BOG TURTLE HEALTH BULLETIN

July 3, 2014

Over the past few years, the U.S. Fish and Wildlife Service (Service) has received sporadic reports of dead and apparently diseased bog turtles from New York, Massachusetts, New Jersey, Delaware, Maryland, and Pennsylvania. In 2014, there have been dead turtles found in Pennsylvania, New Jersey, and North Carolina. Most notably, 14 bog turtles were found dead at 1 site in early May in Pennsylvania while 4 were found at 1 site in New Jersey between mid-April and mid-June. The Service and the two states are still investigating potential causes of death. In August 2009 and May 2013, the Service issued an Advisory Bulletin for all surveyors, researchers, and agency biologists working with bog turtles. This bulletin provides updated information and guidance for collection and processing of specimens and animals should any dead or sick turtles be found at bog turtle sites, and decontamination of clothing and equipment when working at bog turtle sites.

Pursuant to the "Emergency Provisions" of Section 6 Cooperative Agreements the Service has with the State wildlife agencies in the northern range of the bog turtle, "Any employee or agent of the Division who is designated by that Agency for such purposes, may, when acting in the course of his official duties, take federally listed Endangered and Threatened fish or wildlife species without a permit if such action is necessary to: (1) aid a sick, injured, or orphaned specimen; or (2) dispose of a dead specimen; or (3) salvage a dead specimen which may be useful for scientific study . . ." Guidelines for taking such actions, data forms for documentation (attachment A), and decontamination protocols (attachment B) to reduce the risk of disease spread are provided below to support agencies and their agents for addressing these emergency situations.

Reports and collections of sick and dead animals since 2009 have included a variety of symptoms and results. In additions to empty (no soft tissue remaining) shells, some dead bog turtles have been found entirely intact, with no obvious cause of death. Others have been incomplete specimens due to predation, scavenging, and/or decay. On several live bog turtles, a grayish or whitish substance and/or discoloration has been documented on the skin of the head, neck, and limbs, as well as on the claws. In some cases, these appear as skin lesions, scute sloughing, and loss of claws, toes, or limbs.

At this time, no causative agent(s) of many observed symptoms or mortalities has been identified. Test results are often inconclusive due to predation or decay, but some results have shown a variety of potential causative factors including injury, infection, pneumonia, and carcinoma. However, considering the risk of infectious agents such as *Mycoplasma* and *Ranavirus* as a threat to bog turtles, the Service is taking this matter seriously and is seeking the voluntary cooperation of bog turtle surveyors and researchers in implementing the following guidance regarding (1) fresh-dead, (2) carcasses and shells, and (3) sick or injured turtles.

Fresh-dead bog turtle specimens[1] should be collected and sent immediately to one of two locations: the USGS National Wildlife Health Center (NWHC) in Madison, WI, or the Wildlife Health Center at the Wildlife Conservation Society (WCS)/Bronx Zoo in Bronx, NY. For those states actively engaged in health assessment studies with WCS (MA, NY, NJ, DE, PA), the latter is recommended. Also, ship fresh-dead *turtles of any species* from known bog turtle sites (and within 1 km of known bog turtle sites) according to the same protocol.

Complete the attached **Turtle Data Sheet** (attachment A, or a similar data sheet that records the same information) and, if sending to NWHC, the USGS **Specimen History Form** as well. When the specimen is shipped, please also provide an email notification and attach copies of completed forms to: the Service's Regional Office (Alison Whitlock, Bog Turtle Recovery Coordinator), the Service point-of-contact for your state, and the appropriate state agency point-of-contact[2].

Instructions for sending turtles to the Wildlife Health Center at the Wildlife Conservation Society/Bronx Zoo:

a. Wearing disposable gloves, place the remains into a plastic bag or leak-proof container. Place that container into another one. If sending multiple specimens, use separate gloves and submit specimens in separate containers.

b. Label each container with the ID of the animal, person collecting, and contact information for that person. Attach the Turtle Specimen Form.

c. Place the container into a cooler with **cold** packs (not ice packs) as soon as possible or into a **refrigerator** (not a freezer). Keep the samples cool.

d. Contact a veterinarian or pathologist at the Wildlife Health Center at the WCS/Bronx Zoo by calling 718-220-7100 (daytime) or 718-220-5095 (nighttime) for further instructions.

e. The body should be transported within **24 hours** to the Wildlife Health Center at the Bronx Zoo or sent via FedEx or UPS overnight to:

Dr. Bonnie Raphael
Wildlife Health Center
WCS/Bronx Zoo
2300 Southern Blvd
Bronx, NY 10460
ph 718-220-7100

Please write in conspicuous lettering: For Immediate Delivery; keep cold (not frozen)
Prior to transporting an animal or body or sending samples via overnight service, contact a veterinarian or pathologist at 718-220-7100 or 718-220-5095.

[1] This refers to fresh-dead turtles whose cause of death is unknown (*e.g.*, do not ship road-killed turtles). Partially scavenged, fresh-dead turtles should be sent in for analysis, because one cannot confidently conclude that predation was the actual cause of death (*i.e.*, the cause of death is unknown). [2] Contact information for Service and State agency biologists is on pages 5 to 6 of this bulletin.

Instructions for sending to the USGS National Wildlife Health Center in Madison, WI:

a. Wearing disposable gloves, place each animal in a plastic bag, close, and seal the bag. Cover zipper bag closure with strapping or duct tape after sealing zipper. Twist nonzipper bags closed, fold over on itself, and secure with package strapping or duct tape.

b. **See full instructions** and download forms from http://www.nwhc.usgs.gov/mortality_events/reporting.jsp

c. Use **USGS Specimen History Form**

d. **Please note**: Fresh-dead specimens should be shipped for overnight delivery on a Monday, Tuesday, or Wednesday to ensure they will arrive during the same work week at the NWHC. If a specimen is shipped on Thursday or Friday, it will not arrive and be unpacked until the following week, well after the packing ice has melted. Therefore, fresh-dead animals collected Thursday-Sunday should be frozen and shipped on Monday. If shipping late in the week is warranted, please contact the NWHC Field Investigation Team to arrange for a special Saturday delivery.

Unless otherwise specified, the Wildlife Health Centers will ship bog turtle specimen remains (postnecropsy) originating from Massachusetts, Connecticut, and New York to the American Museum of Natural History in New York, and will ship specimens originating from Pennsylvania, Maryland, New Jersey, and Delaware to the Smithsonian Institution in Washington, D.C.

2. Carcasses and shells (*i.e.*, the ones that are NOT fresh-dead and have no tissue remaining) should be collected, labeled, and shipped to your state wildlife agency, in accordance with State Scientific Collector's Permit conditions. Complete the **Turtle Data Sheet** and provide copies to the Service's Regional Office (Alison Whitlock, Bog Turtle Recovery Coordinator), the Service point-of-contact for your state, and the appropriate state agency point-of-contact (see list).

3. Live bog turtles (and turtles of other species) found during routine field surveys and/or research activities at (or < 1 km of) bog turtle sites should be carefully examined. Document any abnormalities[3] on bog turtles via close-up photographs and complete the attached **Turtle Data Sheet**. Submit the data sheet and photographs to the Service's Regional Office. If there is a telemetry study occurring at the site where the affected bog turtle was found, we recommend placing a transmitter on the affected turtle(s) and (1) photo-documenting the affected area(s) of the body every 1 to 4 weeks, and (2) collecting relevant health data (*e.g.*, body weight[4], notes regarding any nasal or eye discharges[5]). Similar information should be collected and reported on other turtle species that appear to be diseased at known bog turtle sites. Please note site conditions (e.g., whether the site is flooded and provides limited turtle basking sites).

[3] In this case, "abnormalities" refers to those that appear to be related to disease or infection. However, also note instances of missing toes, whether they are healed over or not. [4] Weighing animals at least once a week is now recommended. A precipitous decline in weight can indicate dehydration and lack of eating. A slow decline in weight would suggest a chronic disease process/infection or lack of eating. [5] Gently put pressure with your index finger on the top of the head and with your thumb under the chin to expel any pus or discharge that may be in the soft palette, evaluate eyelids for puffiness or discharge, look for missing toenails or swollen feet or digits, and check skin for signs of discoloration or blisters.

Illness or mortality observed among amphibians occurring at bog turtle sites should also be investigated. Live, sick (preferably), or fresh-dead amphibian specimens should be collected and packaged for diagnostic evaluation by the USGS National Wildlife Health Center following the NWHC's Amphibian Research & Monitoring Initiative Standard Operating Procedure (ARMI SOP No. 105, see Web site above for instructions and forms, or contact the NWHC Field Investigation Team).

At this time, we are **NOT** recommending that live, affected bog turtles be removed from their wetland habitat unless the state is engaged with a health assessment study with the Wildlife Conservation Society **OR** has another approved state and Federal agency health emergency plan.

If an obviously ill or injured bog turtle is observed in the field, an assessment should be made as to whether the condition of the animal warrants transfer of the animal out of the field for diagnostics and treatment or whether it should remain in the field. If the animal has a radio transmitter, it should remain in the field after taking detailed notes describing the injury/illness (with photographs). State and Service leads should be contacted for consultation with WCS (or other approved veterinarian/organization) regarding follow-up actions. If the animal cannot be tracked, Service and/or state contacts must be reached ***while in the field*** for consultation and approval before the animal may be removed from the site.

If the condition warrants treatment and approvals have been secured, arrangements should be made to transport the animal, in an appropriate carrier with water and vegetation from the site of collection, to the Wildlife Health Center at the WCS/Bronx Zoo. Veterinarians are available at all times by telephoning 718-220-7100 (daytime) and 718-220-5095 (nighttime). If it is determined not to move the animal, attempts should be made to contact a WCS veterinarian at the same phone numbers to arrange for samples to be collected from the animal. This could be a valuable step in determining whether the population is at risk for an infectious disease.

4. Do **NOT** euthanize any bog turtles.

5. Do **NOT** collect tissue samples from live bog turtles without specific Service authorization.

6. To reduce the risk of spreading the agent(s) responsible for the observed mortality and disease, see Attachment B: *Disinfection Protocol for Bog Turtle Field Research, Surveys and Monitoring* (June 2014). Disinfection protocols include routine equipment procedures for **all field visits** ("Level 2") and more involved protocols ("Level 1") for handling of sick or dead animals at sites of known morbidity or mortality.

Field biologists play a vital role in the early detection of incidents of wildlife mortality and disease. The Service extends its sincere thanks for your cooperation in implementing this guidance.

U.S. Fish and Wildlife Service Contacts

Revised 6/24/14

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Revised 6/24/14

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Attachment A - Turtle Specimen Data Sheet (ver 6-30-14)

(For use in reporting sick or dead turtles)

Observation Date _____ **Time** _____ **Biologist** _____

State ___ **Co** ____ **Site ID Name:** _____

Turtle Species: Bog / Spotted / Wood / Painted / Snapping / Box / Musk / Redbelly / Map

Condition: (circle one) Fresh / dead whole-partial / Shell only / Sick-injured

Turtle notch code: Marked / Unmarked Notch code _____

PIT Tag No. _____ New today? Y / N

Sex: Male / Female / U **Age**: Annuli ___ Still growing / Worn

Reproductive Condition: NA Gravid Not Gravid Not Sure

Location of turtle in Microhabitat (circle all that apply):

On tussock / In Rivulet / In deep water / In short vegetation (<1') / In tall vegetation (>1')

Buried in mud / In upland / Other (describe): _____

Behavior (circle all that apply):

Basking / Feeding / Stationary / Swimming / Mating / Escaping / Nesting / With another turtle

Other (describe): _____

Location of turtle in Study Area/Property: Describe and attach map with location and GPS coordinates if possible:_____

Canopy: ____Open _____Part open (%) _____Closed

Habitat Type (Circle): Marsh / Wet Meadow / Flood plain / Forest / Other: _____

Surroundings (circle all that apply): Ag field / fallow field / development / forest / road / other _____

Measurements: Straight-line Carapace Length: _____mm (Maximum length)

Straight-line Plastron Length: _____mm (Maximum length)

Mass: _____g (to nearest 0.5 g)

Tail Complete? Y N **Limbs**: All limbs present and complete: Y N

Carapace/plastron: Diagram and describe below any abnormal scutellation, gnaw marks, old injuries, ectoparasites, swelling, discoloration, open wounds, remaining tissue, etc. Draw feet/legs if abnormal.

Description:_____

Other signs of disease in sick animals (eye/nose/palette discharge, skin discoloration or blisters.

Description: :_____

Follow-up action

Specimen Sent to/by _____ Date: _____

Contacted: State _____FO_____RO_____

Attachment B

U.S. FISH AND WILDLIFE SERVICE

Disinfection Protocol for Bog Turtle Field Research, Surveys and Monitoring

(revised June 24, 2014)

Isolated reports of bog turtle disease and mortality have been reported in several northern range states, including Massachusetts, New Jersey, New York, Delaware, and Pennsylvania. Some disease and mortality rates appear higher than what is typically reported. Reports and collections of sick and dead animals since 2009 have included a variety of symptoms and results. In addition to empty shells, some dead bog turtles have been found entirely intact, with no obvious cause of death. Others have been incomplete specimens due to predation, scavenging, and/or decay. On several live bog turtles, a grayish or whitish substance and/or discoloration has been documented on the skin of the head, neck and limbs, as well as on the claws. In some cases, these appear as skin lesions. Tumors, scute sloughing, and loss of claws, toes, or limbs have also been observed.

At this time, no single causative agent of many observed symptoms has been identified. Test results are often inconclusive due to predation or decay, but some results have shown a variety of potential causative factors including post-injury infection, pneumonia, *Mycoplasma*, and carcinomas. Considering the low reproductive potential of the bog turtle, any elevated levels of mortality or disease are of concern. Therefore, the risk of infectious agents such as *Mycoplasma* and *Ranavirus* poses a threat to bog turtles that the Service is taking seriously. Consequently, the Service is seeking the voluntary cooperation of bog turtle surveyors and researchers in implementing the following guidance.

To minimize the potential for disease transmission while handling and surveying for bog turtles (between surveyor and turtles, between turtles, and between surveyor and environment), these procedures are recommended. The Service advises implementation of equipment disinfection protocols to reduce the risk of unintentional, human-assisted spread of disease. In addition, we recommend that similar guidelines be used any time people handle wildlife to minimize potential disease-related impacts to wildlife and people. ***Please note that individual states/agencies may have additional permitting requirements above and beyond these general procedures.*** These guidelines may be revised upon review of new information.

CAUTION: Although one or more bog turtles within a wetland may have visible symptoms of a disease, it should not be assumed that all individual bog turtles within the wetland are or will be affected. Therefore, care should be taken not to cross-contaminate specimens by lax handling methods. This is especially true if samples are to be submitted for diagnostic purposes.

PROCEDURES

These procedures are tailored to address the relative level of risk of spreading disease between turtles, between occupied bog turtle sites, and between wetlands.

Disinfectant – Throughout this document, "an appropriate disinfectant"[1] refers specifically to a 10% sodium hypochlorite bleach (*i.e.*, household bleach) solution or a 1% Nolvasan solution[2].

Bleach/water solutions should be diluted to 1 part bleach to 9 parts water in a tub or plastic container; a pump-pressurized applicator[3] is effective for spraying equipment with the bleach/water disinfectant. Because bleach breaks down with exposure to air and sunlight, bleach solutions should be discarded if not used up within 5 days after mixing.

We recommend that all field personnel obtain two pump sprayers (one for bleach, one for rinse water) to have out in the field. After returning from the field, equipment and personal gear should be thoroughly washed and disinfected again before being hung in order to dry completely. Do not forget to thoroughly wash your hands, even though you were wearing nitrile gloves.

Note: NEVER mix bleach and ammonia, or any solutions containing bleach or ammonia. Bleach and ammonia combine to produce a poisonous gas.

For more information on disinfecting field equipment, see NEPARC Publication 2014-02 (http://northeastparc.org/products/disinfection_protocol.htm).

LEVEL 1

Level 1 procedures apply at bog turtle sites (wetlands) where <u>any</u> of the following "triggers" have been observed or reported[4]:

- ≥ 2 dead <u>bog turtles</u> in a single year
- ≥ 1 dead <u>bog turtle</u>/year over 2 or more consecutive years
- ≥ 1 <u>bog turtle</u> with missing toes or claws, where the surrounding tissue appears discolored, ulcerated, or unhealthy
- ≥ 2 dead turtles of <u>any species</u> in a single year
- ≥ 1 diseased, sick or apparently unhealthy turtle of <u>any species</u> (this includes any skin lesions or ulcerations, as well as scute sloughing)

If a site exhibits any of the above "triggers," it is considered **suspect** or **potentially suspect.**

[1] All of the referenced disinfectants are toxic to aquatic life. Therefore, do not apply or dispose of disinfecting solution in or near wetlands.

[2] http://www.valleyvet.com/ct_detail.html?pgguid=30e0778a-7b6a-11d5-a192-00b0d0204ae5&ccd=IFF003&utm_source=google&utm_medium=cpc&utm_campaign=F%2BCat%2BLivestock%2BEquip%2BAnd%2BSupp%2B%286000%29%2BPLA&mr:trackingCode=2AF1A264-3C81-E211-BA78-001B21631C34&mr:referralID=NA&mr:adType=pla&mr:ad=15165184603&mr:keyword=&mr:match=&mr:filter=35193115603&gclid=CKqK5Kb71rYCFUOe4AodEiYAPA

[3] Applicators are typically available at hardware or home-improvement stores as 1-gallon or 2-gallon, hand-pump-pressurized spray applicators for herbicides or pesticides, or as pressure washers. When using bleach mixtures, avoid using applicators with metal parts.

[4] With regard to dead turtles, the trigger only applies in cases where the cause of death is unknown. The trigger does not apply at sites where mortality is clearly due to vehicles (*e.g.*, road-killed turtles) or predation. However, a scavenged turtle is not necessarily one that succumbed to predation; it could have died of disease. If an efficient predator is active at a site, there should be signs of this (*e.g.*, multiple turtle carcasses found near a mammal den, tooth marks on carapace). If the cause of death cannot be clearly attributed to predation, consider the site suspect, especially if multiple mortalities are observed in one year or over consecutive years.

If you have been in a Level 1 site, change clothing, footwear, and gear before going into any other wetlands, including wetlands that are, or may be, occupied by bog turtle then decontaminate clothing and equipment as described below.

Submersible Gear

- For **clothing** – Wash all clothing and any appropriate equipment in a washing machine using the hottest cycle possible for material and conventional detergents. Dry using the hottest setting allowable for the material. If possible, use bleach in the wash cycle to further reduce the risk of spreading disease.

- For other submersible gear (*e.g.*, **small traps, notching files**[5]) – First, clean in warm, soapy water to remove organic debris. Use clean water source for rinsing; avoid using potentially contaminated wetland water source, especially if moving gear between sites. Then, submerse equipment in **an appropriate disinfectant** for a minimum of 5 minutes for bleach and 1 minute for Nolvasan, then rinse and air dry.

Non-submersible Gear

- For non-submersible gear (*e.g.*, **large traps, temporary holding buckets, calipers, radio-transmitters**) – First, clean/wipe off organic debris. Then, disinfect any equipment that cannot be submersed by applying **an appropriate disinfectant** to the surfaces. The disinfectant should be in contact with the equipment for a minimum of 5 minutes for bleach and 1 minute for Nolvasan, then rinse and air dry.

- **Survey pole** – In many cases, surveyors use a wooden or metal pole (or tool/broom handle) for conducting Phase 1 and Phase 2 surveys. Scrub and rinse pole to remove all soil and organic material. Then apply **an appropriate disinfectant**; the disinfectant should be in contact with the equipment for a minimum of 5 minutes for bleach and 1 minute for Nolvasan.

- For **boots** – Boots need to be fully scrubbed and rinsed so that all soil and organic material is removed. Entire rubber and leather boots, including soles and leather uppers, can then be disinfected with **an appropriate disinfectant**; the disinfectant should be in contact with the equipment for a minimum of 5 minutes for bleach and 1 minute for Nolvasan, then rinse and air dry.

Equipment should always be rinsed with water to remove any residual bleach/Nolvasan and avoid negatively impacting aquatic organisms.

Hands

In most cases, gloves will hinder field work, so bog turtles will come in contact with bare hands. Apply hand sanitizer with alcohol (*e.g.*, Purell®) after handling each turtle of any species. Make sure it dries completely before handling the next turtle. If turtles (*e.g.*, dead, diseased, or apparently unhealthy) are found on the substrate surface, use disposable exam gloves to handle the specimen. Use a separate pair of gloves for each specimen.

[5] Notching files can also be sprayed with a disinfecting solution and scrubbed with a brush.

Weighing Bags - Bog turtles should be weighed in single-use, disposable plastic bags (*e.g.*, non-ziplock sandwich baggies). Use a separate bag for each turtle of any species. When using a clip-on scale to weigh turtles, apply **an appropriate disinfectant** to the clip (see instructions above) after each use. After applying the disinfectant, rinse and air-dry before placing the clip on the next turtle.

Holding Buckets - If it is necessary to temporarily hold a bog turtle in a bucket (*e.g.*, to allow the epoxy for a radio transmitter to dry), use a separate holding bucket for each bog turtle. First, rinse bucket with clean water to remove any debris (*e.g.*, soil, vegetation). Then, disinfect the bucket with **an appropriate disinfectant** for at least 5 minutes, then rinse and let air dry before placing another bog turtle in it. Do not place multiple turtles in the same bucket at the same time.

Notching Files - Disinfect file between each turtle (see disinfecting instructions above).

Traps - Disinfect all traps between wetland sites (see disinfecting instructions above).

PIT-tagging - If authorized to PIT-tag bog turtles, only use single-use, sterile, disposable syringes.[6]

LEVEL 2

Level 2 procedures apply at <u>all</u> bog turtle sites (wetlands), as well as <u>all</u> <u>wetlands</u> undergoing Phase 1 surveys, Phase 2 surveys, or trapping surveys. As soon as any Level 1 "triggers" are observed or have been reported, Level 1 procedures become effective immediately.

As a precautionary measure, these procedures are also highly recommended at <u>all</u> wetlands to reduce the risk of spreading both disease and invasive plant species. Similar procedures are already being implemented in stream environments pursuant to an advisory by the American Fisheries Society.

Level 2 procedures follow those as described in Level 1 for the following:

- Boots
- Survey poles
- Traps
- Notching files
- PIT-tagging

Field biologists play a vital role in the early detection of incidents of wildlife mortality and disease. The Service extends its sincere thanks for your cooperation in implementing this guidance.

[6] Reusable syringes are <u>not</u> recommended because they cannot be effectively sterilized or disinfected with 70% isopropyl alcohol. Reusable syringes are acceptable when sterilized using a medical-grade autoclave.