Amphibian Ranavirus Surveillance: Sample Type and Biosecurity

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Abstract: Ranaviruses are pathogens contributing to worldwide amphibian declines. Field researchers conducting surveillance studies are met with challenges regarding what type of samples to collect and how to prevent transmission among field sites. While lethal methods allow for histological examination and testing of internal organs to more accurately assess the degree of infection, non-lethal methods of pathogen surveillance (e.g., tail or toe clips, surface swab) often are desired, especially for threatened species. To compare the accuracy of non-lethal versus lethal test results, we performed polymerase chain reaction (PCR) for ranavirus on swab specimens, tail clips, and fresh-frozen internal organs (liver, kidney) from 96 American bullfrog (*Lithobates catesbeianus*) tadpoles. Tail clip PCR results matched those of internal organs more often than results from swab specimens (75% versus 68%). False-negative results were similar between tail clips and swabs (20% and 22%, respectively), but false-positive results occurred twice as often with swabs (12% versus 6%). Although false-negative results were reduced to 11% when non-lethal samples were combined, the false-positive results remained at 12%. False-positive and false-negative results likely reflect environmental contamination and viral load of the samples, respectively. Due to the occurrence of false test results with non-lethal samples, we recommend that biologists submit whole fresh amphibians when possible to test for ranavirus. For non-lethal sampling, tail clips from tadpoles are preferred over swabs. Regardless of sampling technique, pathogen transmission on field equipment and personnel (e.g., boots) must be minimized. We recently found that chlorhexidine diacetate (Nolvasan) and sodium hypochlorite (bleach) are effective at inactivating ranavirus at concentrations of 0.75% and 3.0%, respectively, with one minute contact time. We recommend that biologists who work in aquatic systems disinfect sampling equipment and footwear to reduce the likelihood of ranavirus transmission among watersheds. Debris and mud must be removed prior to applying disinfectants.

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