

“How do Embryos Assess Risk? Vibrational Cues in Predator-Induced Hatching of Red-Eyed Treefrogs” (2005), by Karen Warkentin

In “How do Embryos Assess Risk? Vibrational Cues in Predator-Induced Hatching of Red-Eyed Treefrogs” (2005), Karen Warkentin reported on experiments she conducted to see how red-eyed treefrog embryos, *Agalychnis callidryas*, can distinguish between vibrations due to predator attacks and other environmental occurrences, such as storms. Though the ability of red-eyed treefrogs to alter their hatch timing had been documented, the specific cues that induce early hatching were not well understood. Warkentin's study demonstrated that, based on vibration signals alone, treefrog embryos can determine whether they are under attack from a predator and respond accordingly.

The ability of embryos to alter their time of hatching, as well as the timing of metamorphosis from juvenile to adult stages, is a main focus of Warkentin's research group. Warkentin holds a joint position as Associate Professor of Biology at Boston University in Boston, Massachusetts, as well as Research Associate at the Smithsonian Tropical Research Institute (STRI). She earned her PhD at the University of Texas in 1998. From 2000 to 2001, Warkentin held a postdoctoral position at STRI, and she continued later research there.

The timing of hatching of an egg is important for embryos that do not receive parental care. These eggs are generally vulnerable to predators, in which case embryos experience a trade-off between the benefits of longer development and the push to hatch early to avoid remaining vulnerable. If an embryo could accurately detect potential risks, such as predators, it could remain in the egg as long as possible unless danger is imminent.

Red-eyed treefrogs are well-known for exhibiting predator- and environment-induced plasticity of hatching time. Females of this species lay groups of eggs, called clutches, on vegetation overhanging ponds, where the eggs may develop undisturbed for six to seven days until they hatch. The eggs in clutches attacked by predators may hatch up to thirty percent earlier, as early as day four of development. Before hatching, these eggs often fall prey to snakes or wasps. Once the treefrog hatches it will fall into the pond below, where the hatchlings may become prey for aquatic predators like fish and shrimp.

Previous work from Warkentin showed that hatchling mortality rates due to aquatic predators vary with hatchling size, with smaller hatchlings more vulnerable to a greater number of predators. Embryos that remain in the egg for longer periods of time may grow to a less vulnerable size, while juveniles that hatch early may leave the egg smaller and more vulnerable compared to juveniles that hatch normally. Thus, while early hatching may increase the chances that hatchlings will survive from arboreal predator attacks, it also exposes hatchlings to a higher risk of predation from aquatic predators.

Warkentin hypothesized that vibrations alone, rather than in concert with visual or chemical cues, would be enough to induce early hatching in treefrog embryos. Vibrations may be passed through air, water, or substrate. Due to the trade-offs associated with hatching too early, Warkentin hypothesized that treefrog embryos could distinguish between the vibrations created by a predator and those that arise from other environmental stimuli.

To test whether vibrational cues were distinguishable and adequate for early hatching, Warkentin conducted a two-part experiment. First, Warkentin recorded and described the differences in vibrations experienced by embryos due to sibling hatching, snake predation, or rain. Second, Warkentin

manipulated the vibrations experienced by each clutch in order to determine what triggers early hatching.

Warkentin collected young clutches of red-eyed treefrogs near Gamboa, Panama, along with two species of snakes to act as predators: cat-eyed snakes and parrot snakes. On day three of egg development, Warkentin mounted clutches on cards attached to rigid jars above cups of water in individual enclosures. Warkentin inserted a tool that measures vibrations by sensing three-dimensional acceleration, called a miniature isotron accelerometer, into these clutches to measure vibrations of the surrounding eggs within the jelly egg mass. Warkentin then induced snake attacks by placing the clutch in a cage with snakes that were conditioned to eat eggs every few days. In order to observe early hatching caused by vibrations during storms, Warkentin positioned cages with clutches outside during storms so that rain could fall directly on the clutch.

Warkentin compared her results between twelve snake-attacked clutches and seven storm-exposed clutches. She analyzed the vibrations for patterns regarding duration, peak acceleration, and frequency, as well as the length of intervals between events. Warkentin found significantly higher and more variable vibrations due to rain than those caused by snake predation. Warkentin then artificially vibrated the clutches using a mini-shaker programmed to imitate either the long-lasting, spaced-out disturbances of snake attacks, or the shortened vibration events with short intervals characteristic of rain storms.

Following the artificial vibration experiment, Warkentin found that snake-like vibrations caused significantly more early hatchings than did rain-like vibrations. These results suggested that red-eyed tree frog embryos can distinguish between vibration signals and that they use this information in order to make behavioral decisions. Warkentin's work suggests that some embryos may have more control over their development than previously thought. Published in a prominent behavior journal and covered by *Scientific American* and *Natural History*, among other media sources, Warkentin's results interested many who were curious about her view of embryos as capable of behaviors and with unexplored control over their own development.

The discovery of early hatching behavior that depends on vibrational signals created the foundation for further research into embryo behavior and perception. Later publications from Warkentin's lab investigated what characteristics make a dangerous signal versus a benign signal and how embryos can avoid reacting to false alarms. Warkentin's 2005 study indicates that embryonic behavior may alter an organism's life history and is more sophisticated than previously thought.

Sources

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