American Association of Zoo Veterinarians Infectious Disease Committee Manual 2013

CHYTRIDIOMYCOSIS DISEASE

Animal Group(s) Affected	Transmission	Clinical Signs	Severity	Treatment	Prevention and Control	Zoonotic
Amphibians	Contact with contaminated water, moist or wet substrates, or infected animals. Crayfish may act as carriers.	Erythematous skin, exces- sive skin shedding, abnormal behavior, sudden death	Outcome of infection ranges from subclinical to fatal	Itraconazole or chloramphenicol baths, elevated temperatures	Isolate affected amphibians	No

Fact Sheet compiled by: Cynthia Stadler

Sheet completed on: 3 August 2011; updated 30 May 2013 **Fact Sheet Reviewed by**: Allan Pessier; Kathryn C. Gamble

Susceptible animal groups: Amphibians

Causative organism: Batrachochytrium dendrobatidis (Bd), a non-hyphal zoosporic fungus.

Zoonotic potential: No

Distribution: World-wide wherever amphibian populations are present. Chytridiomycosis has been implicated as the cause of massive amphibian population declines.

Incubation period: 14-70 days

Clinical signs: Erythematous or discolored skin, abnormal posture, neurologic signs, excessive skin shedding, behavior changes. Clinical signs may not be apparent prior to acute death.

Post mortem, gross, or histologic findings: Gross lesions are often not present but may include increased sloughing of the skin, discolored skin, erosions. Histologic lesions involve focal hyperkeratosis and epidermal hyperplasia with sloughing of the keratin layer. Fungal zoosporangia are found within the keratin layers. The fungal lesions are not evenly distributed on the skin surface. Predilection is noted for the digits, ventral aspect of the hind limbs, inguinal and pelvic regions, and in tadpoles, mouth parts.

Diagnosis: PCR (skin swab best), histopathology. Cytology requires experience. PCR is best for detecting subclinical infection, whereas histopathology and cytology are most useful for clinically significant infection.

Material required for laboratory analysis: Shed skin, skin scraping, skin swab or skin sample (preferably from the ventral pelvic patch) from adults. Mouthpart swabs from live tadpoles, mouthparts from deceased tadpoles. Using fine-tipped swabs (not wooden-handled), gently swab skin 20-30 times. Break swab 2-3 cm from tip and place in screw-top tube, avoiding contact with outside of tube. Allow to air-dry for 5 minutes. Samples can be kept at room temperature or 4 degrees C for 1- 2 weeks or frozen for longer-term storage. Avoid exposure to high temperatures and direct sunlight.

Relevant diagnostic laboratories:

For histopathology, any laboratory that routinely examines amphibian tissues.

For PCR:

Amphibian Disease Laboratory 15600 San Pasqual Valley Road Escondido, CA 92027

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http://www.sandiegozooglobal.org/News/Amphibian_Disease_Laboratory/

Pisces Molecular 1600 Range Street, Suite 201 Boulder, CO 80301 303-546-9300 info@pisces-molecular.com http://pisces-molecular.com

Research Associates Laboratory 14556 Midway Road Dallas, TX 75244

Phone: (972) 960-2221 Fax: (972) 960-1997 http://vetdna.com

Zoologix Laboratories

9811 Owensmouth Avenue, Ste. 4 Chatsworth, CA 91311-3800

Phone: 818-717-8880 Fax: 818-717-8881 info@zoologix.com http://zoologix.com

Treatment: Itraconazole 0.005% (50 mg/liter) diluted with 0.6% saline or amphibian Ringer's solution used as a 5 minute bath applied once daily for 6-10 days. Lower concentration of 0.0025% (25 mg/liter) also has been successful at eliminating the organism. Hygiene is essential during treatment and animals should be returned to a clean disinfected container after EACH treatment. Previously recommended higher concentration of 0.01% itraconazole is toxic to tadpoles and recently metamorphosed amphibians. Other treatments include chloramphenicol baths and elevated environmental temperatures of 37°C for 16 hrs, in those species that are thermo-tolerant. Animals with clinical chytridiomycosis may have issues from hyponatremia and hypokalemia so electrolyte replacement may be helpful.

Prevention and control: Newly acquired amphibians should undergo a minimum of 30 days in quarantine, preferably 60 days. Skin swab PCR testing or prophylactic itraconazole baths should be implemented prior to release from quarantine. All animals that die in quarantine should be necropsied and submitted for histopathology. Enclosures and equipment should be disinfected routinely. However, it is prudent to wear disposable gloves and use separate equipment for different enclosures.

Suggested disinfectant for housing facilities: Bleach, Virkon and quaternary ammonium compounds can be used for enclosures. For surgical instruments, 70% ethanol, glutaraldehyde, and benzalkonium chloride can be used.

Notification: None

Measures required under the Animal Disease Surveillance Plan: Currently none

Measures required for introducing animals to infected animal: Introductions are not recommended until numerous negative PCR tests have been completed.

Conditions for restoring disease-free status after an outbreak: It is recommended to test amphibians by

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PCR to confirm the fungus is no longer present. For disease-free status, there should be serial negative PCR tests over the course of 6 months to 1 year.

Experts who may be consulted:

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References:

- 1. http://www.amphibianark.org/the-crisis/chytrid-fungus/ Accessed 5 August 2013.
- 2. http://amphibiaweb.org/chytrid/chytridiomycosis.html Accessed 5 August 2013.
- 3. http://www.wildlifehealth.org.au/FactSheets.aspx Accessed 5 August 2013.
- 4. Berger, L., R. Speare, and A. Kent. 2000. Diagnosis of chytridiomycosis in amphibians by histological examination. Zoos Print J. 15: 184-190.
- 5. Bishop, P.J., R. Speare, R. Poulter, M. Butler, B.J. Speare A. Hyatt, V. Olsen, and A. Haigh. 2009. Elimination of the amphibian chytrid fungus *Batrachochytrium dendrobatidis* by Archey's frog *Leiopelma archeyi*. Dis. Aquat. Org. 84: 9-15.
- 6. Brannelly, L.A., C.L. Richards-Zawacki, and A.P. Pessier. 2012. Clinical trials with itraconazole as a treatment for chytrid fungal infections in amphibians. Dis. Aquat. Org. 101:95-104.
- 7. Forzan, M.J., H. Gunn, and P. Scott. 2008. Chytridiomycosis in an aquarium collection of frogs: diagnosis, treatment, and control. J. Zoo Wildl. Med. 39(3): 406-411.
- 8. Jones, M.E.B, D. Paddock, L. Bender, J.L. Allen, M.S. Schrenzel, and A.P. Pessier. 2012. Treatment of chytridiomycosis with reduced-dose itraconazole. Dis. Aquat. Org. 99: 243-249.
- 9. Pessier, A.P., and J.R. Mendelson (eds). 2010. A Manual for Control of Infectious Diseases in Amphibian Survival Assurance Colonies and Reintroduction Programs. Conservation Breeding Specialist Group, Apple Valley, MN. 229 pp.
- 10. Pessier, A.P. 2008. Amphibian chytridiomycosis. *In*: Miller, R.E., and M.E. Fowler (eds.). Fowler's Zoo and Wild Animal Medicine: Current Therapy, Vol. 6, Elsevier Saunders, St. Louis, Missouri. Pp. 137-143.
- 11. Pessier, A.P. 2012. Diagnosis and control of amphibian chytridiomycosis. *In*: Miller, R.E., and M.E. Fowler (eds.). Fowler's Zoo and Wild Animal Medicine: Current Therapy, Vol. 7, Elsevier Saunders, St. Louis, Missouri. Pp. 217-223.
- 12. Tamukai, K., Y. Une, A. Tominaga, K. Suzuki, and K. Goka. 2011. Treatment of spontaneous chytridiomycosis in captive amphibians using itraconazole. J. Vet. Med. Sci. 73: 155-159.
- 13. Woodhams, D.C., R.A. Alford, and G. Marantelli. 2003. Emerging disease of amphibians cured by elevated body temperature. Dis. Aquat. Organ. 55: 65-67.
- 14. Young, S., R. Speare, L. Berger, and L.F. Skerratt. 2012. Chloramphenicol with fluid and electrolyte therapy cures terminally ill green tree frogs (*Litoria caerulea*) with chytridiomycosis. J. Zoo Wildl. Med. 43(2): 330-337.