

# GUIDELINES FOR COMPREHENSIVE ELEPHANT HEALTH MONITORING PROGRAM

February 2010

Routine health monitoring should be performed on all elephants on an ongoing basis. Animals should be trained to permit sampling and examination. The following protocol advises that specific baseline laboratory tests be performed for the purpose of evaluating current health status. Additional tests are recommended to increase baseline information on other diseases to determine their significance to elephant health. The final decision for specific procedures should be made by the institutional animal care and veterinary staff based on individual circumstances. For additional information, refer to the Elephant Husbandry Manual, AZA Standards for Elephant Management and Care, and the AAZV Preventive Medicine Recommendations. Additionally, it is recommended that the veterinarian review the behavioral profile of the individual animals with elephant management staff on a regular basis.

Minimum Database:

1. Signalment - age, sex, origin, studbook#, ISIS#; picture of individual (as viewed from the front and sides) should be included in the permanent record.
2. Anamnesis - summary of information regarding health screens, medical problems, diagnostic test results, and treatment over the previous year (complete "Individual Annual Elephant Medical Survey" form and send to SSP Veterinary Advisor – will be available in 2010).
3. Complete physical exam by a veterinarian familiar with elephant health problems. This should include a review of all systems.
4. Body weight – actual weight should be recorded at least annually.
5. Blood collection
  - a. Complete blood count (CBC), serum chemistry panel, fibrinogen, serum protein electrophoresis
  - b. Serologic (ELISA) test for elephant herpesvirus – contact Erin Latimer to submit samples, or Dr. Laura Richman (1, 2).
  - c. Bank a minimum of 10-20 ml serum (duplicate sample for SSP serum bank) – all banked samples should be labeled with species, studbook #, age, sex, and date collected. Use submission form for serum samples sent to the SSP serum bank (maintained at Palm Beach Zoo).
6. Serum progesterone analysis in females – Serial samples should be collected weekly on an ongoing basis to evaluate estrous cycles (3). Assays can be performed at the National Zoo's Conservation & Research Center. Contact Dr. Janine Brown - CRC (National Zoo), for submission instructions (see protocol below).
7. Fecal analyses
  - a. Parasite screen - Fecal samples should be collected at least semiannually; direct, flotation, and sedimentation should be performed on every sample to detect intestinal parasitism.
  - b. Enteric pathogen screen - Aerobic culture of feces for enteric pathogens should include special media for the detection of *Salmonella spp.* Because *Salmonella* organisms may be shed intermittently, at least 3-5 fecal cultures should be performed (may be done on consecutive days) on an annual basis.

8. TB cultures and serological testing - refer to the current USDA Guidelines for the Control of Tuberculosis in Elephants 2008 (4). Protocol can be accessed on the USDA website [http://www.aphis.usda.gov/animal\\_welfare/publications\\_and\\_reports.shtml](http://www.aphis.usda.gov/animal_welfare/publications_and_reports.shtml). Note: new guidelines are planned to be implemented in 2010. Initial official testing will be done in the presence of an USDA inspector.
- Samples for cultures must be collected under the direct supervision of a licensed veterinarian.
  - Three trunk wash samples should be collected on separate days, ideally within a 7 day period. Trunk swabs are no longer acceptable.
  - All samples should be frozen immediately after collection and shipped frozen.
  - Ship by overnight express to NVSL (or other laboratory facility offering comparable procedures). Request mycobacterial culture with speciation (use VS Form 10-4 submission form for NVSL). Serum/plasma sample for serological testing (Elephant TB STAT-PAK®) to be collected during same period as trunk wash cultures.

#### 9. Vaccinations

- Tetanus toxoid \*- current vaccination (within 12 months) with a commercial equine product is recommended. Follow label instructions for product use (usually 1 ml IM). Data are insufficient at this time to determine adequate protective vaccine doses and titers.
- Rabies vaccine\* - current vaccination (within 12 months) with a commercial killed rabies product approved for horses should be considered if the animal resides or will be traveling to an endemic area. Follow label instructions for product use (usually 2 ml IM). Vaccination with Imrab 3 has induced detectable titers to rabies virus in African elephants (5). Biannual vaccination is recommended. Data are insufficient at this time to determine adequate protective vaccine doses and titers.
- Leptospirosis vaccine\* - recent cases of Leptospirosis in elephants have been documented. Although insufficient information is available at this time, current vaccination (within 12 months) with a commercial killed multivalent vaccine product approved for domestic large animals should be considered if there is evidence of potential for exposure. Follow label instructions for product use.
- West Nile Virus vaccine - insufficient information is available at the time of this writing to recommend vaccination of elephants with the commercial equine WNV vaccine. Some institutions have elected to vaccinate following recommendations for horses. If the receiving institution requests that the elephant is vaccinated, pre- and post-vaccination titers should be measured and submitted to Cornell (contact the SSP Veterinary Advisor for more information).

\* Tetanus, rabies, and leptospirosis have been reported to occur in elephants (6,7,, M. Miller, pers. commun.).

#### Additional Preventive Health Recommendations:

1. Serological screening for EMC (encephalomyocarditis virus), leptospirosis (multiple serovars), and WNV (West Nile Virus). Although these tests are not species-specific and have not been validated for elephants, they may detect cross-reactive antibodies in exposed animals. The presence of antibodies does not necessarily denote infection/disease. Encephalomyocarditis virus may cause clinical disease and death in elephants (8). Antibodies to leptospirosis have been detected in both Asian and African elephants (9, 10). At the time of this writing, EMC serologic testing was not available. Insufficient data is available at this time to determine the significance of WNV antibodies in elephants; it is important to include the history of exposure and vaccination to WNV when interpreting results.
2. Serum vitamin E levels – submit heparinized plasma to Michigan State University.
3. Reproductive tract examination – whenever feasible, a complete reproductive examination should be conducted which includes transrectal ultrasound, semen collection and analysis, serum collection (weekly best, or bi-weekly to monthly) for testosterone analysis, cytology and microbial cultures of the lower urogenital tract (to be screened for bacteria, Chlamydia, protozoa, and Herpesvirus). Herpesvirus has been identified in biopsies of vaginal lymphoid patches in an African elephant (2). A high prevalence of uterine leiomyomas has been observed in captive Asian elephants and could be detected via transrectal ultrasound (11). Since both of these conditions have potentially significant effects on reproduction, a careful evaluation is warranted if the animal is being considered for breeding. All elephants (male and female) over the age of 5 years should have both ultrasound and hormonal (testosterone in males; progesterone/LH in females) assessments performed.
4. Urinalysis – fluid and sediment evaluation of clean voided sample; +/- microbial culture.
5. Foot radiographs – baseline radiographs of all feet are strongly recommended; see Gage for description of technique (12). It may be appropriate to annually monitor selected elephants (i.e., those that have a history of chronic foot problems).
6. Ancillary diagnostic tests for tuberculosis –see Guidelines for the Control of Tuberculosis in Elephants 2008 for current recommendations (4).
7. Other vaccination regimens will depend on regional requirements and exposure risks (consider vaccination for equine encephalitis viruses, Clostridial diseases, Leptospirosis). Insufficient information is available at this time to provide a recommendation for West Nile Virus vaccination of elephants. Contact the SSP veterinary advisor for current information.

**Elephant Serum Bank Submission Form**

Institution/owner: \_\_\_\_\_

Submitter: \_\_\_\_\_

Address: \_\_\_\_\_

Tel: \_\_\_\_\_ Fax: \_\_\_\_\_ Email: \_\_\_\_\_

**Animal Information**

Asian  African  ISIS# \_\_\_\_\_ Studbook # \_\_\_\_\_

Name \_\_\_\_\_ Age: \_\_\_\_\_  actual  estimate

Sex:  male  female

**SAMPLE COLLECTION INFORMATION**

Date of sample collection: \_\_\_\_\_ Time of collection : \_\_\_\_\_

Site of sample collection:  ear vein  leg vein  other: \_\_\_\_\_

Health status of animal:  normal  abnormal

Fasted:  no  yes – how long \_\_\_\_\_

Weight \_\_\_\_\_  actual  estimated

Type of restraint:  manual  anesthetized/sedated  behavioral control

Temperament of animal:  calm  active  excited

Type of blood collection tube:

no anticoagulant (red-top)

EDTA (purple)

heparin (green)

other: \_\_\_\_\_

Sample handling:  separation of plasma/serum by centrifugation

(check all that apply)  stored as whole blood

frozen plasma/serum

other – describe \_\_\_\_\_

**TB EXPOSURE STATUS**

Known infected animal

Known exposure to culture positive source within the past 12 months

Known exposure to a culture positive source within the past 1-5 years

No know exposure to a culture positive source in the last 5 years

**TREATMENT INFORMATION**

Is elephant currently receiving any medication or under treatment?  yes  no

If yes, please list drugs and doses: \_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

Time between blood collection and last treatment: \_\_\_\_\_

Ship samples overnight frozen with shipping box marked "PLACE IN FREEZER UPON ARRIVAL"

**Send completed form with samples to:**

Dr. Michele Miller

Palm Beach Zoo, 1301 Summit Blvd, West Palm Beach, FL 33405

W: (561) 833-7130 ext 224; Cell: (561) 727-9630; Fax: (561) 833-7135; email: mmiller@palmbeachzoo.org

## **Instructions for Elephant Endotheliotropic Herpesvirus (EEHV) Sample Submission**

Please submit all samples on plenty of dry ice in a sealed styrofoam container.

\*\*Please send at least 1-2 ml of whole blood (EDTA, lavender top tube). It is best to transfer the blood to a freezer-safe tube (not glass) after thoroughly mixing with EDTA. We have received broken glass tubes in the past.

\*\*Please send 3 serum tubes. Freeze ASAP.

\*\*Please collect the following tissues from deceased animals and place in labeled 50 ml tubes/Whirlpak bags/Ziploc bags. Freeze ASAP.

- hilar and mandibular lymph nodes
- heart
- liver
- spleen
- hidden lung lymphoid nodules
- skin or vestibular nodules

**FedEx to:**

**Erin Latimer/Laura Richman  
Smithsonian, National Zoological Park  
Department of Pathology  
3001 Connecticut Ave. NW  
Washington, DC 20008  
202-633-4252  
703-855-9611**

12/09

# Request for EEHV testing

Department of Pathology  
Smithsonian National Zoo  
3001 Connecticut Ave. NW  
Washington, DC 20008  
(202) 633-4252

Date \_\_\_\_\_

Requestor's name \_\_\_\_\_

Institution \_\_\_\_\_

Address \_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

Phone/email \_\_\_\_\_

Animal Name/ISIS # \_\_\_\_\_

Age \_\_\_\_\_

Species \_\_\_\_\_

Reason for request (i.e. current symptoms of possible EEHV, or possible exposure to

EEHV) \_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

Type of samples being sent \_\_\_\_\_

Before sending samples, please call Erin Latimer 202-633-4252 (W)

703-855-9611 (H)

Please send samples **overnight on dry ice in a sealed styrofoam container**. The best day to send samples is Wednesday, for Thursday delivery.

# SERUM COLLECTION PROTOCOL FOR MONITORING REPRODUCTIVE ACTIVITY IN ELEPHANTS

**Janine L. Brown, PhD**

Elephant SSP Reproductive Advisor  
Endocrine Research Laboratory

## Females

- Blood samples should be collected once weekly to establish if females exhibit normal ovarian cycles (e.g., estrous cyclicity). **Note:** the Elephant SSP requires that females between 8 and 35 years of age be hormonally assessed, and recommends that all elephants be monitored throughout their lifespan.

## Males

- Blood samples should be collected weekly (bi-weekly or monthly if weekly is not possible) to assess testicular steroidogenic activity. **Note:** the Elephant SSP recommends that elephant bulls be monitored throughout their adult lifespan.

## Sample collection and shipment

- For best results, allow blood to clot for ~1 hour at room temperature, or for ~2 hours at refrigerator temperature. Avoid exposing blood to ambient temperatures for longer than 3 hours (blood cells can metabolize progesterone and affect results). Plasma (collected in either EDTA or heparinized tubes) can also be analyzed, although serum is preferred.
- Centrifuge blood (~1000 x g for 10-15 min) and decant serum into a polypropylene vial (best sizes, 12 x 75 mm or 12 x 55 mm) with a tight-fitting cap that pushes or screws on and is flush with the tube (i.e., cap doesn't hang over the side of the tube). We recommend tubes with a frosted writing space and caps from the Sarstedt company (1-800-257-5101). The catalogue number for the tubes is 62.526.003 PP, and for the push caps is 65.809. Smaller tubes are okay, but do not use ones that exceed a 5-ml capacity. Information on the tube should include: animal name, date (mo/day/year), and facility name or abbreviation. Please provide a minimum 1 ml of serum for each sample. Store frozen (-20°C or colder).
- Ship samples in a styrofoam container with dry ice or cold packs. We will return box and any shipping materials. Use an overnight express courier (e.g., Airborne Express or FedEx) and ship only on Mon, Tues or Wed. Never ship on Fri or before a government holiday.
- Include in the shipment a written request as to what hormone analyses are required. If you need results immediately (emergencies only, please), let us know in the paperwork you provide. If you will be requesting Prolactin or LH (or any other protein hormone), call a week in advance so that we can schedule an iodination.
- Address samples to: Nicole Abbondanza, Conservation and Research Center, 1500 Remount Rd., Front Royal, VA 22630. Please notify us when samples are shipped (540-635-6521, ext. 225; [abbondanzan@crc.si.edu](mailto:abbondanzan@crc.si.edu)).
- We will issue an invoice at the time data are sent. Please make checks payable to: Conservation & Research Center Foundation, c/o Janine Brown.
- For our records, please provide the studbook number, name and age of your elephant(s). If you have any questions, please contact Dr. Janine Brown: phone (540) 635-6586, fax (540) 635-6506, email [jbrown@crc.si.edu](mailto:jbrown@crc.si.edu). Lab hours are 8:00 am-5:00 pm est.

## REFERENCES

1. Richman, L.K., R.J. Montali, R.C. Cambre, D. Schmitt, D. Hardy, T. Hildebrandt, R.G. Bengis, F. M. Hamzeh, A. Shahkolahi, and G.S. Hayward. 2000. Clinical and Pathological Findings of a Newly Recognized Disease of Elephants Caused by Endotheliotropic Herpesviruses. *J. Wildl. Dis.* 36:1-12.
2. Zong, J.C., E. Latimer, S.Y. Heaggans, L.K. Richman, and G.S. Hayward. \_\_\_\_\_. Pathogenesis and Molecular epidemiology of Fatal Elephant Endotheliotropic Disease Associated with the Expanding Proboscivirus Genus of the Betaherpesvirinae. \_\_\_\_\_
3. Brown, J.L. 2000. Reproductive endocrine monitoring of elephants: An essential tool for assisting captive management. Special Issue on Elephant Biology. *Zoo Biol.* 19:347-368.
4. Guidelines for the Control of Tuberculosis in Elephants 2008.  
Available on the Internet: [http://www.aphis.usda.gov/animal\\_welfare/publications\\_and\\_reports.shtml](http://www.aphis.usda.gov/animal_welfare/publications_and_reports.shtml)
5. Miller, M., and F. Olea-Popelka. 2009. Serum Antibody Titers Following Routine Vaccination in African Elephants. *J. Am. Vet. Med. Assoc.* 235:978-981.
6. Burke, T.J. 1975. Probable Tetanus in an Asian Elephant. *J. Zoo Anim. Med.* 6:22-24.
7. Wimalaratne, O. and D.S. Kodikara. 1999. First Reported Case of Elephant Rabies in Sri Lanka. *Vet. Rec.* 144:98.
8. Gaskin, J.M., Jorge, M.A., Simpson, C.F., Lewis, A.L., Olson, J.H., Schobert, E.E., Wollenman, E.P., Marlowe, C., et al. 1980. The Tragedy of Encephalomyocarditis Virus Infection in Zoological Parks of Florida. *Proc. Am. Assoc. Zoo Vet.* 1-7.
9. Karesh, W.B., K.H. Smith, F. Smith, M. Atalia, P. Morkel, A. Torres, C. House, W.E. Braselton, and E.S. Dierenfeld. 1997. Elephants, Buffalo, Kob, and Rhinoceros: Immobilization, Telemetry and Health Evaluations. *Proc. Am. Assoc. Zoo Vet.* 296-300.
10. Bhatt, M.N. and R. Manickam. 1998. Detection of Leptospiral Antibodies in the Sera of Elephants. *Indian Vet. J.* 75:201-203.
11. Montali, R.J., T. Hildebrandt, F. Goritz, R. Hermes, K. Porter, and J. Tsibris. 1998. High Prevalence of Uterine Leiomyomas in Captive Asian Elephants and their Implications for Reproductive Soundness. *Proc. Third International Elephant Research Symposium*.
12. Gage, L.J. Radiographic Techniques for the Elephant Foot and Carpus. 1999. In: *Zoo & Wild Animal Medicine Current Therapy 4*, eds. Fowler, M.E. and R.E. Miller. W.B. Saunders, Philadelphia, Pennsylvania. Pp. 517-520.

## APPENDIX

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For Vit E analysis, contact:  
Michigan State University  
Animal Health Diagnostic Laboratory  
Lansing, MI 48909

For Leptospiral and WNV titers, contact:  
Cornell University  
College of Veterinary Medicine  
Diagnostic Lab  
Upper Tower Rd.  
Ithaca, NY 14853  
(607) 253-3900

For EMC titers, contact:  
Texas Veterinary Diagnostic Lab.  
College Station, TX 77841  
(979) 845-3414

February 2010 mm