WHOI-2008-02

Woods Hole Oceanographic Institution

Imaging Procedures for Stranded Marine Mammals

by

Darlene R. Ketten and Eric W. Montie

Biology Department
Woods Hole Oceanographic Institution,
Woods Hole, Massachusetts

Department of Otology and Laryngology
Harvard Medical School
Boston, Massachusetts

January 2008

Technical Report

Funding was provided by the Office of Naval Research through
Contract No. N00244-071-0022.

WHOI-2008-02

Imaging Procedures for Stranded Marine Mammals

by

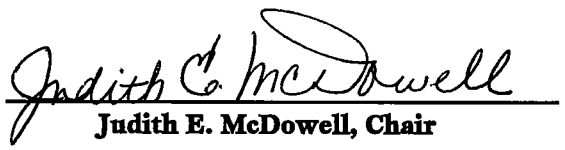
Darlene R. Ketten and Eric W. Montie

January 2008

Technical Report

Funding was provided by the Office of Naval Research through
Contract No. N00244-071-0022.

Approved for Distribution:



Judith E. McDowell, Chair

Department of Biology

INTRODUCTION

This section provides an introduction to biomedical imaging techniques and guidelines for diagnostic imaging of marine mammals to assist with both live examination and necropsy procedures. The procedures described are based on imaging equipment and techniques that are relatively common in human and veterinary facilities and to provide the majority of stranding response groups with the most likely options that will assist their efforts. The imaging techniques described include basic radiography, computed tomography (CT), and magnetic resonance imaging (MRI) and are applicable to both live and post-mortem cases. Special emphasis has been placed on whole body, airway, head and ear imaging procedures. Sub-sections cover basic information on the basic principles and appropriate applications for radiography vs. CT vs. MRI, handling and preparation of live and dead animals in clinical settings, and image and data formats that may be encountered. The protocols are also listed in outline form in order to provide a rapid overview. The introductory discussion of principles behind techniques is not required to employ the protocols but does provide additional information that can aid in deciding which techniques are most efficacious and what the limitations are for interpretation of imaging data. Examples of some pathology imaged with these procedures are also provided.

Biomedical Imaging – An Introduction to Techniques

Radiography

Plain film radiographs, commonly called X-rays, are the most common non-invasive imaging method used by both physicians and veterinarians. Most veterinary clinics and some stranding networks will have X-ray machines. The equipment for plain films is relatively inexpensive and broadly available and the exams are relatively rapid depending upon the size of the animal vs. the tissue type and pathology present, plain film X-rays may be sufficient for diagnosing some conditions, including fractures, hemorrhages, tumors, and foreign object. They are particularly useful for determining the presence of any metallic objects and thus may be an important rapid screening device to use prior to attempting MRI on any animal.

Radiographs are essentially a shadow picture obtained by exposing a photosensitive film with X-ray energy that has passed through an object or animal. Differences in the density of the tissues or objects in the animal being imaged determine how much of the X-ray beam will be attenuated or passed through to the film. The denser a structure is, the fewer X-rays that pass through it. These differences are encoded as grades of exposure ranging from black (e.g. air and therefore full pass of X-ray energy, fully exposed film) to white (fully attenuated or absorbed X-rays). Areas of very low density, such as normal, air-filled lungs, will be dark grey to black with mottling of dark to light grey where there are thicker alveolar walls, bronchioles, etc. The cartilage and soft tissue areas of the trachea will be a lighter gray and all of it will be overshadowed by the brighter, white bone of the ribs. Very dense objects such as bones, teeth, calcium deposits in tumors, and plastic or metallic foreign bodies absorb or reflect the X-ray beam and appear bright white. Muscles and organs (heart, liver, and spleen) are shades of grey.

In a plain film or radiograph, we are actually looking at a 2D composite of all the overlying tissue. They can therefore be difficult to interpret because of the multiple layers of tissues, especially if the animal is very large or if the tissues to be visualized are surrounded by several denser or complex structures. In most cases, on simple radiographs, pathologies are observed as inappropriate darker areas if there is a collection of fluid (hematoma), an air pocket (pneumothorax or emphysema), or bone separation (fractures) or as denser and whiter areas, sometimes called “clouding”, as in the case of advanced pneumonia, consolidated lungs, fibrous tissues, or inflammatory regions. Resolution of a plain film is a balance of the voltages used and length of exposure vs. the mass of the animal and density of tissues, as

well as some other factors, such as screens, film cassette types, and emulsion quality that are related to reducing scatter and improving grey levels. All of these factors should be discussed with the technician or radiologist conducting the exam in order to determine how to obtain the best images in the shortest time for the animal and the pathology you are investigating.

Computed Tomography

"Computed Tomography" refers to the production of sectional images of an animal or specimen. The name derives from the Greek *tomos* meaning slice vs. *topos* meaning surface (figure 1) as well as the use of computers rather than film to acquire the imaging data and reconstruct it as sections. . Sectional imaging is analogous to slicing a loaf of bread and acquiring an image of whatever is in each slice. In standard X-rays, the entire loaf would be visible with all internal structures as overlapping shadows. In one sense, a CT image is a tomogram and a plain film or standard X-ray is a topogram. In comparison to plain film X-rays, tomograms provide far greater detail but they are more time-consuming and generally more expensive. Sectional CT images amount to a digital dissection, allowing us to view internal structures within individual sections in far greater detail because we do not have the interference of the surrounding tissues, but they also therefore require somewhat longer examination times and may involve more exposure to X-rays or use of tranquilizers than do plain films. Most hospitals and an increasing number of veterinary facilities have one or more tomographic machines.

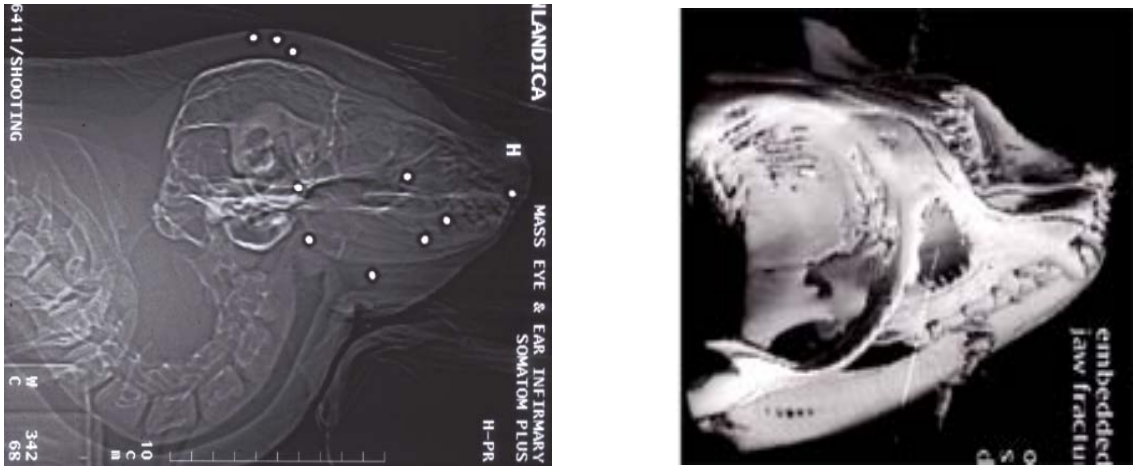


Figure 1. A CT topogram or scout image (left), which is similar to a plain film, of a live seal that had been shot in t he head, showing multiple metal fragments and a 3D reconstruction (right) from the CT images of the animal demonstrating the extent and pattern of the fractures that resulted in relation to the fragments.

The two most common forms of sectional imaging are based on X-ray and on magnetic resonance techniques. Technically, the term computed tomography applies to both, but the phrase and its abbreviation, CT, is most often used to mean sectional imaging from X-rays while magnetic resonance or nuclear magnetic resonance imaging is called MRI or NMR. These two techniques are complementary. CT can image both soft and hard tissues but is especially well suited for any exams requiring information on bones, foreign objects or air filled spaces. MRI provides superior images for soft tissues, but, because the technique depends upon having hydrated tissues, it cannot accurately represent bony or heavily aerated tissues, and because it employs shifting magnetic fields, it is precluded from use in any animal that may have any metal fragments.

CT: X-ray Computerized Tomography

Particular indications for CT imaging include pathologies that involve any form of bone lesion, aerated tissues, and mixed bone and soft tissue evaluations, such as trauma cases, intracranial hemorrhage, fracture evaluation, dislocations of any bony structures, foreign bodies, and diagnosis of primary and secondary neoplasms of the liver, kidney, brain, lung, and bone, as well as tumor staging.

CT images, as noted above, like plain films, are based on X-ray attenuation, but an array of detectors linked to a computer rather than film are used to acquire the image data. A computer controlled table (gurney) moves the patient through a doughnut shaped opening in the housing (gantry) that contains the X-ray tube. As the tube rotates along the arch of the housing and the table moves through the gantry, pulses of X-rays are emitted at multiple positions and recorded at the detectors below the patient. A grid or collimator overlying the detectors assists in reducing scattered beams from being recorded. Analogous to the process in plain films, the resolution of the digital images that are produced is based on a complex combination of the kV; exposure time; number of pulses; collimators; detectors; and table speed. In addition, modern scanners have two basic modes, single slice and spiral. In the former, the table moves in increments from 1 to 10 mm and images are produced at matching thicknesses with one traverse of the tube per slice. In the spiral mode, the table moves at rates of 0.5 to 10 mm per second, with the tube moving, and the data being collected continuously. This method is far more rapid and allows slice reconstruction at thicknesses from 0.1 mm to 10 mm from one data set.

Both forms of acquisition also can be used for imaging soft or hard tissues, which is a parameter determined through the acquisition kernels, a term referring generally to the reconstruction algorithm used to transform the attenuation values into grey scale values. The type of exam, spiral or single, the table speed, and the kV and time are generally determined based on the body area, e.g.; head or abdomen, and the principal tissue type, soft or bone, that is of concern. For example, for CT imaging of the thorax and abdomen, a protocol using a 10-mm slice thickness and soft tissue kernels is common because these structures are large. Lung scans use a 3 mm and specialized protocol to emphasize fine differences in the aerated passages, while exams of the middle and inner ear use an even smaller slice protocol of 0.,5 mm acquisitions with 0.1 mm image reconstruction an high resolution bone kernels. Some of these numbers will vary according to manufacturer of the scanner, but in most facilities, at least some spiral imaging will be available and in all CT facilities, bone and soft tissue kernels for most organ types are available. In addition, CT images can now be reconstructed at virtually any angle required even though they are acquired in all cases in a standard transaxial plane.

As in a standard radiograph, the CT images are shown as grades of grey that relate to the amount of X-ray beam that was attenuated by tissues of the specimen. Attenuation is affected by many factors but is largely determined by density of the exposed tissues. In the images, the higher the attenuation, the denser the object, the whiter the appearance of the structure. In addition to the 256 grayscale used to tint each pixel, a second value, the Hounsfield units (HU), is also available from the CT data. Hounsfield, named after one of the inventors of CT methodology, is a measure of attenuation standardized to water. Air is therefore -1000 HU; water, 0. Higher numbers indicate increasing density. Mammalian tissues typically range between -100 (fats) to +3100 (very dense bone) but even metals can be measured up to approximately 41000 HU on an extended scale. Hounsfield units therefore offer a far greater range of information about the density of and physical characteristics on a per pixel basis than the 256 grey scale used for imaging alone. They are especially useful in diagnosing hyper mineralization, the quality or age of a blood deposit (clots are denser and have higher HU values than fresh, fluid blood), etc.

This brings us to a rather technical area about how images are formed using kernels and look-up tables. It is not necessary to understand these fully but it is important to be aware of them. The attenuation values or coefficients are the raw data of the CT scan. These values are processed into image files by applying a convolution algorithm, which briefly amounts to combining multiple values that are obtained from each X-ray projection, weighting them according to what tissue characteristics are of interest, and then providing a third output that results after this “filtering” has been applied. At the user interface this operation amounts to choosing first a protocol that weights the data for better definition of bone vs. soft tissues and then atop that adding a second weighting that emphasizes a range of Hounsfield values, commonly called windows and centers. Therefore, for any scan session you need to indicate to the

technologist whether you need primarily images of the soft tissue or bony structures or both. Applying the proper kernel is particularly important if you are planning to make measurements of tissue dimensions. Reformats of any CT data set must be done from the raw data set for accurate information, not from the image files, since these are already set by the kernel previously chosen and although the appearance may change, the representation of the anatomy may be distorted. In most facilities, only the image files are archived because the raw data files are both large and time-consuming to copy. However, it is important to archive both raw data as well as the basic image files whenever possible, especially if the animal is rare, has an unusual condition, is a legal case, or if reformats for other tissue types may be needed later.

Contrast agents may be employed in live cases by injecting an agent that is radio-opaque and which assists with tracking fluid accumulations or flow rates. These are most commonly used to determine presence of tumors or for cardiac assessments. Contrast agents require veterinary oversight and although reactions are rare in non-humans, precautions to treat a reaction should be in place before they are administered.

CT Summary

In general, CT provides very high resolution imaging in a relatively short time frame. It is the primary choice for live animals that require quick exams and provides the best images for diagnoses of bone, airway, or foreign body pathologies.

Tranquilization may be required, but for very short spiral CT exams, it may be sufficient to simply restrain the animal, or in the case of a captive animal, to train it to hold a position for 5 minutes or less. The procedure requires exposure to X-rays but for most exams, this does not represent a significant increase over plain film procedures. It is limited to animals under 250 kg., less than 1.7 meters in length and less than 70 cm in diameter. Some larger gantry machines that accommodate greater weights are available but are rare.

Because these scanners are generally located in clinical settings, proper precautions for both live and post-mortem specimens are required to prevent contamination as described in the outlines below. The first scan will be a “topogram” or “scout” of the whole animal or specimen (Figure 1). This image resembles a regular X-ray and is used to determine the regions of the animal that will be examined. Following the scout and the designation of scan sequences, the raw data for generating section images are acquired by either spiral or single section scan sets. The typical whole body exam will require approximately 20 minutes of table time to acquire the data, at which point the animal can be removed from the scanner. Image reconstruction may require typically 30 minutes to 2 hours to obtain complete sets of all image sets depending upon the number of types of tissues and reconstructions required. Both 2D and 3D images are possible, as well as multi-tissue displays and measurements of significant features. Bone or soft tissue kernels or both should be used as appropriate for each case rather than attempting to convert one series to the other by re-windowing from just the image files. Contrast agents may assist with diagnosis of hemorrhage, tumors, or heart conditions, but require careful administration under veterinary supervision. Images are generally exportable as hard-copy films or digital DICOM format images (described below) on CD or other electronic media.

MRI: Magnetic Resonance Imaging

Magnetic resonance imaging (MRI) is another increasingly available method for sectional anatomy imaging. While CT units are still more common than MRI, more clinics have both than before and if available, MRI is extremely valuable for many soft tissue exams. The strength of MRI is the ability to detect differences in soft tissue, whereas radiography and CT which provides excellent examinations of bone and air-filled spaces, provides less resolution than MRI for most soft tissues. For example, MRI is an excellent tool to view the brain and gastro-intestinal or reproductive systems, because it provides good resolution of subtle differences in soft tissue hydration, such as between white matter and grey matter. On the other hand, MR cannot be used to image most bony structures or highly pneumatized areas. It also is

precluded for subjects that have any metal in or on them and because of longer imaging times, generally will require tranquilizers or anesthesia for most veterinary cases.

MR images are acquired by generating a magnetic field across the subject. Hydrogen nuclei (protons) present in the tissue align with this field. Pulses of radio waves are then broadcast into the sample, energizing the hydrogen protons, causing them to resonate and deflect away from the magnetic field. Once the radio pulse stops, the protons decay (i.e. return or "relax") to their original alignment. A receiver coil detects the number of resonating protons and the changes in their spin, which is then translated into a signal intensity value. Tissues with high levels of fluids (e.g. blood, cerebrospinal fluid) have high proton content and produce high signal intensities; those with low fluid content (e.g. lungs or dense bone) produce low signal intensities. Analogous to the process of converting X-ray attenuations in CT, the MR signal intensities are transformed by a convolution process also into grey scales for imaging. High intensity, high fluid content areas are generally imaged as bright white while lower fluid content structures are in graded shades of grey. Bone and air have virtually no fluid content and therefore produce little or no signal and are both black. These parameters can be adjusted however to preferentially enhance imaging of some tissue types. Typically, two types of images can be produced, depending on what stage of decay is being measured (i.e. how long after the radio pulse transmission is stopped): T1-weighted images are taken early in the decay process, while T2-weighted images are taken later (shorter and longer relaxation times). T1 images, which have greater resolution, are most useful for examining anatomic detail. By comparison, T2-weighted images have greater contrast and are most useful in diagnosing gross pathologies such as cysts or some tumors because these pathologies typically contain relatively high amounts of water, which results in high signal intensities that appear bright white.

As with CT, the images are usually provided in sections in coronal, sagittal and axial planes and at 1-10 mm thicknesses. The absolute level of resolution depends partly on the number of signal sequences that are employed and therefore on the total time required for the exam. The more pulse sequences measured and compared, the higher the resolution, the longer the exam for each case study. The average head or abdominal MRI exam will usually require about 30-45 minutes of table time and have a resolution of 2-8 mm per pixel. Resolutions of fewer than 2 mm will normally require upwards of one hour of signal acquisitions and are therefore practical generally for post-mortem cases. As with the kernels in CT, most MRI units have multiple sequence modes that can be employed and should be discussed with the technologist prior to the start of the exam. In addition to the T1 and T2 sequences described above, some of the more commonly available protocols include CISS and fat-suppressions as well as some newer techniques for viewing lungs or cardiac functions.

Contrast agents are also available for use in MRI, and as with CT agents, they should be employed only with veterinary approval and oversight and precautions to treat a reaction should be in place before they are administered.

MRI Summary

MRI provides exceptional soft tissue imaging but requires longer table time for most exams than CT. It is the primary choice for animals or specimens that requires good soft tissue differentiation and provides the best images for diagnoses of brain and abdominal soft tissue pathologies.

Tranquilization is likely to be required in all live cases. Size limitations depend upon the unit as well as whether a body, head or surface coil will be used. In general, animals must be less than 200 kg and the exam area will less than 1 meter in length and less than 55 cm in diameter.

Because these units are generally located in clinical settings, proper precautions for both live and post-mortem specimens are required to prevent contamination as described in the outlines below. As with CT, MRI requires a scout acquisition to determine orientation and regions to be imaged. Following this process, the full MR sequences are applied. The typical whole body exam will require approximately 40 minutes of table time for each sequence or mode chosen and 30 minutes to 2 hours to obtain complete sets of images, depending upon the number of types of tissues and reconstructions required. Both 2D and 3D images are possible. Contrast agents may assist with diagnosis of hemorrhage, tumors, or heart conditions,

but require careful administration under veterinary supervision. Images are generally exportable as hard-copy films or digital DICOM format images (described below) on CD or other electronic media.

PRACTICAL GUIDELINES

Choosing the Right Imaging Modality

There are numerous factors to consider when determining which imaging technique(s) to use for a stranded marine mammal. These include equipment availability, amount of time available, size of the animal, specimen condition, and suspected pathology type. Below are guidelines for each modality as well as procedures for preparing the animal or specimen. It should be noted that the times listed do not include specimen preparation or positioning prior to scanning. Dimension and weight tolerances vary by scanner manufacturer; those listed are typical for most recent units. For post-mortem material, limits include all containers, packaging, and fluids surrounding the specimen when scanned.

Procedural Outlines and Size Limits

Species size categories employed in the protocols below assume the following limits:

- Small
  - Weight < 250 kg
  - Maximum diameter < 58 cm
  - Maximum girth < 80
- Medium
  - Weight < 400 lb
  - Maximum height < 70 cm
  - Maximum girth > 80 cm
- Large
  - Weight > 400 lb
  - Maximum height > 70 cm
  - Maximum girth > 80 cm

Plain film X-rays

- Time required: Seconds per exposure for each X-ray
- Size restriction: NONE
  - Weight: No weight limits; Resolution decreases with body mass in the field of view
- Specimen condition
  - Live
  - Fresh dead
  - Moderate to advanced decomposition
- Particular applications
  - Fractures
  - Hemorrhages
  - Pneumothorax
  - Tumors
  - Foreign bodies, esp. metallic objects
  - Pre-screening for MRI

CT

Time required: 0.5 to 10 min depending upon body region examined
Size restrictions: small to medium species for whole animals or parts of small, medium and large species that fit within the following limits
- Weight: <250 kg
- Maximum dimension: <70 cm (diameter)
- Maximum length: 1.7 meter in length

Specimen condition
- Live
- Fresh dead
- Moderate to advanced decomposition

Particular applications
- Trauma
- Gunshot wound
- Fractures
- Middle and inner ear pathology
- Skull and spinal column pathology
- Osteolytic disease
- Joint pathology
- Air emboli
- Foreign bodies
- Abdominal organ injury
- Hemorrhage
- Tumors

MRI

Time required: 15-45 minutes per scan
Size restrictions: small species for whole animals or parts of small, medium and large species that fit within the following limits
- Weight: <200 kg
- Maximum dimension
- Head coil: 80 cm circumference (girth) or ~20 cm diameter
- Tunnel opening: 58 cm diameter
- Maximum length: 1 meter

Specimen condition
- Live
- Fresh dead

Particular applications
- Trauma
- Neurological symptoms (aberrant behavior, seizures, tremors, circling)
- Soft tissue damage
- Brain lesions
- Hemorrhage
- Tumors
- Circulatory system function
- Cysts

Biomedical Imaging Procedures for Stranded Animals

Applicable imaging approaches differ depending upon many factors, including whether the animal is

alive or dead, whether sedation is feasible, what is the specimen size and weight, and what imaging options are available. As outlined above, both CT and MRI have weight and aperture limits that may vary by manufacturer but in general limit the exams to small to mid-size cetaceans and pinnipeds or to post-mortem exams of parts of larger animals

If all imaging options are available and there is time, the optimal scenario would be to perform a screening radiograph of the head and body of most specimens and then pursue CT or MRI scans of the body and head for high resolution imaging of any areas of interest. The recommendations below therefore provide procedures for screening with plain films followed by CT and MRI. If CT is available, the plain films may be eliminated. If MR is available, either plain films or CT should be used first to screen the specimen or animal to be certain no metallic components are present in the gut or embedded in any tissues prior to MRI use.

We recommend employing CT rather than MRI for full body scanning because MR imaging of the whole body requires extensive table time and has imaging length limits that make it impractical for all but the smallest post-mortem specimens. It also will not provide adequate resolution of many structures in the head, thorax or abdomen (e.g. ears, lungs or other bony or gas-filled regions) that are important for assessing strandings. When possible, we do recommend performing MRI of the head, because this modality provides the best resolution of the brain.

Prior to the examination, it is advised that you contact the facility, inform them of the species, size category and exact gross condition of the animal. Basic details they will require are live vs. dead, whether intact or with external trauma, and if dead, the state of decomposition. Handling for each of the stages in post-mortems is discussed below.

Animal/Specimen Preparation:

All Live Animals

Transport Preparations

a. Document the condition of the animal with photographs and notes.

b. Clean animal to the extent feasible to remove superficial sand and mud as the minerals in these particles create image artifacts and may cause injury in MRI exams. It is advisable to photograph again after cleaning and prior to transport.

c. Transport to radiography facility.

Any carrier normally used for animal transport is acceptable but care should be taken to consider both the animal's health and that of other patients in the facility. It is recommended that an arrival time be agreed upon, and if it is a clinical facility that entrance location and access to a transport gurney or cart be arranged in advance. It may also be advisable to alert the security services in larger hospitals in order to arrange parking for any large transport vehicles.

NOTE: Photography in the facility of the procedure is also recommended however, **it is imperative to get permission from any clinical facility, particularly a human radiology unit before any photography is done**. Although most facilities will allow photographs, they may be understandably limited or forbidden for reasons of patient privacy protection.

d. Prepare the animal for placement on the imaging table and examination.

Anesthetize or tranquilize the animal based on best veterinary practice, anticipating up to 10 min of sedation needed for handling for plain films and 30 minutes minimum for one CT or MRI procedure. During the exam an attendant or veterinarian should be in the room and wearing proper shielding for CT or MRI during the exam in order to observe the condition of the sedated animal and to notify the scanner technologist of any motion while scanning.

All Post-mortem Animals and Extracted Tissue Preparation:

Most of the preparation of postmortem material is the same as for live described above, with the exception of course of sedation and the added need for proper sealed containers for any decomposing material.

Chilling, Freezing, Fixation

Preparation and scanning of the animal or tissues should be done as soon as possible post-mortem or post-extraction, preferably without the need to hold for more than a few hours. . In most cases, there will not be a preparation area connected to the scanner facility therefore preparation will need to be done on the beach or in the necropsy facility.

All bodies should be cooled as soon as possible. If several hours holding are required, it is reasonable to consider placing dry ice if available in the mouth cavity and blowhole to cool the brain and in the rectum to chill the abdominal area. The body may also have ice packs or cold packs applied to the abdomen and head regions.

If the animal or tissues must be held longer, they should be packaged (described in the next section) in a container with as little air surrounding the specimen as possible and placed in a chiller at approximately +4 degrees C. If a chiller is not available, the bagged animal can be placed in a vat of ice or in a second bag with ice between the layers. The unpackaged animal should not be immersed directly into water or an ice bath in order to prevent artifactual water deposits or bloating and dilution by osmotic processes. Animals may be held in a chiller for several days if necessary. If chilling is not possible, freezing is a reasonable alternative but can create artifacts. As with chilling, the animal should be sealed in a bag with little air surrounding it and placed in the freezer by suspending or on cushioning to avoid compression of major organs during the chilling and freezing process. If frozen, the animal may be X-rayed or CT scanned without thawing. However, because MRI depends upon fluid molecular mobility, all material must be fully thawed before MRI scanning.

Formalin or other fixed materials should be handled with the usual procedures for fixation and then packaged with the same precautions against leakage or contamination as outlined below. The imaging facilities should also be contacted to ascertain if they have any regulations related to fixatives. Above all, care should be taken to prevent leaks or fumes. Small specimens can generally be scanned in their fixative in all modalities and may not require removal from containers.

Packaging, Transport Preparations

a. Photo document and clean the animal (see live above) to the extent feasible to remove superficial sand, sediments, and mud,

b. Packaging and transport to the imaging facility.

Proper packaging to prevent odor or fluid leaks during transport and imaging is the most important step in this process. Fortunately, most animals can be placed in plastic bags or body bags that may be used in a scanner and therefore can be prepared and sealed prior to entering the clinic or scanner area.

The basic rule is transport in one or more bags and examine in a triple bag. The following is an ideal situation, modify as needed:

Place the clean animal in a clear or translucent thick plastic bag with disposable diapers or other absorbent material and seal the bag securely with non-metallic tape or ties, then place it in a second sealed bag for transport, again placing some absorbent material between the bags. If available, place the double bagged animal in a body bag or other sturdy container, with sealed icepacks surrounding the animal, for transport. The main idea is to keep the animal cool and to avoid leaks from the inner or outer bags. Loose ice should

be avoided so that pooling does not occur that will interfere with MRI and make the specimen harder to handle. Tear resistant paper bags, sometimes called Grainger bags, are also available from some industrial suppliers that may be used for MRI. Be sure to check however that they do not contain any ferrous staples or other metallic elements.

As noted above, it is recommended that an arrival time be agreed upon, and if it is a clinical facility that entrance location and access to a transport gurney or cart be arranged in advance. It may also be advisable to alert the security services in larger hospitals in order to arrange parking for any large transport vehicles. It is also helpful to assure the facility that you are aware of the need for both proper containment of the specimen and consideration of their patients.

c. Prepare the animal for placement on the imaging table and examination

In general, the animal can be examined inside the double bagging. Some body bags have non-metal zippers that do not interfere with either CT or MRI and therefore will not have to be removed. In most cases, however, the animal will have to be removed from the outer packaging, but can be left in the two plastic bags. It is advisable to have clean bags to use at the scanner if needed. If performing MRI, the absorbent materials may need to be removed to reduce artifact, therefore come prepared also with gloves and trash bags to handle waste removal and minimize odor.

Scanning and Imaging Protocols:

The following procedural outlines apply to imaging both live and post-mortem material for most odontocetes, pinnipeds, and extracted mysticete and sirenian tissues. Longer sequences and higher X-ray dosages are of course possible in post-mortem or extracted tissues and multiple sequences for different tissue types should be employed for optimizing images particularly of the head areas or ears whenever time and costs allow.

Plain film radiography:

Exposures depend heavily on the thickness of the area being imaged and the relative densities of material within the image field. In the absence of prior experience with marine mammals, for small to medium animals, it is recommended that an initial image be obtained using parameters for human abdomen or for large dogs if a veterinary facility. For Large marine mammals, substantially greater voltages and times will be required for the head and lower abdominal regions. Projections to employ are as follows:

Head – Dorsal-ventral and oblique, orbit to occiput; lateral for skull base trauma.
Thorax - Dorsal-ventral
Abdomen - Dorsal-ventral and lateral for genitourinary track and for any suspicious areas.

CT

Scouts should be obtained to cover the regions of interest in as few sequences as possible. Transaxial scans should be obtained using a spiral scan protocol, particularly for live animals, and images should be formatted in both soft and bone windows. Rapid reconstruction images obtained during scanning can be used to determine whether the proper areas and detail were obtained and the animal can then be removed from the scan area while full resolution 2D and 3D images are produced and reviewed.

The following values should be considered as general suggestions and modified as necessary to provide best image quality. More than one spiral set may be required to cover the entire body. Overlap sequential spiral sets by one full section. The following are suggested limits for image slice thickness for proper diagnostic surveys of each region:

Head:

3 mm increments through the entire head.
3mm feed (pitch of 1)
1 to1.5sec rotation time
180 mAs, 120kv

Bone reconstruction:

1 to 3mm U90s , w4000 c1000

Soft tissue reconstruction:

1 to 3mm H30 or H40 , w500 c50

Brain:

1 mm increments, reconstructed as for soft tissue above

Ears:

0.5mm slice increments
0.5mm feed (pitch of 1)
1.0sec rotation time
180 mAs, 120kv

Bone reconstruction:

0.1 to 0.3 mm U90s, w4000 c1000, smallest FOV possible

Thorax/Abdomen:

3-5 mm increments through the entire head.
5 mm feed
1 to1.5sec rotation time
180 mAs, 120kv

Bone reconstruction:

1 to 3mm U90s , w4000 c1000

Soft tissue reconstruction:

1 to 3mm H30 or H40 , w500 c50

Lung Image reconstruction:

If available use specialized lung kernel or reconstruct with windows near +1200 and centers near -200.If 180 eff. mAs is not available, request exposures to produce ~160 mAs. Lower voltages may decrease image quality particularly in odontocetes and increase artifacts in the images, especially in sections with dense bony elements. In addition to the image parameters noted above which are typical for odontocetes, the following values should be tried to optimize imaging of any animal:

Bone window reconstructions:

High resolution 80-95 kernel, w 2800 to 4000; c 600 to 1000

Soft tissue reconstructions:

Mid to soft tissue 30-40 kernel, w 200 to 500; c 50 to 150

Lung reconstruction:

Note: If any tissues are extracted and fixed for further examination it is recommended that they are rescanned as isolated tissues prior to further dissection or histological processing to optimize imaging of the substructure of the tissues of interest. The same parameters as listed above apply but the field smallest FOV possible (generally $1/10^{th}$ the maximum image field) that will encompass the specimen should be used.

MRI

MRI is recommended for the head if brain lesions or hemorrhage are suspected and for suspected abdominal lesions. **If there is suspicion of metallic fragments or contaminants present in the animal, MRI should not be attempted, regardless of whether the exam is on live or dead tissue**. Injury to a live animal or attending personnel or to the equipment can occur if ferrous material is present in the

magnetic field. Any specimen larger than 80 cm in circumference cannot be imaged in most MRI units. To optimize imaging of the brain, head coils if available should be used. In some cases, larger coils may be available by special order from the scanner manufacturer but may require a special purchase.

Before scanning the head, particularly if it is large and the brain images are the focus of the exam, the blubber, nuchal fat, and semispinalis muscle may need to be removed to decrease the circumference to fit into the head coil. If possible, scan the head outside the plastic bag and in a Grainger bag as noted above, but be certain to provide a waterproof table cover and to avoid contamination of the head coil. Excess fluid inside the bag and/or pooling around the head can cause signal artifacts, so it is important to remove standing liquid prior to scanning. Rapid reconstruction images obtained just after the acquisitions are complete can be used to determine whether the proper areas and detail were obtained and the animal or specimen can then be removed from the scan area while full resolution 2D and 3D images are produced and reviewed.

The following values should be considered as general suggestions and modified as necessary to provide best image quality. A scout should be completed to ensure that the entire region of interest, e.g., the brain or lungs will be captured in the planned MRI scan sequence. Initial images should be acquired, at a minimum, in the sagittal and coronal planes. The following are suggested limits for image slice thickness for proper diagnostic surveys of the brain:

Two-dimensional proton density (PD) and T2-weighted images acquired using a fast spin-echo sequence.

Parameters:
TE = 15/106 ms for PD and T2 respectively
TR = 8000 to 9000 ms
slice thickness = 2mm
Flip angle = 180^{o}
FOV = 240 x 240mm
Matrix = 256 x 256
Voxel size = 0.9 x 0.9 x 2.0 mm.

For thorax and abdomen, similar acquisitions may be used but slice thickness may be increased to 5-8 mm and the field of view and voxel size increased as necessary.

Viewing, Copying, Exporting, and Archiving Images and Exam Data

Plain film radiographs may be printed on X-ray film or in larger facilities are now available as on-screen, digital images in the same formats as MRI and CT images. The digital image form most commonly encountered is a DICOM format. DICOM stands for Digital Imaging and Communications in Medicine, and is a standard developed by the American College of Radiology Manufacturers Association to define the connectivity and communication protocols of medical imaging devices. It is the current, open architecture standard for biomedical images. CT and MR images in both 2D and 3D reconstructions are now generally exportable directly from the scanner as DICOM copies on CD or magneto-optical disks as well as hard-copies on film. In some cases, TIFF, or JPEG formats may also be available for export or available through post-processing at the facility or through third-party software now available for both MAC and PC computers. Some of these programs will work to a limited degree with the standard RAM in most computers, but in general 1 gB or more is recommended for full functionality of these programs.

It is strongly recommended that at a minimum all images produced during an exam be archived in DICOM formats by the facility onto CD, external hard drive, and magneto-optical disks at the imaging facility. At present, there is no common archive for marine mammal images, but several have been proposed and may become available in the near future. Raw scan data are not usually retained or archived

by the original scan facility, but it is highly recommended that raw data copies be requested in addition to the image files for all significant cases and particularly for all research or legal cases. The raw scan data are critical for any reformats, data based magnifications, or accurate HU analyses that may be required at a future date. Some facilities may be unable to archive raw data or may charge additional costs for this archiving.

Interpretation of Images

Radiographic, CT, and MR images should be interpreted by a veterinary radiologist or other professional who specializes in specific anatomical structures (e.g. head and neck trauma, ears, brain, airways, lungs, abdomen, extremities, genitor-urinary). We advise avoiding any interpretation without specific training or consultation because many conditions will produce similar grey scale appearances, and the proper interpretation depends heavily on a comprehensive, integrated analysis of multiple features including shape, position, attenuation characteristics, HU values, etc. In addition, marine mammal anatomy is sufficiently different from that of humans and most common veterinary cases that even well-trained conventional radiologists may find these images challenging. Below, we provide contact information for a list of individuals who have extensive experience in interpreting marine mammal radiographs. The listings of these individuals are an unsolicited recommendation by the authors of this manual and do not indicate an agreement for consultation by any of the listed individuals.

• Dr. Darlene Ketten, Woods Hole Oceanographic Institution, Woods Hole, MA, (508) 289-2731, dketten@whoi.edu
  Head and ear structures, lungs
  Blast trauma, fractures, bone lesions
  Image processing, 3-D reconstructions
• Dr. Eric Montie, Woods Hole Oceanographic Institution, Woods Hole, MA, (508) 289-3501, emontie@whoi.edu
  Brain, volume analyses
  Anthropogenic chemicals, biotoxins, parasites
  Image processing, 3-D reconstructions, measurements of brain regions
• Dr. Lori Marino, Neuroscience and Behavioral Biology Program, Emory University, Atlanta, GA, (404) 727-7582, lmarino@emory.edu
  Brain, volume analyses
• Dr. Sam Ridgway, Department of Pathology, University of California, San Diego, CA, (858) 534-0455, ridgway@spawar.navy.mil
  Brain pathologies
• Dr. Ted Cranford, Department of Biology, San Diego State University, San Diego, CA, tcranfor@mail.sdsu.edu
  Airways and melon
• Dr. Joy Reidenberg, Mount Sinai School of Medicine, New York City, NY, (212) 241-7563, joy.reidenberg@mssm.edu
  Larynx, airways
• Dr. Frank Fish, Department of Biology, West Chester University, West Chester, PA, (610) 436-2460, ffish@wcupa.edu
  Limb and propulsive structures

Data Analysis

Sectional anatomy imaging (CT and MRI) offers many advantages. It creates a permanent archive of images of anatomical structures, without the disruption of dissection, and allows us to view the anatomy of freshly dead, stranded specimens at a much finer scale rapidly. CT and MR images also allow us to

produce three-dimensional models of anatomical structures to examine spatial relationships and provide accurate volumetric details. Furthermore, the images can be used to guide dissections for sampling smaller pathologies that may have been missed in conventional necropsy.

CT and MRI Scans Completed in Marine Mammals

Tables below list species for which CT or MRI scans have been completed by WHOI researchers for which images are available for comparison with future exams. Access to these images can be obtained via the WHOI CSI website (www.whoi.edu/csi) as well as through links to other facilities and research laboratories listed on the site. Many other examples may be available through other laboratories or other facilities, and we encourage both the formation of a centralized data base and exchange of cross-listings of available image data sets.

Table 1. CT Scans and Archived Images of Marine Mammals
WHOI Computerized Scanning and Imaging Center (www.whoi.edu/csi)

Classification	Scientific Name	Common Name	Tissue	N
Fissipedia	Enhydra lutris	Sea otter	Head	8
Mysteceti	Balaenoptera acutorostrata	Minke whale	Head	4
Mysteceti	Balaenoptera acutorostrata	Minke whale	Ears	2
Mysteceti	Balaenoptera acutorostrata	Minke whale	Periotics	2
Mysteceti	Balaenoptera acutorostrata	Minke whale	Mandibles	1
Mysteceti	Balaenoptera musculus	Blue whale	Ear	2
Mysteceti	Balaenoptera physalus	Fin whale	Larynx	1
Mysteceti	Eubalaena glacialis	Right whale	Ear	16
Mysteceti	Eubalaena glacialis	Right whale	Vertebra	1
Mysteceti	Eubalaena glacialis	Right whale	Ribs	1
Mysteceti	Eubalaena glacialis	Right whale	Sternum	1
Mysteceti	Eubalaena glacialis	Right whale	Mandible	3
Mysteceti	Eubalaena glacialis	Right whale	Peduncle	1
Mysteceti	Eubalaena glacialis	Right whale	Pectoral flipper	1
Mysticeti	Eschrichtius robustus	Grey whale	Head	1
Mysticeti	Eschrichtius robustus	Grey whale	Ear	1
Mysticeti	Eschrichtius robustus	Grey whale	Periotics	2
Mysticeti	Megaptera novaeanglia	Humpback whale	Ears	9
Odontoceti	Cephalorhynchus hectori	Hectors dolphin	Whole body	1
Odontoceti	Delphinus delphis	Common dolphin	Head	6
Odontoceti	Delphinus delphis	Common dolphin	Whole body	11
Odontoceti	Delphinus delphis	Common dolphin	Ears	7
Odontoceti	Delphinus delphis	Common dolphin	Fluke	5
Odontoceti	Delphinus delphis	Common dolphin	Flipper	2
Odontoceti	Delphinus delphis	Common dolphin	Dorsal fin	2
Odontoceti	Delphinus delphis	Common dolphin	Peduncle	1
Odontoceti	Delphinus delphis	Common dolphin	Lungs	1
Odontoceti	Tursiops truncatus	Bottlenose dolphin	Ears	40

Odontoceti	Tursiops truncatus	Bottlenose dolphin	Head	17
Odontoceti	Tursiops truncatus	Bottlenose dolphin	Scapula	1
Odontoceti	Tursiops truncatus	Bottlenose dolphin	Vertebra	1
Odontoceti	Tursiops truncatus	Bottlenose dolphin	Sternum	1
Odontoceti	Tursiops truncatus	Bottlenose dolphin	Whole body	1
Odontoceti	Tursiops truncatus	Bottlenose dolphin	Fluke	5
Odontoceti	Tursiops truncatus	Bottlenose dolphin	Flipper	3
Odontoceti	Tursiops truncatus	Bottlenose dolphin	Dorsal fin	2
Odontoceti	Phocoena phocoena	Harbor porpoise	Ears	17
Odontoceti	Phocoena phocoena	Harbor porpoise	Head	25
Odontoceti	Phocoena phocoena	Harbor porpoise	Whole body	15
Odontoceti	Phocoena phocoena	Harbor porpoise	Dorsal fin	4
Odontoceti	Phocoena phocoena	Harbor porpoise	Fluke	7
Odontoceti	Phocoena phocoena	Harbor porpoise	Flipper	7
Odontoceti	Delphinapterus leucas	Beluga whale	Ear	2
Odontoceti	Delphinapterus leucas	Beluga whale	Brain	1
Odontoceti	Delphinapterus leucas	Beluga whale	Head	1
Odontoceti	Feresa attenuata	Pygmy killer whale	Ear	1
Odontoceti	Grampus griseus	Risso's dolphin	Ear	5
Odontoceti	Grampus griseus	Risso's dolphin	Head	8
Odontoceti	Grampus griseus	Risso's dolphin	Fluke	2
Odontoceti	Grampus griseus	Risso's dolphin	Dorsal fin	1
Odontoceti	Grampus griseus	Risso's dolphin	Whole body	1
Odontoceti	Globicephela macrorhyncus	Short-finned pilot whale	Ear	31
Odontoceti	Globicephela macrorhyncus	Short-finned pilot whale	Flipper	1
Odontoceti	Globicephela macrorhyncus	Short-finned pilot whale	Fluke	1
Odontoceti	Globicephela macrorhyncus	Short-finned pilot whale	Head	1
Odontoceti	Globicephala melaena	Long-finned pilot whale	Head	3
Odontoceti	Globicephala melaena	Long-finned pilot whale	Ear	2
Odontoceti	Globicephala melaena	Long-finned pilot whale	Vertebra	7
Odontoceti	Kogia breviceps	Pygmy sperm whale	Head	9
Odontoceti	Kogia breviceps	Pygmy sperm whale	Ear	5
Odontoceti	Kogia breviceps	Pygmy sperm whale	Fluke	2
Odontoceti	Kogia breviceps	Pygmy sperm whale	Dorsal fin	2
Odontoceti	Kogia breviceps	Pygmy sperm whale	Flipper	2
Odontoceti	Kogia breviceps	Pygmy sperm whale	Whole body	1
Odontoceti	Kogia simus	Dwarf sperm whale	Whole body	1
Odontoceti	Kogia simus	Dwarf sperm whale	Pectoral flipper	1
Odontoceti	Kogia simus	Dwarf sperm whale	Ear	3
Odontoceti	Kogia simus	Dwarf sperm whale	Dorsal fin	1
Odontoceti	Lagenorhynchus acutus	Atlantic white-sided dolphin	Ear	7
Odontoceti	Lagenorhynchus acutus	Atlantic white-sided dolphin	Whole body	21
Odontoceti	Lagenorhynchus acutus	Atlantic white-sided dolphin	Head	5

Odontoceti	Lagenorhynchus acutus	Atlantic white-sided dolphin	Throat	1
Odontoceti	Lagenorhynchus acutus	Atlantic white-sided dolphin	Lungs	1
Odontoceti	Lagenorhynchus acutus	Atlantic white-sided dolphin	Brain	1
Odontoceti	Lagenorhynchus acutus	Atlantic white-sided dolphin	Flipper	1
Odontoceti	Lagenorhynchus acutus	Atlantic white-sided dolphin	Fluke	1
Odontoceti	Lagenorhynchus acutus	Atlantic white-sided dolphin	Dorsal fin	1
Odontoceti	Lagenorhynchus albirostris	White-beaked dolphin	Ear	3
Odontoceti	Mesoplodon bidens	Sowerby's beaked whale	Head	8
Odontoceti	Mesoplodon bidens	Sowerby's beaked whale	Whole body	2
Odontoceti	Mesoplodon bidens	Sowerby's beaked whale	Ear	1
Odontoceti	Mesoplodon bidens	Sowerby's beaked whale	Tooth	1
Odontoceti	Mesoplodon bidens	Sowerby's beaked whale	Periotic	1
Odontoceti	Mesoplodon densirostris	Blainville's beaked whale	Head	4
Odontoceti	Mesoplodon densirostris	Blainville's beaked whale	Ears	2
Odontoceti	Mesoplodon europaeus	Gervais' beaked whale	Head	5
Odontoceti	Mesoplodon europaeus	Gervais' beaked whale	Ears	4
Odontoceti	Mesoplodon mirus	True's beaked whale	Head	1
Odontoceti	Monodon monoceros	Narwhal	Head	2
Odontoceti	Monodon monoceros	Narwhal	Ears	2
Odontoceti	Mesoplodon sp.	Beaked whale	Head	2
Odontoceti	Mesoplodon sp.	Beaked whale	Ears	1
Odontoceti	Physeter catadon	Sperm whale	Ears	9
Odontoceti	Physeter catadon	Sperm whale	Head	1
Odontoceti	Physeter catadon	Sperm whale	Vertebra	1
Odontoceti	Physeter catadon	Sperm whale	Ribs	2
Odontoceti	Physeter catadon	Sperm whale	Humerus	1
Odontoceti	Physeter catadon	Sperm whale	Chevron bone	1
Odontoceti	Peponocephala electra	Melon-headed whale	Fluke	1
Odontoceti	Stenella attenuata	Pantropical spotted dolphin	Ears	3
Odontoceti	Stenella attenuata	Pantropical spotted dolphin	Head	1
Odontoceti	Steno bredanensis	Rough-toothed dolphin	Head	1
Odontoceti	Steno bredanensis	Rough-toothed dolphin	Tooth	1
Odontoceti	Steno bredanensis	Rough-toothed dolphin	Ears	1
Odontoceti	Stenella clymene	Clymene dolphin	Ears	1
Odontoceti	Stenella coeruleoalba	Striped dolphin	Ears	2
Odontoceti	Stenella coeruleoalba	Striped dolphin	Head	2
Odontoceti	Stenella coeruleoalba	Striped dolphin	Fluke	1
Odontoceti	Stenella coeruleoalba	Striped dolphin	Flipper	1
Odontoceti	Stenella frontalis	Atlantic spotted dolphin	Whole body	1
Odontoceti	Stenella longirostris	Spinner dolphin	Ears	1
Odontoceti	Ziphius cavirostris	Cuvier's beaked whale	Tympanic	1
Odontoceti	Ziphius cavirostris	Cuvier's beaked whale	Periotic	2
Odontoceti	Ziphius cavirostris	Cuvier's beaked whale	Ears	3
Odontoceti	Ziphius cavirostris	Cuvier's beaked whale	Head	2

Odontoceti	Ziphius cavirostris	Cuvier's beaked whale	Liver	1
Pinnipedia	Cystophora cristata	Hooded seal	Flipper	1
Pinnipedia	Callorhinus ursinus	Northern fur seal	Ears	1
Pinnipedia	Callorhinus ursinus	Northern fur seal	Head	1
Pinnipedia	Mirounga angustirostris	Northern elephant seal	Head	7
Pinnipedia	Mirounga angustirostris	Northern elephant seal	Temp bone	1
Pinnipedia	Mirounga angustirostris	Northern elephant seal	Skull	1
Pinnipedia	Mirounga angustirostris	Northern elephant seal	Flipper	2
Pinnipedia	Phoca groenlandica	Harp seal	Head	3
Pinnipedia	Phoca groenlandica	Harp seal	Flipper	1
Pinnipedia	Phoca groenlandica	Harp seal	Whole body	2
Pinnipedia	Phoca vitulina	Harbor seal	Head	17
Pinnipedia	Phoca vitulina	Harbor seal	Ears	5
Pinnipedia	Phoca vitulina	Harbor seal	Whole body	4
Pinnipedia	Phoca vitulina	Harbor seal	Flipper	3
Pinnipedia	Zalophus californianus	California sea lion	Ears	10
Pinnipedia	Zalophus californianus	California sea lion	Skull	1
Pinnipedia	Zalophus californianus	California sea lion	Head	13
Pinnipedia	Zalophus californianus	California sea lion	Flipper	2
Sirenia	Dugong dugon	Dugong	Head	3
Sirenia	Dugong dugon	Dugong	Whole body	1
Sirenia	Trichechus manatus	West Indian manatee	Head	2
Sirenia	Trichechus manatus	West Indian manatee	Ears	2
Sirenia	Trichechus manatus	West Indian manatee	Skull	1
Sirenia	Trichechus manatus	West Indian manatee	Flipper	1
Sirenia	Trichechus manatus	West Indian manatee	Fluke	1

Table 2. Marine Mammal Brain MRI's
E. Montie, 2005

Field ID	Species	Common Name	Sex	Length
CCSN04-120-Pv	Phoca vitulina	harbor seal	m	86
CCSN04-177-Dd	Delphinus delphis	common dolphin	m	209
CCSN04-186-Hg	Halichoerus grypus	grey seal	m	113
CCSN04-191-Dd	Delphinus delphis	common dolphin	f	207
CCSN04-195-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	m	192
CCSN04-217-Dd	Delphinus delphis	common dolphin	f	210
CCSN04-218-Dd	Delphinus delphis	common dolphin	m	139
CCSN04-219-Dd	Delphinus delphis	common dolphin	f	169
CCSN05-014-Dd	Delphinus delphis	common dolphin	f	205
CCSN05-014-Fetus-Dd	Delphinus delphis	common dolphin	m	41
CCSN05-037-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	f	206
CCSN05-038-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	f	208
CCSN05-039-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	f	211
CCSN05-039-Fetus-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	m	44
CCSN05-040-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	f	204
CCSN05-040-Fetus-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	m	54
CCSN05-084-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	m	156

CCSN05-231-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	f	137
CCSN05-232-La	Lagenorhynchus acutus	Atlantic white-sided dolphin	f	185.5
CCSN05-236-Dd	Delphinus delphis	common dolphin	m	179
CCSN05-237-Dd	Delphinus delphis	common dolphin	m	166

Example Pathologies Imaged by CT and MRI

The following figures provide examples of scans that show normal and pathologic anatomy in marine mammals scanned by CT and MRI. They also illustrate the comprehensive detail that can be gained by combining biomedical imaging, necropsy and dissection, and histopathological analysis.

Figure 2. CT scans of a beaked whale (Mesoplodon densirostris) head with bilateral mandibular fractures. Left**:** coronal 2D view showing the major transverse mandibular fracture. Right: A ventral view of the 3D reconstruction of the same animal shows the position of multiple mandibular fractures and the position of dislodged bone fragments. Close examination of the 2D images showed little bleeding in the fracture zones which suggested these were post-mortem injuries, which was later confirmed on necropsy and by histology of the adjacent bone and soft tissues. (Images copyright D. Ketten, 2005)

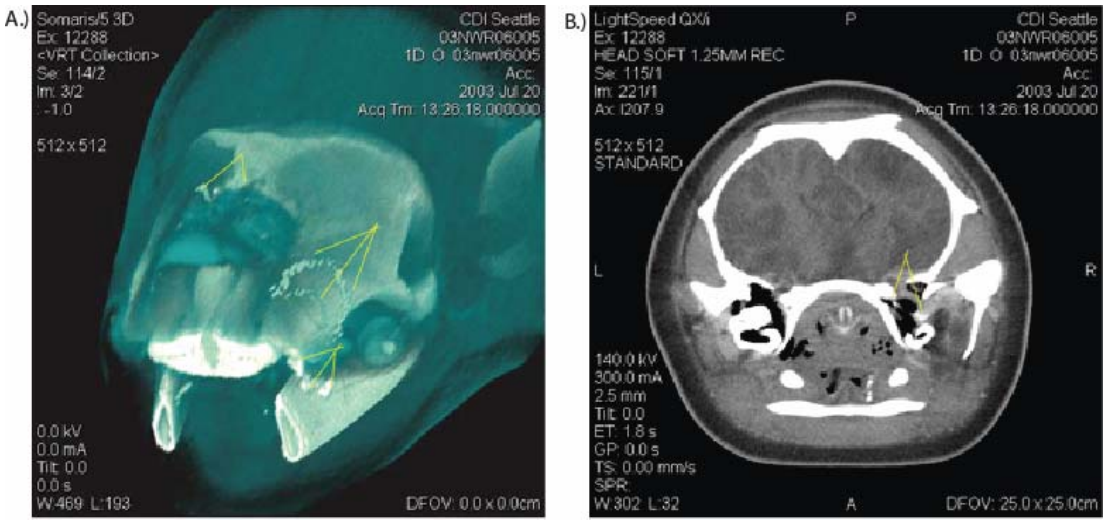


Figure 3. CT scans of a harbor porpoise with calcified parasites. Left: A three-dimensional reconstruction of the mid-region of the head shows multiple extensive calcified parasites (white tracks, yellow arrows) that were located near the right eye and extended over the melon. Right: A two-dimensional transaxial image at the level of the ears shows a peribullar soft tissue mass with small calcified inclusions that represent a group of nematodes and calcified cysts, (yellow arrows). Additional cysts are evident as bright white inclusions in the tissues on the right of the esophagus. Parasites are a common finding in this species in the middle ear and retrobullar sinuses, but the extensive calcified tracks are an unusual finding. (Images copyright D. Ketten, 2005)

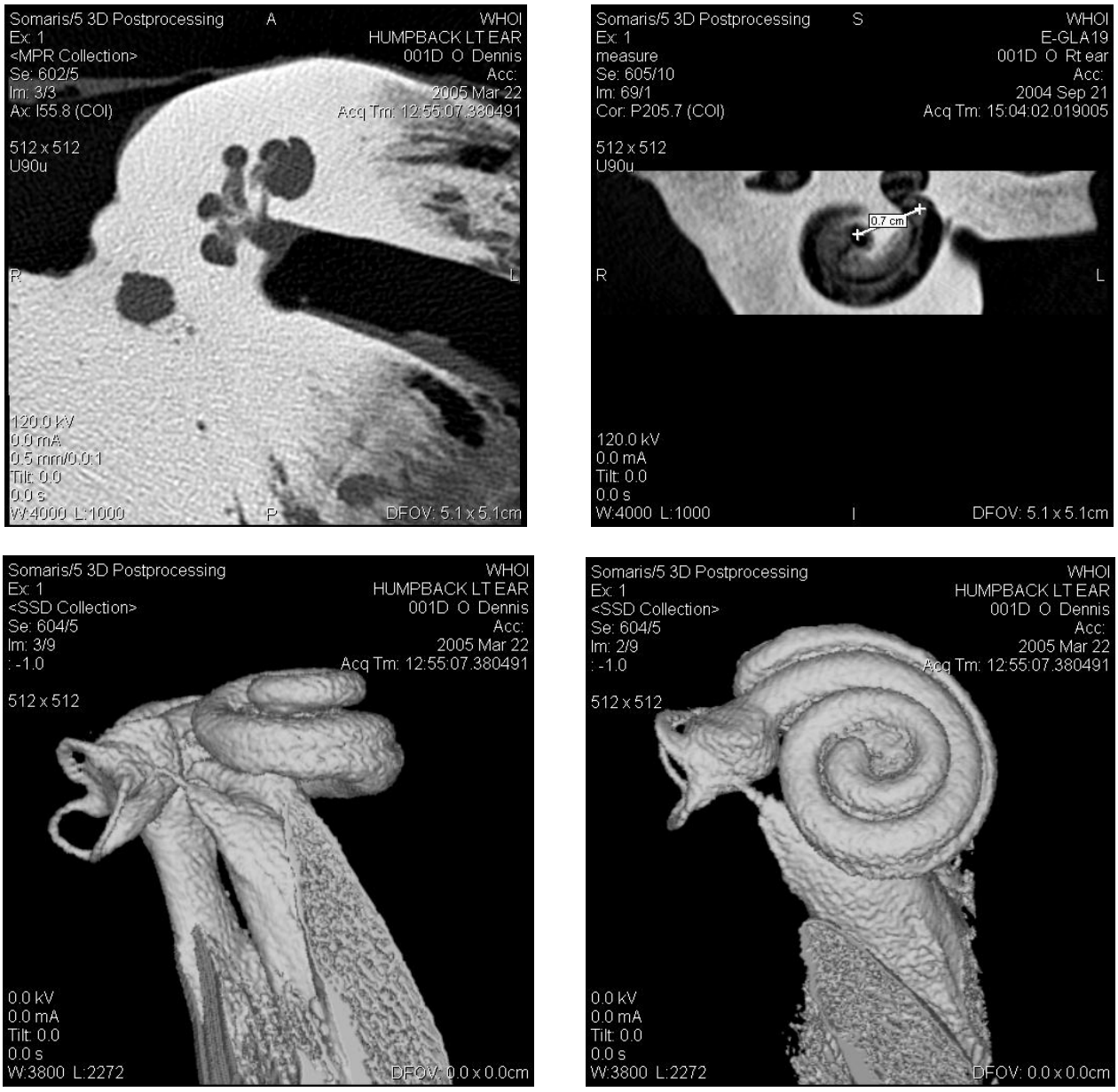


Figure 4. Mysticete Inner Ears. Two-dimensional CT images (top) show mid-modiolar and coronal views of the inner ear of a right whale (Eubalaena glacialis) that illustrate the number of turns (left) as well as soft tissue neural tracts (right). Three-dimensional images of a humpback whale ear (Megaptera novaengliae) in similar orientations show the dimensions and angular relationship of the vestibular canals as well as the cochlear canal. (Images copyright D. Ketten, 2004)

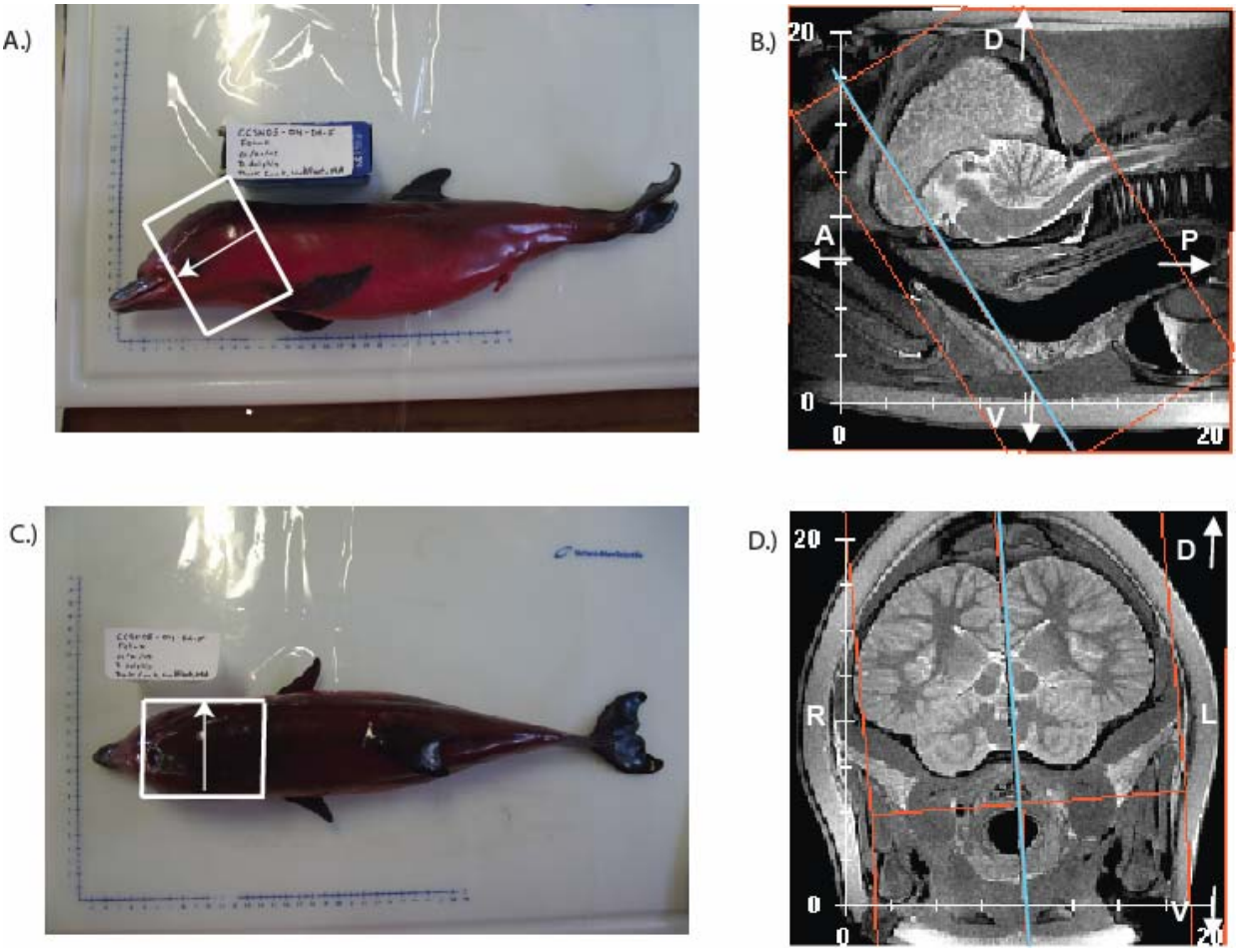


Figure 5. Photographs of a common dolphin fetus and MR images of an Atlantic white-sided dolphin sub adult brain. A-B.) Coronal orientation (from occipital lobe to frontal lobe). The plane of section is parallel to both the posterior boundaries of the occipital lobe and the cerebellum. C-D.) Sagittal orientation (from left hemisphere to right hemisphere). The plane of section is mid-sagittal. The blue line indicates the plan of section in panels B and D. D = dorsal; V = ventral; A = anterior or towards snout; P = posterior or towards fluke; L = left; R = right.

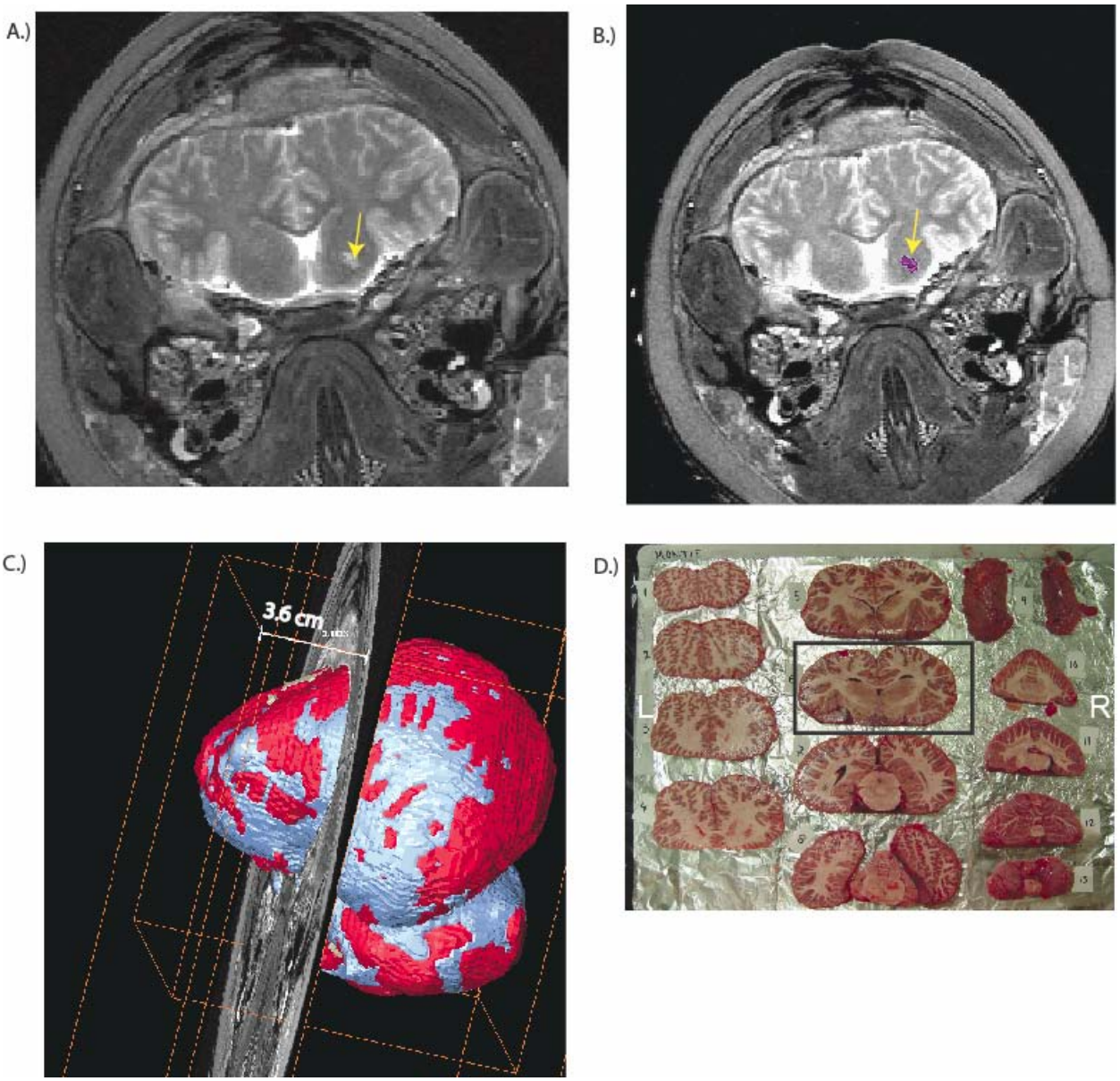


Figure 6. 3D models of the brain in conjunction with virtual reconstructions of lesions as a guide to small lesions. A.) The brain lesion in this Atlantic white-sided dolphin (indicated by the yellow arrow) was identified by MR imaging. A 3D model of the brain was constructed to measure the distance of the lesion (purple) from the anterior boundary of the frontal lobe (3.6 cm). Red = grey matter; blue = cerebrospinal fluid. This distance was measured on the gross brain to find the lesion. The black rectangle indicates the brain section that contained the lesion. (Images copyright E. Montie, 2006)

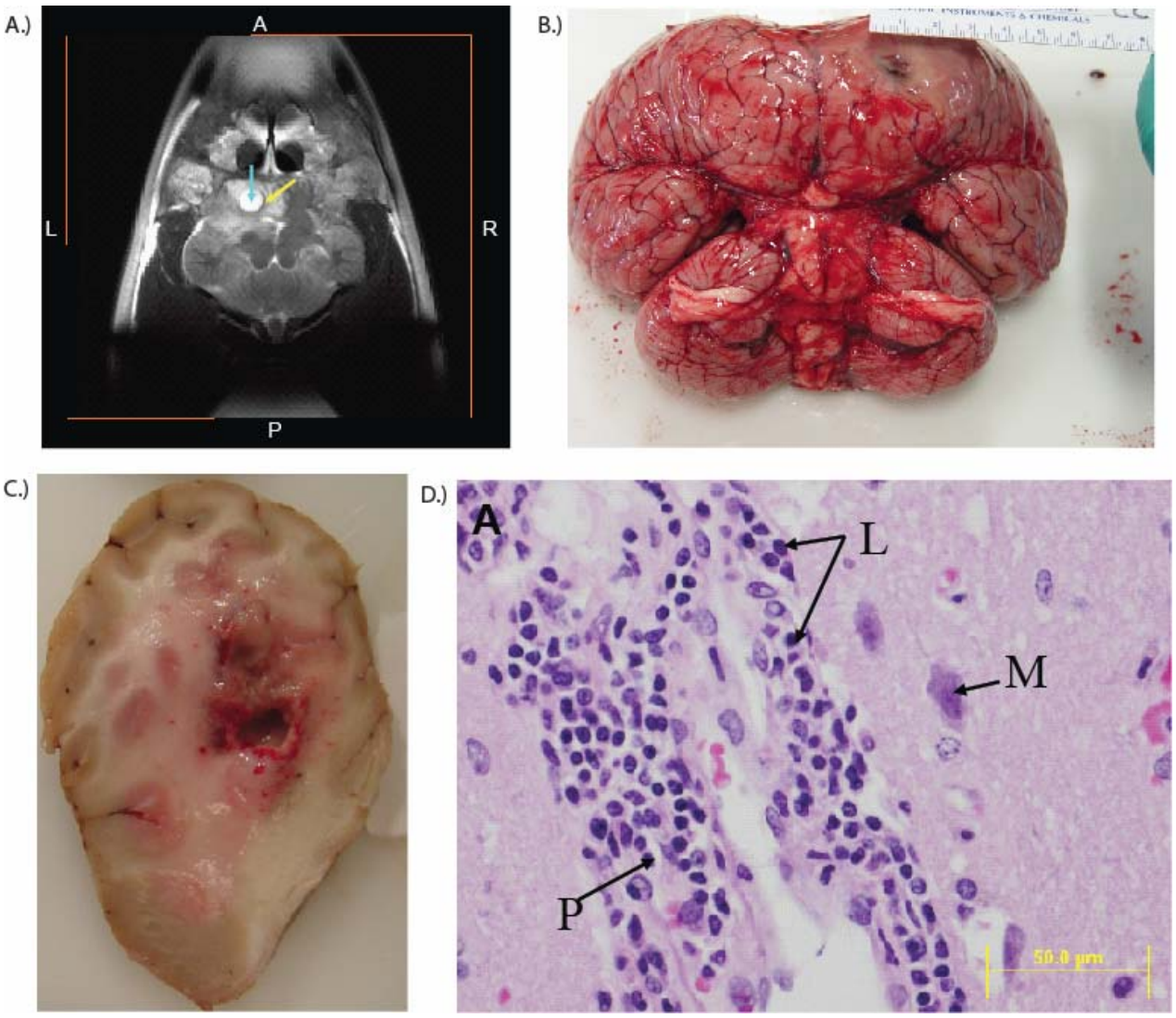


Figure 7. Brain lesion in CCSN04-177-Dd common dolphin. A.) The MR image indicated a 2 cm spherical abscess identified in the left frontal lobe. A halo of damaged tissue is noted by the yellow arrow. A fluid filled necrotic core is indicated by the turquoise arrow. B) Brain removal revealed pus in the left frontal lobe. C.) The lesion was revealed during the dissection. D.) Histological analysis revealed abundant perivascular lymphocytes (L) and plasma cells (P), and activated microglial cells (M). (Images copyright E. Montie, 2006. Histology courtesy, Dr. David Rotstein, Univ. of Tennessee).

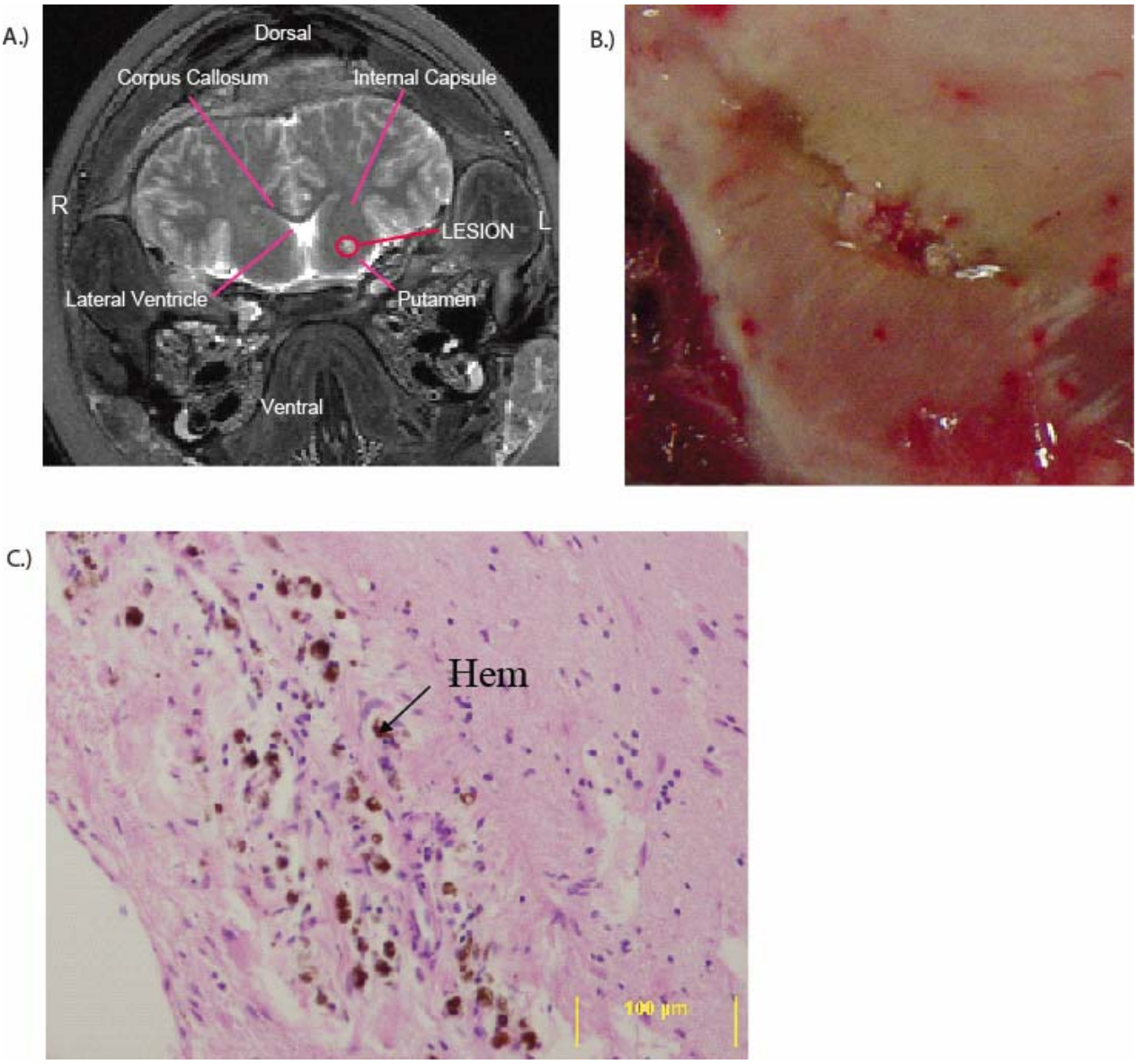


Figure 8. Brain lesion in CCSN04-191-Dd common dolphin. A.) The MR image indicated a small lesion in the region of the putamen and globus pallidus or collectively termed the lentiform nucleus. B) The lesion was revealed during the dissection. C.) Histological analysis revealed irregular tracts of rarefied neuropil, hemosiderin-laden macrophages, and the accumulation of extracelluar hemosiderin (Hem) black-brown pigment. (Images copyright E. Montie, 2006. Histology courtesy, Dr. David Rotstein, Univ. of Tennessee).

50272-101

REPORT DOCUMENTATION PAGE	1. REPORT NO. WHOI-2008-02	2.	3. Recipient's Accession No.

4. Title and Subtitle
Imaging Procedures for Stranded Marine Mammals

5. Report Date
January 2008

6.

7. Author(s) Darlene R. Ketten and Eric W. Montie

8. Performing Organization Rept. No.

9. Performing Organization Name and Address

Woods Hole Oceanographic Institution
Woods Hole, Massachusetts 02543

10. Project/Task/Work Unit No.

11. Contract(C) or Grant(G) No.
(C)N00244-071-0022
(G)

12. Sponsoring Organization Name and Address

Office of Naval Research

13. Type of Report & Period Covered
Technical Report

14.

15. Supplementary Notes
This report should be cited as: Woods Hole Oceanog. Inst. Tech. Rept., WHOI-2008-02.

16. Abstract (Limit: 200 words)

This report provides an introduction to biomedical imaging techniques and guidelines for diagnostic imaging of marine mammals to assist with both live examination and necropsy procedures. The procedures described are based on imaging equipment and techniques that are relatively common in human and veterinary facilities and to provide the majority of stranding response groups with the most likely options that will assist their efforts. The imaging techniques described include basic radiography, computed tomography (CT), and magnetic resonance imaging (MRI) and are applicable to both live and post-mortem cases. Special emphasis has been placed on whole body, airway, head, and ear imaging procedures. Sub-sections cover basic information on the basic principles and appropriate applications for radiography vs. CT vs. MRI, handling and preparation of live and dead animals in clinical settings, and image and data formats that may be encountered. The protocols are also listed in outline form in order to provide a rapid overview. The introductory discussion of principles behind techniques is not required to employ the protocols but does provide additional information that can aid in deciding which techniques are most efficacious and what the limitations are for interpretation of imaging data. Examples of some pathology imaged with these procedures are also provided.

17. Document Analysis **a. Descriptors**

marine mammals
computed tomography (CT)
Magnetic Resource Imaging (MRI)

b. Identifiers/Open-Ended Terms

c. COSATI Field/Group

18. Availability Statement

Approved for public release; distribution unlimited.

19. Security Class (This Report)
UNCLASSIFIED

20. Security Class (This Page)

21. No. of Pages
26

22. Price

(See ANSI-Z39.18) *See Instructions on Reverse*

OPTIONAL FORM 272 (4-77)
(Formerly NTIS-35)
Department of Commerce