



August 24, 2006

Contact Person:
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*"Nurturing
wildlife
and enriching
the human
experience"*

Honorable City Council
c/o City Clerk
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Los Angeles, California 90012

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California 90027
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Subject: **NECROPSY REPORT ON ASIAN ELEPHANT**

On June 21, 2006, a Motion (LaBonge-Rosendahl) was introduced directing the Zoo Department to report immediately to the Council with the results of the necropsy on the Asian elephant, "Gita", who passed away on June 10, 2006 (C.F. 06-1448)

Antonio R. Villaraigosa
Mayor

In response to this Motion, the Zoo Department hereby transmits the final necropsy report (attached in its entirety) received from the California Animal Health & Food Safety (CAHFS) on August 23, 2006. CAHFS is a part of the School of Veterinary Medicine at the University of California, Davis. They have five regional labs throughout California, and the Zoo used the facility in San Bernardino, California. Each regional lab has a staff of veterinary pathologists and technicians that perform the work on-site.

Tom LaBonge
Council Member
4th District

Zoo Commissioners

Kimberly Marteau
President

Karen B. Winnick
Vice President

The following provides a summary of the report, other findings, and our conclusions relative to Gita's condition.

Shelby Kaplan Sloan

SUMMARY

Mark D. Persaud

Bernardo Silva

Richard Lichtenstein
ex officio member

As you know, Gita passed away on the morning of June 10, 2006 and was immediately transported to CAHFS for a full and complete necropsy. When an animal as large as an elephant passes away, the necropsy is more time consuming than other animals due to the sheer size of the animal. CAHFS carefully worked on this necropsy for over two months, obtaining specific biological specimens such as tissues and cultures when indicated. The tissues were then examined histopathologically under the microscope to see if a determination on her collapse and cause of death could be made.

John R. Lewis
Zoo Director

The initial findings illustrate the comprehensive nature of this necropsy. Thirty-eight (38) findings during the gross, or initial examination, were identified and examined further in the laboratory to determine which were specific and relevant to her collapse and death versus those that were due to her age and life events. The complete analysis indicates that Gita died of cardiac failure associated with thrombi, or blood clots, blocking the right chambers and major vessels of her heart. The cause of the clots was a systemic coagulation disorder which began three to five days before her death. The cause of this coagulation disorder could not be determined.



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Unfortunately the necropsy can not explain definitively why Gita collapsed although it may have been caused by generalized body weakness related to the blood clots in her system. The necropsy did, however, explain why she couldn't get up. After falling, Gita's weight damaged her rear legs causing a problem called "compartment syndrome". The compartment syndrome referenced in the report basically means that her rear legs were no longer functioning so she could lift herself. This inability to use her rear legs was observed by Staff on June 10th as she was unable to support herself on her rear legs even after she was lifted with a crane. While the compartment syndrome prevented Gita from getting up it is not associated with her cause of death.

As you know, there was a delayed response time on Friday, June 9th, when Gita was initially observed in a sitting down position. It is not possible from the results of the necropsy to determine whether the time lag contributed to her death. However, as I have previously reported, I have taken corrective actions to see that such a delay does not occur in the future if a similar event should happen with any animal residing here.

OTHER FINDINGS

There were a few other findings of note which I would like to point out.

- There was no evidence of a systemic infection in Gita.
- There was no explanation of the cause of the abscesses on her tail and hip.
- The surgical and medical treatment to arrest and cure the osteomyelitis in her left front foot was successful.
- She had some kidney disease which may have been age related.
- In addition to the joint disease in her feet and legs, Gita also had arthritis in her spine which would have added to her difficulty in lying down and getting up.
- Gita was free of tuberculosis which is an important finding in captive elephants.
- Gita was in a good nutritional state with adequate fat reserves.

ZOO CONCLUSIONS

The Los Angeles Zoo and Botanical Gardens provides educational and recreational opportunities for the citizens of Los Angeles. In addition to the visitor experience, the scientific aspect of our operation is equally as vital. The Zoo is constantly learning about its animals through research projects, births, husbandry techniques and in death. From the necropsy finding the Zoo concludes:

- There were no clear histopathological explanations for the sequence of events that led to her death;
- Foreknowledge of these findings would not have changed the course of Gita's care and treatment;
- The Veterinary and Elephant Care Staff will discuss these findings and consider whether there were any signs prior to her death that would have indicated a weakening state due to the presence of blood clots; and
- The Los Angeles Zoo & Botanical Gardens has proven that surgery and aggressive long term treatment can be a viable solution for digital osteomyelitis in elephants. This information will be shared with other zoo and wildlife professionals through professional meetings and publications.

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In closing let me add that Gita was a special animal and no time or expense was spared in treating or caring for her, particularly in the past year. During that time, many people went above and beyond their normal duties to care for her.

I am available to discuss this report with you in further detail at your earliest convenience, and can be reached at (323) 644-4261.

Sincerely,

A handwritten signature in black ink, appearing to read "John R. Lewis". The signature is fluid and cursive, with a long horizontal stroke extending to the right.

JOHN R. LEWIS, General Manager
Zoo Department

JRL:DMV/dmt

Attachment

(This report supersedes all previous reports for this accession)

* Emailed Copy. *
* A signed original is on file. *

California Animal Health & Food Safety
Laboratory System (CAHFS) - San Bernardino
P.O. Box 5579
San Bernardino, CA 92412
(909) 383-4287

ACCESSION#:S0607485
District: 6
County: LOS ANGELES
Case Coordinator: DREAD

Submitter
STEPHEN KLAUSE DVM
LOS ANGELES ZOO HLTH CTR
5333 ZOO DR

Owner:
LOS ANGELES ZOO
5333 ZOO DRIVE

LOS ANGELES, CA 90027

LOS ANGELES, CA 90027
(323) 644-6018

Agent or Collector:
Reference Number: AE600000SM

Species: OTHER MAMMAL
Herd/Flock ID: 2006-0039
Date Taken: 06/10/06
Date Received: 06/10/06

1 Specimens submitted: CARCASS OF ASIAN ELEPHANT

Approved by: Deryck Read, BVSc, PhD, Dip.

,ACVP

L A B O R A T O R Y F I N D I N G S / D I A G N O S I S

Adult Asian female elephant submitted with a long-term (from Sept, 2005) history of surgical removal of an osteomyelitic 5th toe of left forefoot; a short-term (May-June, 2006) history of tail-head abscess and right hip seroma; and an acute history (overnight) of "dog sitting" recumbency, collapse and death.

Findings

1. Extensive, necro-suppurative, chronic-active, severe, septic, emphysematous, cavitatory, cellulitis-myositis of dorso-caudal sacral ("tail-head") area with:
-extensive peripheral and ventral edema
2. Extensive, subacute, mildly infected fibrinohemorrhagic seroma of right hip area, incidental.
3. Surgically exposed corium of sole of right forefoot, adjacent to toe of digit 4, incidental.
4. Chronic fibrosis, edema and hyperemia of R.axillary and L.inguinal lymph nodes...histologically mild hyperplastic lymphadenitis, incidental.
5. Bilateral polycystic ovaries and cystic hyperplasia of endometrium.
6. Hepatic and splenic pigmentation, incidental. (See finding #38).
7. Acute extensive occlusive thrombosis of right chambers of heart and

- associated great vessels (aorta, vena cava, pulmonary arteries and veins)
8. Cytologic and histopathologic findings in joint synovial membranes compatible with Degenerative Joint Disease.
 9. No bacteria detected in liver and kidney by aerobic culture.
 10. Insignificant growth or no growth of bacteria in lymph nodes and joint fluids by aerobic culture.
 11. Streptococcus dysgalactiae subsp. equisimilis isolated from tail-head and R.hip lesions.
 12. No salmonella detected in liver by culture.
 13. No acid fast bacilli detected in uterine lesions by ZN-stained smears.
 14. No evidence of heavy metal toxicosis or trace mineral deficiency.
 15. No Clostridium chauveii, novyi, septicum or sordelli detected by FA on smears from tail-head lesion.
 16. Very severe degenerative joint disease of right radioulnar-carpal joint.
 17. Severe to moderate degenerative joint disease of: left and right intercarpal; carpometacarpal; metacarpophalangeal; and stifle joints
 18. Mild degenerative joint disease of: left and right shoulder and elbow joints; left radioulnar-carpal joint; left and right hip, tibiotarsal and tarsometatarsal joints, incidental.
 19. Acute deep vein thrombosis of all 4 legs and vertebral canal (C3-L3).
 20. Focal proliferative vertebral osteodermatitis with focal, non-compressive stenosis of vertebral canal at T17:18, L1:2 and L2:3; opposing spinal cord at T17:18 is normal microscopically, incidental.
 21. Chronic-active sole ulcers adjacent to toes of digit 2 and 5 of right foreleg (additional to finding #3 above), incidental.
 22. Diffuse hyperkeratosis and hyperemia of mucosa of cheeks and tongue, with rare acute ulceration, incidental.
 23. Abnormal tooth wear in left maxillary arcade with caries, incidental.
 24. Missing caudal molar teeth of left maxillary and right mandibular arcades, incidental.
 25. Multiple calcinosis circumscripta lesions in lateral subcutis of left hindleg, incidental.
 26. Multiple leiomyomas of wall of uterus, incidental.
 27. Chronic membranoglomerulonephritis with proteinuria, probable cause of hypoalbuminemia reported in history.
 28. Replacement of phalanx 3,2 and distal 4/5 of phalanx 1 of digit 5 of left front foot by healthy reparative fibro-osseous tissue with:
 - a) 1.5cm long, 2-3mm diameter, chronic-inactive sinus tract, perpendicular to skin surface and opposite the surgical site, measured from subcutis surface.
 - b) chronic, resolved, inactive, arthritis of metacarpophalangeal joint 5.
 - c) multiple, microscopic spicules of necrotic bone sequestra, some within small sterile abscesses admixed with areas of healthy new bone (osseous metaplasia).
 - d) small single subchondral sterile bone cyst involving distal metacarpal 5
 29. Fibrosis and calcinosis circumscripta of digital cushion of left hind foot, incidental
 30. Chronic septic sole ulcer at toe of digit 4 of left hind foot, incidental.
 31. Mixed heavy growth of anaerobic bacteria isolated from tail-head lesion.
 32. Acute, locally-extensive, ischemic-type necrosis of skeletal muscle, popliteal region, LH leg.
 33. Subacute, locally-extensive, mild necrosis of skeletal muscle,

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- popliteal region, RH leg, incidental.
34. Acute to subacute (~3 days) thrombosis of blood sinuses in an a small incidental uterine anomalous mass, non-septic.
 35. Chronic, solitary, microscopic, submucosal cyst in esophagus, incidental.
 36. Chronic, multiple, mineralized microscopic foci involving pituitary gland, incidental.
 37. Mild cystic glandular hyperplasia of endometrium, incidental.
 38. Diffuse hemosiderosis and/or lipofuscinosis of spleen, liver, adrenal cortex and myocardium, incidental.

For summary of major pathologic findings and their clinical correlations, please see Accession Summary on page 4.

A C C E S S I O N S U M M A R Y

06-12-06: In view of the duration and type of recumbency and the size of the animal, careful rule in/out of compartment syndrome of rear legs will be performed.

Since the tail head cellulitis-myositis smelt "anaerobic", smears made at the time of necropsy will be evaluated for clostridial involvement by FA, culture and histopathology.

If you wish to be present during dissection of the RF and LF foot let me know. I plan to begin these after brain and cord removal-probably this Thursday/Friday.

Cultures of body organs are in process in order to attempt rule in/out of septicemia.

06-20-06: Septicemia is ruled out by the negative aerobic cultures of liver and kidney. Immediate cause of death was occlusive thrombosis of the right chambers of the heart, as well as major veins and arteries to and from heart. Determination of the presence or the absence of disseminated micro-thrombosis (DIC) is pending additional histopathology. Thrombogenesis may have been triggered by the extensive synovial tissue damage in joints or bacterial toxins from the cellulitis lesions. Cause of the severe lameness in RF could have been due to the very severe (rough bone on rough bone) DJD of radioulnar-carpal joint. Cause of limb paresis may have been due to the deep vein thrombosis in the legs as well as the occlusive thrombosis of the right heart and aorta. E-images are on file.

06-27-06:

The gross appearance of the surgical site in the LF (foot from which most of digit 5 had been surgically removed) indicated that that the reparative tissue consisted of mature, healthy granulation tissue containing islands of healthy bone. Histopathology of this tissue will confirm/refute this gross assessment. The bone within the fibrous tissue may be new bone (from osseous metaplasia) or pre-existing bone fragments trapped in the repair process. Again, histopathology should be able to distinguish between these 2 outcomes. No pus pockets or bone sequestra were seen (pending further dissection after decalcification of 3 to 4 weeks).

The metacarpo-phalangeal joint of digit 5 of the LF foot was involved by a chronic ulcerative arthritis which appeared resolved and non-active (pending histopathology).

No gross lesions were found in the RF foot to explain the acute onset of lameness in this foot ~3 days before death.

An infected sole ulcer involved the LH foot adjacent to the toe digit 4. This was a subtle slit on the surface of the sole but, deeper in it was an active developing abscess in the supradjacent corium.

No mass lesion was found in the digital cushion of the RF foot that would correspond to the large oval dense mass seen in the CT scan (histopathology pending).

Several nodular gritty-white and boney lesions involved the digital cushion of the LH foot (?calcinosis circumscripta).

08-18-06: SUMMARY OF MAJOR PATHOLOGIC FINDINGS (from chronic to acute):

1) Extensive, chronic-active, severe to very severe Degenerative Joint Disease involving right radioulnar-carpal joint (very severe); and right and left intercarpal, carpometacarpal, metacarpophalangeal and stifle joints (severe).

2) Extensive, necro-suppurative, chronic-active, severe, septic (mixed-bacterial, non-specific), cavitatory cellulitis-myositis of "tail-head" area.

3) Subacute (approximately 3 days old) thrombosis of blood sinuses in an (incidental) uterine anomaly, non-septic. (This finding provides important evidence of the time-course of the coagulation disorder).

4) Extensive acute (hours old), severe, occlusive thrombosis of right chambers of heart, including major in-flow and out-flow tracts (e.g vena cava, aorta, pulmonary artery and vein) and of distant blood vessels, namely, vertebral venous sinuses, extra-dural sinuses of cranial vault, deep veins caudal to both elbows and both stifles, and digital veins and arteries, non-septic.

5) Locally-extensive, acute (hours old), ischemic-type necrosis of musculature deep in the left hind leg caudal to stifle joint (popliteal region).

6) No evidence of bacteremia.

7) Healthy resolution of surgical site in left fore foot.

8) Chronic bilateral membranoglomerulonephritis with secondary proteinuria; weeks to months old; non-specific pathogenesis, probably immune-complex in type.

CORRELATION OF MAJOR PATHOLOGIC FINDINGS WITH CLINICAL HISTORY:

1) The reason why Gita "went down" initially was not determined but it may have been caused by generalized body weakness related to the systemic

coagulation disorder which began approximately 3 days before death.

2) The reason why Gita could not "get up" on the Saturday morning was because of deep vein thrombosis of the legs, particularly the left hind leg which had secondary acute ischemic-type necrosis of musculature deep in the thigh behind the stifle ("Compartment Syndrome").

3) A major lethal event contributing to Gita's death was acute (hours old) occlusion of the right chambers of the heart by massive thrombi.

4) The nature of the "arthritis" was typical of "Degenerative Joint Disease", a chronic (months to years old), non-infectious disorder commonly found in zoo elephants.

5) The cause of the coagulation disorder was not determined.

6) The cause of the hypoalbuminemia was probably protein loss from the inflamed damaged renal glomeruli.

E-images of necropsy and gross lesions on file. These will be sent to you on CD via US post.

G R O S S P A T H O L O G Y

Necropsy findings: Necropsy of an adult female Asian elephant began at 3:00 p.m., June 10, 2006. ID verified by Dr. D. Read June 10, 2006.

External examination:

Skin of dorsal midline caudosacral region ("tail head") had an ovoid area (~15x20cm) of erythema and alopecia. Associated region was moderately swollen. Within this region and ~5cm cranial to the coccygeal-sacral articulation, skin had a transverse, 20cm long opening (compatible with a surgical lance performed ~8 days ago according to history). The opening contained several, irregularly-shaped, protruding masses of grey malodorous tissue. Cut surfaces revealed red-purple-grey mottled, firm tissue (compatible with inflamed granulation tissue). The tissue was covered by grey necrotic suppurative surfaces. The opening was continuous with an extensive cavity (from the level of the anus to the level of the hips (~30cm) and from the midline to ~20cm left lateral of midline. The cavity extended (at its deepest part) ~10cm deep (to the skin) where it involved superficial musculature. The cavity was lined by irregular necrotic tissue similar to that described at the opening. Approximately 10-15cm inside the cavity surfaces were covered by copious yellow pus. Several large masses (3x4x5cm) within the cavity were loose. The cavity was highly malodorous compatible with a heavy mixed aerobic-anaerobic bacterial infection. Subcutaneous connective tissue and fat peripheral to the cavity was involved by yellow gelatinous edema. The edema extended ventrally to involve the perineum, the L. and R. flank and L. and R. caudal thighs.

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Skin in the region of R.hip had a ~10cm long straight slit-like opening (compatible with a surgical lance performed a few days previously according to history). The opening oozed a little serous fluid. The opening was continuous with extensive subcutaneous underrunning (~20cm dia.). Surfaces of the underrun tissue were mottled grey and dark red, shiny, fibrinous and non-malodorous (compatible with early, minimally inflamed granulation tissue).

Sole of right forefoot had a ~2cm dia. opening ~3cm volar to digit 4 (compatible with surgical removal of sole corn ~3 days previously according to history). The corium was exposed. The opening oozed a little serous fluid.

Internal examination:

Carcass was in good nutritional condition and in a fresh state of post mortem preservation. Fat reserves were adequate. Liver parenchyma was dark brown and had an accentuated lobular pattern. Adrenal cortices were pale yellow-brown.

Lumens of right chambers of heart, distal posterior and anterior vena cava, pulmonary arteries and veins and proximal aorta (to mid-thoracic level) were occluded by terminal thrombi. L. and R. ovary were polycystic (L=9 cysts, 2 0-45cm dia; 2/9 empty; 2 semisolid pedunculated ovoid bodies. R=7 cysts; 4/7, 8-11cm diam.; 3/7, 2-3.5cm dia., 1 solid ovoid pedunculated body). R.axillary lymph node was enlarged (5x3x3cm), oozed yellow gelatinous fluid and on cut surface, had multifocal, small (2-3mm dia.) grey foci surrounded by a red line (hyperemia). R. L. inguinal lymph node had similar lesions.

Additional necropsy findings to follow as dissections are completed over the next 1-2 weeks.

06-20-06: ADDENDUM

On 6-13-06, the brain was removed intact (by use of chain saw) dissected, sampled and immersed in NBF. Spinal column was removed from carcass and divided into ~40cm long segments (by chain saw). No gross lesions were seen in brain, cranium or cranial vault. Internal iliac lymph nodes were not found. Lumens of terminal aorta and origins of femoral arteries had no occlusive (saddle) thrombus. Uterus had multiple, variably-sized (0.5-2.0cm dia.) yellow caseous or fleshy grey spherical masses in wall; mucosa had numerous small (~0.5cm) cysts in endometrium.

On 6-14-06, left-foreleg, including shoulder, elbow, radiocarpal, and inter-carpal joints were examined. Gross lesions of degenerative joint disease (DJD) involved these joints in varying severity: mild in shoulder, elbow,

radiocarpal and moderate in inter-carpal. Common DJD gross lesions were: diffuse, pale, yellow, soft fibrillated articular cartilage; occasional, small (2-5mm dia.) articular ulcers with dark red beds; extensive periarticular spur/shelf-like exostoses (5-10mm long); excess blood-tinged synovial fluid; sometimes containing small blood clots; and red villonodular proliferative synovial membranes. Ligaments, tendons and muscles had no gross lesions. Deep great veins volar to elbow had variegated tan-current jelly, large thrombi. Foot (with intact carpo-metacarpal joint and associated bones) was stored at 4c.

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On 6-14-06, trachea, cervical esophagus, larynx, thyroid gland, buccal cavity, trunk, buccal surfaces of teeth, spinal column and spinal cord were also examined.

Trachea, larynx, cervical esophagus, pharynx, tonsil, trunk airways and thyroid had no gross lesions.

Mucosa of cheeks and tongue had extensive confluent areas of diffuse hyperkeratosis and subkeratotic intense reddening. The excess keratin was grey-white, soft, cracked and easily removed by digital scrape. When removed it revealed a bright red rough mucosal surface. Submaxillary, mandibular

and retropharyngeal lymph nodes were not found. Upper (maxillary) left tooth ascade had a midsagittal, deep (22mm), 100 mm long 30mm wide groove worn shiny smooth by a ridge-shaped bearing surface of the opposing lower (mandibular) arcade. In the mid-segments, 2 holes (caries) both ~10mm dia. and 12 and 15mm deep, respectively, penetrated the arcade in a maxillary direction. Probing the holes revealed hard bases. Upper right arcade was normal except for several (4) small boles, 2-3mm wide and deep. The left maxillary and right mandibular arcades were ~ 4cm shorter in length than their corresponding arcades due to an absence of caudal molars.

Ventral extremity of the articulation of dorsal spinous process of T17:18 was enlarged by a two, grey-green, fibro-osseous, conical masses protruding 12-14mm ventrally into the vertebral canal. Similar (but less conical and less marked) boney enlargements protruded ventrally into the vertebral canal at L1:2 and 2:3 at the level of the cauda equina. These rounded enlargements had red streaks in overlying fibrous tissue; and, in one enlargement, there was early lipping of bone. No compression of cauda equina at L1:2, 2:3, occurred. Apposing spinal cord at T17:18 was not obviously compressed but felt focally soft by palpation. Dorso-lateral, extra-dural venous sinuses contained tan thrombi continuous from C4 to L3.

On 6-15-06, left and right hindlegs and right foreleg, including stifle, tibiotarsal, tarsometatarsal, shoulder, elbow, radiocarpal, intercarpal and carpometacarpal joints, deep veins and muscles and sciatic nerves were examined. Similar gross lesions to those seen in the joints of the left foreleg were present in these legs. Mild to no gross lesions involved hips, tibio-tarsals, tarso-metatarsals, shoulder, elbow and intercarpal joints. Stifle joints of both hindlegs had severe lesions; and, radiocarpal joint of LF had very severe lesions (extensively, chronically excoriated, rough articular surfaces and dark red bloody synovial fluid). Deep veins of all legs contained large variegated tan-current jelly thrombi. Deep musculature plantar to stifle had areas of equivocal pallor (? compressive/ischemic myopathy). Sciatic nerves at the level of the stifle had streaky hemorrhages in perineurium (? ischemic neuropathy). Sole of RF had 3 chronic-appearing ulcers, each immediately volar (~0.5-1.0cm) to toe of digits 2, 4 and 5. They were irregularly ovoid, ~3x2cm not underun and ulceration of corium was superficial. Feet (3) were stored at 4 C.

Dissection methods, organ weights and dimensions:

- 1) Brain (fresh)
 - a) Total = 5.14kg
 - b) Cerebrum = 3.56kg (cranial to caudal colliculus)
 - c) Cerebellum = 1.40kg (removed by transection of cerebellar peduncles)
 - d) Brain stem = 0.20 kg (caudal to caudal colliculi)
 - e) Cerebrum: Cerebellum ratio = 3.6:1.4=2.5:1=5:2

Brain (fresh) was further dissected on 6/13/06 as follows:

- a) Approximately 100g of frontal cerebral cortex (removed by coronal T/S) was added to ~40g of lateral cerebellar cortex (removed by L/S) and submitted for toxicologic analysis.
 - b) A single 4x5x5cm cube of diencephalon was removed (by section) to include both thalami, hypothalami and pre-and supra-optic chiasma areas. This was placed in NBF.
 - c) Small-pieces of cerebral and cerebellar cortex, brain stem and midbrain were placed in VTM and stored at -80 C.
 - d) Remaining brain was immersed in NBF.
- 2) Spinal cord. Spinal cord was removed on 6-14-06 according to standard protocol after paramedian bandsaw section. Entire cord (C1 to cauda equina) was immersed in NBF.
 - 3) Thyroid (fresh) = 500g
 - 4) Teeth arcades:

Arcade ID	Length (cm)	Width (cm)	Comment
Left, maxillary	13	5	Caudal ~5cm missing
Right, maxillary	19	5	-
Left, mandibular	16	7	-
Right, mandibular	12	6	Caudal ~4cm missing

06-26-06: ADDENDUM

On 6/23/06, 3 feet were examined with Dr. Klause: LF,RF and LH.

Methods: Skin was removed to skin-horn junction. Distal row of carpal bones were removed from LF and RF. The site of the Sept '05 surgery (LF, digit 5) was superficially explored by multiple 3-5mm, serial sections by knife and sterile handsaw parallel to skin surface. Digital structures, metacarpal bones, sesamoid bones, joints, digital cushion and sole (foot pad) were examined by making mid-sagittal bandsaw sections through each digit and adjoining metacarpal/tarsal bones.

The following samples were taken for histopathology:

- 1) joint capsule with synovial membrane from carpometacarpal, metacarpophalangeal of LF and LF; metatarsophalangeal of LH.
- 2) distal 2cm of metacarpal and proximal 1cm of P1 of digit 5 of LF, serially and sagittally bandsaw sectioned at ~2mm intervals.
- 3) distal 9x10x5cm of where digit 5 pre-existed before most of it was removed surgically from LF. (this tissue consisted of scar tissue and areas of fibro-osseous tissue).
- 4) sole ulcer at toe of digit 4 of LH.

Findings:

Left front foot:

The subcutaneous aspect of the surgical site area (digit 5) was slightly rounded and hard on palpation. The sections parallel to the skin surface revealed a pale yellow, narrow (~2mm dia) tract of tissue perpendicular to skin surface. No trace of the tract was found deeper than 1.5cm from the subcutis, nor was any abscess or sequestra of necrotic bone found in deeper tissues. A small volume (~0.1ml) of pale yellow exudate was collected from the tract on a culturette swab for aerobic culture and Gram staining.

Proximal and distal dorsal periarticular margins of distal row of carpal bones had extensive, marked (3-6mm long), sharp boney spurs (exostoses). The distal margins apposing P1 of digit 2, 3 and 4 were the most severely involved. Adjacent synovial membranes were dark red. Synovial fluid was dark red.

Metacarpophalangeal: joints had mild (insignificant) lesions, except for digit 5 which had reddening and hypertrophy of synovial membrane and periarticular scarring.

The area where most of digit 5 pre-existed was extensively involved by grey, glistening, firm fibrous tissue within which were embedded several, irregularly-shaped, variably-sized (2x3x1cm to 5x5x3cm) areas of pink-off white cancellous bone. No yellow (necrotic) bone, foreign body or abscess was detected grossly. This entire area (distal to metacarpal P1 joint 10cm to toe, 9cm to side of foot and 5cm thick was removed and placed in decalcifying solution for further dissection.

Distal articular surface of metacarpal 5 had a locally-extensive, obliquely-oriented, trough of deep ulceration (~1-3mm deep) involving ~1/4 of the articular surface. The edges of the ulcer were rounded by white reactive cartilage (indicating chronicity and resolution) and the bed of the ulcer was pink and healthy-appearing.

Associated sesamoid bone was normal. Serial bandsaw section of distal metacarpal 5 revealed a solitary, small (3mm dia), spherical cavity lined by a thin red membrane ("bone cyst") situated ~4mm beneath articular surface (overlying articular surface was not ulcerated).

Right fore foot:

Proximal and distal joints of the distal row of carpal bones had similar lesions to those involving the left front foot.

Metacarpophalangeal: joints 2 to 5 had moderately pronounced lesions of DJD (bloody excess synovial fluid with mucoid blood clots; dark red synovial membranes, and mild periarticular exostoses).

Sagittal bandsaw section of all digits revealed no additional lesions. Particular attention was given to the digital cushion (looking for a large ovoid central area of increased density as seen on CT scan (per Dr. Wynne)) but none was detected except for subtly increased nodularity and increase in connective tissue. Samples were taken for histopathology.

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Left hind foot:

Metacarpophalangeal: joints of digit 2 and 5 had moderate lesions: digit 2 had blood clots in joint fluid and digit 5 had excess yellow, but otherwise normal-appearing, joint fluid.

Digital cushion had several, spherical to ovoid, relatively large (~3-4cm across) masses of white material embedded at random ~6cm proximal to sole in the plantar half of the foot. Some were putty-like in consistency whereas others were gritty or bone-like (? calcinosis circumscripta, fat necrosis, and/or osseous metaplasia).

Sole had a slit-like ulcer ~1cm plantar to toe of digit 4. On sagittal section, adjacent sole horn was crumbly, black, moist and fetid. Supra-adjacent corium was cavitated and suppurating with dark red hyperemic margins. Proximal margin of cavity was ~15mm distant from P3.

H I S T O P A T H O L O G Y

Tissues examined: R. axillary and L. inguinal lymph nodes.

Lesions present:

- 1) Chronic mild hyperplastic lymphadenitis characterized by lymphoid hyperplasia, mild mature fibrosis and multifocal osseous metaplasia.

ADDENDUM: 06-20-06 Additional histopathology:

Tissues examined: spinal cord at T17:18, thrombi from chambers of right heart and major blood vessels at base of heart (4), thrombi from minor blood vessels near base of heart (27), adrenal gland, L.papillary m. of heart, liver, lung, kidney, tail-head lesion (blocks 9,15,17,19), R.hip lesion (block 20), joint-capsule (LF intercarpal), spleen, stomach, intestine (4) and uterus (4).

Lesions present:

- 1) Acute thrombosis, non-septic, right chambers of heart and major and minor vessels near heart.
- 2) Chronic-active, severe, necrotizing, septic cellulitis-myositis with superficial and deep colonization by coccal bacteria, superficial emphysema, poor granulation, collagen necrosis and multiple thrombosis, tail-head lesion.
- 3) Chronic-active, moderate, fibrinosuppurative, cellulitis with superficial colonization by bacterial cocci, R.lip lesion.
- 4) Chronic-active, severe, fibrinohemorrhagic, ulcerative, non-septic arthritis, left intercarpal.
- 5) Chronic membranoproliferative glomerulonephritis with proteinuria.

Incidental findings:

- 1) Lipofuscinosis of myocardium, adrenal cortex and motor neurons.
- 2) Hemosiderosis of liver and spleen.
- 3) Calcinosis circumscripta of skin, left hind leg.
- 4) Leiomyomas of uterus.

ADDENDUM: 7-25-06 Additional histopathology:

Tissues examined (block #'s are in parenthesis):

LF foot sinus tract (37); LF, MC-P1, digit 5 joint capsule (38-40); bronchial LN (41); cheek mucosa (42); L. (43) and R. (44) sciatic nerves; L. tibial n. (45); dorsal mid-brain (46), ventral mid-brain (47); dorso-median occipital (48), temporal (49) and frontal (50) cortex; spinal cord at C3 (51), C7 (52), T6 (53), L4 (54), S1 (55); submaxillary LN (56); sublumbar LN (57); tongue (58); cheek mucosa (59,60); tonsil abscess (61); L. elbow deep thrombus (62) and joint capsule (63); extra-dural vertebral venous sinus at C3 (64) and L3 (65); L. popliteal region venous thrombosis (66) and musculature (67); R. popliteal region venous thrombus (68) and musculature (69); R. stifle joint capsule (70); L. stifle joint capsule (71-73).

Lesions present (block #'s are in parenthesis):

- 1) Chronic, inactive, resolved sinus tract, LF foot, operative site (37).
- 2) Chronic, inactive, resolved, arthritis (subsynovial aggregates of large numbers of plasma cells) and trapped fragments of loose degenerate articular cartilage in synovial crevices (38-40), LF, MC-P1 joint.
- 3) Diffuse parakeratosis of mucosa of oral cavity with focal acute ulceration (diffuse parakeratosis, epithelial hyperplasia, focal ulceration with superficial sepsis and diffuse hyperemia and congestion (non-specific).
- 4) Acute deep vein thrombosis of L. elbow, R. and L. popliteal regions and extra-dural vertebral sinuses (C3 to L4).
- 5) Acute skeletal muscle necrosis (swelling and sarcoplasmic flocculation of ~90% myofibers, intramuscular hemorrhage and fibrin deposition, interstitial infiltration of neutrophils and mononuclear leukocytes), popliteal region of LH leg.
- 6) Chronic-active, diffuse, villonodular, severe, ulcerative, plasmacytic arthrit

is

(synovial cell hyperplasia, multifocal ulceration, proprial infiltrates of large numbers of plasma cells with small numbers of lymphocytes and macrophages, engulfed fragments of articular cartilage and collagen, hyperemia and edema, focal hemorrhage, fragments of articular cartilage debris in crevices of synovial membrane), R. and L. stifle (70,71-73).

Incidental findings:

- 1) Slight anthracosis and single-cell calcification, bronchial lymph node
- 2) acute intra-adipose tissue hemorrhage, submaxillary
- 3) Slight chronic tonsillitis (intra-crypt inflammatory cell debris)
- 4) Slight subacute skeletal muscle necrosis (swelling and sarcoplasmic flocculation of ~5% of myofibers with intra-myofiber accumulation of macrophages), R. popliteal region
- 5) Lymphoid hyperplasia of lymph node, L. popliteal

ADDENDUM: 08-18-06 Additional histopathology:

Tissues examined (block #'s are in parenthesis): R. intercarpal joint capsule (74); RF sole ulcers, digit 2(75), 4(76), and 5(77), L. auricle and pharyngeal fold (78), R. auricle (79), R. papillary m. of heart (80), esophagus (81), non-glandular stomach (82), arytenoid fold (83), pituitary gland and cerebral arteries (84, 90), subpituitary venous sinus (84), uterine anomaly (84, 125-128), dorsal vermis of cerebellum (85), epiglottis (86), obex of medulla oblongata and basilar artery (87,89), endometrium (88,91), LF carpometacarpal joint capsule (92); LF, distal end of MC5 (93); LF, D5, proximal P1 (94-95) and surgical site beneath proximal P1 (96-98); LF, MC-P1, joint capsule D1 (99), D2 an

d

3(100), D4(101); LF, digital cushion (102-103); RF, joint capsules, carpometacarpal (104-105), MC-P1, D1(106), D2(107), D3(108), D4(109), D5(110); RF digital cushion (111-113); LH digital artery (114), LH, joint capsules, MC-P1, D1,2,3 pool (115), D4 and 5 (116); LH digital cushion (117-118); LH, D4(119); T17:18 dorsal (120, T17:18) and ventral (121) vertebral junction masses, medial vertebral canal wall; L2:3, dorsal (122) and ventral (123) vertebral junction masses, medial vertebral canal wall; thyroid gland (124).

Liver, spleen, adrenal and heart specially stained by Perl's Blue for iron and PAS and ZN for lipofuscins and complex carbohydrates.

Lesions present:

- 1) Severe chronic-active, ulcerative fibrinous arthritis (slight mononuclear leukocyte infiltration of propria, diffuse adherence of fibrin on naked propria, ischemic necrosis of some villi, no pus, no sepsis), R. intercarpal (74).
- 2) Sole ulcers, chronic-active (focal loss of sole horn, colonization of degenerate horn/exposed corium by bacteria, congestion and granulation of corium, RF (digits 2,4, and 5) (75-77) and LH (digit 4) (119).
- 3) Acute thrombosis of extra-dural venous sinus at base of pituitary gland (84), digital vein in LF (99) and digital artery in LH (114).
- 4) Chronic-active thrombosis of a localized small uterine anomaly (a mesonephric duct remnant in wall of uterus resembling corpus cavernosum of penis). Most blood sinuses were acutely thrombosed; a few sinuses were occluded by organizing thrombi (fibroplastic invasion of thrombi), approximately 3 days old (84, 125-128).
- 5) Joint capsule findings: a) Severe chronic active ulcerative fibrinous arthritis (diffuse loss of synovial membranes, replaced by extensive layers of cell-poor fibrin containing fragments of degenerate cartilage, large numbers

of

red cells and small numbers of neutrophils in some joints; pannus formation in occasional joints; variable numbers of infiltrating plasma cells in propria; no pus or bacterial sepsis), RF: intercarpal (74), carpometacarpal (104,105) and metacarpophalangeal (d5) (110) LF: carpometacarpal (92); LH: metacarpophalangeal (d1,2, and 3 pool, 1/3) (115,116). b) No lesions RF: metacarpophalangeal (d1,2,3, and 4) (106-109) LF: metacarpophalangeal (d1,2,3, and 4) (99-101); LH: metacarpophalangeal (d1,2, and 3 pool 2/3; d4 and 5) (115-116).

- 6) Findings in proximal articular remnant of surgically manipulated bone (P1, d5, LF) (94,95).

Sagittal section revealed extensive replacement of articular cartilage by mature fibrous tissue (pannus) with sub-adjacent healthy bone beneath. Not significant.

- 7) Findings in distal metacarpal 5 of LF (which articulated with proximal articular remnant of P1, d5) (93).

Sagittal section of MC5 revealed focal pannus and a solitary, small, non-inflamed subarticular bone cyst. Not significant.

- 8) Findings in gross fibro-osseous tissue (distal to proximal remnant of surgically manipulated P1 of d5 of LF) (96-98).

Multiple sections of this area revealed extensive healthy mature granulation tissue within which were fragments of healthy pre-existing bone undergoing osteoclastic resorption, areas of healthy new bone growth (osseous metaplasia) and several (~10) small (0.1-1mm) spicules of pre-existent necrotic bone, some within small non-septic abscesses, well walled-off, no sepsis.

- 9) Findings in digital cushions:
- a) LF: No lesions (101-103).
 - b) RF: No lesions (111-113).
 - c) LH: Chronic locally-extensive areas of necrosis of adipose tissue with secondary calcification, fibrosis, minimal inflammation, chondroid metaplasia and deposition of amorphous-hyaline eosinophilic (117-118).

Incidental Findings:

- 1) Solitary, microscopic, submucosal esophageal cyst.
- 2) Rare microscopic mineralized foci in pituitary gland.
- 3) Mild cystic hyperplasia of endometrial glands.
- 4) Focal, proliferative, degenerative vertebral osteodermatitis involving bone and ligaments of vertebral bodies T17:18 and L2:3, mediodorsal and medioventral.
- 5) Pigmentation of liver, adrenal, spleen and heart.

Tissue	PBlue	PAS	ZN
Liver	-	++	-
Spleen	+++	+++	-
Adrenal	-	+++	-
Myocardium	-	-	-

Conclusion: A mixture of hemosiderin and lipofuscin involve spleen; and lipofuscin involves liver and adrenal, and possibly myocardium.

T O X I C O L O G Y

The tissues had the listed metals in acceptable concentrations when compared to normal ranges for equine species. MDL = method detection limit (lowest concentration detectable by our test method).

*** CHOLINESTERASE

Specimen Information	Result	MDL
Id Type		
2006-0039 BRAIN	2.2 uM/g/min	0.1 uM/g/min

HEAVY METAL SCREEN

CAHFS #F
08/21/06

ACCESSION#: S0607485
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Specimen Type KIDNEY
Metal Arsenic Cadmium Copper Iron Lead
MDL 1 PPM 0.3 PPM 0.1 PPM 0.2 PPM 1 PPM
2006-0039 < 1 PPM PP 35.0 PPM 2.3 PPM 103 PPM < 1 PPM PP

Metal Manganese Mercury Molybdenum Zinc
MDL 0.04 PPM 1 PPM 0.4 PPM 0.1 PPM
2006-0039 0.52 PPM < 1 PPM PP < 0.4 PPM 52.2 PPM

Specimen Type LIVER
Metal Arsenic Cadmium Copper Iron Lead
MDL 1 PPM 0.3 PPM 0.1 PPM 0.2 PPM 1 PPM
2006-0039 < 1 PPM PP 7.7 PPM 15.2 PPM 207 PPM < 1 PPM PP

Metal Manganese Mercury Molybdenum Zinc
MDL 0.04 PPM 1 PPM 0.4 PPM 0.1 PPM
2006-0039 2.17 PPM < 1 PPM PP 3.4 PPM 143 PPM

C L I N I C A L P A T H O L O G Y

Cytology

Specimens examined: joint fluid (L.stifle, L.intercarpal, R.tibiotarsal).

Findings	L.stifle	L.inter-c.	R.tt.
Chondroid/cytes	+	-	+++
Red blood cells	+++	+++	+++
Granulocytes	++	+++	+
Mononuclear cells	-	+	++
Micro-organisms	-	-	-

Interpretation: Degenerative joint disease (3/3)

B A C T E R I O L O G Y

Antigen Detection:

Direct FA stain on exudate from "tail-head" cellulitis-myositis lesion was negative for Clostridium septicum, Chauvei, Novyi and Sordelli.

CAHFS #F
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ACID FAST STAIN

Specimen Information		Results
ID	Type	
2006-0039 A	UTERUS-SMEAR	No Acid-fast Organisms detected
2006-0039 B	UTERUS-SMEAR	No Acid-fast Organisms detected

*** BACTERIAL AEROBIC CULTURE

Specimen Information		Results
ID	Type	
2006-0039	TAIL BUTT	Streptococcus dysgalactiae subsp. equisimilis Lge# Mixed flora Lge#
2006-0039	R AXILLARY LN	No growth - In 48 Hrs.
2006-0039	L INGUINAL LN	Mixed flora Rare
2006-0039	R HIP	Streptococcus dysgalactiae subsp. equisimilis Mod# Aeromonas sp. Mod#
2006-0039	KIDNEY	No growth - In 48 Hrs.
2006-0039	LIVER	No growth - In 48 Hrs.
2006-0039	CARPUS-LEFT	Mixed flora Rare
2006-0039	HOCK-RIGHT	No growth - In 48 Hrs.
2006-0039	LEFT KNEE	Mixed flora Sm#
2006-0039	JOINT-LEFT M/	Mixed flora Rare
2006-0039	LF TRACK	No growth - In 48 Hrs.

BACTERIAL ANAEROBIC CULTURE

Specimen Information		Results
ID	Type	
2006-0039	TAIL BUTT	Mixed flora - Lge#

GRAM STAIN

Specimen Information		Results
ID	Type	
2006-0039	R HIP	Gram-positive cocci Mod#
2006-0039	L INGUINAL LN	No Organisms detected
2006-0039	R AXILLARY LN	No Organisms detected
2006-0039	TAIL BUTT	Gram-positive cocci Mod# Gram-variable rod Mod#
2006-0039	LF TRACK	No Organisms detected
2006-0039 A	UTERUS-SMEAR	No Organisms detected
2006-0039 B	UTERUS-SMEAR	No Organisms detected

SALMONELLA CULTURE - MAMMALIAN

Specimen Information		Results
ID	Type	
2006-0039	LIVER	No Salmonella sp. detected

C L I N I C A L H I S T O R Y

See attached history on submission form.

This elephant went down and was down overnight for an unknown duration; treatment with Banamine/Ketoprofen/and solu-delta were done before any lift attempt to minimize reprofusion toxins/injury. 20-30 minutes after lift attempts - she went into vascular collapse/toxic shock; CPR was instituted

(R side thorax) and drugs given. CPR 20 minutes and stopped.

Conditions suspected: Many years ago was housed for a period of time with/a TB+ animal. For many years 2x/yr TB trunk washes have been neg.

06-20-06: Additional history (per Dr. Wynne's telephone call 6-14-06) Gita did a lot of hard rubbing of tail head abscess region against posts etc which would account for the traumatic dislodgement of the large loose pieces of inflammed tissue seen at post mortem. Blood was taken for CBC and aerobic and anaerobic blood cultures on 6-9-06 and sent to Antech.

06-27-06: Additional history (per Dr. Wynne's email):
Just a couple of pieces of information to add to the necropsy.

Gita's keepers left between 3-3:30 p.m. Gita was observed down and quiet at 8:45, so she went down at some time between 3:30 and 8:45. There was some evidence that she may have spent some time attempting to get up (disturbed areas of ground). Based on this information, she was down from 12 to 17 hours. I don't know if this will help with determining if the extensive clots were the cause of her going down, or a possible consequence. I think we would all like to know your best guess on this.

Other history I've been reviewing-her albumin tended to run low. It's been 2.2-2.5 for many months. Normal should be 3.2. The day before she died it dropped to 1.7. Previous urinalysis showed only trace protein. Based on the glomerular inflammation you saw, I would guess that she was loosing albumin in her urine in the last few days of her life. Her WBC never showed significant change.

C O N T A C T L O G S U M M A R Y

Report	Date Reported
Preliminary 3	06/27/06-FAX
Preliminary 2	06/21/06-FAX
Preliminary 1	06/12/06-FAX

S P E C I M E N S U M M A R Y

Specimen Type	Breed	ID	Age	Sex	Qty
CARCASS	ASIAN ELEPHAN	2006-0039	49 YEARS	FEMALE	1