resistance to damage and group traits that may reduce uncertainty in predictions of and expose mechanisms that explain disparity in butterfly damage.

Three evaluators conducted this survey to quantify the extent of damage to representative field-planted *Cycas* species at Nong Nooch Tropical Botanical Garden in Thailand. The survey included 85 species growing in homogeneous abiotic and insect pressure conditions. Two arthropods exhibit chronic pest pressures on the *Cycas* plants in this garden, *Chilades pandava* and *Aulacaspis yasumatsui* Takagi. The butterfly can feed on several tissue types, but for every substrate, it requires expanding tissue for ovipositioning and caterpillar food. The various groups of *Cycas* species produce vegetative flushes at varied times of the year, and some exotic *Cycas* taxa produce leaves throughout the year. These characteristics provide the resident butterfly population with a sustained supply of caterpillar food. Therefore, the pest pressures are relentless and constant throughout the year so that no matter when a *Cycas* has a flush of leaves, it is subjected to adult butterfly ovipositioning. *A. yasumatsui*, the second resident *Cycas* pest in our garden, is an armored scale that infests any organ that presents live exposed surfaces. Unidentified biological control organisms keep the widespread *A. yasumatsui* infestations from erupting. Confusing the plant damage imposed by these two arthropods is not possible; therefore, identifying butterfly damage is unambiguous.

Individual plants were ranked for butterfly damage in Mar. 2012. We assigned 0 for a plant exhibiting no visible damage and 10 for a plant with butterfly damage on every leaf. A total of 673 plants in 85 species, representing all five *Cycas* sections, were evaluated. A Poisson regression model was fitted on log-transformed damage evaluation scores using SAS GLIMMIX procedure in Version 9.3 (SAS Institute Inc., Cary, NC). A Type III test of fixed main effects was used to determine significance of evaluators and species. The damage differences among the species were determined using pairwise comparison of least squares means on log scale.

Results and Discussion

The mean damage score among species was significant based on Type III test ($P \leq 0.0001$) after removing the evaluator differences. The log-transformed mean damage scores were back-transformed and presented as a LSMEANS table (Table 1). Butterfly damage of the 85 *Cycas* species in this common garden setting exhibited an overall mean of 2.093, and mean damage ranged 8.7-fold among the species (0.5947 to 5.1502). The damage scores for these species partitioned into 18 overlapping groups. Clearly, whether *Cycas* plants succumb to or resist damage by chronic butterfly pressure is governed by differences expressed at the species level.

Phylogeny. *Cycas micronesica*, *C. petraea* A. Lindstr. & K.D. Hill, *C. revoluta*, *C. seemannii* A. Braun, *C. tansachana* K.D. Hill & S.L. Yang, and *C. thouarsii* R.Br. ex Gaudich. emerged as the most damaged

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Fig. 1. Relationship of *Cycas* species with the specialist herbivore *Chilades pandava*. (A) Extent of herbivory damage for 85 *Cycas* species sorted into the five Cycadaceae sections, mean $\pm$ SD. (B) Female butterfly ovipositioning eggs on *Cycas revoluta* leaf. (C) *C. revoluta* plant showing typical extensive butterfly damage to leaflets. Damage score ≈ 9 . (D) *C. pachypoda* plant with undamaged leaves. Damage score 0. (E) Production beds of the susceptible *C. micronesica* showing plant mortality and heterogeneous growth. (F) Production beds of the resistant *C. chamaoensis* showing homogeneous growth.

species and *C. aculeata* K.D. Hill & T.H. Nguyễn, *C. circinalis* L., *C. condaoensis* K.D. Hill & S.L. Yang, *C. pachypoda* K.D. Hill, *C. pruinosa* Maconochie, *C. taitungensis* C.F. Shen, K.D. Hill, C.H. Tsou & C.J. Chen, *C. tuckeri* K.D. Hill, and *C. wadei* Merrill exhibited the least amount of damage among the 85 species (Table 1). Exploring characteristics that explain these two extreme groups may establish a foundation for understanding the controlling mechanisms of the disparity in damage within the genus. Two obvious group traits to explore for this purpose are phylogenetic sections and native biogeographic range.

The 107 species (Osborne et al., 2012) that comprise the Cycadaceae are classified into five sections (Hill, 2004) based primarily on morphological characters. Mean damage score for these sections exhibited a 2-fold non-significant range (Fig. 1A). The section Asiorientales, distinguished by tomentose ovules, provides an example of the immense

variation within each of these sections. Asiorientales exhibited a mean score that was greater than the overall survey mean, contained *C. taitungensis* in the least damaged group, and *C. revoluta* in the most damaged group. These major *Cycas* sections do not adequately inform our understanding of what underlies the differences in butterfly damage among the 85 species that we observed.

Our results do not broadly correspond with phylogeny. However, the non-significant lower damage scores in this garden survey for the species in the Wadeanae section indicate that direct bioassays may show this section to be more resistant than the other sections. Furthermore, one smaller group of species emerges as highly damaged relative to the overall list of *Cycas* species. The Rumphiae subsection of the Cycas section (Hill, 2004) is distinguished by spongy tissue within the seed sclerotesta. When fully developed, this tissue enables oceanic seed dispersal (Dehgan and Yuen,

1983). The mean for the eight species from this subsection in our survey was 3.1281, and half of the *Cycas* species ranked as most damaged (Table 1) were from this small subsection. An interesting example that underscores these observations is the Philippine island of Culion, where the strict endemic *C. wadei* (Wadeanae section without floating seeds) and the widespread *C. edentata* de Laub. (Rumphiae subsection within Cycas section with floating seeds) live on the main island and its barrier islands (Lindström et al., 2008). Damage to *C. edentata* was three times greater than that to *C. wadei*, and it is ranked among the top 10 most damaged species (Table 1). Although this efficient long-distance seed dispersal mechanism enables colonization of remote islands to establish an extensive indigenous range, dispersal to an unfested location may preclude or at least delay the butterfly or any other specialist from establishing in the new location.

Table 1. Extent of *Chilades pandava* herbivory among 85 *Cycas* species growing at Nong Nooch Tropical Botanical Garden in Thailand, native biogeographic range, and Cycadaceae section based on Hill (2004).^z

Species	Origin	Section	Minimum	Maximum	Mean	
<i>C. revoluta</i>	Japan	Asiorientalis	2	8	5.1502	A ^y
<i>C. seemannii</i>	South Pacific Islands	Cycas*	1	9	5.0062	A
<i>C. tansachana</i>	Thailand	Indosinensis	3	8	4.7055	A
<i>C. micronesica</i>	Palau, Yap, Mariana Islands	Cycas*	1	10	4.5935	A
<i>C. thourarsii</i>	East Africa	Cycas*	1	8	4.2826	AB
<i>C. petraea</i>	Thailand	Indosinensis	2	8	3.9958	ABC
<i>C. lacrimans</i>	Philippines	Cycas	2	5	3.3591	BCD
<i>C. bougainvilleana</i>	South Pacific Islands	Cycas*	1	9	3.1673	BCDE
<i>C. edentata</i>	Widespread in SE Asia	Cycas*	1	7	3.1003	BCDE
<i>C. multipinnata</i>	China, Vietnam	Stangerioides	1	7	3.0969	BCDE
<i>C. rumphii</i>	Indonesia, Papua New Guinea	Cycas*	1	7	3.0796	BCDE
<i>C. couttsiana</i>	Australia	Cycas	1	6	3.0522	BCDEF
<i>C. shanyangensis</i>	China	Stangerioides	2	4	3.0475	BCDEF
<i>C. megacarpa</i>	Australia	Cycas	2	5	2.9309	BCDEF
<i>C. conferta</i>	Australia	Cycas	1	5	2.8601	BCDEFG
<i>C. falcata</i>	Indonesia	Cycas	1	7	2.7982	BCDEFGH
<i>C. macrocarpa</i>	Thailand	Cycas	1	8	2.7584	BCDEFGHI
<i>C. desolata</i>	Australia	Cycas	1	6	2.7579	CDEFGHI
<i>C. dolichophylla</i>	Vietnam	Stangerioides	2	4	2.5761	CDEFGHIJ
<i>C. fairylakea</i>	China	Stangerioides	1	4	2.5592	CDEFGHIJ
<i>C. furfuracea</i>	Australia	Cycas	2	5	2.5054	CDEFGHIJ
<i>C. collina</i>	Vietnam	Stangerioides	1	5	2.4689	CDEFGHIJ
<i>C. vespertilio</i>	Philippines	Cycas	1	7	2.4379	CDEFGHIJ
<i>C. szechuanensis</i>	China	Stangerioides	2	4	2.4243	CDEFGHIJ
<i>C. arnhemica</i>	Australia	Cycas	1	4	2.3653	CDEFGHIJK
<i>C. campestris</i>	Papua New Guinea	Cycas	1	5	2.2848	DEFGHIJK
<i>C. sphaerica</i>	India	Cycas	1	4	2.2572	DEFGHIJK
<i>C. maconochie</i>	Australia	Cycas	0	5	2.2545	DEFGHIJK
<i>C. indica</i>	India	Cycas	1	5	2.2242	DEFGHIJK
<i>C. semota</i>	Australia	Cycas	1	5	2.2165	DEFGHIJK
<i>C. badensis</i>	Australia	Cycas	1	6	2.1874	DEFGHIJK
<i>C. angulata</i>	Australia	Cycas	1	5	2.1722	DEFGHIJKL
<i>C. micholitzii</i>	Vietnam	Stangerioides	1	4	2.1687	EFGHIJKL
<i>C. zeylanica</i>	Indian Ocean	Cycas*	0	4	2.1556	EFGHIJKL
<i>C. silvestris</i>	Australia	Cycas	1	6	2.1299	EFGHIJKL
<i>C. pranburiensis</i>	Thailand	Cycas	1	7	2.0725	EFGHIJKL
<i>C. guizhouensis</i>	China	Stangerioides	1	3	2.0351	EFGHIJKL
<i>C. inermis</i>	Vietnam	Cycas	1	4	2.0013	EFGHIJKLM
<i>C. ophiolitica</i>	Australia	Cycas	1	7	1.9543	EFGHIJKLM
<i>C. glauca</i>	Indonesia	Cycas*	1	3	1.9169	FGHIJKLM
<i>C. schumanniana</i>	Papua New Guinea	Cycas	1	3	1.9087	FGHIJKLM
<i>C. hoabinensis</i>	Vietnam	Stangerioides	1	3	1.8521	FGHIJKLM
<i>C. simplicipinna</i>	Widespread in SE Asia	Stangerioides	1	3	1.8388	FGHIJKLMN
<i>C. apoa</i>	Indonesia, Papua New Guinea	Cycas	1	3	1.8235	FGHIJKLMNO
<i>C. clivicola</i>	Widespread in SE Asia	Indosinensis	1	6	1.8231	FGHIJKLMNO
<i>C. debaoensis</i>	China	Stangerioides	1	3	1.8067	FGHIJKLMNO
<i>C. media</i>	Australia	Cycas	1	4	1.7710	GHIJKLMNOP
<i>C. yorkiana</i>	Australia	Cycas	1	4	1.7582	GHIJKLMNOP
<i>C. changiangensis</i>	China	Stangerioides	1	7	1.7560	HIJKLMNOP
<i>C. calcicola</i>	Australia	Cycas	1	3	1.7242	HIJKLMNOP
<i>C. pectinata</i>	Widespread in SE Asia	Indosinensis	1	8	1.6787	IJKLMNQP
<i>C. riuminiana</i>	Philippines	Cycas	1	4	1.6767	IJKLMNQP
<i>C. ferruginea</i>	Vietnam	Stangerioides	1	3	1.6386	IJKLMNQP
<i>C. papuana</i>	Papua New Guinea	Cycas	1	3	1.5974	IJKLMNQP
<i>C. xipholepis</i>	Australia	Cycas	1	3	1.5496	IJKLMNQP
<i>C. nitida</i>	Philippines	Cycas	1	2	1.5207	IJKLMNQP
<i>C. taiwaniana</i>	China	Stangerioides	1	3	1.5133	JKLMNQP
<i>C. lindstromii</i>	Vietnam	Indosinensis	1	3	1.5104	JKLMNQP
<i>C. sexseminifera</i>	Vietnam	Stangerioides	1	3	1.4920	JKLMNQP
<i>C. elongata</i>	Vietnam	Indosinensis	1	3	1.4774	JKLMNQP
<i>C. scratchleyana</i>	Widespread in S. Pacific	Cycas	1	4	1.4647	JKLMNQP
<i>C. beddomei</i>	India	Cycas	0	4	1.4509	JKLMNQP
<i>C. montana</i>	Indonesia	Cycas	1	2	1.4507	JKLMNQP
<i>C. diannanensis</i>	China	Stangerioides	1	3	1.4076	JKLMNOPQ
<i>C. brunnea</i>	Australia	Cycas	1	2	1.3830	KLMNOPQ
<i>C. nathorstii</i>	Sri Lanka	Cycas	1	3	1.3789	KLMNOPQ
<i>C. hainanensis</i>	China	Stangerioides	1	3	1.3772	KLMNOPQ
<i>C. tropophylla</i>	Vietnam	Stangerioides	1	3	1.3759	KLMNOPQ
<i>C. platyphylla</i>	Australia	Cycas	1	2	1.3604	KLMNOPQ
<i>C. aenigma</i>	Philippines	Wadeanae	1	2	1.3542	KLMNOPQ
<i>C. elephantipes</i>	Thailand	Indosinensis	0	4	1.3182	KLMNOPQ
<i>C. chevalieri</i>	Vietnam	Stangerioides	1	2	1.3138	KLMNOPQ
<i>C. nongnoochiae</i>	Thailand	Indosinensis	1	4	1.3054	KLMNOPQ

(Continued on next page)

Table 1. (Continued) Extent of *Chilades pandava* herbivory among 85 *Cycas* species growing at Nong Nooch Tropical Botanical Garden in Thailand, native biogeographic range, and Cycadaceae section based on Hill (2004).^z

Species	Origin	Section	Minimum	Maximum	Mean	
<i>C. balansae</i>	Vietnam	Stangerioides	1	2	1.2935	KLMNOPQ
<i>C. cairnsiana</i>	Australia	Cycas	1	2	1.2931	KLMNOPQ
<i>C. panzhihuaensis</i>	China	Asiorientalis ^w	0	3	1.2769	LMNOPQ
<i>C. chamaoensis</i>	Thailand	Indosinensis	1	3	1.2647	MNOPQ
<i>C. tuckeri</i>	Australia	Cycas	1	2	1.1559	MNOPQR
<i>C. pruinosa</i>	Australia	Cycas	1	2	1.1144	NOPQR
<i>C. circinalis</i>	India	Cycas	1	2	1.0967	OPQR
<i>C. wadei</i>	Philippines	Wadeanae	1	2	1.0598	OPQR
<i>C. pachypoda</i>	Vietnam	Indosinensis	0	2	1.0592	PQR
<i>C. condaoensis</i>	Vietnam	Indosinensis	0	2	0.9792	PQR
<i>C. taitungensis</i>	Taiwan	Asiorientalis	0	1	0.8389	QR
<i>C. aculeata</i>	Vietnam	Stangerioides	0	1	0.5947	R

^zMeans of three evaluators, 0 = no damage, 10 = extensive damage.

^yMeans with the same letter are not significantly different.

^xSubsection Rumphiae.

^wRecent evidence supports change to Stangerioides section.

Biogeography. *Cycas* is a widespread genus comprised of species that range geographically from strict endemics to some widespread species (Table 1). Native geographic range at the country or regional level is also not useful for explaining the differences that we found among these 85 species. Although the gross geographic region of origin does not appear to be useful for understanding the differences in butterfly damage among *Cycas* species, there may be explanations for the diversity in damage noted within a country or region. We propose that identifying the *Cycas* species possessing a native habitat that is shared with the butterfly may be informative. Four subspecies of this cycad specialist Lycaenidae species are currently recognized. *Chilades pandava lanka* is known from Sri Lanka, *Chilades pandava peripatria* occurs exclusively in Taiwan, *Chilades pandava vapidula* occurs in the Philippines, and the widespread *Chilades pandava pandava* is found in mainland China and Southeast Asia (Hsu, 2002; Igarashi and Fukuda, 2000; Wu et al., 2010). The precise genotype of the population we studied at Nong Nooch Tropical Botanical Garden is not known but is likely an admixture of *Chilades pandava pandava* introductions from numerous locations. The closely related Lycaenidae specialist *Theclines thes onycha* occurs in Australia (Braby, 2004; Forster and Machin, 1994). The eight *Cycas* species that assemble as the least damaged species (Table 1) occur within these prescribed geographic ranges.

For the Australian *Cycas* species exhibiting greater than average butterfly damage in our survey, these may occur outside the constrictions of Queensland and New South Wales that comprise the contiguous native range of *T. onycha* (Braby, 2004). Furthermore, any evolved reactions of a *Cycas* species to *T. onycha* herbivory may not translate to how that *Cycas* reacts to a different genus or species of herbivore. A full understanding of why some *Cycas* species are susceptible to butterfly damage despite their nativity within known butterfly range is more elusive. An example is *C. tansachana*, which is heavily damaged by butterfly relative to the entire list of *Cycas* (Table 1). *Cycas tansachana* is

endemic to a small limestone outcrop with extremely high surface temperatures. We suspect that this butterfly cannot thrive in this habitat despite its location within the indigenous range for the butterfly. The details of field conditions to validate these habitat-specific relationships will require direct observations of butterfly behavior in situ.

Mechanisms and caveats. The specificity of lepidopteran larvae to feed on particular host plant genotypes is well studied (Ehrlich and Raven, 1964). We have discussed potential underlying mechanisms that explain the disparity in butterfly feeding behavior in relation to *Cycas* species (Marler et al., 2012). One mechanism may be an information-mediated system that is founded in differences in constitutive or induced leaf volatiles that searching female butterflies may use as advertisements for suitable tissue for larval development (Bernays and Chapman, 1994). This mechanism could be tested with oviposition choice tests whereby reproductive female butterflies are simultaneously provisioned with expanding leaves of least-damaged vs. most-damaged *Cycas* species. An alternative mechanism may be a resource-mediated system that is founded in differences among the *Cycas* species for leaf palatability and defense traits. This mechanism may be expressed within the most damaged species as greater abundance or diversity of phagostimulants for feeding caterpillars and/or slow development of leaf toughness as leaves expand. Alternatively, it may be expressed within the least damaged species as constitutive or induced plant substances that inhibit caterpillar feeding and/or rapid leaf growth out of the soft, vulnerable tissue stage. Relevance of this resource-mediated mechanism could be tested with caterpillar bioassays that determine survival and growth rates among caterpillars reared on expanding leaf tissue of various *Cycas* species. We note that many species have individuals exhibiting no discernible butterfly damage (indicated by 0 in Table 1) despite high-density butterfly population pressure and our observation of ovipositioning on all *Cycas* species. Therefore, these assays should include first instar performance because this stage is highly vulnerable for many Lepidoptera species (Zalucki et al., 2002).

The extant literature illuminates several issues that deserve mention as possible relevant factors for interpreting the results of our study. For this *Cycas*–*Chilades* system, we have mentioned (Marler et al., 2012) that tritrophic relations may partly mediate the relationship of the herbivore with the *Cycas* plant (e.g., Harrewijn et al., 1994). Activity of parasitoids or other natural enemies of butterfly eggs or caterpillars may interplay to explain the highly heterogeneous extent of herbivory at the *Cycas* species level. In addition, the step of ovipositioning by itself may suppress emission of both constitutive and herbivore-induced volatiles in some plants (Peñaflor et al., 2011). Therefore, disparity in timing of ovipositioning among *Cycas* species within close proximity may influence the attraction of subsequent gravid female butterflies. Moreover, genotype of neighboring plants may influence herbivore abundance on focal plants (Genung et al., 2012). Consequently, herbivory of an individual *Cycas* tree within a common garden setting may be influenced by layout of the species within the planting. Our understanding of how these sorts of interactions affect plant–pest relations in cycad biology is nil.

The current literature is also revealing more about belowground and aboveground interactions in relation to arthropod herbivory. For example, leaf herbivory may directly influence root microbiota (Landgraf et al., 2012), yet root microbiota may directly influence aboveground volatile infochemicals produced by plants (Pineda et al., 2012; Schausberger et al., 2012). The *Cycas* plant rhizosphere is characterized by three-way relations among plant roots, mycorrhizae, and cyanobacteria (Fisher and Vovides, 2004; Muthukumar and Udaiyan, 2002; Norstog and Nicholls, 1997). Little is known about the genotype diversity and function of the microbiome of *Cycas* in a common garden setting. Preliminary work indicates that cyanobacteria genotype within each coraloid root complex is homogeneous, but within one plant's root system, each coraloid root can be colonized by different genotypes (Costa et al., 1999). The relationship among an in situ population of the butterfly, its host *Cycas* population, and the natural root symbiont

populations is not influenced by anthropogenic introductions of non-native organisms.

The caveats discussed here illuminate some of the challenges involved in interpreting research conducted in botanic garden settings. For example, a botanic garden in which leaf damage by the butterfly is highly heterogeneous among closely planted *Cycas* individuals provides a situation in which these feedback loops between aboveground and belowground relations may become a part of the mechanisms that foster sustained disparity in foliar damage among the *Cycas* species. Moreover, fertilization and irrigation regimes in managed landscapes may influence the relations of cultivated *Cycas* plants in a manner that deviates from that in natural habitats. In the current study, microsite variation throughout the garden and disparity in age and size of the trees are among other potentially confounding factors.

We have focused exclusively on one Lycaenidae pest in this discussion, but in the real world, a cycad plant is attacked by multiple herbivores. In Guam, for example, *C. micronesica* is threatened by a growing list of invasive and native specialist arthropods living in sympatry (Marler and Muniappan, 2006; Marler et al., 2011). The armored scale *A. yasumatsui* deserves mention as a rapidly invading pest that infests *Cycas* populations throughout much of the native and invasive *Chilades pandava* range. Most *Cycas* trees in Thailand, including within our garden setting, experience minor infestations of this scale pest. An understanding of butterfly herbivory cannot be fully attained if the influences of *A. yasumatsui* and other pests are ignored in the process. For example, Gómez et al. (2012) used individual and combined attack of *Tsuga canadensis* (L.) Carrière by a scale insect and a gall-forming insect to show that feeding by one arthropod herbivore altered the tissue quality for and damage by the other herbivore.

Horticulture and conservation. Native and invasive populations of *Chilades pandava* affect horticultural decisions in several ways. First, the aesthetic appeal of susceptible *Cycas* trees can be entirely removed by the damage imposed on susceptible species (Fig. 1C). In some locations this pest cannot be ignored by landscape managers. Second, the desired homogeneous growth rates needed to reduce costs in any horticultural production system are difficult to achieve for the susceptible *Cycas* species when this pest is present (Fig. 1E). The growth rates of resistant *Cycas* species are homogeneous among plants despite chronic butterfly pressure (Fig. 1F). These two factors underscore the importance of understanding the biology of the pest for the best horticultural outcomes and defining the most appropriate means of control. Third, horticulturists would do well to acknowledge their role in conservation, and this case study provides a good example of the complications in fulfilling that role. Introducing non-native *Cycas* trees into landscapes close to *Cycas* habitats, especially

those of endemic *Cycas* species, has the potential of disrupting the specialist dependence of the habitat's natural butterfly population on its co-occurring *Cycas* species. For example, Wu et al. (2010) reported that horticultural plantings of *C. revoluta* in Taiwan lead to irruptions of the native *Chilades pandava* that in turn increased the threats to the endemic, endangered *C. taitungensis* populations. Cycad conservation plans may be more effective if the role of cycad trees in the life histories of other native organisms is prominently figured into the plans.

Expanding leaves on all *Cycas* plants in a garden may be frequently sprayed with insecticides for protection against butterfly damage as one means of managing the pest. At Nong Nooch Tropical Botanical Garden, these expensive sprays are applied at weekly intervals. Our results and field observations indicate that one means of improving efficiency would be to eliminate a fixed spray regime on the most resistant *Cycas* species. If the *Cycas* species comprising the least damaged taxa could be positioned together in the garden landscape, then this section of the garden could be avoided during sprays applied on a fixed schedule. This may result in substantial reduction in pesticide costs and would also allow limited human resource effort to focus more heavily on protecting the susceptible species.

Some or all of the disparate mechanisms and caveats that we discussed here may mediate the relationships between *Chilades pandava* and comingled *Cycas* species in a garden landscape. The real situation likely is a conflation of multiple mechanisms.

Literature Cited

- Bernays, E.A. and R.F. Chapman. 1994. Host-plant selection by phytophagous insects. Chapman and Hall, New York, NY.
- Braby, M.F. 2004. The complete field guide to butterflies of Australia. CSIRO Publishing, Collingwood, Australia.
- Costa, J.-L., P. Paulsrud, and P. Lindblad. 1999. Cyanobiont diversity within coralloid roots of selected cycad species. FEMS Microbiol. Ecol. 28:85–91.
- Dehgan, B. and C.K.K.H. Yuen. 1983. Seed morphology in relation to dispersal, evolution and propagation of *Cycas* L. Bot. Gaz. 144:412–418.
- Ehrlich, P.R. and P.H. Raven. 1964. Butterflies and plants: A study in coevolution. Evolution 18: 586–608.
- Fisher, J.B. and A.P. Vovides. 2004. Mycorrhizae are present in cycad roots. Bot. Rev. 70:16–23.
- Forster, P.I. and P.J. Machin. 1994. Cycad host plants for *Lilioceris nigripes* (Fabricius) (Coleoptera: Chrysomelidae) and *Theclines thes onycha* (Hewitson) (Lepidoptera: Lycaenidae). Aust. Entomol. 21:99–102.
- Genung, M.A., G.M. Crutsinger, J.K. Bailey, J.A. Schweitzer, and N.J. Sanders. 2012. Aphid and ladybird beetle abundance depend on the interaction of spatial effect and genotypic diversity. Oecologia 168:167–174.
- Gómez, S., C.M. Orians, and E.L. Preisser. 2012. Exotic herbivores on a shared native host: Tissue quality after individual, simultaneous, and sequential attack. Oecologia 169:1015–1024.
- Harrewijn, P., A.K. Minks, and C. Mollema. 1994. Evolution of plant volatile production in insect–plant relationships. Chemecology 5: 55–73.
- Hill, K.D. 2004. Character evolution, species recognition and classification concepts in the Cycadaceae, p. 23–44. In: Walters, T. and R. Osborne (eds.). Cycad classification: Concepts and recommendations. CABI Publishing, Cambridge, MA.
- Hsu, Y.F. 2002. Butterflies of Taiwan. National Fonghuanggu Bird Park, Nantou, Taiwan.
- Igarashi, S. and H. Fukuda. 2000. The life histories of Asian butterflies. Tokai University Press, Tokyo, Japan.
- Landgraf, R., R.S. Schaarschmidt, and B. Hause. 2012. Repeated leaf wounding alters the colonization of *Medicago truncatula* roots by beneficial and pathogenic microorganisms. Plant Cell Environ. 35:1344–1357.
- Lindström, A.J., K.D. Hill, and L.C. Stanberg. 2008. The genus *Cycas* (Cycadaceae) in the Philippines. Telopea (Syd.) 12:119–145.
- Marler, T.E. and J.H. Lawrence. 2012. Demography of *Cycas micronesica* on Guam following introduction of the armored scale *Aulacaspis yasumatsui*. J. Trop. Ecol. 28:233–242.
- Marler, T.E., A. Lindström, and L.I. Terry. 2012. Information-based or resource-based systems may mediate *Cycas* herbivore interactions. Plant Signal. Behav. 7:760–762.
- Marler, T.E. and R. Muniappan. 2006. Pests of *Cycas micronesica* leaf, stem, and male reproductive tissues with notes on current threat status. Micronesica 39:1–9.
- Marler, T.E., L.S. Yudin, and A. Moore. 2011. *Schedorhinotermes longirostris* (Isoptera: Rhinotermitidae) on Guam adds to assault on the endemic *Cycas micronesica*. Fla. Entomol. 94: 699–700.
- Moore, A., T.E. Marler, R. Miller, and R. Muniappan. 2005. Biological control of cycad aulacaspis scale on Guam. The Cycad Newsletter 28:6–8.
- Muthukumar, T. and K. Udaiyan. 2002. Arbuscular mycorrhizas in cycads of southern India. Mycorrhiza 12:213–217.
- Norstog, K.J. and T.J. Nicholls. 1997. The biology of the cycads. Cornell Univ. Press, Ithaca, NY.
- Osborne, R., M. Calonje, K. Hill, L. Stanberg, and D.W. Stevenson. 2012. The world list of cycads. Mem. N. Y. Bot. Gard. 106:480–508.
- Peñaflor, M.F., M. Erb, C.A. Robert, L.A. Miranda, A.G. Werneburg, F.C. Dossi, T.C. Turlings, and J.M. Bento. 2011. Oviposition by a moth suppresses constitutive and herbivore-induced plant volatiles in maize. Planta 234:207–215.
- Pineda, A., R. Soler, B.T. Weldegergis, M.M. Shimwela, J.J.A. van Loon, and M. Dicke. 2012. Non-pathogenic rhizobacteria interfere with the attraction of parasitoids to aphid-induced plant volatiles via jasmonic acid signaling. Plant Cell Environ. (in press).
- Schausberger, P., S. Peneder, S. Jürschik, and D. Hoffmann. 2012. Mycorrhiza changes plant volatiles to attract spider mite enemies. Funct. Ecol. 26:441–449.
- Wu, L.-W., S.-H. Yen, D.C. Lees, and Y.-F. Hsu. 2010. Elucidating genetic signatures of native and introduced populations of the Cycad Blue, *Chilades pandava* to Taiwan: A threat both to Sago Palm and to native *Cycas* populations worldwide. Biol. Invasions 12:2649–2669.
- Zalucki, M.P., A.R. Clarke, and S.B. Malcolm. 2002. Ecology and behavior of first instar larval Lepidoptera. Annu. Rev. Entomol. 47:361–393.