Computerized Tomography (CT) Techniques for Analysis of Trauma and Disease in Marine Mammals

Cramer, S. R.[1], Ketten, D. R.[1,2], Arruda, J.[1,2], Prahl, S.[3], Williams, S. R.[4], Dunnigan, B.[4]

(1) Biology Department, Woods Hole Oceanographic Institution, Woods Hole, MA, USA (2) Harvard Medical School, Boston, MA, USA (3) FTZ, University of Kiel, Forschungs- u. Technologiezentrum, Kiel, Germany (4) National Marine Life Center, Bourne, MA, USA

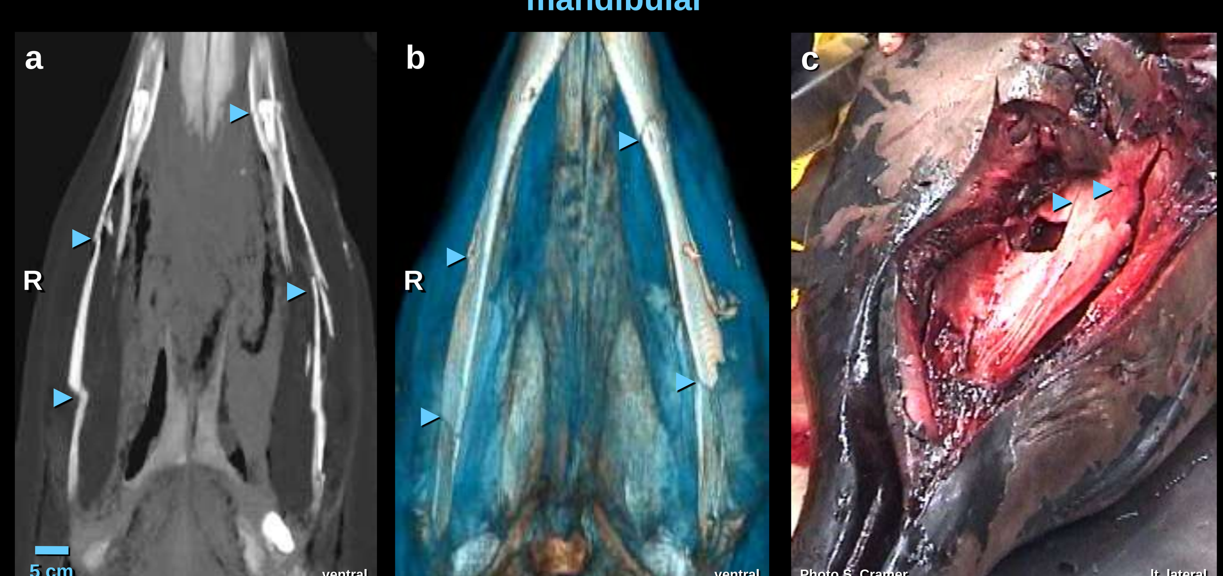
INTRODUCTION: Reports of biomedical imaging use for marine mammal investigations have been increasing in the last two decades for research (Ketten, 1984; Nordoy and Blix, 1985; Cranford, 1988; Hillman, 1991; Ponganis *et al*., 1992; Endo *et al*., 1999; Montie, 2006; Arruda *et al*., 2007) and for clinical applications (Haulena *et al*., 1998; Van Bonn *et al*., 2001; Williams and Dunnigan, 2005). However, clinical methods are not as well documented for marine mammals as for domestic species (Van Bonn *et al*., 2001). Further, most clinical CT scanners have exposure controls that compound artifacts when scanning larger specimens that impair diagnoses (Prokop, 2003). Over the past decade, we have examined more than 600 specimens (57 species, 17 families, 3 orders) and explored multiple protocols balancing higher dose levels and longer exposure times to achieve proper tissue penetration and improve imaging of larger, more massive animals, such as smaller mysticetes and whole, larger odontocetes, pinnipeds, and sirenians. Below we describe pathologies and techniques developed for CT imaging of both large and small marine mammals. Our objective is to establish standard protocols for CT for diagnostic imaging, forensics, and research on both live and post mortem marine mammals. Representative cases for a range of abnormalities and the protocols employed are described below.

MATERIALS & METHODS: Marine mammal specimens described here were obtained from local stranding networks, fisheries services, aquaria, and marine mammal rehabilitation centers in accordance with state and federal regulations under the amended Marine Mammal Protection Act (MMPA) of 1972. Live animals were scanned under the guidance of licensed veterinarians. CT scans were acquired at the Computerized Scanning and Imaging Facility (CSI) of Woods Hole Oceanographic Institution (WHOI), and Massachusetts Eye & Ear Infirmary (MEEI), using a Siemens® Volume Zoom CT unit. Animals were scanned craniocaudal, prone. All were scanned using ultra-high resolution spiral protocols. Effective mAs and KV ranged 200 to 300 mAs, with 120 KV for smaller animals and 300 to 700 mAs, with 120 to 140 KV for larger ones. Transaxial slices were obtained at 8 mm slice thicknesses through whole animals, 3 mm for head, thoracic, and abdominal exams, and 0.5 to 1.0 mm through ear and brain regions. High resolution images were reformatted at slice increments ranging from 0.1 mm to 10 mm (Arruda *et al*., 2007). Shaded Surface Display (SSD), Volume Rendering Technique (VRT), and target tissue segmentation 3D views were reconstructed from tissue attenuation data using multiple 3D visualization software applications. Images of these and other cases as well as a description of equipment and protocols are available at the WHOI CT facility website: www.whoi.edu/csi

RESULTS:

FRACTURES

mandibular



cranial

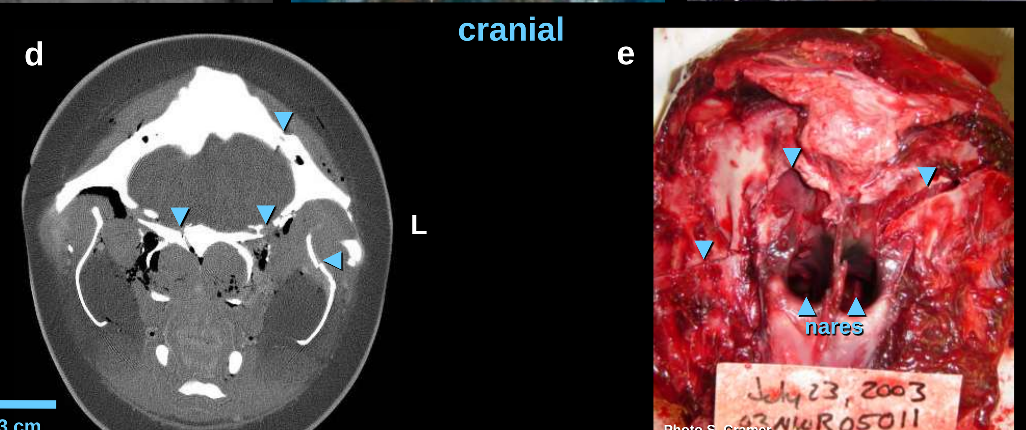


Figure 1. Coronal CT (a) (bone window; 1 mm slice) of a Blainville's beaked whale (*Mesoplodon densirostris*) with bilateral compound mandibular fractures (arrows) (Ketten, 2002). VRT reconstruction (b) (1 mm slice; 3 mm spiral CT) of the head highlighting the fractures (Ketten, 2002). Jaw fractures were confirmed during necropsy (arrows) (Specimen courtesy Caribbean Stranding Network).
Transaxial CT (d) (soft tissue window; 3 mm slice) through the head of a harbor porpoise (*Phocoena phocoena*) with multiple longitudinal and comminuted fractures of the skull and left mandible (arrows). Photograph (e) of the porpoise during dissection (Specimen courtesy NMFS/NOAA).

DISCUSSION: FRACTURES – Necropsy of the beaked whale confirmed multiple bilateral, parallel mandibular fractures (Ketten, 2005a). Adjacent contusions on the ventral surface suggested blunt trauma from post mortem handling. The etiology of the fractures seen in the harbor porpoise (*P. phocoena*) is unknown but is consistent with blunt trauma and possibly vessel strike (Norman *et al*., 2004).

BLUNT TRAUMA

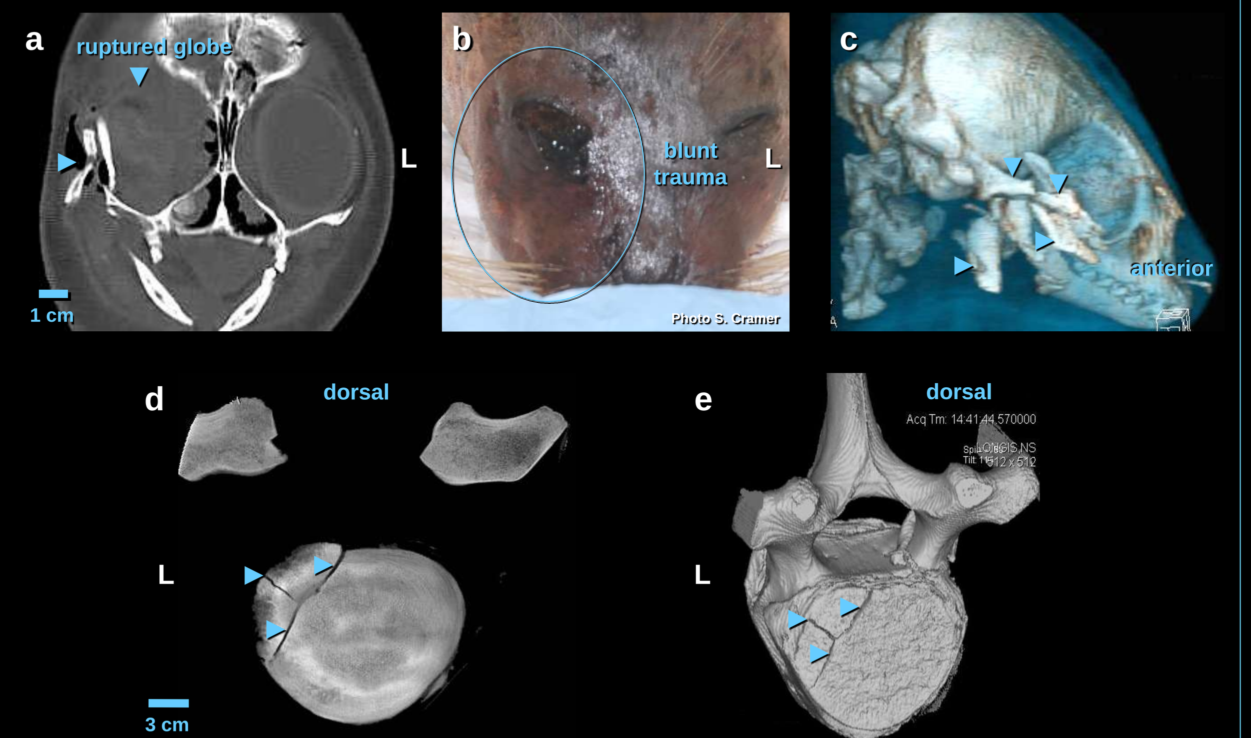


Figure 2. Transaxial CT (a) (bone window; 3mm slice) through the head of a harp seal (*Phoca groenlandica*) with multiple fractures of the right mandible and orbit, and right eye rupture. Seal (b) prior to CT. Note edema and ecchymosis along the right mandible and eye and vitreous humor oozing of the right eye. VRT reconstruction (c) (1 mm slice; 3 mm spiral CT) of the head showing the fractures (arrows) of the zygoma, mandible, and malar bones.
Transaxial CT (d) (bone window; 3mm slice) through a northern right whale (*Eubalaena glacialis*) vertebra with fractures (arrows) of the anterior thoracic vertebral facet. SSD reconstruction (e) (1 mm slice; 3 mm spiral CT) of the vertebra (Specimens courtesy Cape Cod Stranding Network and New England Aquarium Right Whale Project).

DISCUSSION: BLUNT FORCE TRAUMA – The harp seal (*P. groenlandica*) presented post mortem with evidence of blunt force trauma to the side of the head: a ruptured eye, swelling, and contusion. CT scans confirmed underlying fractures. The vertebral fracture pattern from this northern right whale (*E. glacialis*) may have resulted from severe flexing of the back during ship collision (Hamilton, 1995).

BLAST TRAUMA

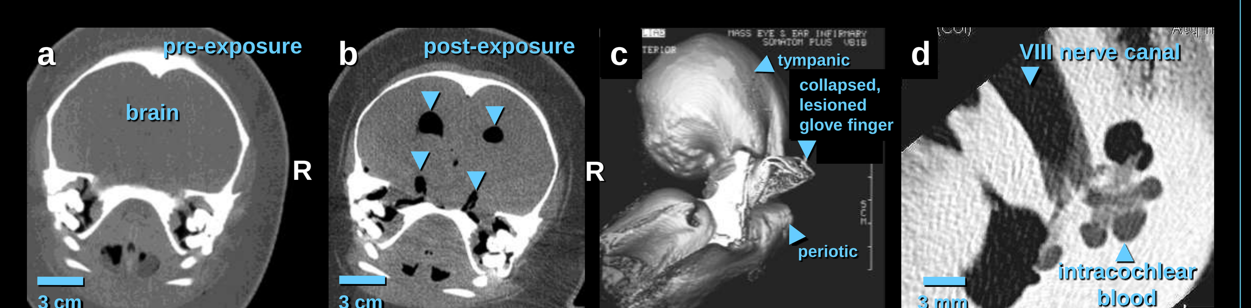


Figure 3. Transaxial CTs (a and b) (soft tissue windows; 3 mm slices) through the head of a harbor porpoise (*P. phocoena*) pre and post exposure to an underwater blast.
SSD reconstruction (c) (1mm slice; 3 mm spiral CT) of a humpback whale (*Megaptera novaeangliae*) ear with glove finger lesions. Transaxial CT (d) (soft tissue window; 0.5 mm slice) through the cochlea of the ear (Specimens courtesy New England Aquarium and J. Lien).

DISCUSSION: BLAST TRAUMA – A harbor porpoise (*P. phocoena*) cadaver was exposed to 100 psi peak pressure in an underwater blast experiment to test correlations between received pressures and blast trauma types. Post exposure CT and necropsy confirmed multiple injuries including brain emboli that would likely have been fatal (Ketten *et al*., 1999; Ketten *et al*., 2005). The humpback whale (*M. novaeanglia*) ear was extracted from an animal that died shortly after a series of underwater explosions. The injuries observed indicated classic auditory trauma consistent with blast injuries: collapsed, lacerated glove finger; blood in inner ear spaces (Ketten, 1995; Todd *et al*., 1996).

GUNSHOT WOUNDS

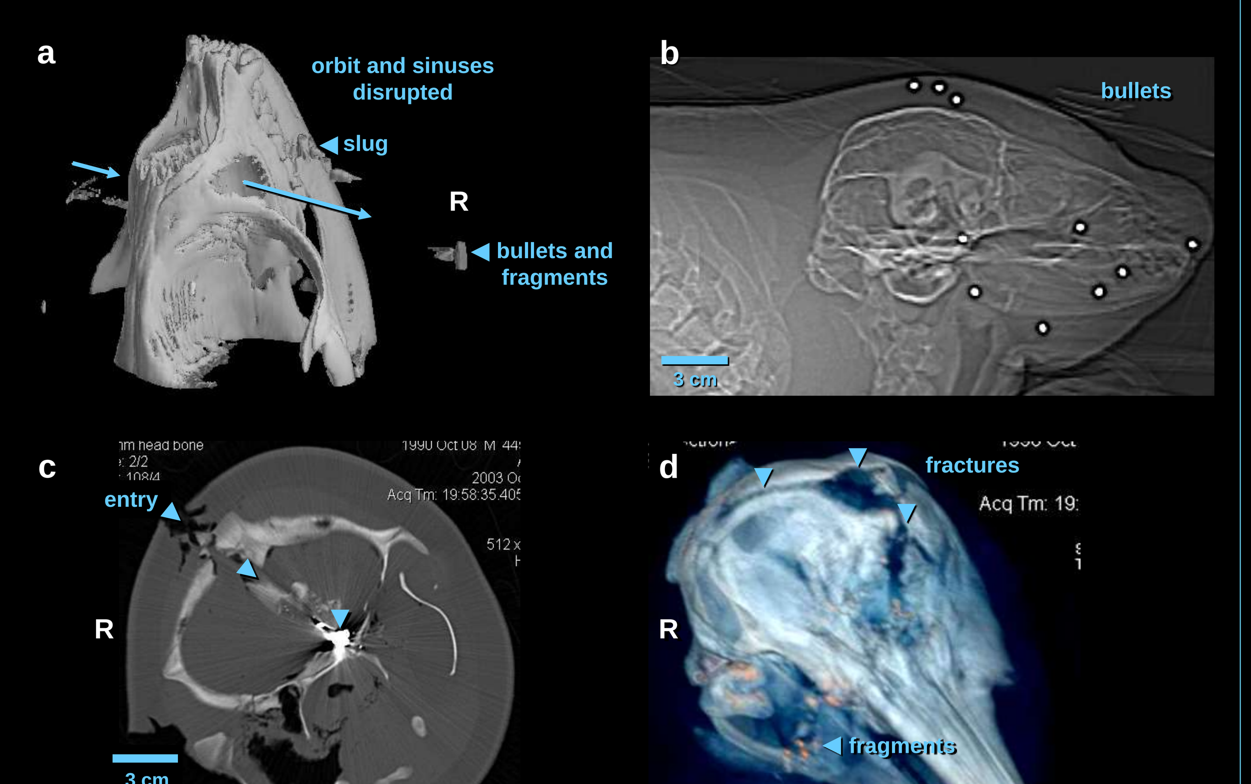


Figure 4. SSD reconstruction (a) (1 mm slice; 3 mm spiral CT) of a harp seal (*P. groenlandica*) with bullets. Topogram (b) of the seal showing the slugs (bright white dots) throughout the head.
Transaxial CT (c) (bone window; 3 mm slice) through the head of a harbor porpoise (*P. phocoena*) with 2 entry wounds to the head. VRT reconstruction (d) (1 mm slice; 3 mm spiral CT) of the porpoise (Specimens courtesy NMFS/NOAA and the German government).

DISCUSSION: GUNSHOT WOUNDS - The metal fragments shown in (a) are slugs from repeated shots at multiple angles. Bone fragments and injury patterns in the 3D reconstruction demonstrate the bullet trajectories. The porpoise was a stranding shot twice with a high velocity, small caliber rifle, once near the blow hole with muzzle contact and once in the right mandible from a greater distance.

TEMPORAL BONE PATHOLOGY

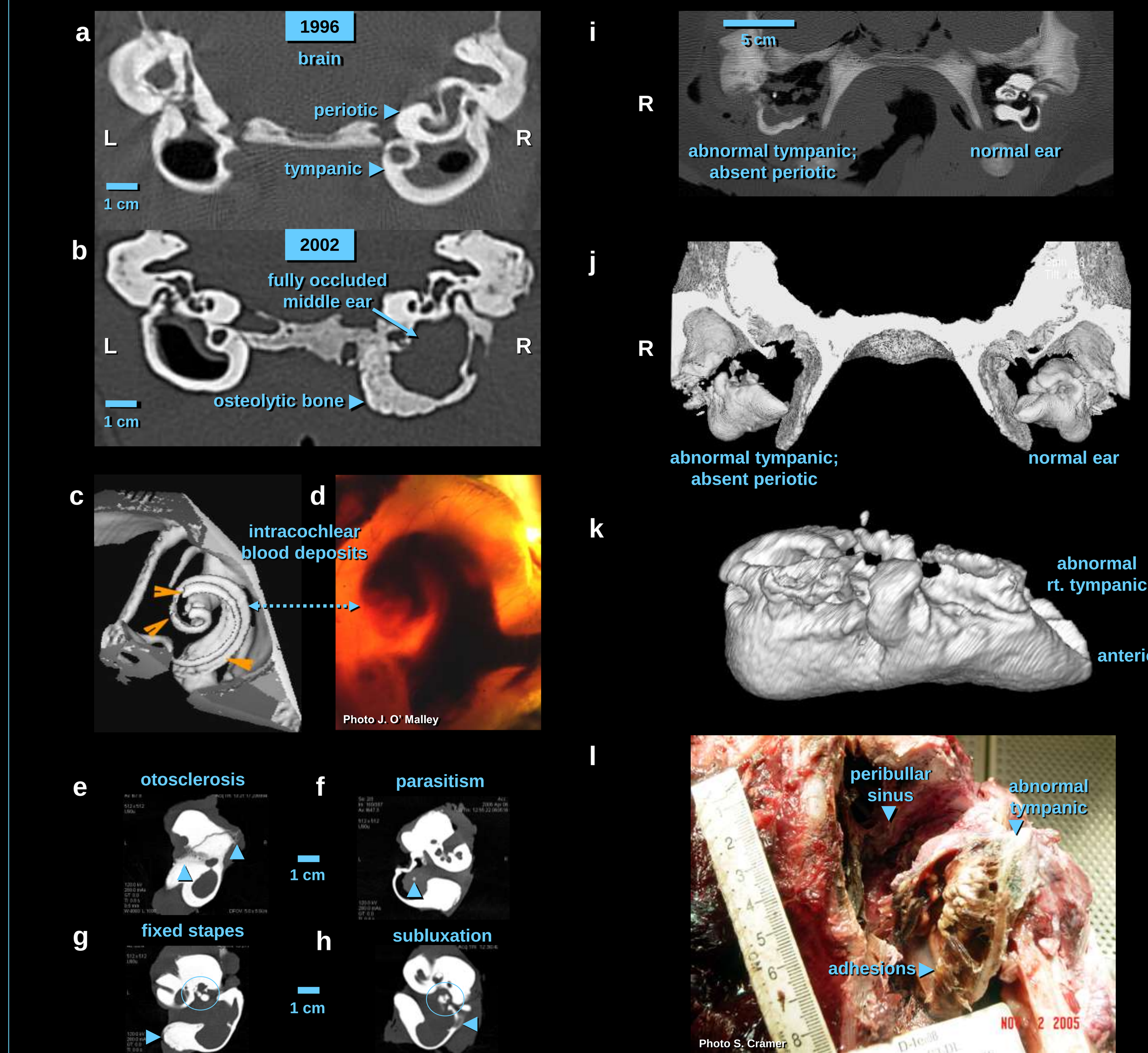


Figure 5. Transaxial CTs (a and b) (bone windows; 0.5 mm slices) through the head of a live harbor seal (*Phoca vitulina*) with a progressive osteolytic dissolution of the right tympanic bone and skull base and occlusion of the middle ear.
SSD reconstruction (c) (0.1 mm slice; 1 mm spiral CT) of a Cuvier's beaked whale (*Ziphius cavirostris*) ear scanned in the head with 3D segmentation for blood deposits. Photograph of the ear (d) after extraction and celloidin embedding.
Transaxial CTs (e – h) (bone windows; 0.5 mm slices) through the ears of four short-finned pilot whales (*Globicephala macrorhynchus*).
Transaxial CT (i) (bone window; 1 mm slice) through the head of a post mortem beluga whale (*Delphinapterus leucas*) with a right ear anomaly (Ketten, 2005b), including absence of a right periotic bone. SSD reconstruction (j) (0.5 mm slice; 1 mm spiral CT) of the right and left ears *in situ*. SSD reconstruction (k) (0.5 mm slice; 1 mm spiral CT) of the right tympanic bone. Right tympanic bone (l) during necropsy (Specimens courtesy Woods Hole Aquarium, Center for Whale Research, D. Rotstein, and New England Aquarium).

DISCUSSION: AUDITORY PATHOLOGY - This harbor seal (*P. vitulina*) presented live with chronic right ear suppuration. CT scans confirmed otitis media (Williams and Dunnigan, 2005). Scans of the beaked whale ear showed a bloody effusion in two inner ear chambers but membranes and scalae were intact (Ketten, 2005a). Scans of the four pilot whale (*G. macrorhynchus*) ears illustrate multiple pathologies: otosclerosis (e), calcified parasitic cysts in the middle ear (f), tympanic sclerosis (arrows) and fixed stapes footplate (g), and ossicular disarticulation (h) (circle) in association with a tympanic wall fracture (arrow). The absence of a right periotic bone in this beluga whale (*D. leucas*) may be due to non-development or a lytic process. The normal left ear argues against but does not rule out a congenital defect. The absence of the periotic bone, the pitted and corrupted right tympanic, the extensive calcified adhesions, and dense granular material in the peribullar sinus suggest protracted infection (Ketten, 2005b).

LABYRINTHITIS

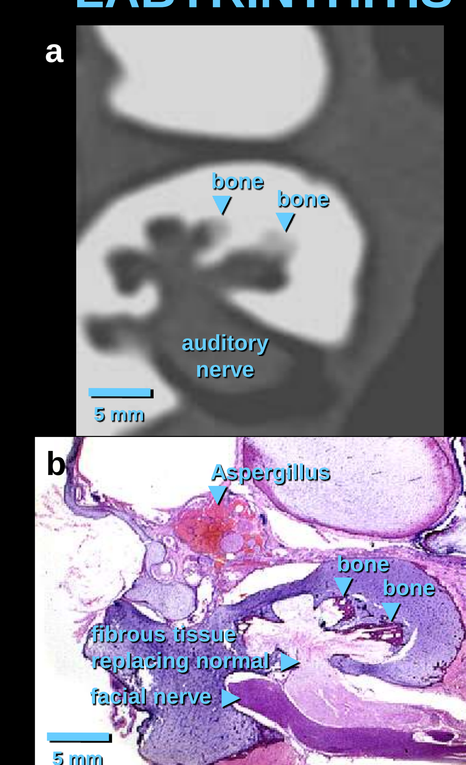


Figure 6. Transaxial CT (a) (bone window; 0.5 mm slice) of an adult female bottlenose dolphin (*Tursiops truncatus*) ear. Midmodiolar histological section (b) of the ear (Specimen courtesy S. Ridgway).

DISCUSSION: LABYRINTHITIS OSSIFICANS – This adult captive female dolphin while alive was unresponsive to sounds in her pool or from trainers. Post mortem scans revealed a total loss of cochlear structure with fibrous and bony tissue filling the cochlear and neural canals (a) which was confirmed histologically (b). This is consistent with labyrinthitis ossificans. There is also evidence now of middle ear Aspergillosis.

BONE PATHOLOGY

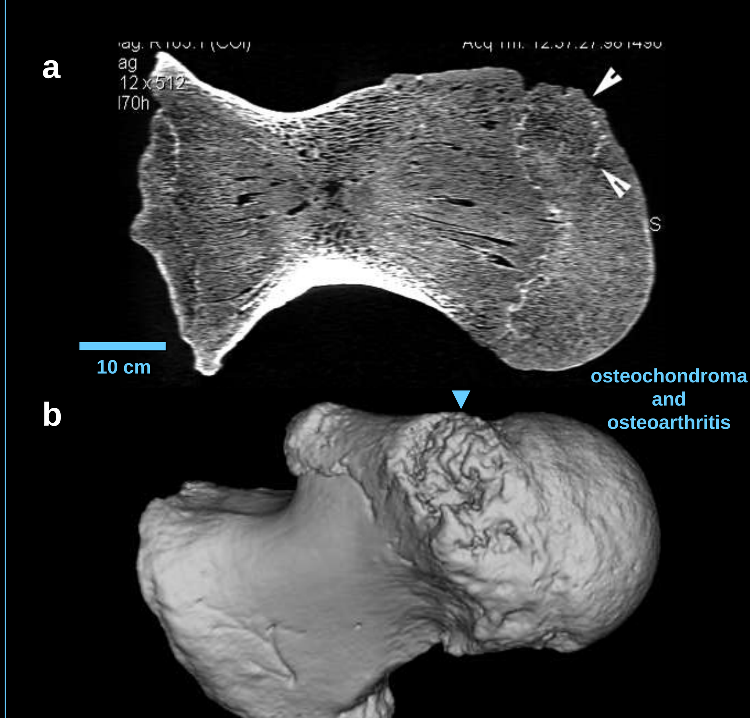


Figure 7. Saggital CT (a) (bone window; 0.5 mm slice) through the humerus of a sperm whale (*Physeter macrocephalus*) and the corresponding SSD reconstruction (b) (0.5 mm slice; 1 mm spiral CT) (Specimen courtesy Cape Cod Stranding Network).

DISCUSSION: CHRONIC OSTEOARTHRITIS & OSTEOCHONDROMA - The humerus was extracted from a juvenile sperm whale (*P. macrocephalus*) found to have numerous bone lesions throughout the skull and axial skeleton. CT and histology demonstrated erosion of articular cartilage and outgrowths of marginal osteophytes (b) (arrow) consistent with chronic osteoarthritis and a benign cartilaginous neoplasm, covered with a rim of proliferating cartilage cells (a) typical of osteochondroma.

LUNG PATHOLOGY

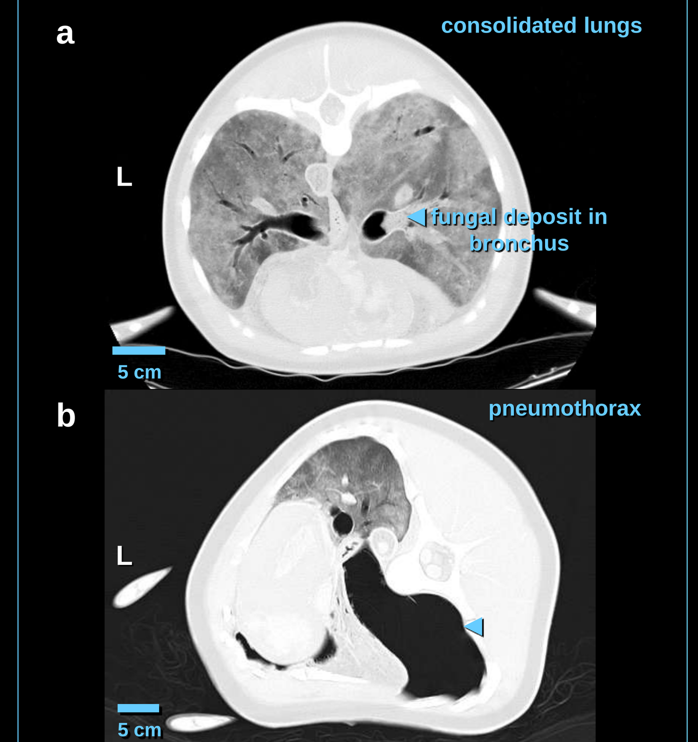


Figure 8. Transaxial CTs (soft tissue windows; 1 mm slices) through the thoracic cavities of a harbor porpoise (a) (*P. phocoena*) with evidence of parenchymous inflammation and bilateral lung consolidation and an Atlantic white-sided dolphin (b) (*Lagenorhynchus acutus*) with a pneumothorax (Specimens courtesy Cape Cod Stranding Network and NWMFS).

DISCUSSION: LUNG PATHOLOGY: – A significant proportion of cetaceans diagnosed with bacterial or viral respiratory tract problems exhibit pulmonary abscesses subsequent to heavy parasite infestation (Dunn, 2001). Pneumonia and or pneumothorax may also be secondary pathologies to inhalation of chemicals, trauma to the chest wall, and a small minority to rickettsiae, fungi, and yeasts.

EMBOLI

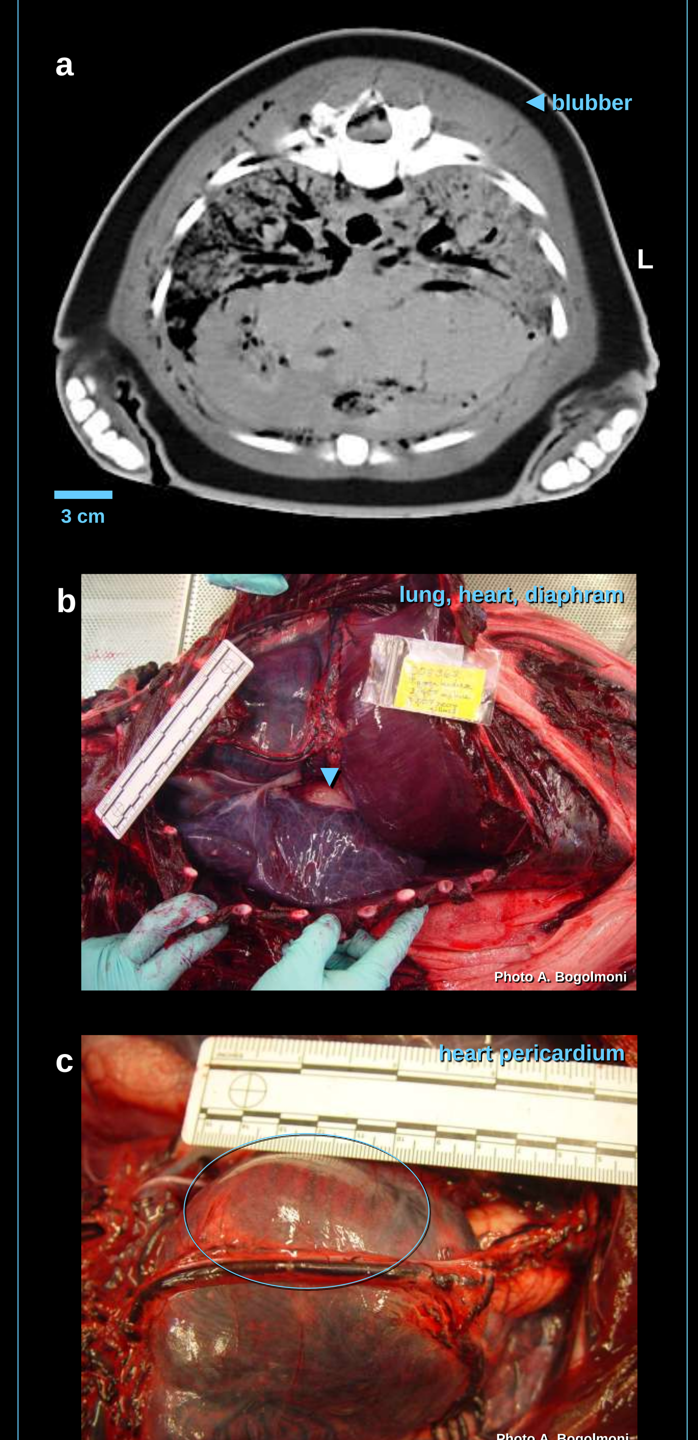


Figure 9. Transaxial CT (a) (soft tissue window; 3 mm slice) through the thorax of a harp seal (*P. groenlandica*). Gas bubbles (b and c) (arrow/circle) were observed in blood vessels, muscle, lungs, heart, brain, and kidney (Bogomolni, 2007) (Specimen courtesy NMFS/NOAA).

DISCUSSION: EMBOLI - This harp seal (*P. groenlandica*) was entangled in a commercial fisheries gillnet (NOAA/NMFS, 2007). The seal was scanned within a few hours of its recovery. No anaerobic organisms were reported from thoracic and abdominal tissues (Bogomolni, 2007). It cannot be determined whether these emboli formed pre or post-mortem.

INTRACRANIAL HEMORRHAGE

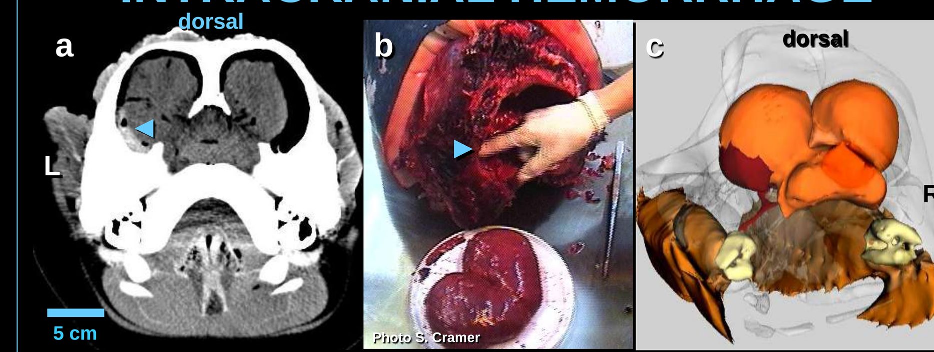


Figure 10. Transaxial CT (a) (soft tissue window; 3 mm slice) through the head of a Blainville's beaked whale (*M. densirostris*) with a left temporal fossa intracranial hemorrhage (arrow) (Ketten, 2005a). Removal of the brain (b) confirmed the hemorrhage. Tissue reconstruction (c) of the head using 3D Slicer® software shows the hemorrhage (red), the brain (orange), the ear bullae (beige), and the jaw fats (bronze) (Ketten, 2005a). (Specimen courtesy Bahamas Stranding Network and NMFS/NOAA).

DISCUSSION: HEMORRHAGE – This animal died in a stranding associated with a Naval sonar exercise. The mechanism behind this hemorrhage is unknown at this time but has been reported in beaked whales stranding under similar circumstances.

PARASITES

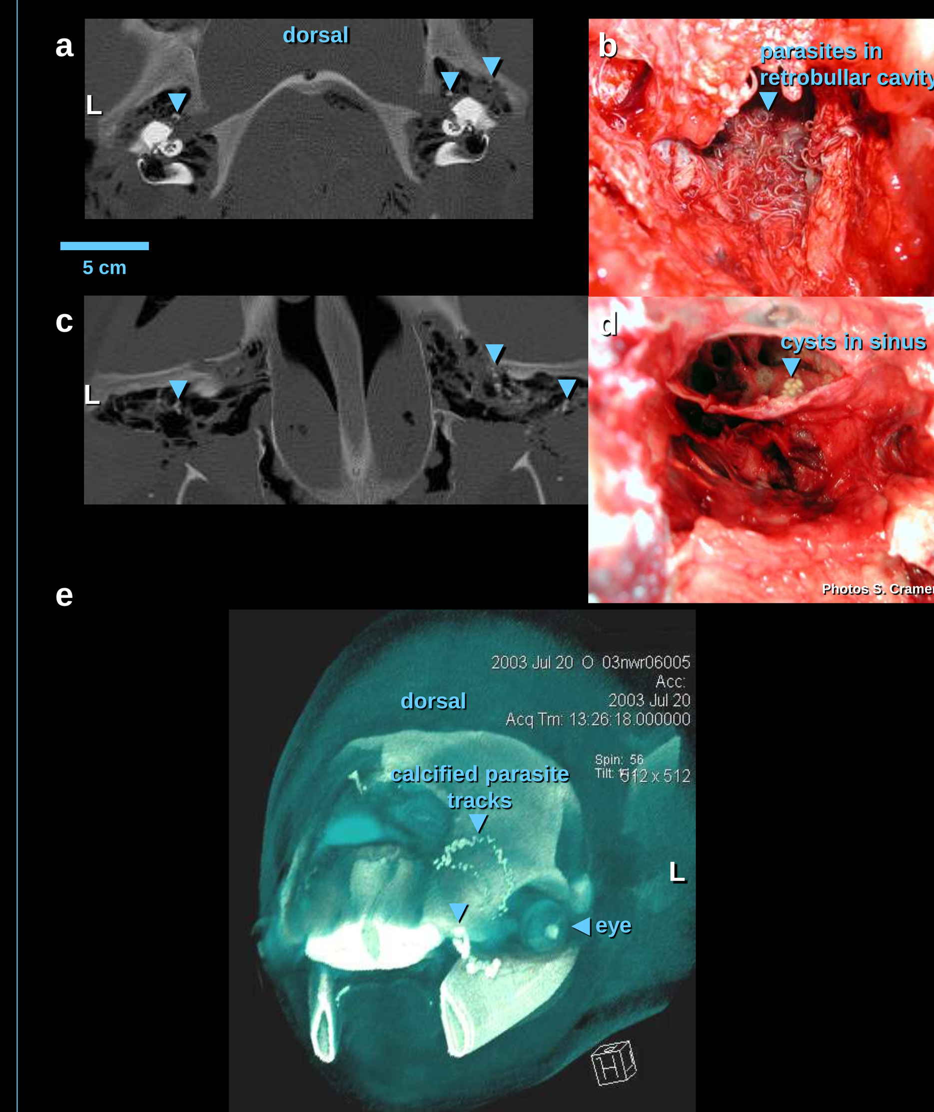


Figure 11. Transaxial CT (a) (bone window; 0.5 mm slice) through the head of a post mortem Risso's dolphin (*Grampus griseus*) with a parasitic infestation (arrows) of the peribullar sinuses. Extraction of the right ear (b) revealed nematodes and cysts (arrow) in the retro bullar cavities. Transaxial CT (c) (bone window; 0.5 mm slice) through the sinus region of the same animal. Dissection revealed calcified cysts (d) (arrow) in the pterygoid sinuses.
VRT reconstruction (e) (1 mm slice; 3 mm spiral CT) of the head of a harbor porpoise (*P. phocoena*) with a > 0.5 m calcified parasitic track invading the blubber, muscle, melon and orbital tissues (arrows) (Norman *et al*, 2004) (Specimens courtesy Woods Hole Aquarium and NWMFS).

DISCUSSION: PARASITES - The parasites observed in the Risso's dolphin (*G. grisseus*) were identified as two species of *Metastrongyles spp.* (T.P. Lipscomb, AFIP). Zucca *et al*., (2004) also reported findings of *Crassicauda grampicola* infestation in the pterygoid sinuses and peribullar regions of a Risso's dolphin (*G. griseus*). In the harbor porpoise (*P. phocoena*) parasite species identification is pending.

CONCLUSION: CT is an increasingly valuable tool for non-invasive, fast, and accurate imaging for research and clinical exams. It offers an opportunity to assess normal and abnormal anatomy with the fidelity required for accurate diagnoses. If more studies team modern imaging with gross observation, dissection, and histological analyses, we can substantially improve our ability to apply this technology to the diagnosis and treatment of conditions in rehabilitation cases. Resolution and speed are improving and with the increasing availability of wider bore machines, better imaging of larger species is becoming feasible for both *in vivo* and necropsy studies.

LITERATURE CITED: Full citations are included in the handout.

ACKNOWLEDGEMENTS: We are grateful for the support and assistance of Woods Hole Oceanographic Institution (WHOI), New England Aquarium (NEAq), Cape Cod Stranding Network (CCSN), National Oceanic and Atmospheric Administration (NOAA), National Marine Fisheries Service (NMFS), and Caribbean Stranding Network. This research was supported by Woods Hole Oceanographic Institution (WHOI), National Institutes of Health (NIH), and the Office of Naval Research (ONR).

Case Images: http://www.whoi.edu/csi