# Waterproof eggs let insects conquer dry land

Membrane that protects eggs from drying out freed critters from need to stay close to water.

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Insects were among the first animals to invade the land around 400 million years ago, and they have diversified so greatly that they now account for three-quarters of all animal species.

This success, according to a new study, depended to a great extent on the serosa – a membrane that makes insect embryos waterproof as they develop inside the eggs. By genetically removing the serosa from embryos of the red flour beetle (*Tribolium castaneum*), Maurijn van der Zee at Leiden University in the Netherlands and his colleagues show that it is crucial in preventing them from drying out or becoming waterlogged. The study is published in the *Proceedings of the Royal Society B*<sup>1</sup>.

The serosa secretes a cuticle of hard, impermeable chitin below the eggshell, which restricts the flow of water through the egg. In an earlier experiment, van der Zee had used a technique called RNA interference to block the activity of a gene called *Tc-zen1*, which red flour beetle embryos need to make a serosa<sup>2</sup>. To his surprise, the embryos survived without the membrane. "That made us question why they go to the effort of making the serosa at all," says Chris Jacobs, another author on the study also at Leiden University.



Peggy Greb/ARS/USDA

Drought-resistant eggs may have been crucial for insects such as the 3-millimetre-long red flour beetle to thrive on land.

### Waterproof

But the embryos could have survived only because of the ideal laboratory conditions of the experiment. In the new work, the Leiden team again blocked *Tc-zen1* expression, and then tried exposing the eggs to changes in humidity. They found that in dry environments with less than 5% humidity, just 6% of their serosa-less eggs hatched compared to 75% in more favourable conditions. And in extremely humid environments, embryos without a serosa failed to develop properly because their eggs absorbed too much water.

Jacobs speculates that the evolution of the serosa enabled ancient insects to lay eggs anywhere on land, without having to restrict themselves to aquatic habitats like their crustacean relatives. In this respect, the serosa is like the amnion, an embryonic membrane that has enabled some vertebrates to break the tie with water, whereas their relatives the amphibians did not. "Conquering land is about resisting desiccation," says Jacobs.

"This is a remarkably clever set of experiments, with clear functional interpretations of an insect structure that has been a mystery for a long time," says Art Woods, a physiological ecologist at the University of Montana in Missoula.

Almost all insects have a serosa, except for primitive groups such as the silverfish, and a group of flies that evolved relatively recently and includes houseflies and fruit flies, *Drosophila melanogaster*. Because many embryonic cells contribute to the serosa, Jacobs speculates that the flies lost it as an adaptation for faster growth. "If you don't have to make a serosa, you have twice as many cells to make an embryo from, so you might be able to develop much quicker," he says. These flies lay their eggs in moist habitats such as soil, food, rotting vegetables or living tissues.

Other land-living arthropods, the taxonomic group to which insects and crustaceans belong, lack a serosa, and either lay their eggs in moist habitats or have evolved other adaptations. Spiders, for instance, wrap their eggs in silk, and scorpions give birth to live young. With no such restrictions, insects were free to exploit the full range of terrestrial niches.

But Woods says that the serosa is unlikely to be a "silver-bullet explanation for insect success". Insects have complex life cycles with several stages, and their conquest of land probably involved significant changes in all of these. Other factors, such as flight and their co-evolution with flowering plants, were probably important too.

# References

- 1. Jacobs, C. G. C., Rezende, G. L., Lamers, G. E. M. & van der Zee, M. *Proc. Roy. Soc. B* http://dx.doi.org/10.1098/rspb.2013.1082 (2013).
- 2. Van der Zee, M., Berns, N. & Roth, S. Curr. Biol. 15, 624–636 (2013).