Biologists Discover Amphibian Eggs Defend Themselves Against Water Molds

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Boston University (BU) scientists have discovered that several species of amphibians use defense mechanisms to protect themselves against deadly water molds found in vernal pools of New England.

Using both field observations and laboratory experiments, Ivan Gomez-Mestre, a research associate in Professor Karen Warkentin's laboratory in the Department of Biology at BU, describes the various methods used by the spotted salamander, wood frog, and American toad to help avoid and survive water mold infections. The results appear in the October issue of the journal *Ecology*.

"Certain water molds cause substantial mortality for aquatic eggs of a wide range of fish and amphibian species throughout the world," said Dr. Gomez-Mestre. "The observations and results of this study demonstrate that there are both parental and embryonic-stage traits that defend egg clusters against water mold infections in three species of amphibians found in the Northeast."

In the Northeast, vernal pools fill with the rising water table of fall or winter or with the melting and runoff of winter and spring snow and rain. Many vernal pools in the region are covered with ice in the winter months and contain water for a few months in the spring and early summer. Typically by late summer, most vernal pools are completely dry.

To assess the incidence and impact of water mold on eggs of the three

study species, Gomez-Mestre and the team – which included Dr. Warkentin and Justin Touchon, a graduate student in the Warkentin lab – surveyed nine vernal pools in Lynn Woods Reservation (Lynn, MA) in spring 2005. The team surveyed the ponds twice weekly from late-March through early-June before the pools dried for the summer. All egg clutches found were flagged and monitored until they hatched except for several that were collected for laboratory experiments.

In order to determine the presence or absence of water molds in the ponds, the researchers sank five tea bags filled with sterilized hemp seeds into each pond as water mold baits. After 10 days, the bags were retrieved and plated on a sterilized cornmeal agar medium on Petri dishes and grown in incubators. Water mold was found in all ponds surveyed. Eight of the nine had high infection rates, with 66 – 86 percent of baits infected, but baits from the largest pond with the thickest tree canopy and the lowest temperature showed only 16 percent infection rate.

“We visually estimated the degree of mold infection within each clutch and considered clutches to be infected when mold had grown over 5 percent or more of the clutch and increased over subsequent observations,” said Touchon.

According to the results, all three amphibian species display behaviors that help protect them from or survive infections by water molds. These defense mechanisms are carried out either by the adult females when laying eggs or by the developing embryos themselves.

Spotted salamanders wrap their eggs in a protective jelly layer that prevents mold from reaching the embryos. Wood frog egg clusters have less jelly, but are laid while ponds are still cold and mold growth is slow. Eggs of the American toad experience the highest mold infection levels since they are surrounded by only a thin jelly coating and laid when

ponds have warmed and mold grows rapidly. However, eggs of all three species are capable of hatching early if mold reaches them and this response was strongest in the American toads. Toad embryos hatched as much as 36 percent prematurely, before they could even move, suggesting accelerated development and the use of enzymes to aid hatching.

“This is quite a dramatic change. If you compared it to humans, this would be like a six months premature baby,” said Warkentin.

In the case of the salamanders, early hatching occurred only after the protective jelly coating was removed in the lab and mold was able to reach the eggs. Although this jelly works well as an anti-mold defense for spotted salamanders near Boston, in upstate New York the species suffers high mortality from water mold infections.

“We don’t yet know if the mold is different in Boston, or the eggs,” said Gomez-Mestre.

Another interesting finding from the study is despite being potential toad hatchling predators, wood frog tadpoles can have a positive effect on toad eggs by eating mold off infected toad clutches which increases their survival rates.

Water molds are a common threat for aquatic embryos, but the study species all demonstrated traits that function to reduce the risk of infection, as well as responses once infection occurs. Parental traits, including breeding early in the year or providing eggs with a protective jelly, decrease the risk of infection to a clutch while hatching early enables embryos to escape a clutch already infected with mold.

“These defenses appear to reduce the overall impact of water mold, so that massive egg mortality is normally associated with a several stressors.

Furthermore, interactions with other species may further reduce mortality rates of infected clutches," explained Gomez-Mestre. "The impact of water molds on a given species therefore depends not only on the effectiveness of the its own defenses, but also on the community composition and the ecological interactions at work in the pool."

Source: Boston University