

Nasal Discharge in Tortoises

Thomas H. Boyer, DVM, DABVP (Reptile & Amphibian Practice)
Pet Hospital of Penasquitos, San Diego, CA, USA

Nasal discharge is a common presenting clinical sign in tortoises and some turtles and is always abnormal, even if chronic. Many clients are not aware of this and fail to seek treatment, or consider discharge not a problem because of its long standing nature. Clients have even speculated the coastal marine layer in southern California causes nasal discharge; not true! There are many potential causes. Mycoplasmosis remains the most common cause of nasal discharge but the differential diagnoses in chelonians also includes colonic obstruction, herpes virus, ranavirus, adenovirus, intranuclear coccidiosis, chlamydiosis, reovirus, paramyxovirus, picorna-like viruses, foreign bodies, oronasal fistulas and even hypovitaminosis A, except in tortoises.

First distinguish nasal discharge from gastric reflux. Gastric reflux or regurgitation occurs with colonic obstruction or stasis and is easy to demonstrate on radiographs (DV, lateral and anterior posterior). Look for distended bowel loops in the lung fields on the horizontal lateral and AP views, and bowel distention, foreign bodies or masses, such as cystouroliths, on the DV view. Dyschezia and bloating may or may not be present but anorexia, constipation or obstipation, are typically present. Green to brown saliva or macerated partially digested greens may be in the oral cavity or discharging from the mouth or nares. Colonic impaction can result from gravel, sand, fibrous material, plastics, wire, coins, nails, screw, yard trash, bladder stones, tumors, gastrointestinal infection, or hypothermia. Colonic obstruction is beyond the scope of this discussion, but be sure nasal discharge is just that, and not gastric reflux, before proceeding. See What You Need to Know to Treat Tortoises for more information.

The most common cause of nasal discharge is upper respiratory tract disease, or mycoplasmosis. Several species of Mycoplasma, *Mycoplasma agassizii*, *Mycoplasma testudineum*, and perhaps several yet to be identified Mycoplasma species, are causative. *Mycoplasma testudinus*, isolated from the cloaca of a Greek tortoise, is not thought to cause disease. Mycoplasmosis has been described in an ever widening arc of tortoise species (especially *Gopherus*, *Testudo* and *Geochelone* species) as well as box turtles, bog and spotted turtles. All terrestrial turtles are likely susceptible. Mycoplasmosis is widespread in wild *Gopherus agassizii* and *Gopherus polyphemus* but was not found in wild *Gopherus berlanderi*. However 80% of 39 *G. berlanderi* were seropositive in a Texas rehabilitation facility. In *G. polyphemus* disease incidence is higher in sexually mature adults, especially males, compared to juveniles. Contacts through courtship, mating and male agnostic behavior may be required for direct horizontal transmission. Initial mortality is high; those that survive have low mortality and high morbidity.

To understand mycoplasmosis we must appreciate nasal sinus anatomy. *Mycoplasma* bacteria attach to the surface of epithelial cells in the nasal sinus and choana (internal nares) which causes an over-exuberant host response. The nasal mucosa suffers from loss of ciliated epithelium, mucosal hyperplasia, and infiltration of lymphocytes and histiocytes. Normal olfactory mucosa becomes replaced with proliferating mucosal epithelial cells and proliferating basal cells, heterophils, and histiocytes. The lesions slough large amounts of epithelial and inflammatory cells, which create the nasal discharge and over time form caseous material filling and occluding the nasal sinus. Clinical impression is that most tortoises don't eat well because they use olfaction to find and select food and mycoplasmosis decreases olfaction.

Clinical signs include clear serous to tenacious mucoid bubbling to mucopurulent nasal discharge, sometimes also from the eyes, conjunctivitis and palpebral edema. Clinical signs may appear within 1-2 weeks of exposure, it takes *G. polyphemus* 6-8 weeks to develop an immune response. Antibiotics against *Mycoplasma* spp. do not appear to provide protective immunity. *Mycoplasma* is spread directly via nasal exudates and is very contagious as tortoises often greet one another nose to nose. Many tortoises are chronically infected. Infected tortoises are often inappetent, lethargic with clear nasal discharge, bubbling from the nares or have clogged nares and have lost weight or feel light. Chronic cases can have erosion of the nares and rhampotheca; hatchlings can have enlargement and distortion of the snout (reminiscent of

atrophic rhinitis in swine). If nasal discharge is not apparent during clinical examination, pushing the tortoise's head straight back into its shell will often produce nasal discharge.

Several diagnostic options are available. Chelonian *Mycoplasma* culture is extremely difficult due to slow growth (4–6 weeks for *Mycoplasma agassizii*), small size (colonies not visible without dissecting microscope) and fastidious culture requirements (lower temperature, special expensive culture media and with serum added). In Jacobson, *et al*, 1994 paper less than a quarter of infected tortoises cultured positive even with a dedicated university *Mycoplasma* laboratory. *Mycoplasma* lacks a cell wall which makes life outside the host precarious at best. PCR diagnosis is also problematic because *Mycoplasma* numbers fall over time due to host response and good nasal flush samples are hard to obtain without anesthesia. Nasal flushing is more sensitive than nasal swabbing. A negative PCR nasal flush doesn't necessarily mean the tortoise doesn't have *Mycoplasma*, it just wasn't in that sample. Serology is the test of choice for *Mycoplasma* infections in other species and ELISA testing has been validated for desert and gopher tortoises to detect anti-*Mycoplasma* antibodies. The disadvantage of ELISA is that it only detects antibodies which documents exposure but not necessarily infection. None-the-less this is an important screening test when importing chelonians to a closed collection. Whole blood or serum can be shipped on ice packs or dry ice via Fed Ex Priority Overnight Service sent to: Mycoplasma Testing Lab, University of Florida, Department of Pathobiology, 1600 SW Archer Road - BSB 350, Gainesville, FL, 32610 (contact lab beforehand, at 352-392-4700 x 3968, to request a sample submission form, samples must arrive on a weekday) or through various commercial laboratories. There is a statistically significant positive correlation between severity of clinical signs and serum antibody ELISA status. Most symptomatic tortoises the author has tested are positive, clients often balk at the cost of testing, in which case the author proceeds to treatment without confirmation.

Four major classes of drugs are used to systemically treat *Mycoplasma* include fluoroquinolones, macrolides, tetracyclines and chloramphenicol. Aminoglycosides are not indicated. Tetracyclines and chloramphenicol are not commonly used in chelonians, probably because of a lack of pharmacokinetic data. Tetracyclines are used in crocodylians with mycoplasmosis, Jarchow recommended 6 mg/kg oxytetracycline IM q 24 hrs x 10–14 days for tortoises. Fluoroquinolones options include enrofloxacin (Baytril 100, 100 mg/ml, or Baytril 2.27%, 22.7 mg/ml, Bayer Corp, Shawnee Mission, KS, 5 mg/kg SC q 24 hrs for 3–6 weeks, vary injection site for Baytril 2.27%), or danofloxacin mesylate (A180, 180 mg/ml, Pfizer Animal Health, NY, NY, 6 mg/kg SC q 48 hours for 3–6 weeks) or clarithromycin (Biaxin, 50 mg/ml, Abbott Labs, Abbott Park, IL, 15 mg/kg PO q 48–72 hours for 3–6 weeks). Palatability is horrible with clarithromycin but in some cooperative chelonians it can be given orally. Antibiotics alone, even long term, do not seem to clear *Mycoplasma*, perhaps because *Mycoplasma* spp. live on the nasal epithelial surface and do not penetrate into tissue. One study, Rettenmund, *et al*, 2014, treated *Mycoplasma* PCR positive asymptomatic Forsten's tortoises, *Indotestudo forstenii*, and Sulawesi forest turtles, *Leucocephalon yuwonoi*, with 20 mg/kg clarithromycin PO q 2–3 days for 3 months. Clarithromycin failed to suppress *Mycoplasma* shedding in 9 out of 10 PCR positive animals.

Another retrospective preliminary study of 10 symptomatic single desert tortoises showed promising results in eliminating clinical signs. No diagnostics were performed. Jarchow 2004, combined nasal flushing with systemic enrofloxacin and found 8 out of 10 desert tortoises remained asymptomatic while still being monitored, for 11–78 months, and 2 out of 10 became symptomatic again at 12 and 30 months post treatment. Treatment consisted of 5 mg/kg enrofloxacin (2.27% or 100 mg/ml) IM in the brachial muscle q 72 hrs (with a range of 48 to 96 hrs) for a total of 3 to 5 treatments and, in addition, flushing the nasal sinus with a solution of 3.0 mg enrofloxacin (2.27%), 0.12 mg dexamethasone (not DexNaP), and 0.8 ml 0.9% sodium chloride, q 72 hrs (with a range of 48 to 96 hrs), also for 3 to 5 treatments. Nasal flushes were applied by placing the (unsexed) tortoise in dorsal recumbency, opening the mouth and flooding the choanae with the flush solution. The mouth was then closed and the solution forced through the nares by applying digital pressure on the intermandibular tissue, pushing the tongue into the choanae. The process was then repeated and the tortoise returned to sternal recumbency before again applying pressure to the intermandibular tissue. The nasal cavity was flushed repeatedly in this manner until only the flushing solution, with no obvious mucus, was expelled from the nares. Treatment was usually one time past resolution of clinical signs, five treatments was typically needed.

For the first flush the author likes to flush from the nares to choanae in a sedated tortoise (unless small or cooperative enough that sedation isn't needed) in dorsal recumbency with the head extended, mouth open and the glottis packed off by several cotton balls clamped to a hemostat to prevent aspiration. Each nare is flushed with 12 cc saline to force thick mucus, solid cellular debris or foreign bodies out the choanae and mouth, opening up the nasal sinuses, before slowly instilling the enrofloxacin/steroid/saline solution as above. Nasal sinus flush can be used for PCR testing. After treatment, most patients have improved appetite, activity and no further nasal discharge.

Severely underweight anorexic animals benefit from an esophagostomy tube. Most cases are still eating, or were recently eating, and in fair body weight, so an esophagostomy tube isn't indicated. Keep patients between 73° (low at night) to 86°F (high during day).

Untreated patients develop chronic disease and may accumulate solid cellular debris or thick mucus in the nasal sinuses leading to weight loss and slow decline. In captivity tortoises can survive indefinitely, in the wild they may fail to make it through hibernation or die within a few years. Gopher tortoises on Sanibel Island, FL, declined by 25 to 50% over ten years and similar declines were observed in desert tortoises at the Desert Tortoise Natural Area, in Kern County, CA.

Mycoplasma positive tortoises should be isolated from *Mycoplasma* negative tortoises as the disease is extremely contagious. *Mycoplasma* spp. survive poorly in the environment without a cell wall, cages can be rinsed and disinfected with 0.15% sodium hypochlorite. PCR samples of burrows with affected animal did not detect *Mycoplasma*. Remember nasal discharge is always abnormal and should be treated. Don't ignore it just because the owner says the tortoise has always had nasal discharge.

Several other maladies can infect tortoises, and cause nasal discharge, including herpes virus, ranavirus, adenovirus, intranuclear coccidiosis, chlamydiosis, reovirus, paramyxovirus, foreign bodies, and oronasal fistulas. The difference here is that tortoises often die quickly after the owner noticing something amiss (except for the last two), which is atypical for mycoplasmosis.

Herpes virus is the next most common cause of nasal discharge. A wide variety of tortoises are affected, especially the common captive chelonians in the *Gopherus*, *Testudo* and *Geochelone* genera. Herpesvirus is characterized by stomatitis-rhinitis. Stomatitis with focal to multifocal to coalescing, white to yellow, diphtheritic plaques, appear about 11 to 12 days after exposure. Nasal discharge initially is serous but becomes mucopurulent later. Severe conjunctivitis and blepharodema may cause the eyes to swell shut with aqueous or white to yellow mucoid discharge. Hyper salivation, glossitis, dyspnea, dehydration and central nervous system signs, such as head tilt and circling, weight loss, cachexia, may be variably present. In the author's experience outbreaks often occur after introduction of Russian tortoises, *Testudo horsfieldi*, which may be carriers or symptomatic.

Two serotypes are known, which can complicate PCR testing, which can be specific for one, or both, depending on the test. Fresh diphtheritic plaques on the tongue or caudal oral cavity make this easy to distinguish from mycoplasmosis and are good for PCR testing. ELISA serology is available to screen tortoises for herpes virus, once exposed tortoises are likely carriers for life. Tortoises produce anti-herpes virus antibodies 4 to 7 weeks post-infection. Infected Mediterranean tortoises had lymphocytosis, heterophilia, elevated AST and alpha globulin fractions, significantly higher than tortoises without herpesvirus. Necropsy with an experienced reptile pathologist can confirm herpes. Impression smears of the tongue may show eosinophilic intranuclear inclusions with Giemsa or hematoxylin & eosin stains. Oral lesions with ranavirus and picorna-like virus infections can look quite similar. Keep in mind that bacterial or fungal stomatitis almost never happens in chelonians, unlike squamates. Direct horizontal transmission is typical from virus shed in respiratory secretions, saliva or feces. Virus can persist for months in soil. Prompt aggressive treatment with acyclovir (80 mg/kg PO SID-TID x 21 days), antibiotics, fluid and nutritional support via esophagostomy tube, and broad-spectrum antibiotics are important.

Ranavirus is an iridovirus known for causing mass mortalities in fish and amphibians that may have crossed over into turtles and other reptiles from frog virus 3. Ranavirus has caused a rash of epizootics in chelonians since the late 1990's, especially in box turtles, aquatic turtles and tortoises. It can cause death in a wide variety of chelonians from tortoises to soft shelled turtles. Clinical signs include lethargy, anorexia, dyspnea, nasal discharge, conjunctivitis, oral ulcerations, severe subcutaneous cervical

edema, ulcerative stomatitis, and “red-neck disease”. Like herpes virus it seems to hit the respiratory and gastrointestinal tracts very hard, histologically, infected animals have hepatitis, enteritis, and pneumonia. PCR testing is available on combined oral and cloacal swabs, as well as whole blood, as well as ELISA testing. Transmission is poorly understood at this time but outbreaks are often associated with amphibians.

A 2009 adenovirus outbreak in over a hundred illegally imported Sulawesi tortoises, *Indotestudo forsteni*, killed the majority of them despite intensive veterinary care at multiple institutions. Since then several other tortoise and box turtles species have been infected. Tortoises had nasal and ocular discharge, mucosal ulcers and palatine erosions. Pathological findings in infected tortoises were multifocal hepatic necrosis, amphophilic to basophilic intranuclear inclusions and diffuse hepatic lipidosis, myeloid necrosis in bone marrow and severe necrotizing enterocolitis. PCR testing is available for tissue, nasal flushes, cloacal swabs or serum. Virus is shed in feces and oronasal secretions and persists in the environment making fomite or keeper transfer possible.

Testudine intranuclear coccidiosis (TINC) can also result in high morbidity and mortality in a wide variety of tortoises and box turtles and was first identified in radiated tortoises, *Geochelone radiata*, in 1990. The causative coccidian and lifecycle still have not been identified. Clinical signs may include severe lethargy, rapid weight loss, weakness, gasping respiration, conjunctivitis, nasal discharge, oronasal fistulas and swollen erythematous vents with ulceration. Thick choanal mucus is present on gross necropsy. The parasite is extremely contagious and seems to permeate all tissues in tortoises. Death follows onset of clinical signs within days unless treated with ponazuril or toltrazuril. Diagnosis is typically by post-mortem histopathology or PCR of combined conjunctival, choanal and cloacal swabs. The University of Florida has a 24 test TINC panel which is great for screening collections for this highly contagious disease. Ponazuril, at 20 mg/kg, PO EOD x 3 months or toltrazuril (Baycox 5% Oral Suspension, Bayer Vital GmbH, Deutschland) at 15-20 mg/kg PO EOD x 3 months results in less mortality but it remains unknown if it cures tortoises. Red gelatin capsules make long term treatment of multiple animals more doable. Treat all in contact and symptomatic animals, as well as any PCR positive animals.

A group of research desert tortoises were examined after unexplained morbidity and mortality. Clinical signs included nasal discharge, ocular discharge, conjunctivitis, loose feces, fecal staining around the vent and mucoïd feces. Several tortoises were positive for *Mycoplasma* and *Chlamydophila*-like organisms.

In tortoises, a reovirus has only been isolated in one case from a spur-thighed tortoise, *Testudo graeca*. The tortoise was cachectic and had a necrosis of the epithelium of the tongue. Two different squamate Paramyxoviruses were recently identified by PCR in a leopard tortoise that died with copious nasal discharge and severe consolidated lung lobes.

Picorna-like viruses, previously named virus “X”, have been isolated from various *Testudo* spp and leopard tortoises. Clinical signs included diptheroid-necrotising stomatitis and pharyngitis (very similar to Herpes), conjunctivitis, rhinitis, pneumonia and ascites. Picorna-like viruses have also been isolated from healthy tortoises without clinical signs, the significance of this virus in causing disease remains undetermined.

A variety of foreign bodies can also cause nasal discharge, such as foxtails or blades of grass, in the nasal sinus or choanae, and sticks in the roof of the mouth. One sulcata tortoise had a bottle cap stuck in the roof of his mouth. Another case of chronic unilateral nasal discharge in a Russian tortoise, *Testudo horsfieldi*, had an oronasal fistula discovered while flushing the nasal sinuses. Overheating, such as a hatchling tortoise flipped over in full sun, will cause nasal discharge for several hours that resolves without treatment if allowed to cool down. Vitamin A deficiency can cause blepharodema with nasal discharge as well, but tortoises rarely get hypovitaminosis A (greens being rich in β -carotene, a vitamin A precursor), unlike aquatic turtles and box turtles which are very susceptible. Remember a good oral exam is always indicated with nasal discharge. Although *Mycoplasma* is the primary differential, do not overlook the many other potential causes.

REFERENCES

1. Jacobson ER, Gaskin JM, Brown MB, Harris RK, Gardiner CH, LaPointe JL, Adams HP, Reggiardo C. Chronic upper respiratory tract disease of free-ranging desert tortoises (*Xerobates agassizii*). *J Wildlife Ds*. 1991;27(3):296–316.
- Brown MA, Schumacher IM, Klein PA, Harris K, Correll T, Jacobson ER. *Mycoplasma agassizii* causes upper respiratory tract disease in the desert tortoise. *Infection and Immunity*. 1994;62(10):4580–4586.
- Feldman SH, Wimsatt JW, Marschang RE, Johnson AJ, Brown W, Mitchell JC, Sleeman JM. A novel *Mycoplasma* detected in association with upper respiratory tract disease syndrome in free-ranging eastern box turtles (*Terrapene carolina carolina*) in Virginia. *J Wildlife Ds*. 2006;42(2):279–289.
- Wellehan J. *Mycoplasma*. In: Mayer J, Donnelly T, eds. *Clinical Veterinary Advisor: Birds and Exotic Pets*. St. Louis, MO: Elsevier Saunders; 2013:118–119.
- Wendland L, Brown D, Klein P, Brown M. Upper respiratory tract disease (mycoplasmosis) in tortoises. In: Mader DR, ed. *Reptile Medicine and Surgery*. 2nd ed. St Louis, MO: Saunders Elsevier; 2006:931–938.
- Johnson A, Morafka D, Jacobson E. Seroprevalence of *Mycoplasma agassizii* in captive desert tortoises (*Gopherus agassizii*) from the Greater Barstow area, Mojave Desert, California. *J Arid Environments*. 2006;67:192–201.
- Tristan T. Seroprevalence of *Mycoplasma agassizii* in wild caught and rescued Texas tortoises (*Gopherus berlanderi*) in South Texas. *JHMS*. 2009;19(4):115–118.
- Wendland L, Wooding J, White L, Demcovitz D, Little R, Remish J, Ozgul Oli K, Klein P, Christman M, Brown M. Social behavior drives the dynamics of respiratory disease in threatened tortoises. *Ecology*. 2010;91(5):1257–1262.
- Collection and preparation of blood samples from gopher tortoises for determining exposure to *Mycoplasma*. Florida Fish and Wildlife Commission PDF, http://myfwc.com/media/290176/PW_urtd_info.pdf.
- Schumacher IM, Brown MA, Jacobson ER, Collins BR, Klein PA. Detection of antibodies to a pathogenic *Mycoplasma* in desert tortoises (*Gopherus agassizii*) with upper respiratory tract disease. *J Clinical Microbiol*. 1993;31(6):1454–1460.
- Wimsatt J, Johnson J, Mangone B, Totill A, Childs J, Peloquin C. Clarithromycin pharmacokinetics in the desert tortoise (*Gopherus agassizii*). *J Zoo Wildl Med*. 1999;30(1):36–43.
- Origgi F. Herpesvirus in tortoises. In: Mader DR, ed. *Reptile Medicine and Surgery*. 2nd ed. St Louis, MO: Saunders Elsevier; 2006:814–821.
- Origgi F, Klein P, Tucker S, Jacobson ER. Application of immunoperoxidase based techniques to detect herpesvirus infection in tortoises. *J Vet Diagn Invest*. 2003;14:133–140.
- Muro J, Ramis A, Pastor J, Velarde L, Tarres J, Lavin S. Chronic rhinitis associated with herpesviral infection in captive spur-thighed tortoises from Spain. *J Wild Dis*. 1998;34:487–495.
- Johnson A, Pessier A, Wellehan J, Childress A, Norton T, Stedman N, Bloom D, Belzer W, Titus V, Wagner R, Brooks J, Spratt J, Jacobson E. Ranavirus infection of free-ranging and captive box turtles and tortoises in the US. *J Wildl Dis*. 2008;44(4):851–863.
- Marshang RE. Viruses infecting reptiles. *Viruses*. 2011;3(11):2087–2126.
- DeVoe R, Geissler K, Elmore S, Rotstein D, Lewbart G, Guy J. Ranavirus associated morbidity and mortality in a group of captive eastern box turtles (*Terrapene carolina carolina*). *J Zoo Wildl Med*. 2004;35:524–543.
- Johnson AJ, Wendland L, Norton TM, Belzer B, Jacobson ER. Development and use of an indirect enzyme-linked immunosorbent assay for detection of iridovirus exposure in gopher tortoises (*Gopherus polyphemus*) and eastern box turtles (*Terrapene carolina carolina*). *Vet Microbiology*. 2010;142:3–4, 160–167.
- Rivera S, Wellehan JFX, McManamon R, et al. Systemic adenovirus infection on Sulawesi tortoises (*Indotestudo forstenii*) caused by a novel Siadenovirus. *J Vet Diagn Invest*. 2009;21:415–426.
- Jacobson ER, Schumacher J, Telford SR, Greiner EC, Buergelt CD, Gardiner CH. Intranuclear coccidiosis in radiated tortoises (*Geochelone radiata*). *J Zoo Wildl Med*. 1994;25(1):95–102.
- Johnson JD, Garner MM, Braun J, Bradway D. Chlamydiosis in a research population of desert tortoises (*Gopherus agassizii*). *Proc ARAV*. Abstract, 2012:111.
- Jarchow J. Treatment protocol for upper respiratory tract disease of desert tortoises. *7th Intl Symp Path Med Reptiles & Amph*. Berlin, Germany. 2004.
- Rettenmund C, Boyer D, Orric OW, Parker S, Wiles R, Pare J. Long-term oral clarithromycin administration to subclinical Forsten's tortoises, *Indotestudo forstenii*, and Sulawesi forest turtles, *Leucocephalon yuwonoi*. *Proc ARAV*. Orlando, FL, 2014:43.
- Jacobson ER, Brown MB, Wendland L, Brown D, Klein P, Christopher M, Berry K. Mycoplasmosis and upper respiratory disease of tortoises: a review and update. *Vet Journal*. 2014;201(3):257–264.
- Papp T, Seybold J, Marshang R. Paramyxovirus infection in a leopard tortoise (*Geochelone pardalis babcocki*) with respiratory disease. *JHMS*. 2010;20(2–3):64–68.

Marschang R, Ruemanapf T. Virus "X": characterizing a new viral pathogen in tortoises. *Proc ARAV*. 2002;101-102.
Gibbons P, Steffes Z. Emerging infectious diseases of chelonians. *Vet Clin Exot Anim Pract*. 2013;16:303-317.