WHY FLY NOW?
PUPA BANKS, APOSEMATISM, AND OTHER FACTORS THAT MAY EXPLAIN OBSERVED MOTH FLIGHT ACTIVITY
BY
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Abstract – This paper addresses factors that affect insect flight activity. It presents a 5-year time series of nightly activity at a site in Clarke County, Georgia for *Epimecis hortaria*, Tulip-tree Beauty (Geometridae: Ennominae); *Nigetia formosalis*, Thin-winged Owlet (Erebidae: Scolecocampinae), and *Dryocampa rubicunda*, Rosy Maple Moth (Saturniidae: Ceratocampinae). These species exemplify three seasonal flight patterns, here defined as *diffuse*, *synchronized*, and *complex*. I propose that *diffuse* flight patterns are typical of many cryptic species and that s*ynchronized* ones are typical of aposematic species and species restricted by the phenology of their hosts. The *complex* pattern of *D. rubicunda* shows variation in when individuals broke pupal diapause and eclosed. Because some insects have *pupa banks*, similar to seed banks in plants, their observed flights and generations may be decoupled. I caution against using terms such as brood, generation, or voltinism to describe observed seasonal adult activity. Instead, I propose that we use the term *flight* to describe their activity.

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Introduction – Many biotic and abiotic factors interact to affect the seasonal flight activity of moths and our ability to sample them accurately (Tauber et al. 1986; Valtonen et al. 2011). They include processes fundamental to each species' natural history and life cycle. By what means do they disperse, avoid natural enemies, find mates, lay eggs on or near hosts, avoid natural enemies, and in the grand scheme of things, survive for millions of years beyond the next mega-drought, warming period, and ice age? Science has barely begun to explore the complexity of these questions. Understanding when moths fly is a frontier that could answer much about how insects will respond to a changing world.

This is the second of a series of articles that presents results from Discover Life's *Mothing* project (www.discoverlife.org/moth).   In the last SLN issue I gave an overview of *Mothing* and invited readers to participate (Pickering, 2015). Here I present nightly data on seasonal moth activity at our Blue Heron site in Georgia and focus on two factors that may help to explain these observations – '*pupa banks'* and '*aposematic vs cryptic coloration'*. In future articles I plan to consider how latitude, thermoregulation as a function of body size, moon phase, weather, sampling time, and other factors affect seasonal and nightly flight observations.

Pupa banks – The multitude of species-specific genetic and environmental factors that control metamorphosis, potential diapause, and pupal eclosion are key to understanding adult flight patterns. However, there is too little appreciation in entomology for the factors that induce long-term insect diapause and those that can vary the time at which individuals eclose as adults.

Here I develop the concept of a *pupa bank*, similar in function to a seed bank which are widespread and important in plants (Kalisz and McPeek 1993, Salguero - Gomez et al. 2015, Stott et al. 2010). Prolonged diapause — defined as diapause of over a year — is widespread in insects, particularly in taxa with pupae (Danks 1987). Powell (1986) lists 91 species in 16 families of Lepidoptera in which it is known. He suggests that there are probably many more species that use prolonged diapause to survive drought and unpredictable food resources. Because pupae are difficult to study under natural conditions, most studies of insect populations ignore them. There is no systematic study of the role pupae play in the dynamics of wild populations. Most studies focus only on changes in adult or larval numbers. Many insect species overwinter as diapausing pupae, and hence, could be considered by definition to have a pupa bank. Worth (1979) reports that 24.5% of 94 reared pupae of *Citheronia regalis*, Regal Moth (Saturniidae: Ceratocampinae), remained dormant beyond a year. Thus, the role of pupa banks could be large.

Observed differences in adult numbers across flights could reflect differences in the size of pupa banks and proportion of individuals eclosing from them rather than in changes in population size per se. Because diapausing individuals can skip sequential flights, generations and flights can be decoupled. When we do not know the genetic structure of a population and the magnitude of gene flow across flights, both within and across years, I propose that we use the term *flight* to describe their seasonal activity.

Aposematic vs cryptic coloration – In the results and discussion below, I hypothesize and present some evidence that

diffuse flight patterns are typical of many cryptic species and that s*ynchronized* ones are typical of aposematic species or of species restricted by the phenology of their host stages.

Methods – Since February 2010, a team of 16 individuals has taken 182,000 photographs to document the activity of creatures attracted to lights at my house at 275 Blue Heron Drive, Athens, Georgia (latitude 33.8882°N, longitude 83.2973°W).

Site description – This Clarke County site is within Georgia's Piedmont region. Its landscape contains a mix of pasture, agricultural fields, and forest patches. The house is on a 9 hectare property in a low-density neighborhood. There are no street lights and little light pollution from other sources. In front of the house there is a successional stand of trees dominated by pines (*Pinus*) and sweetgum (*Liquidambar*). Behind the house there is a mature stand of hardwoods dominated by oaks (*Quercus*) and hickories (*Carya*), a grassy area that includes *Andropogon* and *Eupatorium*, and a 1-hectare dammed pond above a forested floodplain including *Acer*, *Betula*, *Carpinus*, *Fraxinus*, *Ulmus*, and invasive *Ligustrum*. On the property there are a total of approximately 50 woody species, numerous wildflowers, including *Hexastylis*, *Podophyllum*, *Tipularia*, *Trillium* and *Sanguinaria*, and a profusion of lichens, including *Cladonia*, *Parmotrema*, and *Usnea*. Since at least 1995 there has been no spraying of pesticides on the property, although some neighbors spray for mosquitoes.

Lights – Each night we run three porch lights (23 watt compact-fluorescent bright white bulbs, 1,650 lumens, Utilitech #0073511), one on the front porch and two on the back. In May 2010 we started running a black light (15 watt 22-inch T8 fluorescent, Utilitech #283498) on the front porch on alternate nights. We switch lights on before dusk. The house walls are white stucco. We photograph moths in a set area around the lights up to a height of approximately 3m.

Photography – We use Nikon D50 cameras with AF Micro Nikkor 105mm 1:2.8 D lens to record all moths and other creatures larger than approximately 3mm in length. We use the camera's built-in flash and typically shoot at a shutter speed of 1/500 second and an aperture of F32. See discoverlife.org/ed/tg/Using_Digital_Camera/nikon_d50.html for other camera settings. In most photographs we include a mm ruler to enable us to measure specimens and help in their identification. In the first half of 2010 and 2011, we typically photographed specimens between 10PM and 2AM; in July of these years we switched to photographing them between 4AM and dawn. Currently, since 2011, we photograph after 4:00AM. Each night, before starting, we photograph the time and date on a cell phone and then the lights and wall. These photographs serve as quality controls to help us manage the images and know whether the moths were on the front or back porch. Since 28 November, 2010, we have sampled every night through February, 2016, with the exception of one night, 22 June, 2012.

Images – Photographers bulk upload images to their personal albums on Discover Life. After automated processing in which the system adds a unique identification number to each image, we tag the associated data records with where and when information. All images and associated data are available through *Mothing's* results page: discoverlife.org/moth/report.html. One can view and link to individual images as explained on Discover Life's help page: discoverlife.org/nh/id/20q/20q_help.html#services_images. For example, discoverlife.org/mp/20p?see= I_JP70255 shows a *Plagodis fervidaria* with ruler and associated data.

Identification – Using Discover Life's local identification guides and annotated checklists (see Pickering 2015), we have developed a rapid workflow to determine specimens within photographs and add their name(s) to each image's *title* field. This field supports multiple names, a count of each species, and additional information such as sex, the number of mites on a specimen, or whether two specimens were mating. Once identified, up to 6,000 images and associated titles can be reviewed simultaneously and '*blessed*' or '*rejected*' by an expert. The system also allows users to link multiple photographs of the same specimen into a group so that they are correctly tabulated as only one specimen. This feature is particularly useful in genera such as *Spilosoma* for which more than just a dorsal view of the forewing is needed for identification.

Depending their taxa, we determine specimens to '*valid species*', '*species group*' (typically groups of 2-4 species that we cannot easily tell apart from photographs), or '*morphospecies*' (typically microlepidopterans, many of which may be undescribed). For species groups we use the convention of separating members with a '--' (double dash), for example, *Hypagyrtis esther--unipunctata*. We use *Gn_* and *sp_* to name morphospecies, for example, *Papaipema sp_new_species_3*. For more details on our naming conventions see discoverlife.org/nh/id/20q/20q_help.html# Scientific_names.

Tabulation – Each night the system tabulates across albums the number of specimens for each taxon by site and date. It tabulates specimens to the date of the night's dusk, even if they were photographed after midnight and technically recorded on the following date. The system makes summary tables and refreshes information used to build maps and produce seasonal graphs for users. discoverlife.org/mp/20m?plot=3&la=33.9&lo=-83.3 presents an interactive graph of the species accumulation since 2010 and allows users to query by date(s) the abundance of species.   For a table of the number of each species recorded by month see discoverlife.org/moth/data/table2_33.9_-83.3. html.   For each species click '*details*' to see a nightly phenology graph across years, including data from *Mothing's* other 22 sites.

Results & Discussion

As of February, 2016, we have identified (or misidentified!) 143,156 of 144,603 (99.0%) of the Blue Heron site's lepidopteran images. The accumulated number of species is 1,254 (1,240 moths; 14 butterflies), including 65 *morphospecies* (1,602 specimens) and 48 *species groups* (6,054 specimens).   Of the 1,254 taxa, 196 are represented by over 150 specimens each.

Seasonal flight patterns – I now consider the moths' seasonal flight activity, using three species to illustrate different patterns. These species are *Epimecis hortaria*, Tulip-tree Beauty (Geometridae: Ennominae) with a *diffuse* pattern; *Nigetia formosalis*, Thin-winged Owlet (Erebidae: Scolecocampinae) with a *synchronized* pattern, and *Dryocampa rubicunda*, Rosy Maple Moth (Saturniidae: Ceratocampinae) with a *complex* pattern. Adults of these species appear in Fig. 1, which contrasts the cryptic coloration of *E. hortaria* in the top two images with the contrasting black and white pattern of *N. formosalis* and pink and yellow of *D. rubicunda.*   Brou (2003, 2010) shows multiple morphs of *E. hortaria* and presents a graph of its seasonal flight pattern accumulated across years in Louisiana.   Brou (2008) shows a similar graph of flight activity for *D. rubicunda.* The subsequent three figures present five years of nightly abundance for these species at Blue Heron Drive.

Fig. 1. Cryptic and aposematic moths. **The top two images are of *Epimecis hortaria*. The one on the left shows it camouflaged on a tree. In contrast, the lower images show *Nigetia formosalis* [left] and *Dryocampa rubicunda,* which clearly are not cryptic.**

Diffuse – Fig. 2 presents the season activity of 713 specimens of *E. hortaria*. These specimens did not exhibit tight seasonal peaks but were spread out over their flight seasons, here observed between late February and early November. This *diffuse* pattern is exemplified in other cryptic species, such as some gray geometrids (e. g., *Iridopsis larvaria*, Bentline Gray; *Anavitrinella pampinaria*, Common Gray; *Protoboarmia porcelaria*, Porcelain Gray; *Hypomecis umbrosaria*, Umber Moth); *Hypena scabra*, Green Cloverworm Moth (Eribidae: Hypeninae), and *Pselnophorus belfragei*, Belfrages Plume Moth (Pterophoridae), as graphed in the last issue (Pickering, 2015).
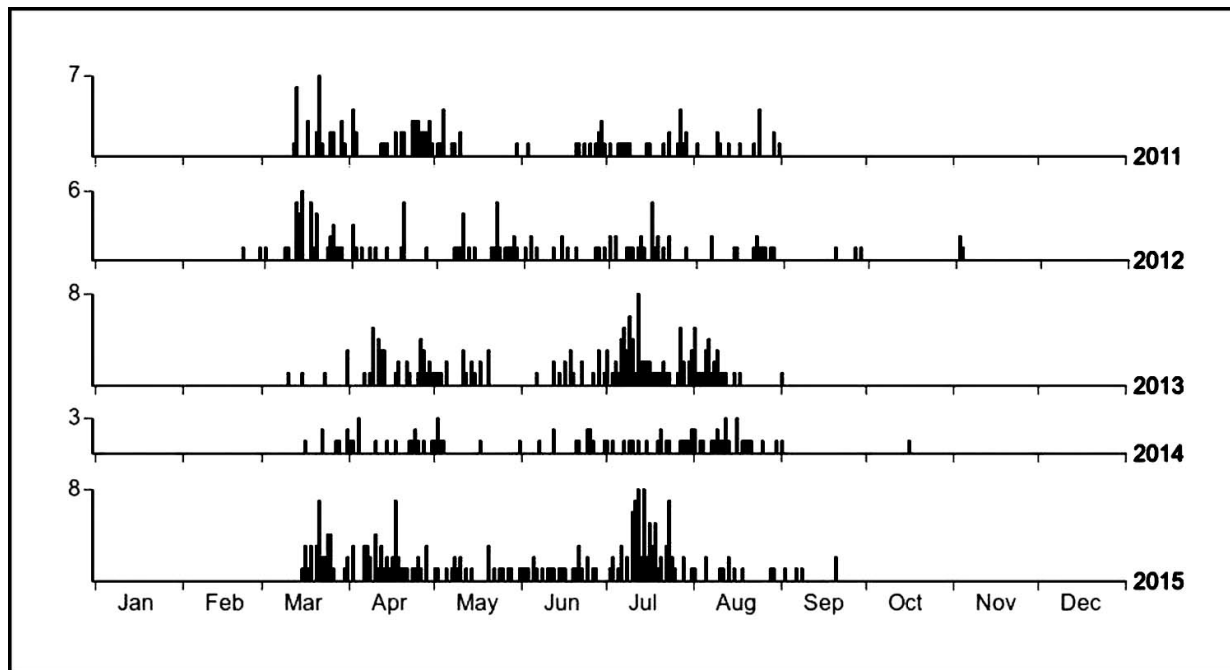
Fig. 2.   Diffuse flight pattern. Activity of *Epimecis hortaria*, Clarke County, Georgia, 2011-2015.   For details see discoverlife.org/mp/20q?search=Epimecis+hortaria

A possible exception to cryptic species having a *diffuse* flight pattern is *Glenoides texanaria*, Texas Gray (Geometridae: Ennominae), which had two fairly distinct flights in the summer and fall of 2013, 2014, and 2015.

Synchronized – Fig. 3 presents the seasonal activity of *Nigetia formosalis*. The 769 recorded specimens had two distinct flight peaks in each year without any specimens between the peaks. Other examples of this *synchronized* pattern with two flights are *Hypoprepia fucosa*, Painted Lichen Moth (Erebidae: Arctiinae), which is red, yellow, and black, and *Cisthene plumbea*, Lead-colored Lichen Moth (Erebidae: Arctiinae), which is yellow and black. Species such as the orange and black *Cisthene packardii*, Packard's Lichen Moth, displayed three such *synchronized* peaks.

Such *synchronized* flight activity may be typical of aposematic species that cluster together in time and space, warning birds and other visual predators that they may be distasteful. Natural selection may work against temporal outliers and favor ones that cluster together. However, an exception of an aposematic species that does not have *synchronized* flights is *Atteva aurea*, Ailanthus Webworm Moth (Yponomeutoidea: Attevidae), which is orange, black and white. It flies both at day and night. We recorded its *diffuse* flight activity between late March and early December.

Another category of species with *synchronized* flights appears to be species that have larvae that depend on a host stage with a restricted phenology. Two examples of such species that are not aposematic and have one *synchronized* flight per year are *Cissusa spadix*, Black-dotted Brown (Erebidae: Erebinae), the young larvae of which feed on young oak leaves (Coyle et al. 2013), and *Malacosoma americana*, Eastern Tent Caterpillar Moth (Lasiocampidae: Lasiocampinae), the young larvae of which feed on young leaves of *Prunus serotina*, Black Cherry (Abarca & Lill 2015).

Complex – Fig. 4 shows the seasonal activity of 225 specimens of *D. rubicunda*. I term this a *complex* pattern because there appear to be several things driving it. In 2014 and 2015, *D. rubicunda* had short flights in April and May that were too close together to be separate generations. These flights were followed by a longer flight starting in late June. The short April and May flights may show that *D. rubicunda* has a split emergence pattern following winter pupal diapause.   Willis et al. (1974) documented that autumn collected cocoons of *Hyalophora cecropia*,
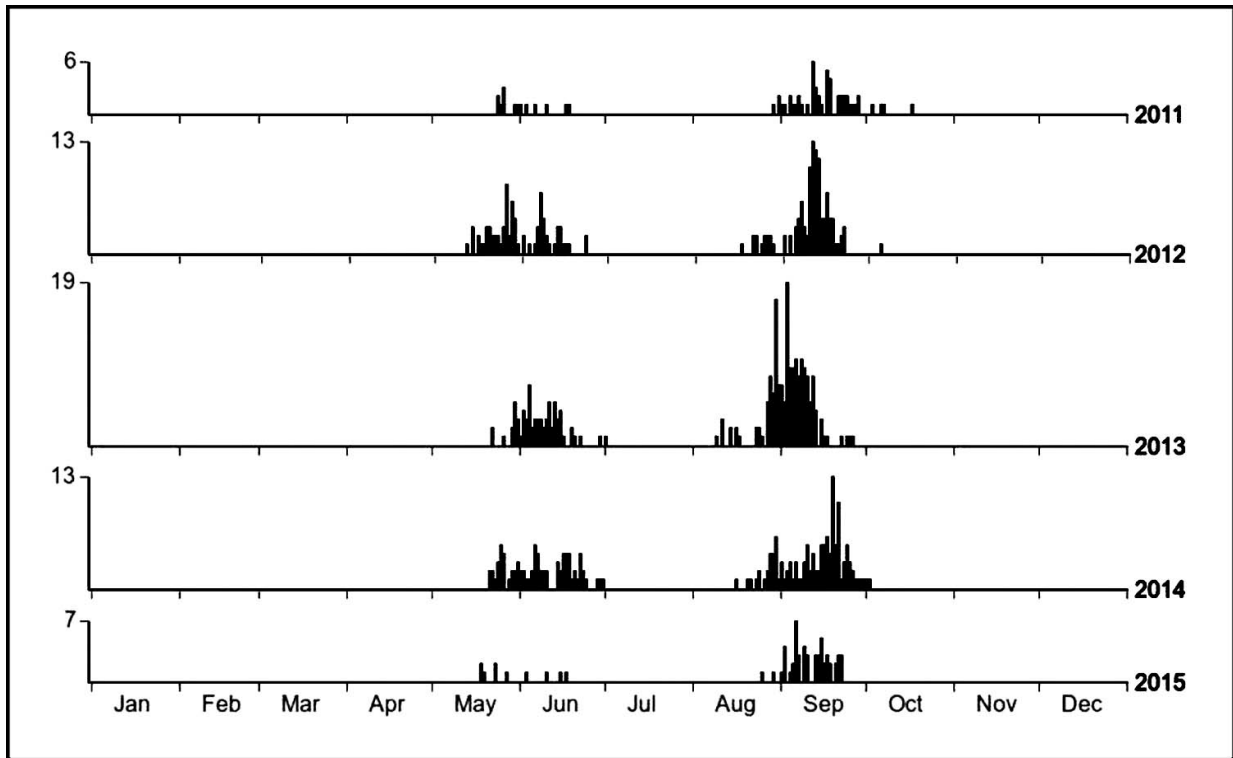
Fig. 3. Synchronized flight pattern. Activity of *Nigetia formosalis,* Clarke County, Georgia, 2011-2015. For details see discoverlife.org/mp/20q?search=Nigetia+formosalis
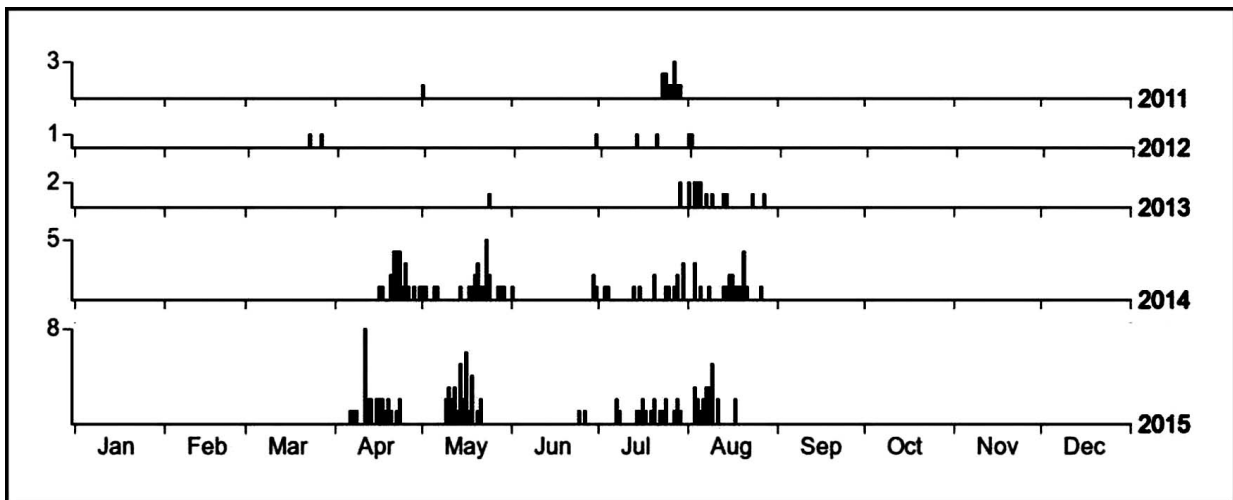

Fig. 4. Complex flight pattern. Activity of *Dryocampa rubicunda,* Clarke County, Georgia, 2011-2015. For details see discoverlife.org/mp/20q?search=Dryocampa+rubicunda

Cecropia Moth (Saturniidae: Saturniinae), in Illinois broke diapause in two distinct morphs, 8% eclosing mostly in late May and the remainder in late June.

The two *D. rubicunda* specimens recorded in March, 2012, probably reflect an early emergence that year after a warmer winter than in the other four years.

Conclusions

A multitude of factors affect the flight activity of insects. Here I have proposed the concept of a *pupa bank,* the importance of which is unknown for most species. If pupa banks are found to play a major role, they will profoundly change our understanding of the population dynamics and long-term survival of insects. Until we know more about the genetical structure of populations, as affected by long-term pupal diapause, I recommend that we use the term *flight* to describe adult seasonal activity and refrain from using brood, generation, and voltinism.

The contrast between the *diffuse* flight pattern presented for *E. hortaria* and the *synchronized* flight pattern of *Nigetia formosalis* may suggest that aposematic species are favored to cluster together temporally and that cryptic species are favored to avoid each other. More data and analyses are clearly needed to test this hypothesis.

References

Abarca, M. & J. Lill, 2015. Warming affects hatching time and early seasonal survival of eastern tent caterpillars. *Oecologia* **169**:901-912.

Brou, V.A., 2003. The many faces of the tulip tree moth *Epimecis hortaria* Fabricius. *S. Lep. News* **25** (1):6.

Brou, V.A., 2008. *Dryocampa rubicunda* (F.) in Louisiana. *S. Lep. News* **30** (2):51.

Brou, V.A., 2010. Adult phenotypes of *Epimecis hortaria* (Fabricius, 1794) (Lepidoptera: Geometridae) from St. Tammany Parish, Louisiana . *S. Lep. News* **32** (2):56-57.

Coyle, D.R., J. Pickering, K. A. Dyer, F. R. Lehman, J. E. Mohan and K.J. K. Gandhi, 2013. Dynamics of an unprecedented outbreak of two native moth species, *Cissusa spadix* and *Phoberia atomaris* (Lepidoptera: Noctuidae), on oak trees (*Quercus* spp.) in southeastern U.S.A. *American Entomologist* **59**:78-90.

Danks, H.V., 1987. *Insect dormancy: an ecological perspective*. Entomological Society of Canada.

Kalisz, S. and M.A. McPeek, 1993. Extinction dynamics, population growth and seed banks: An example using an age-structured annual. *Oecologia* **95**:314-320.

Pickering, J., 2015. Find your dark side: Invitation to join Discover Life's *Mothing* project. *S. Lep. News* **37** (4):205-208.

Powell, J.A., 1986. Records of a prolonged diapause in Lepidoptera. *Journal of Research on the Lepidoptera* **25**:83-109.

Salguero-Gomez et al., 2015. The compadre Plant Matrix Database: an open online repository for plant demography. *J. Ecol.* **103**:202-218.

Stott, I., M. Franco, D. Carslake, S. Townley, and D. Hodgson, 2010. Boom or bust? A comparative analysis of transient population dynamics in plants. *J. Ecol.* **98**:302-311.

Tauber, M.J., C.A. Tauber, and S. Masaki, 1986. *Seasonal Adaptations of Insects*. Oxford University Press, New York.

Valtonen, A. and M.P. Ayre, 2011. Environmental controls on the phenology of moths: predicting plasticity and constraint under climate change. *Oecologia* **165**:237-248.

Willis, J.H., G.P. Waldbauer, and J.G. Sternburg, 1974. The initiation of development by the early and late emerging morphs of *Hyalophora cecropia*. *Entomologia Experimentalis et Applicata* **17**:219-222.

Worth, C.B., 1979. Doubly overwintering *Citheronia regalis* Fabricius (Lepidoptera, Saturniidae). *J. Lepid. Soc.* **33**:166.

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