

# Starvation effects on ecdysteroid titers and the metamorphic commitment in *O. lignaria*



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## Abstract

The timing of metamorphosis is influenced by both intrinsic and extrinsic factors, however, the indicator triggering metamorphosis differs across different species<sup>3,4</sup>. The timing of when solitary bees, such as the blue orchard bee (*Osmia lignaria*), undergo this change is unknown<sup>1,2</sup>. To understand when pupation occurs in *Osmia*, hormonal titers of juvenile hormone III (JH III) and 20-hydroxyecdysone (20-E) and the gene expression of those involved in the 20-E pathway were analyzed.

Analysis showed that ecdysone genes were upregulated after 24 hours of starvation. Also, individuals did not experience the second peak of 20-E even after 96 hours after starvation. In conclusion, a surge of ecdysone is prolonged while genes are expressed; this delay will need to be considered in future studies of the development of other insects.

Knowing how nutrient availability, affect gene expression and therefore the developmental hormones is an important component of characterizing the development of solitary bees who contribute to the pollination of many natural and agricultural crops.

## Hypothesis

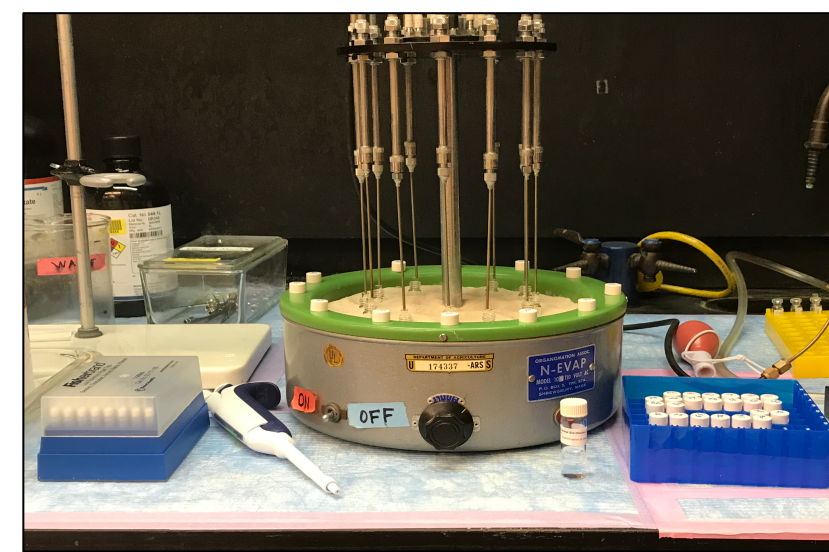
Starvation causes an early onset of metamorphosis because it initiates hormonal cascades in solitary bees.

- H1: The absence of food elicits the decline of JH which begin a cascade of hormonal events culminating in growth cessation and metamorphosis, as in *Manduca sexta*.
- H2: The absence of food could cause termination of growth through direct stimulation of ecdysone levels disregarding JH titers such as in *Drosophila*.

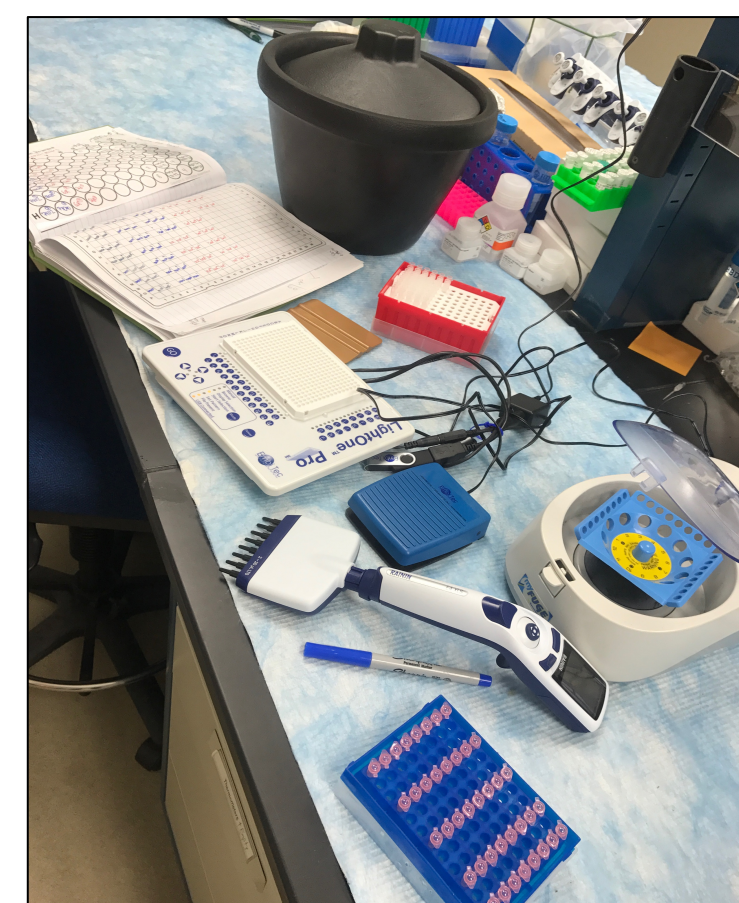


## Materials and Methods

High-performance liquid chromatography (HPLC) analyzed extracted hemolymph samples from fifth instar *O. lignaria* larvae from Washington and Utah populations from a fed and starved treatment.



Quantitative polymerase chain reaction (qPCR) measured the gene expression of those involved in the 20-E signaling pathway in the larvae remains: EcR, Shd, Sad, and BrC.



*O. lignaria* (n=6 per time point)

Fed	Daily after day 5									
Starved (hours after food removal)	1	2	4	8	12	24	36	48	72	90

## Results

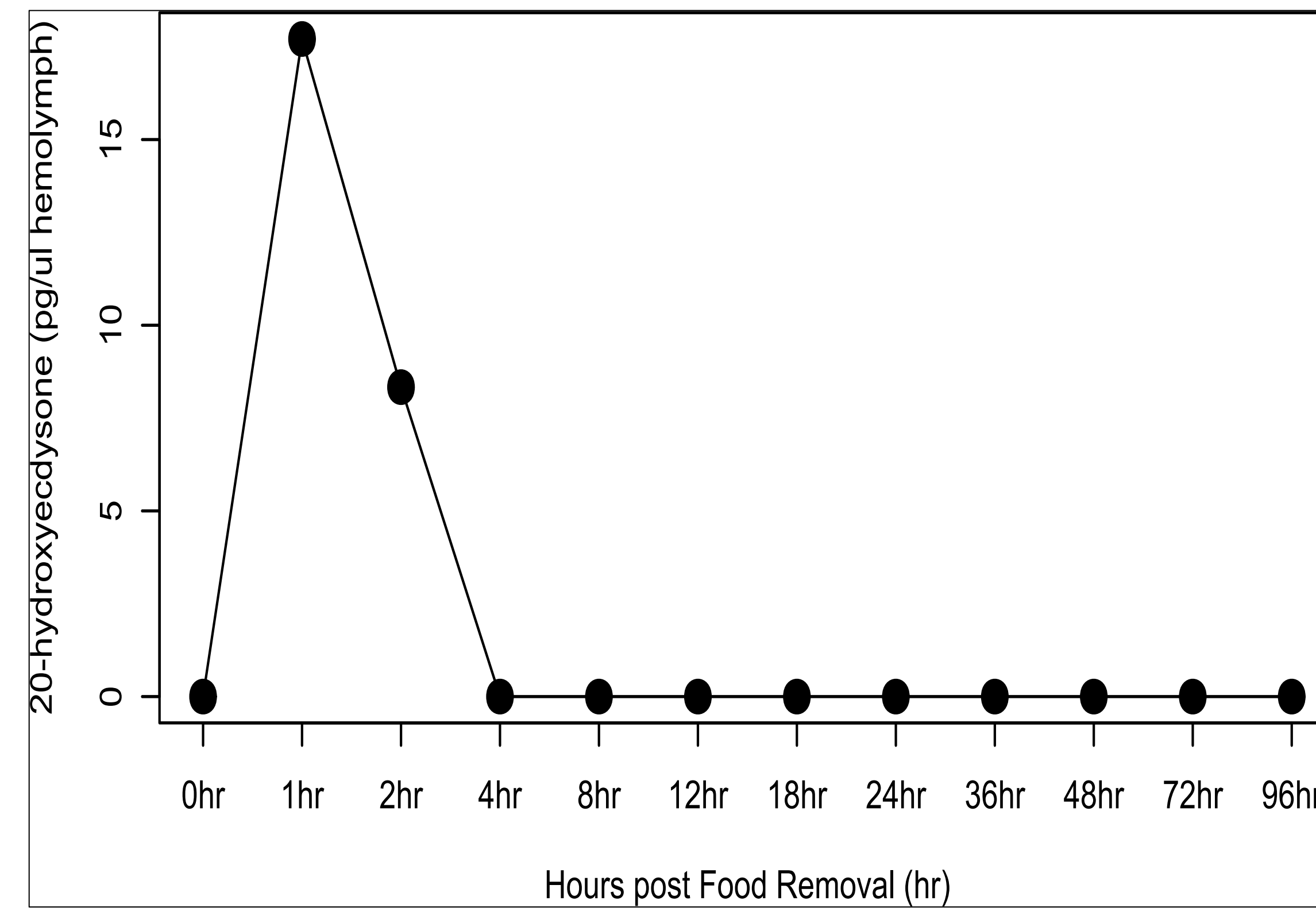


Figure 1. Ecdysone titers (via HPLC) in *O. lignaria* after food removal. The highest surge of 20-E occurs in the first hour of food removal; however, the second surge of ecdysone did not occur even after 96 hours of starvation.

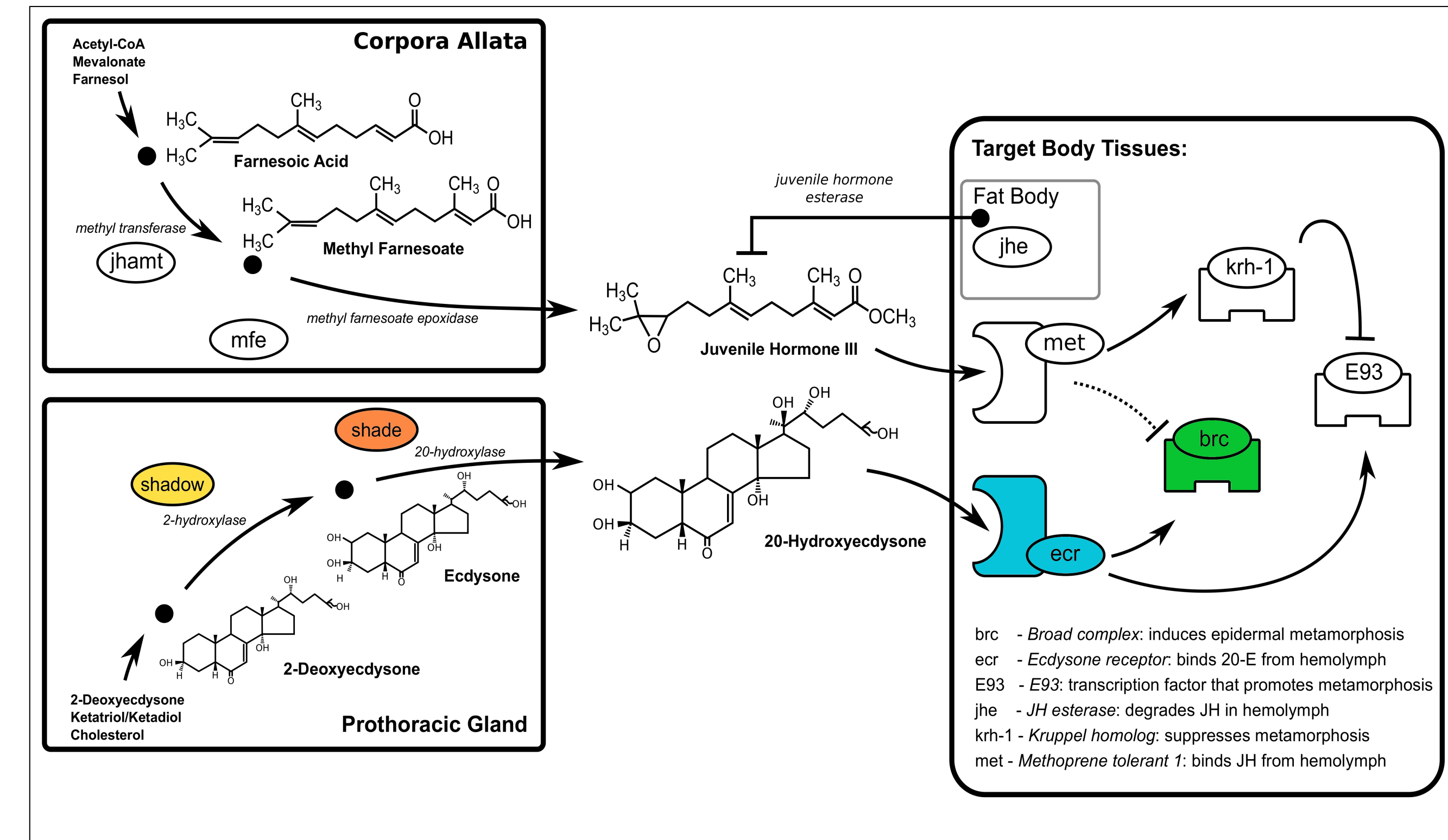


Figure 2. The signaling pathway of 20-E and JH III with respective genes and receptors for ecdysone highlighted in various colors.

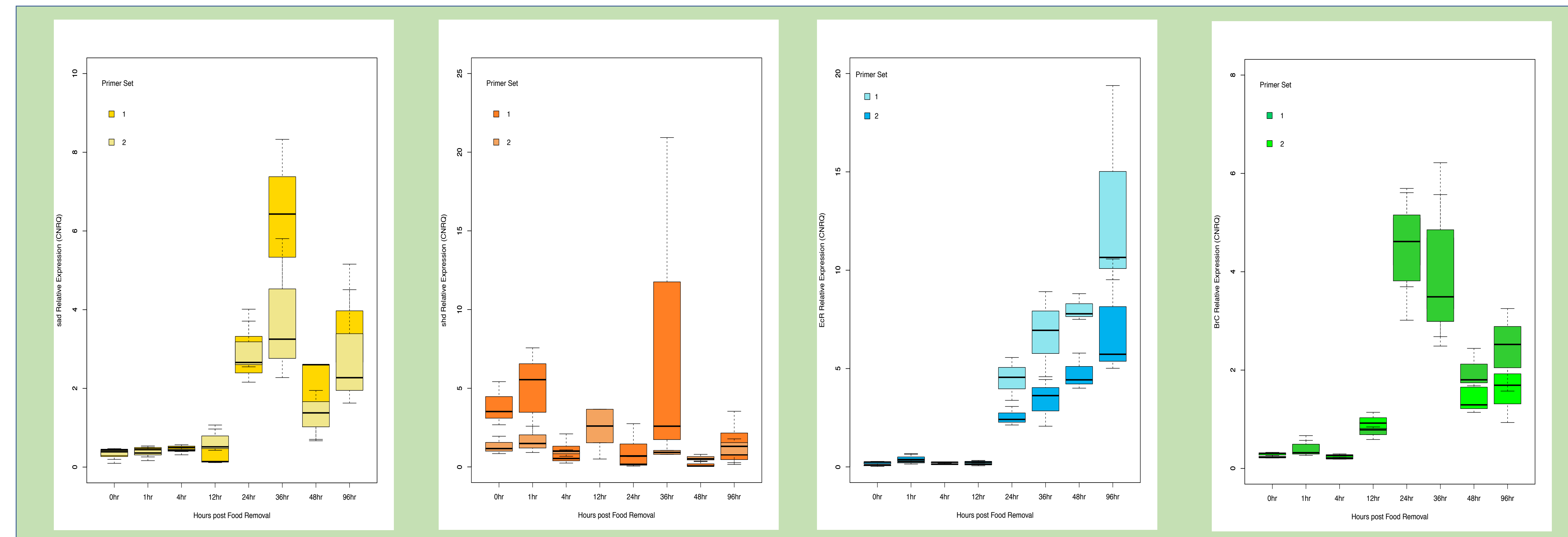


Figure 3. Relative quantity of gene expression of shadow (sad) in yellow, shade (shd) in orange, EcR in blue, and BrC in green with two different primer sets for each. Generally, there was more expression as time of food removal progressed.

## Conclusion

- After 24 hours of starvation, sad, shd, EcR, and BrC are upregulated.
- 20-E is present after the first hour of starvation, but is not present even after 96 hours of starvation.

## Future Work

- When does the late, second peak of 20-E occur?
- Does *O. lignaria* undergo diapause in the summer?
- How does diapause affect the upregulation of genes/hormones?
- How is the network of other genes, receptors, and transcription factors regulating genetic expression?

## Acknowledgements

This would not have been possible without the help of Marnie Larson, Dacotah Melicher, Jason Holthusen, Arun Rajamohan, the USDA, and the NSF

## References

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