Identifying Critical Metabolic and Developmental Transitions in Apple Maggot Flies

John Fuller, Dr. Greg Ragland and Dr. Daniel Hahn, Department of Entomology and Nematology, University of Florida

Background

- Metabolic pathways and regulatory mechanisms are conserved among animals.
- Extreme models of metabolic regulation can help us understand metabolic disorders and diseases in humans.
- The apple maggot fly *Rhagoletis pomonella* enters a state of deep metabolic depression during winter. (>90% metabolic decrease, i.e. diapause).
- *R pomonella* recently radiated from hawthorns, its native host, to apples, which fruit 3 weeks earlier.

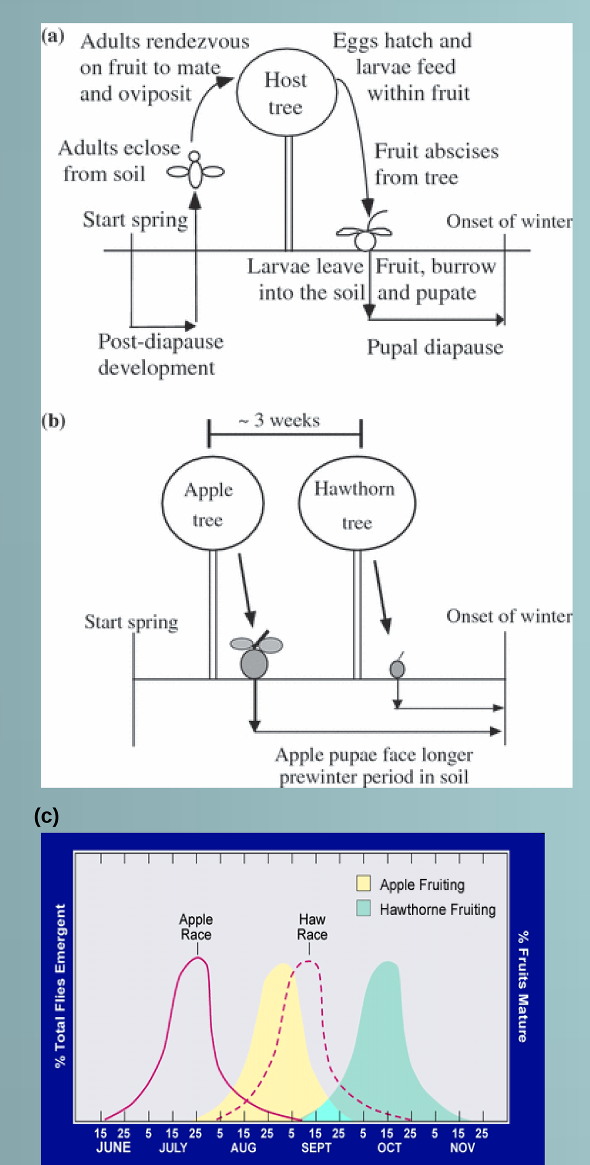

Figure 1.a) Life cycle of Apple Maggot Fly. b) timing difference between two populations. c) Seasonality of apple and hawthorn fruiting, and emergence of adult flies.

- Adaptation to apples has caused flies to hibernate earlier in the year, increasing metabolic demands. This has caused partial speciation into apple- and hawthorn-preferring populations, allowing us to study both speciation and metabolic regulation.

Goals of this study:

1) Characterize metabolic depression in diapause and the transition to post-diapause development.

2) Identify when diapausing flies transition from pupa to adult by identifying when the pupal cutucle becomes separated from adult epidermis.

Experiment 1

Using respirometry, the metabolic rates of pupae were observed as they broke diapause. At the same time, the head regions of uncapped pupae were checked for morphological changes.

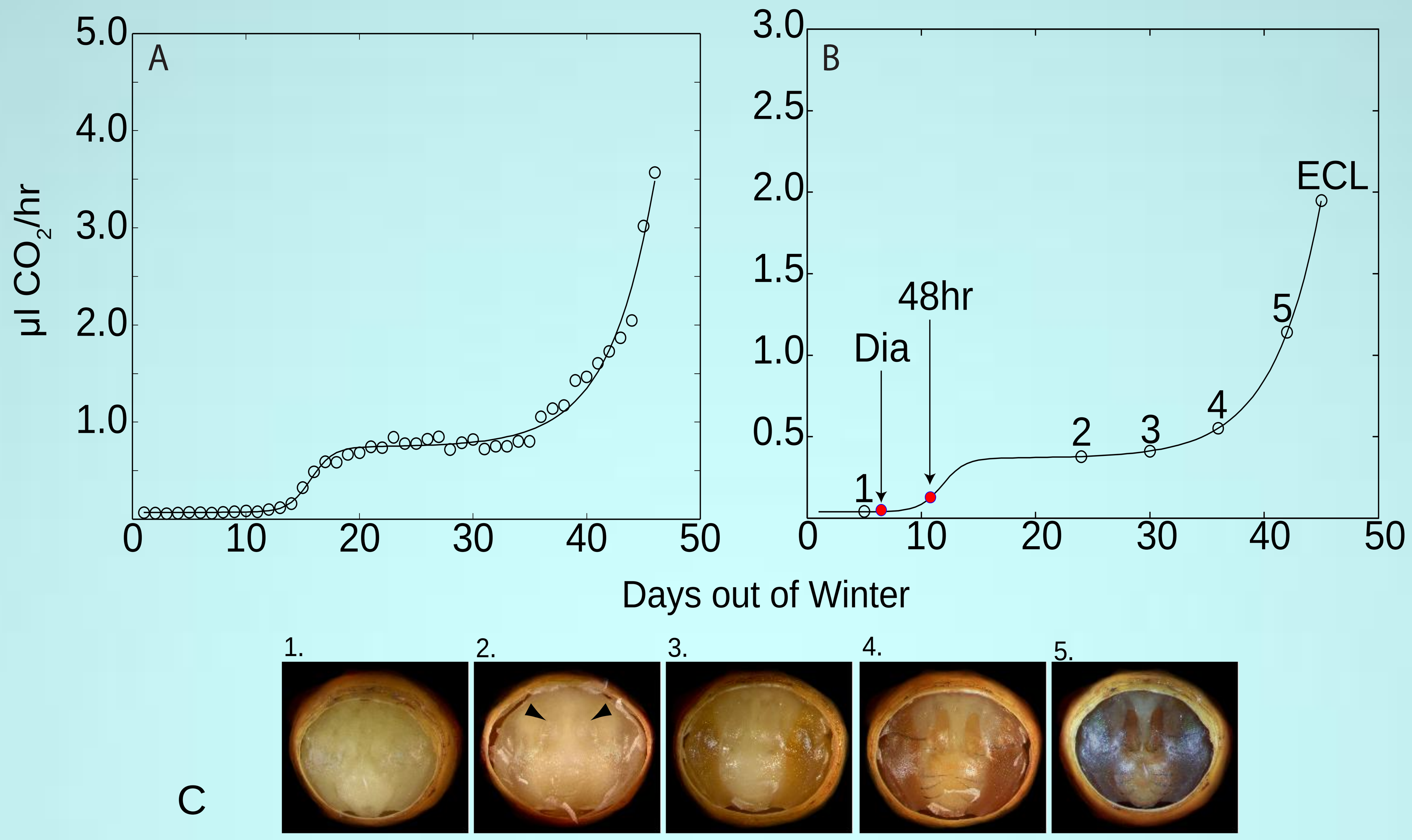

Figure 2. a)Fitting respirometric data of developing pupae to biphasic curve. b) Final, idealized curve. c) major developmental stages of pupae after diapause break.

Experiment 2

Using respirometry, pupae were identified that had broken diapause within the last 3 to 6 days. Another group was identified that was still well within diapause. The two groups were fixed, embedded in paraffin, sectioned, and compared, to determine whether pupal cuticle separation correlated with diapause break.

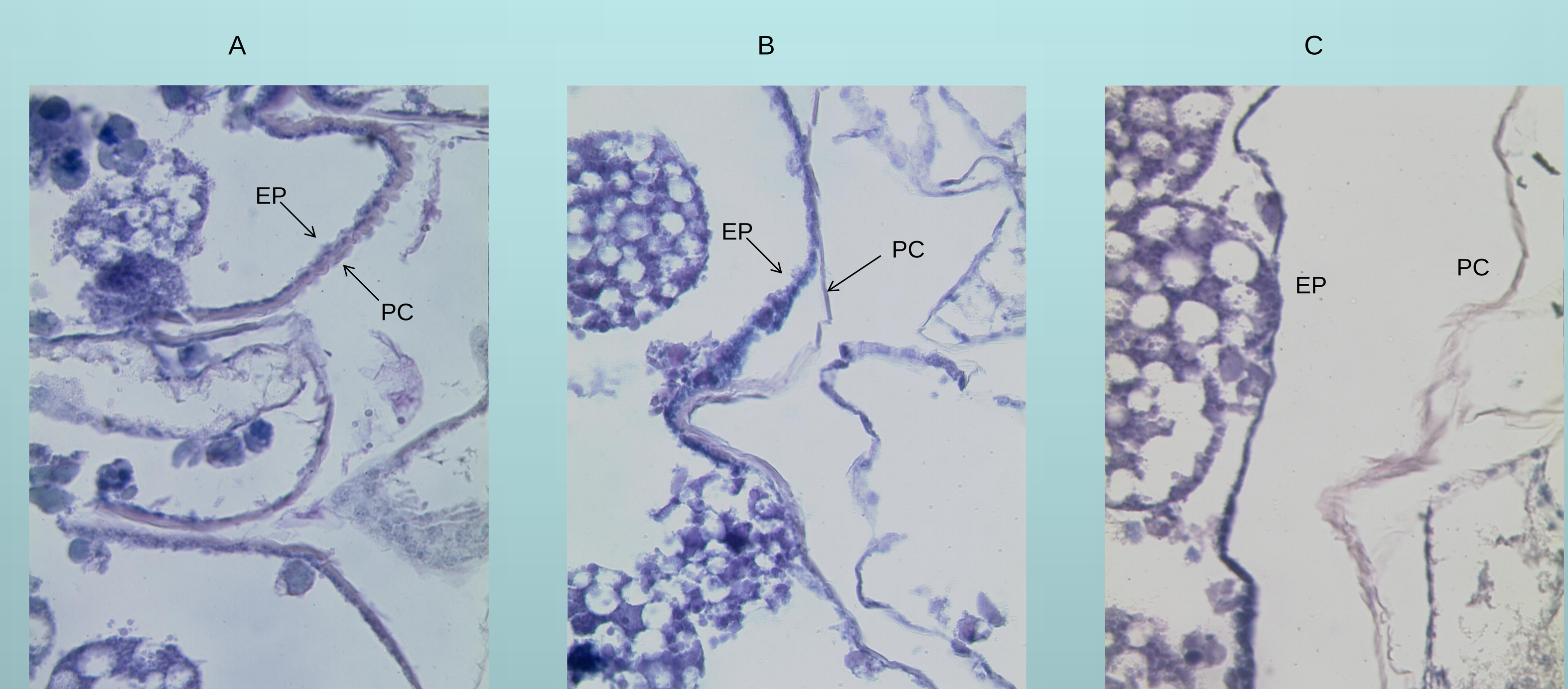

Figure 3. Paraffin sections of a) diapausing pupa, with no cuticle separation, b) breaking pupa, with cuticle starting to separate. c) breaking pupa with fully separated cuticle. EP= epidermis, PC= pupal cuticle.

Results

In experiment 1, we found that pupae undergo a biphasic increase in metabolic rate as they break diapause. Numerous developmental landmarks were connected to specific points along this curve, but diapause break occurred long before any morphological changes occurred.

In Experiment 2, pupal cuticle separation was identified in all breaking pupae, but was absent among diapausing pupae. The transition from pupa to adult must occur within ~72 hours of diapause break.

Together, these experiments show that the resumption of metabolic activity and the continuation of adult development both start at about the same time.

What's next?

The metabolic curve lets us pinpoint critical transitions in metabolism and morphology. Now that we know when these transitions occur, we have taken samples from these times and are identifying the actual genes and proteins that control these changes.

References

Ragland, GJ, J Fuller, JL Feder, and DA Hahn. "Biphasic metabolic rate trajectory of pupal diapause termination and post-diapause development in a tephritid fly." *Journal of Insect Physiology* 55.4 (2009): 344-50.