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# LARVAE OF IO MOTH, AUTOMERIS IO, ON THE CORAL BEAN, ERYTHRINA HERBACEA, IN FLORIDA—THE LIMITATIONS OF POLYPHAGY

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**ABSTRACT.** The coral bean, *Erythrina herbacea*, is reported as a new host for *Automeris io* in north-central Florida, based on a single batch of larvae found on this plant in nature and reared through on it in the laboratory. However, the consequent laboratory rearing showed a high cost associated with using this host plant. The mortality of young larvae of the F-2 generation reared on *E. herbacea* was very high (over 90%), and much higher than that of control larvae, which were reared on mature leaves of *Quercus nigra* (14–38%). The leaves of *E. herbacea*, known for their toxic alkaloids, were extracted with methanol, made into a water solution, applied on the mature leaves of *Q. nigra*, and fed to 1st instars of *A. io*. This produced no negative effect on the caterpillars. While the mature leaves of *Q. nigra* produced low mortality in young caterpillars, the young terminal leaves of this plant were as lethal to *A. io* as leaves of *E. herbacea*. Additionally, it was noted that rearing larvae on *E. herbacea* (a diet with a higher N2 content) led to faster larval development and smaller adult moths. The *A. io* larvae in this study developed during 63-100 days and underwent seven larval instars, which contradicts many popular accounts of the fifth instar being the final in *A. io*. Finally, *Prunus angustifolia* proved to be an unsuitable hostplant for *A. io*, as feeding on this species led to the arrested development of larvae and their eventual death.

Additional key words: alkaloids, caterpillar, community ecology, nutrition, plant and herbivore defense

On 13 September 2011 in Gainesville, Florida, I found a cluster of neonate caterpillars of Automeris io Fab. (Saturniidae) feeding on leaves of the coral bean, *Erythrina herbacea* L. (Fig. 4A, B). Automeris io is a highly polyphagous species, with over 100 host plants listed just for the state of Florida (Heppner 2003). Among them are plants such as *Citrus, Solanum, Salix,* and *Prunus* that contain toxic compounds in their leaves. Therefore, it is not surprising to find yet another host plant to be palatable to the *A. io* larvae, even if this host plant is laced with alkaloids (e. g., Powell & Westley 1993). However, considering how few Lepidoptera are able to utilize this widely available plant as their host, this discovery seemed to constitute a notable expansion in the diet of *A. io*.

Indeed, in Florida, where ca. 3,000 Lepidoptera species are found, the only ones that were recorded to feed on *Erythrina herbacea* are a psychid, *Cryptothelea* gloverii (Packard), a tineid, *Opogona sacchari* (Bojer), lycaenids, *Celastrina ladon* Cramer and *C. neglecta* Edwards, a pyralid, *Trachylepidia fructicassiella* Ragonot, three crambids, *Epicorsia oedipodalis* Guenée, *Terastia meticulosalis* Guenée and *Agathodes designalis* Guenée (Heppner 2003), and a lyonetiid, *Leucoptera erythrinella* Busck. During my extensive field work in north central Florida that involved searching multiple *E. herbacea* bushes for caterpillars, I observed only the latter three species feeding on this plant (Sourakov 2011; 2012).

Genus Automeris has been known to utilize various other species of *Erythrina* in Central America. For instance, Automeris metzli (Sallé) was found to feed on *Erythrina costaricensis* Micheli; *Automeris banus* Boisd. and *A. zugana* Druce - on *Erythrina lanceolata* Standl. (Janzen & Hallwachs 2012); *Automeris illustris* (Walker) - on *Erythrina cysta-gali* L. and *E. falcata* Bentham (Specht et al. 2006). Hence, the genes responsible for the ability to detoxify *Erythrina* alkaloids are not novel in *Automeris*, but whether they have migrated to the *A. io* population in Florida from more tropical populations remains to be shown. Alternatively, of course, *A. io* may have always occasionally utilized *Erythrina* as one of its host plants, and this has simply not been heretofore discovered.

Closely related species and even populations within species can vary in detoxification abilities, as was shown in Papilio glaucus L./Papilio canadensis Rothschild & Jordan species complex and the luna moth (e. g., Lindroth et al. 1986, Lindroth 1989), and this can even become a driving force for speciation (e.g., Mercader et al. 2009, Zvereva et al. 2010). Some plant compounds can have metabolic repercussions for the insect in the form of delayed larval development (e.g., Lindroth et al. 1988a). Such compounds can be present in different quantities in different, even closely related, species of plants, and can determine the growth pattern and ability to complete metamorphosis in generalist moth species (e. g., Ruuhola et al. 2001). Herbivores frequently develop adaptations to plant-specific combinations of defensive compounds and these compounds can determine their choice of food (e.g., Tahvanainen et al. 1985). Other types of plant compounds, such as lectins, were shown to negatively affect larval growth, survival, and pupation, as well as adult emergence and fecundity

(Machuka et al. 1999, Fitches et al. 1997, Fitches et al. 2001, Shukle & Murdock 1983). Lectins are also known to be a deterrent of oviposition behavior (e. g., Michiels & Smagghe 2010). Plant lectins were shown to have a species-specific effect on insects, causing high mortality in some herbivores, while simultaneously being completely neutral to others (e. g., Machuka et al. 2000).

Larval growth rate can also be positively affected by an increased nitrogen contents in plants (e. g., Manuwoto & Scriber 1985). Leaves of different plant species can seasonally differ in value as host plants to the same species of herbivore, and, remarkably, such variation is not universal, but unique for each host plant considered (e. g., Lindroth et al. 1988b, Finke & Scriber 1988). In the latter study, the low water content of ingested leaves was found to be responsible for decreasing the growth rate of larvae, even when the nitrogen levels in leaves remained adequate. The nitrogen and water in poplar trees, for example, as well as glycosides responsible for suppressing herbivores, decline in the fall, but such fluctuations vary between individual trees (Lindroth et al. 1987).

If *Erythrina herbacea* is not among the normal host plants for Automeris io, the question is: How and why did the A. io female choose to oviposit on it? Studies on butterflies suggest that contact chemoreception can guide oviposition choices (e. g., Frankfater & Scriber 2003). Oviposition preference can also be on a genotypic level, not only of insects but of plants as well (Bossart & Scriber 1995). It is not the nutritional value, but rather presence of repellent compounds that may be responsible for a host plant not being regularly utilized by polyphagous herbivores. Frankfater and Scriber (1999) showed, for example, that red bay leaf extracts deter oviposition by Papilio glaucus, even though the host plant might otherwise be suitable for larval development. Larvae of many other Lepidoptera species can survive on host plants on which they would never oviposit in the wild (e. g., Dowell et al. 1990). Studies (e. g., Sétamou et al. 2002; Hogervorst et al. 2008; Bell et al. 2004) suggest that the detrimental effect of insecticidal plant compounds can be passed on to the next trophic level-the parasitoids. Thus, it is possible that the generalist parasitoids avoid searching E. herbacea plants for hosts, because there are few potential hosts that feed on it and because those that do might reduce the fitness of the parasitoid's offspring as a result of the ingested toxins. During my previous studies, which involved collecting and rearing numerous caterpillars of crambid moths off of E. herbacea (Sourakov 2011; 2012), not a single larva of these moths was found to be parasitized. Hence in Florida, where

the larval parasitism by generalist tachinid flies is frequent and can reach as high as 90% (e. g., Sourakov 2009), oviposition on a toxic plant, such as *E. herbacea* by a polyphagous species, such as *A. io*, while possibly a complete "accident", could also prove to be an example of exploiting an enemy-free space, in which a tradeoff exists between high larval mortality and low parasitism rates.

In the present study, following the initial discovery of *Automeris io* larvae feeding on *Erythrina herbacea* in the wild, I examined the relative suitability of this host plant as a host of *A. io* by comparing its costs and benefits to those of other diets in controlled settings and attempted to provide an explanation to the resulting observations.

# MATERIALS AND METHODS

**Experiment 1:** Rearing a brood of field collected *A. io* larvae on different hostplants

Sibling first instar larvae of Automeris io (N=14) were found in the wild feeding as a group near their empty egg shells on E. herbacea foliage on 13 September 2011(Fig. 4A, B). They were maintained as a group inside a plastic bag feeding on *E. herbacea* until the end of the 3rd instar (3 October 2011), when they were split into five groups and from this point raised in separate plastic bags. Two groups of three larvae were continued on E. herbacea, the third group of three larvae was switched to the black oak (Quercus nigra L.), the fourth group of three larvae to the chicksaw plum (Prunus angustifolia Marsh.), and the remaining two larvae to the blackberry (Rubus flagellaris Willd.). Bags and host plant cuttings were replaced three times a week, and an effort was made to use conspecific plants and leaves of similar age. Fresh brown tissue paper was placed in each bag to absorb excessive moisture. Towards the end of larval development, larvae that reached the final (7th) instar were maintained individually and pupated inside cocoons spun among tissue paper and hostplant leaves. Bags with pupae were kept on an ambient light cycle at ca. 75°F until mid-December, when they were transferred to a non-heated garage, where conditions reflected ambient variations in temperature and humidity. In February 2012, the bags with pupae were brought back into the laboratory, where they were kept inside the plastic bags until emergence in May. Drops of water were periodically added to keep the pupae from desiccating. Live adults were weighed upon emergence and their right forewing was measured using calipers.

**Experiment 2:** Rearing a lab-obtained brood 1 of *A*. *io* on *E*. *herbacea* and *Q*. *nigra* 

A female and a male raised on *Erythrina herbacea* during Experiment 1 emerged on 6 May and 8 May

2012 respectively, and were allowed to mate in a cage. The resulting 332 eggs proved to be fertile and the first instar larvae were offered pieces of Quercus nigra and E. herbacea onto which they crawled at will. These pieces of host plants were then transferred into separate plastic cups where clippings of respective host plants were added and larvae were raised using the feeding protocol outlined in Experiment 1. A total of 18 such cups with first instar larvae ranging in number from 14 to 30 were initially set up, and were treated as separate replications in the statistical analysis of the data. Ten cups contained Erythrina herbacea (mean number of larvae per cup = $20 \pm 5$  (SD); N (total)=199), and eight cups contained *Q. nigra* (mean number of larvae per  $cup = 18 \pm 3$  (SD); N(total)=133). During this experiment, only older foliage was provided as food, corresponding to the type of foliage on which larvae were found in the field and maintained during Experiment 1. Larval mortality was assessed three times a week during the change of host plant clippings and cups.

**Experiment 3:** Rearing a lab-obtained brood 2 of *A*. *io* on *E*. *herbacea* and *Q*. *nigra* (leaves with variable water contents)

By crossing a male raised on oak during the Experiment 1 and female raised on blackberry (both emerged on 6 June 2012), a brood of 278 larvae was obtained. These larvae hatched on 9 July, were split into groups, and reared using protocol similar to Experiment 2. This time however, several types of foliage were used: very young tender leaves of the terminal leaf growth, firm but still immature leaves of slightly older terminal leaves (light green in color), and old firm and dark green leaves of *Quercus nigra* were placed in separate cups. Similarly, older (dark green) leaves of Erythrina herbacea were placed in one of the cups, while other cups contained tender terminal leaves (light green). There were a total of 14 cofee cups: 7 with larvae feeding on young E. herbacea, 7 with larvae feeding on Q. nigra (two of which contained terminal tender leaves, four terminal firm leaves, and one mature leaves). Larval mortality was assessed during the change of hostplant material.

**Experiment 4:** Determining the relative toxicity of *E. herbacea* leaf compounds to young *A. io* larvae

The mature leaves of *E. herbacea* and *Q. nigra* were collected in June, dried at room temperature, and ground up separately using a coffee maker. Three grams of the leaf powder resulting from each of the species were separately extracted using 20 ml of cold methanol in an ice-bath for 25 minutes. The mixture was filtered, resulting in a bright green solution of leaf chemicals, which was allowed to evaporate under the hood. The glass beakers with the resulting residue of dry plant compounds were then stored at -23°C until use. The residue was diluted with 10 ml of water and the leaves of *Q. nigra* were dipped into the solutions and air dried prior to feeding to A. io caterpillars. Fourteen first instar A. *io* larvae in separate plastic cups were fed on leaves dipped into each of the solutions. Leaves dipped in tap water were used as the control. Mortality was assessed over the span of one week, during which caterpillars fed continuously on the leaves.

The water contents of leaves was measured by clipping them at the base from the plant, weighing them fresh, and then drying them in the lab at room temperature and appx. 50% humidity prior to reweighing. A sample of host plant leaves was dried, ground-up into powder, and analyzed for nitrogen content using an Eager 200 CHN analyzer.

## RESULTS

**Experiment 1:** Rearing a brood of field collected *A*. *io* larvae on different hostplants

On *Erythrina herbacea*: All six larvae of *Automeris io* continued developing normally; one male pupated in 46 days; five females pupated simultaneously after 63 days of development. In *A. io*, males are smaller, hence faster development and smaller larva/pupa in males is to be expected. On *Quercus nigra*: two larvae (one male, one female) pupated after 65 days, and one female pupated after 70 days. Despite the small sample size, time to pupation of *A. io* females raised on *E. herbacea* (= 63±0 days (N=5)) differed significantly (P < 0.05) from that of those raised on *Q. nigra* (= 67.5±3.5 days; (N=2)). *Automeris io* females raised on *E. herbacea* were

TABLE 1. Water and nitrogen contents in leaves of *Erythrina herbacea* and *Quercus nigra* and the corresponding larval mortality of *Automeris io*.

	Erythrina herbacea- young leaves	<i>Quercus nigra-</i> young leaves	<i>Erythrina herbacea-</i> old leaves	<i>Quercus nigra-</i> old leaves
Nitrogen (%)	N/A	N/A	2.62.8%(Jun); 2.02.1%(Sep)	1.2-1.3%
Water (%)	78.3%	57.9-67.5%	59.2-62.4%	40.1%
Mortality	High	High	High	Low

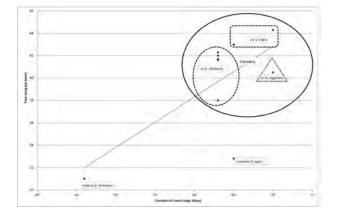


FIG. 1a. Experiment 1: Correlation between larval stage duration and size of resulting adult *Automeris io* moths found on *Erythrina herbacea* in the wild as 1st instar and subsequently raised on *E. herbacea*, *Quercus nigra* and *Rubus flagellaris*.

smaller than those raised on Q. nigra (FW length=40.8±1.9mm; N=4 vs. FW length=43.7±0.7mm; N=2). Correlation between size of the resulting adults and length of larval development can be observed in Figure 1a. On *Rubus flagellaris*: One larva developed at a normal rate, but died after 30 days as an early fifth instar; the other (female) pupated after 70 days. It appears that development on this host plant occurred at a similar rate to that on Q. nigra, but the resulting female was smaller. On *Prunus angustifolia*: One larva continued feeding after transfer from *Erythrina*, but

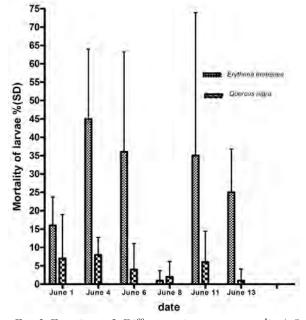


FIG. 2. Experiment 2: Differences in average mortality (±St. Dev.) of young *Automeris io* larvae raised on *Erythrina herbacea* vs. *Quercus nigra*.

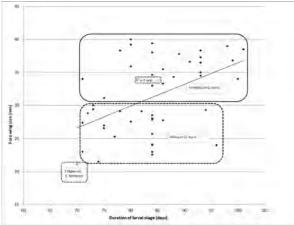


FIG. 1b. Experiment 2: Correlation between larval stage duration and size of resulting adult *Automeris io* moths raised on *Erythrina herbacea* vs. *Quercus nigra*.

showed completely arrested development, and died 71 days later at the 4th instar without visibly gaining in size. The other two larvae died at the 6th instar after 85 and 110 days of feeding. Their growth was noticeably slower than the growth of those that fed on other host plants.

Seven larval instars: The present study allowed, among other things, for a detailed assessment of *Automeris io* biology. The larvae underwent seven larval instars, which contradicts many popular accounts of the fifth instar being the final (e. g., Wikipedia 2013). Such discrepancy is quite common, partly because it is frequently erroneously assumed that the final instar is the fifth one. Only by thoroughly observing and collecting head capsules at every molting (Fig. 4D) is it possible to determine the true number of instars in Lepidoptera.

**Experiment 2:** Rearing a lab-obtained brood 1 of *A*. *io* on *E*. *herbacea* and *Q*. *nigra* 

Figures 2 and 3a illustrate the difference in survival dynamics of larvae fed on Erythrina herbacea and Quercus nigra. Young Automeris io larvae survived much better when raised on Q. nigra than on E. herbacea. After the first three days, the average mortality on *E. herbacea* was twice as high,  $=16\pm8\%$ ( $\pm$ SD), versus 7 $\pm$ 12% on Q. nigra (P<0.05). More importantly, dead larvae were found in every one of the 10 cups containing E. herbacea, while larvae experienced mortality in only three of the cups containing Q. nigra. Between 1 June and 4 June, the trend continued, with mortality on E. herbacea being much higher at an average of  $45\pm19\%$ , vs. only  $8\pm5\%$ when feeding on Q. nigra (P<0.0006). Between 4 June-6 June, after nine days of feeding, at which point some larvae reached the 2nd instar, larvae feeding on E.

herbacea experienced 36±27% average mortality vs. only  $4\pm7\%$  on Q. nigra (P<0.004). This occurred in every cup containing E. herbacea, while 100% survival occurred in 5 out of 8 cups containing Q. nigra. Relative mortality was calculated only for cups which still retained six larvae or more, and hence was not assessed following June 13, by which date all E. herbacea test cups contained fewer than six larvae. The initial mortality on Q. nigra plateaued, while on E. herbacea larvae continued to die off throughout the first two instars. Larvae developed to pupal stage faster on E. herbacea  $(70.3\pm0.6 \text{ days } (N=3))$  than on Q. nigra (86±7.5 days (N=41)) (P<0.001). The 51 adults that emerged in 2012–13 showed the same trend as in Experiment 1: adults raised on *E. herbacea* were smaller than those raised on *Q. nigra* (Figure 1b).

**Experiment 3:** Rearing a lab-obtained brood 2 of *A. io* on *E. herbacea* and *Q. nigra* (leaves with variable water content)

Figure 3b illustrates the difference in survival dynamics of larvae fed on leaves of E. herbacea and on young and old leaves of *Q. nigra*. At the end of July, the water content in mature *Erythrina herbacea* was 59.2%, while the water content in mature Quercus nigra was only 40.1%. However the firm terminal young leaves of Q. nigra had a water content of 57.9%, while the terminal young immature (tender) leaves were 67.5% water. Leaves of E. herbacea collected at the end of September contained 62.4% of water, while Q. nigra leaves contained just 41.4%. In mid-June, the nitrogen content in E. herbacea was 2.8%, in contrast to the nitrogen content of *Q. nigra*—1.2%. Towards the end of September, nitrogen in E. herbacea leaves dropped to 2.0%, while it remained the same (1.2%) in Q. nigra (Table 1).

Larvae started feeding successfully and evenly on *Q*. nigra in all 15 cups. With the exception of the cup containing old Q. nigra leaves where larvae reached second instar three days earlier, the larvae developed at similar rates. Mortality was first observed in cups containing tender Q. nigra leaves, with all larvae in these cups dying off. During molting leading to the second instar, high mortality was observed in all cups, except for the cup with old *Q. nigra* leaves (Figure 3b). This mortality correlated with an apparent bloating of the larvae (Fig. 4H) when compared to the larvae fed on old oak leaves (Fig. 4G). Also, the normal ability of the larvae to stay in tight-knit groups when migrating from leaf to leaf, feeding, or molting (Figs. 4 B, C, F, I) was lost, with larvae spreading out to feed solitarily or in small groups of 2-3.

**Experiment 4:** Testing the relative toxicity of *E. herbacea* leaf compounds to young *A. io* larvae

Larvae developed normally in all three cups and 0% mortality was observed in cups containing *Q. nigra* leaves treated with *E. herbacea* extract, *Q. nigra* extract, and the control leaves.

# DISCUSSION

It appears that the principals postulated by Scriber and Feeny (1979) concerning individual leaf qualities and those proposed by Lindroth (1989) concerning individual broods/populations being the key to the survival of larvae apply to the usability of *Erythrina herbacea* as a host plant of *Automeris io*. The same applies to *Quercus nigra*: successful as I was in Experiments 1 and 2 when raising *A. io* on that plant, this success vanished when leaves with a higher water

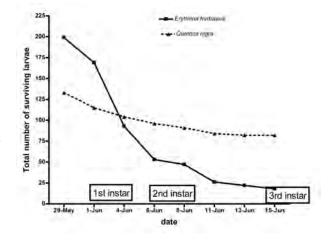


FIG. 3a. Experiment 2: Survival of young *Automeris io* larvae when raised on *Erythrina herbacea* and *Quercus nigra*.

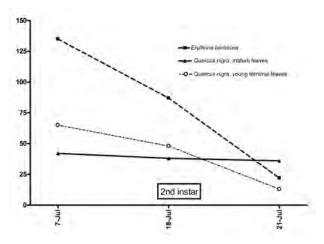


FIG. 3b. Experiment 3: Survival of young *Automeris io* larvae when raised on old leaves of *Erythrina herbacea* (water contents 59%), young *Quercus nigra* leaves (water contents 58%) and old *Q. nigra* leaves (water contents 40%).

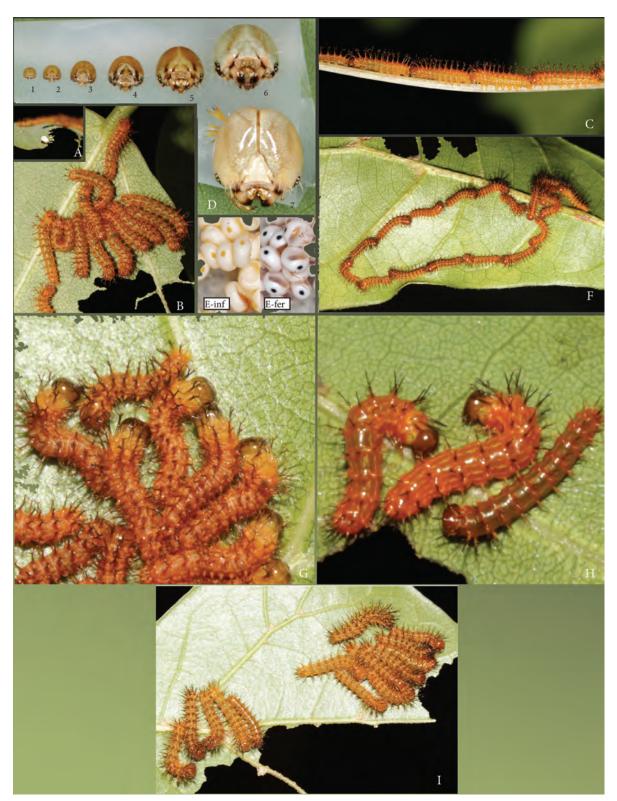


FIG. 4. Early stages of *Automeris io*: (A-C) A batch of 1st instar larvae found feeding on the Coral Bean, *Erythrina herbacea*, Gainesville, FL, 23 Sept 2011; (D) Head capsules of the seven instars that these larvae underwent prior to pupation; (E) Fertile and infertile eggs; (F) 1st instar of F-2 generation feeding on the Water Oak, *Quercus nigra*; (G) same as (F) molting into 2nd instar; (H) larvae fed on *E. herbacea* showed a characteristic bloating when molting into 2nd instar (see text for details); (I) late 1st and early 2nd instars feeding together on *Q. nigra*.

content were introduced. The dynamics of larval survival then became very similar to those seen on E. herbacea leaves. This, combined with the fact that extracts of E. herbacea leaves applied to Q. nigra leaves did not cause larval mortality and larvae fed on high-H<sub>2</sub>O containing leaves displayed a bloated appearance, leads me to conclude that the high water content of the leaves may have been detrimental to the larvae. This conclusion can appear contradictory to accounts of rearing A. io on young leaves of other plants, specifically the black cherry, Prunus serotina Ehrh. (e. g., Eric Anderson, pers. com.; Manley 1990 & 1993), and perhaps individual properties of leaves and plants are important for larval survival. As a side note, the larvae of Agathodes designalis—the only large larvae that specialize on E. herbacea leaves-have a thin, transparent cuticle, perhaps designed to deal with the hostplants' high water content by allowing evaporation. The observed acceleration in development of A. io larvae on *E. herbacea* is most likely due to the nitrogen content, which was shown to be twice as high in this hostplant as in *Q. nigra*.

The present study showed that, although Automeris io has a rather remarkable ability to survive feeding on *E. herbacea*, the cost (in the form of larval mortality) of ovipositing on this hostplant can be high, which probably leads to selective pressure against the adaptation of this hostplant as a norm by *A. io.* The mechanism by which *A. io* deals with the alkaloids of this plant (via sequestration, or elimination straight from the gut, etc.) is yet to be determined. It is clear from this study, however, that the polyphagous nature of *A. io* has its limitations, even though this species has an incredible ability to adapt to feeding on a wide variety of, sometimes toxic, host plants.

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## References

- BELL, H.A., A. E. KIRKBRIDE-SMITH, G. C. MARRIS, J. P. EDWARDS, & A. M. R. GATEHOUSE. 2004. Oral toxicity and impact on fecundity of three insecticidal proteins on the gregarious ectoparasitoid *Eulophus pennicornis* (Hymenoptera: Eulophidae). Agric. For. Entomol. 6(3): 215–222.
- BOSSART, J. L. & J. M. SCRIBER. 1995. Genetic variation in oviposition preferences in the tiger swallowtail butterfly: interspecific, interpopulation, and interindividual comparisons. *In* Scriber J. M., Y. Tsubaki & R. C. Lederhouse (eds.) Swallowtail Butterflies: Their Ecology and Evolutionary Biology, Scientific Publishers, Gainesville, pp.183–194.

- DOWELL, R. V., J. M. SCRIBER, & R. C. LEDERHOUSE. 1990. Survival of Papilio rutulus Lucas (Lepidoptera: Papilionidae) larvae on 42 potential hostplants. Pan-Pac. Entomol. 66: 140–146.
- FINKE, M. D. & J. M. SCRIBER. 1988. Influence on larval growth of the eastern black swallowtail butterfly *Papilio polyxenes* (Lepidoptera: Papilionidae) of seasonal changes in nutritional parameters of Umbelliferae species. The Am. Midl. Nat. 119(1): 45-62.
- FITCHES, E. C., A. M. R. ĜATEHOUSE, & J. A. GATEHOUSE. 1997. Effects of snowdrop lectin (GNA) delivered via artificial diet and transgenic plants on the development of tomato moth (*Lacanobia oleracea*) larvae in laboratory and glasshouse trials. J. Insect. Physiol. 43: 727–739.
- Fitches, E.C., S. D. Woodhouse, J. P. Edwards, & J. A. Gatehouse. 2001. In vitro and in vivo binding of snowdrop (*Galanthus nivalis agglutinin*; GNA) and jackbean (*Canavalia ensiformis*; ConA) lectins within tomato moth (*Lacanobia oleracea*) larvae; mechanisms of insecticidal action. J. Insect. Physiol. 47: 777–787.
- FRANKFATER, C. & J. M. SCRIBER. 1999. Florida red bay (*Persea borbonia*) leaf extracts deter oviposition of a sympatric generalist herbivore, *Papilio glaucus* (Lepidoptera: Papilionidae). Chemoecology 9: 127-132.
- . 2003. Contact chemoreception guides oviposition of two Lauraceae-specialized swallowtail butterflies (Lepidoptera: Papilionidae). Holarctic Lepid. 7(2): 33-38.
- HEPPNER, J. B. 2003. Lepidoptera of Florida, Part 1: Introduction and catalog. Arthropods of Florida and neighboring land areas. Florida Department of Agriculture, Gainesville.
- HOGERVORST, P. A. M., F. L. WÄCKERS, J. WOODRING & J. ROMEIS. 2008. Snowdrop lectin (*Galanthus nivalis agglutinin*) in aphid honeydew negatively affects survival of a honeydew- consuming parasitoid. Agric. For. Entomol. 11(2): 161-173.
- JANZEN, D. H. & W. HALLWACHS. 2012. Area de Conservación Guanacaste (ACG), northwestern Costa Rica: Caterpillars, pupae, butterflies & moths database. http://janzen.sas.upenn.edu/ (last accessed 6-1-2012)
- LINDROTH, R. L., 1989. Chemical ecology of the luna moth. J. Chem. Ecol. 15(7): 2019-2029.
- LINDROTH, R. L., M. T. S. HSIA & J. M. SCRIBER.1988. Seasonal patterns in the phytochemistry of three *Populus* species. Biochem. Syst. Ecol. 15: 681-686.
- . 1987. Seasonal patterns in the phytochemistry of three *Populus* species. Biochem. Syst. Ecol. 15(6): 681-686.
- LINDROTH, R. L., J. M. SCRIBER & M. T. S. HSIA. 1986. Differential responses of tiger swallowtail subspecies to secondary metabolites from tulip tree and quaking aspen. Oecologia 70: 13-19.
- \_\_\_\_\_. 1988. Chemical ecology of the tiger swallowtail: mediation of host use by phenolic glycosides. Ecology 69: 814-822.
- MACHUKA, J., É. J. M. VAN DAMME, W. J. PEUMANS & L. E. N. JACKAI. 1999. Effect of plant lectins on larval development of the legume pod borer, *Maruca vitrata*. Entomol. Expt. App. 93: 179–187.
- MACHUKA, J., O. G. OKEOLA, M. J. CHRISPEELS & L. E. N. JACKAI. 2000. The African yam bean seed lectin affects the development of the cowpea weevil but does not affect the development of larvae of the legume pod borer. Phytochemistry 53(6): 667-674.
- MANLEY, T. R. 1993. Diapause, voltinism, and foodplants of Automeris io (Saturniidae) in the southeastern United States. J. Lepid. Soc. 47(4): 303-321.
- . 1990 (1991). Heritable color variants in Automeris io (Saturniidae). J. Res. Lepid. 29(1-2): 37-53.
- MANUWOTO, S. & J. M. SCRIBER. 1985. Differential effects of nitrogen fertilization of three corn genotypes on biomass and nitrogen utilization by the southern armyworm, *Spodoptera eridania*. Agric. Ecosyst. Environ. 14: 25-40.
- MERCADER, R. J., M. L AARDEMA & J. M. SCRIBER. 2009. Hybridization leads to host-use divergence in a polyphagous butterfly sibling species pair. Oecologia 158: 651-662.
- MICHIELS, K., E. J. VAN DAMME & G. SMAGGHE. 2010. Plant-insect interactions: what can we learn from plant lectins? Arch. Insect Biochem. Physiol. 73(4): 193–212.
- POWELL, M. H. & S. B. WESTLEY (edits). 1993. Erythrina Production

and Use: A Field Manual. Published by the Nitrogen Fixing Tree Association. Paia, Hawaii, 55pp.

- RUUHOLA, T., O-P. TIKKANEN & J. TAHVANAINEN. 2001. Differences in host use efficiency of larvae of a generalist moth, *Operophtera brumata* on three chemically divergent *Salix* species. J. Chem. Ecol. 27: 1595-1615.
- SCRIBER, J. M. & P. FEENY 1979. Growth of herbivorous caterpillars in relation to feeding specialization ant to the growth form of their food plants. Ecology 60(4): 829-850.
- SÉTAMOU, M., J. S. BERNAL, J. C. LEGASPI & T. E. MIRKOV. 2002. Effects of Snowdrop lectin (*Galanthus nivalis agglutinin*) expressed in transgenic sugarcane on fitness of *Cotesia flavipes* (Hymenoptera: Braconidae), a parasitoid of the nontarget pest *Diatraea saccharalis* (Lepidoptera: Crambidae). Ann. Entomol. Soc. Am. 95(1): 75-83.
- SHUKLE, R. H., L. L. MURDOCK. 1983. Lipoxygenase trypsin inhibitor, and lectin from soybeans: effects on larval growth of *Manduca sexta* (Lepidoptera: Sphingidae). Environ. Entomol. 12(3): 787-791.
- SPECHT, A., A. C. FORMENTINI & E. CORSEUIL. 2006. Biologia de Automeris illustris (Walker) (Lepidoptera, Saturniidae, Hemileucinae). Rev. Bras. Zool. 23(2).

- SOURAKOV, A. 2009. Notes on the biology of the Gulf Fritillary Agraulis vanillae (Lepidoptera: Nymphalidae) in north-central Florida. J. Lepid. Soc. 63(2): 127.
- . 2011. Niche partitioning, co-evolution and life histories of *Erythrina* moths, *Terastia meticulosalis* and *Agathodes designalis* (Lepidoptera: Crambidae). Trop. Lepid. Res. 21(2): 84-94.
- SOURAKOV, A. 2012. On the biology of moths that feed on *Erythrina* in Florida. Trop. Lepid. Res. 22(2): 110-118.
- \_\_\_\_\_. 2012. On the biology of moths that feed on *Erythrina* in Florida. Trop. Lepid. Res. 22(2): 110-118.
- TAHVANAINEN, J., R. JULKUNEN-THITO & J. KETTUNEN. 1985. Phenolicglycosides govern the food selection pattern of willow feeding beetles. Oecologia 67: 52–56.
- WIKIPEDIA. 2013. Automeris io. (http://en.wikipedia.org/wiki/Automeris\_io) (last accessed 2-19-2013)
- ZVEREVA, E. L., M. V. KOZLOV & M. HILKER. 2010. Evolutionary variations on a theme: host plant specialization in five geographical populations of the leaf beetle *Chrysomela lapponica*. Popul. Ecol. 52: 389-396.
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### ADDENDUM

Following the acceptance of this manuscript, I conducted two additional experiments involving Erythrina herbacea and A. io. The eggs from cage-mated A. io were obtained in June 2013. When a cluster of ca. 70 neonate larvae were simultaneously offered blackberry and E. herbacea leaves, they quickly dispersed and started feeding on both hosts. A day later, the ca. 35 larvae that had been feeding on E. herbacea were observed to fall into a temporary "comatose" state: they interrupted their feeding, were not moving or responding to external stimuli, and were in unnatural positions as if in a spasm. They were presumed to have been poisoned, but the following day, they resumed normal feeding and continued growing. However, while the control batch of larvae that were feeding on blackberry underwent normal development, the larvae feeding on E. herbacea mostly died at molting, with only four larvae surviving past the first instar, and two surviving past the second instar. These two larvae developed faster (in 72 and 79 days) than their ten siblings raised on blackberry, the average development time of which was  $89 \pm 8$  days.

From a different egg batch, 20 late-penultimate and earlylast-instars raised on blackberry were switched to *E. herbacea*. These larvae also readily accepted the new hostplant, but underwent high mortality: only eight larvae (40 %) reached the pupal stage. Death was preceded by an interruption in feeding followed by a discharge of clear fluids, but otherwise the larvae appeared normal. The symptoms began after the larvae had been feeding on *E. herbacea* for several days and up to 3 weeks, so it is unclear if the mortality was due to the cumulative effect of gradually ingested toxins or to the ingestion of a lethal dose contained within particular leaves. These trials confirm the experiments described in the main body of the manuscript: while there may be a limited benefit to feeding on *E. herbacea* in the form of faster development, very few *A. io* larvae are able to take advantage of it.

During the summer of 2013, while searching various plants for caterpillars in the Gainesville, FL, area, I found four additional batches of A. io eggs (16-20 eggs each). One, found on Prunus serotina, was parasitized by Trichogramma. Three others were found on Crotalaria pumila and Crotalaria pallida, which are, like E. herbacea, legumes high in nitrogen and rich in alkaloids that very few larvae can detoxify. The neonates fed on leaves of C. pumila and C. pallida, if offered an alternative (cherry or oak) immediately switched, and if not offered an alternative, died before reaching the second instar. These findings, in addition to my earlier finding of A. io on E. herbacea begin to form a pattern that requires an explanation other than "accidental" or "mistaken" oviposition. The first possible reason is that A. io females, while searching for hostplants, lay small batches of eggs on nitrogen-rich plants regardless of their toxicity and, shall it be unsuitable, rely on larvae to disperse to a different hostplant. So far all five batches found in nature have been 14-20 eggs, so the risk to an individual female resulting from losing a batch or two is relatively small. The second, more farfetched yet interesting hypothesis is that A. io females favor toxic plants for oviposition to avoid egg parasitoids and to offer potential enemy and competition-free space to their progeny and added protection to caterpillars via secondary plant chemicals. More fieldwork will be required to prove either of these hypotheses.