# The Examiner's Guide to the National Marine Mammal Tissue Bank Form

## Introduction.

The purpose of this document is to clarify the protocol for completing the National Marine Mammal Tissue Bank (NMMTB) Form (NOAA Form ??-????) for the collection of specimens from marine mammals in the United States. This measure will standardize the field data sent to the National Institute of Standards and Technology (NIST). NIST maintains the NMMTB at the Marine Environmental Specimen Bank at the Hollings Marine Laboratory in Charleston, South Carolina. The NMMTB provides a resource of samples that have been collected in a systematic and well-documented manner for comparing results over time to identify whether environmental trends exist, provides for future retrospective analyses for new analytes of interest, and allows for future analyses of samples collected today using improved analytical techniques of tomorrow. Many of the fields on this form may have several interpretations. For consistencies in information and accuracy of the database, please use this guide as the common convention in understanding and completing the form.

### **Background.**

In 1989 the National Oceanic and Atmospheric Administration (NOAA) and the National Marine Fisheries Service (NMFS) initiated the development of the National Marine Mammal Tissue Bank (NMMTB) as a result of a massive die-off of bottlenose dolphins (Tursiops truncatus) in 1987-88. A large number of animals stranded along the Atlantic coast of the United States and although it was concluded that a naturally occurring toxin, brevitoxin, was likely the cause of death (Geraci, 1989), environmental pollution was suspected due to high levels of contaminants. It was also determined then that baseline data of anthropogenic contaminants was relatively unknown but needed for future reference. The NMMTB was developed to provide this reference data as well as other valuable information by collecting and banking marine mammal tissues for long-term storage for retrospective analyses. The guidelines for the NMMTB were based on the already established goals and protocols of the Alaska Marine Mammal Tissue Archival Project (AMMTAP). This project was established in 1987 through an agreement between NOAA's National Ocean Service (NOS), the National Institute of Standards and Technology (NIST) and the Minerals Management Service (MMS) to help determine contaminant levels of marine mammals that were taken primarily during native subsistence hunts in Alaska (Becker, et al., 1988; 1991). Tissues were also collected and banked for long-term archival using standardized protocols. Currently the AMMTAP is conducted by the United States Geological Survey (USGS) Biological Resource Division in cooperation with NOAA's NMFS and NIST.

In 1990 a demonstration phase of the NMMTB began evaluating the collection protocols that were developed in order to obtain tissues from animals taken during incidental catches and strandings from coastal locations throughout the U.S. The New England Aquarium (NEA), Boston, MA., assisted NOAA/NMFS and NIST personnel in the initial collection of two species of marine mammals, the harbor porpoise (*Phocoena phocoena*), taken during incidental catches, and the pilot whale (*Globicephala melaena*), taken during mass strandings. Protocols were developed that identified the criteria to determine whether an animal was suitable for sampling as well as the logistics and procedures for collecting and processing samples from these events. Efforts have been extended to sampling on the west coast, as well as the southeast Atlantic and Gulf of Mexico regions. Animals are collected through incidental catches, single and mass strandings as well as subsistence hunts in Alaska through the

AMMTAP. The species list has also expanded to include other cetaceans as well as some pinnipeds, and fissipeds.

In 1992, the NMMTB program was formally established by the Marine Mammal Health and Stranding Response Act (Public Law 102-587) and was expanded and combined with the Marine Mammal Stranding Network to become a larger program that resulted in several components; Stranding Networks, the NMMTB, and Monitoring and Quality Assurance. This expansion is now known as the Marine Mammal Health and Stranding Response Program (MMHSRP) and is coordinated by the NMFS in cooperation with the USFWS. The MMHSRP is focused on animal health assessment, real-time contaminant monitoring, specimen banking, response to strandings and mass mortalities, quality assurance/quality control of analytical results, and the management of a nationwide database on the health of marine mammal populations. The NMMTB and the quality assurance program are administered by NIST. In 1995 the quality assurance program was formalized and became the National Marine Analytical Quality Assurance Program (NMAQAP). More information about the NMAQAP can be found at http://www.nmfs.noaa.gov/pr/health/aqa.htm.

After the demonstration phase the New England Aquarium continued to collect tissues for the NMMTB. In addition, other federal and non-federal partners have been formally trained by NIST personnel and collect samples for the NMMTB. These partners include:

Alaska Department of Fish and Game Alaska Nanuuq Commission Alaska Sea Life Center Cape Cod Stranding Network Hubbs Sea World Research Institute Marine Mammal Center Minerals Management Service Mote Marine Laboratory National Marine Fisheries Service (NMFS) National Oceanic and Atmospheric Administration (NOAA)/National Ocean Service (NOS) Center for Coastal Environmental Health and Biomedical Research (CCEHBR) Laboratory New England Aquarium NOAA, Panama City Laboratory North Slope Borough, Department of Wildlife Management United States Fish and Wildlife Service (USFWS) University of Alaska-Fairbanks University of North Carolina at Wilmington US Geological Survey/Biological Resources Division

Over 1,000 animals have been collected from 44 species of cetaceans, pinnipeds, and fissipeds. There have been approximately 1,517 marine mammal tissue specimens collected from 31 species outside of Alaska and 1,700 tissue specimens collected from 17 species in Alaska. Blubber or other adipose tissue, liver and kidney tissues are the primary tissues collected from each animal for the NMMTB. In the past muscle samples were collected for animals that were killed for subsistence uses.

The goal of the NMMTB was initially established for the purposes of anthropogenic chemical evaluations by:

1. archiving samples for future retrospective analyses for new analytes of interest;

2. archiving samples for future analyses using improved analytical techniques and;

3. providing a resource of samples that have been collected and stored in a systematic and well documented manner for comparing results over time to identify if environmental trends exist.

Initial sample collection and archival protocols were developed for chemical contaminants (Becker, *et al.*, 1999) and were based on the AMMTAP for Alaska species (Becker *et al*, 1988; 1991). Although there have been some changes and updates made over time, the goals and focus of the NMMTB have remained primarily on chemical contaminants and non-essential elements. Recently the NMMTB added archival protocols for blood collection for chemical analyses. Additional protocols reflect those additions.

Tissue or fluid samples are primarily obtained from stranding network personnel, subsistence hunting collaborators, or biologists or veterinarians for live capture release programs that have been trained by NIST personnel to collect and process tissues for the NMMTB. Animals and tissues or fluids selected for collection and archival must meet specific criteria before they can be collected. The following is the criteria that an animal must meet before it is considered for archival in the NMMTB as a representation of a 'normal' animal:

Dead animal

1. Animal appears to be 'normal' and 'healthy' as defined by lack of evidence of chronic disease, evidence of recent human interaction (ship strike, entanglement) or incidental to other human activities, or good robust body condition with no known cause of death; 2. Animal is Code 2 (fresh dead):

3. There is no obvious scavenger damage;

4. Body cavity is intact and has not been breached; and

5. Elapsed post-mortem time is less than 24 hours or the animal has been kept

refrigerated or frozen for 48 hours post mortem.

The NMMTB Specimen Access Policy was developed in order to outline the procedure for requesting use of tissues/fluids in the NMMTB, to prioritize the use and release of tissues/fluids from the NMMTB, and to track such tissue/fluid distribution. A NMMTB Tissue Request Form and research proposal must be submitted to the MMHSRP Program Manager, Office of Protected Resources, NMFS for review of the request. A copy of this access policy as well as the specimen request form can be found at http://www.nmfs.noaa.gov/pr/health/tissue/.

After tissues/fluids are collected following the NMMTB protocols, they are shipped frozen to and maintained by the NIST in two locations, the National Biomonitoring Specimen Bank (NBSB) in Gaithersburg, MD and the Marine Environmental Specimen Bank (Marine ESB) in Charleston, SC at the Hollings Marine Laboratory. Currently all samples, including the A and B subsamples of each tissue, are shipped frozen to the Marine ESB in Charleston, SC where they are stored in liquid nitrogen vapor phase freezers (-150 °C). Subsample A is stored for long term archival and Subsample B is available for analysis. The 2 subsamples of each tissue are stored in separate freezers for security purposes so if a freezer breaks down or overfills with liquid nitrogen, the entire sample is not compromised.

#### **Reporting.**

One of the goals of the NMMTB is to provide a resource of samples that have been collected and stored in a systematic and well-documented manner for comparing results over time to identify if environmental trends exist. In order to achieve this goal, data associated with the collection, processing and storage of the samples must be reported. Reporting burden for this collection of information is estimated to average 45 minutes per animal collection, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information, including completing the paper form and entering the information into the NMMTB Database. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing this burden, to National Marine Fisheries Service, Office of Protected Resources, Marine Mammal and Turtle Conservation Division, 1315 East-West Highway, Silver Spring, MD 20910.

Protocols developed by NIST for collecting and archiving tissues/fluids are designed to: (1) provide sufficient material for multiple analyses, (2) minimize the possibility of sample change and/or loss during storage, (3) minimize inadvertent contamination during sample handling and ensure sample integrity, (4) provide for long-term sample stability through cryogenic techniques, and (5) track and maintain a record of sample history.

Sources of specimen from dead animals include fresh stranded animals, animals taken incidental to fishing activities, and animals taken for subsistence uses.

Indicator species include: harbor seal (*Phoca vitulina*), California sea lion (*Zalophus californianus*), northern fur seal (*Callorhinus ursinus*), ringed seal (*P. hispida*), pilot whale (*Globicephala melas*), harbor porpoise (*Phocoena phocoena*), Atlantic white-sided dolphin (*Lagenorhynchus acutus*), pygmy sperm whale (*Kogia breviceps*), bottlenose dolphin (*Tursiops truncatus*), rough-toothed dolphin (*Steno bredanensis*), common dolphin (*Delphinus delphis*), beluga whale (*Delphinapterus leucas*), bowhead whale (*Balaena mysticetus*), polar bear (*Ursus maritimus*). Additional species have also been included. See Table 1 for a list of all species collected as a part of the NMMTB.

Marine Mammal Healt hand Stranding Response Program (MMHSRP)	Alas ka Marine Mammal Tissue Archival Project (AMMTAP)
Harbor Porpuise (Phocoena phocoena )	Ringed Seal (Phoc a hispida)
Pilot Whale (Clobic aphala malas)	Harbor Seal (Phoca vitulina)
Short Finned Pilot Whale ( <i>Clobicephala macrorym</i> hus)	SpottedSeal(Phocalargha)
Pygmy Sperm Whale (Kogia breviceps)	Bearded Seal (Erignathus barbatus)
Dwarf Sperm Whale ( <i>Ko gia simus</i> )	Elephant Seal (Mircunga angustivostris)
North Atlantic Right Whale (Eubalaena glacialis)	Stellar Sea Lion (Eurre topias jubatus)
Blainville's Beaked Whale (Mesoplodon densirostris)	Northern Fur Seal (Callorhinus ursinus)
Gervais Beaked Whale (Mesoplodon suropasus )	Pacific Walnus (Cilobenus rosmanus divergens)
Bottlenose Dolphin ( <i>Tursiops truncatus</i> )	Beluga Whale (Delphinapterus laucas)
Striped Dolphin ( <i>Stenella cce nie oalba</i> )	Bowhead Whale (Balas na mysticatus)
Dolphin (Stenel la spp.)	Polar Bear (Ursis maritimus)
Risso Dolphin (Grampus griseus)	SeaOtter (Enhydralutris)
White Side dDolphin (Lagenorhynchus acutus)	Harbor Porpoise (Phocogna phocogna)
California Sea Lion (Zalophus californianus)	Gray Whale (Eschrichtius robustus)
Cammom Dolphin ( <i>Delphinus del phis</i> )	Ribbon Seal ( <i>Phoca fasciata</i> )
Hooded Seal ( <i>Cystophona cristata</i> )	Steineger's Beaked whale (Mesophpdon st ginegri)
Harbor Seal ( <i>Phoca vitulina</i> )	Humpback Whale (Magaptera nouse anglize)
Ringed Seal (Phoca hispida)	
HarpSeal ( <i>Phoca groenlandica</i> )	
GreySeal (Hali choerus grypus)	
Rough-Toothed Dolphin ( <i>Seno bredznensis</i> )	
Fin Whale (Balaenoptera physalus )	
Curier's Beaked Whale (Zphius cavirostris)	
Atlantic Spotted Dolphin (Stenella frontalis)	
Eastern Spinner Dilphin (Stenella longirostris orientalis)	
Pantropical Spotted Dolphin ( <i>Stenella attenuata</i> )	
Chyne re Dolphin ( <i>Stenel la clymene</i> )	
Mirke Whale (Balae napt era acutorostrata)	
Beluga Whale (Delphinaptorus loucas)	
Melon-headed Whale (Peponccephala electra)	
Bryck's Whale ( <i>Balaenoptera edeni</i> )	
Pygny Killer Whale (Føresa attenuata)	

#### Table 1. Species of Marine Mammals Collected as a Part of the National Marine Mammal Tissue Bank(NMMIB).

Under Title IV of the Marine Mammal Protection Act (MMPA), Section 407, the Secretary of Commerce shall make provision for the storage, preparation, examination, and archiving of marine mammal tissues/fluids. These tissues/fluids will be archived in the "NMMTB". In addition, the Secretary shall consult with the Marine Mammal Commission, the Secretary of Interior, and individuals with knowledge and experience in marine science, marine mammal science, marine mammal veterinary and husbandry practices, and conservation to issue guidance for analyzing tissue samples as a means to monitor and measure overall health trends in representative species or populations of marine mammals after the public has an opportunity to review and comment.

The MMHSRP working with the federal partners is developing an integrated information management system which will serve to assist the Secretary of Commerce and Interior to meet their goals of the Act for the following:

- Facilitate the collection and dissemination of reference data on the health of marine mammals and health trends of marine mammal populations in the wild;
- Correlate the health of marine mammals and marine mammal populations with available data on physical, chemical and biological environmental parameters; and
- Coordinate effective responses to unusual mortality events by establishing a process in the Department of Commerce in accordance with Section 404 of the Marine Mammal Protection Act.

Under Section 407 (c) of Title IV, the Secretary of Commerce shall maintain a central database that provides an effective means for tracking and accessing data on marine mammal tissues collected for and maintained in the NMMTB. This database should contain reference data on the health of marine mammals, their populations and species that are subject to unusual mortality events. In addition, the Secretary shall consult with the Secretary of the Interior to establish criteria for access to marine mammal tissues in the tissue bank; analyses conducted; and marine mammal information within the database after the public has an opportunity to review and comment.

### NIST SAMPLE PROCESSING

Field ID:	Other ID Number:
Common Name:	Genus species:

**Field ID** is the unique identifier assigned to the animal from which the tissue(s) or fluids were obtained. The format is open to each agency's requirements; however, please remain consistent within your agency.

<u>**Other ID Number</u>** is any other identifier(s) related to this animal Examples include: previous field ID numbers if this animal previously stranded; ID numbers assigned by other organizations (including authorized rehabilitation facilities to which the animal is transferred), former identification numbers from scientific research projects, etc.</u>

Common Name is the commonly used name for this species (e.g., harbor porpoise).

Genus, Species is the Latin or scientific names for the species using standard binomial nomenclature.

Collection       Single Strand       Biopsy       Incidental Take       or         Type:       Mass Strand       Repeat Event       Incidental Take       or         UME       Live Capture Release       Other       Other       (choose one)         Rescuel       Other (specify):       (specifiy):       (specifiy):	Clubbed Bow/Arrow Gunshot Other (specify):
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<u>Collection Type</u> is the description of the event(s) or circumstances pertaining to the animal from which the samples are taken and the collection method used. Examples include: strandings (single or multiple); unusual mortality event, repeat stranding event, rescue, live capture release, biopsy, etc.

<u>Single Strand</u>: Check this box if one animal stranded and was responded to by your organization or another.

**Mass Strand**: Check this box if the animal from which the samples were collected was part of a mass stranding. A "*mass stranding*" is more than 2 cetaceans that strand, other than cow-calf pairs. A "*cow/calf pair*" would be two animals stranding where one is the mother and the other is the offspring (a mom/pup pair would also qualify).

<u>UME</u>: Check this box if the animal was part of an official "*unusual mortality event*" (UME). An "*unusual mortality event*" is a die-off that has been officially designated as a UME by the Working Group on Marine Mammal Unusual Mortality Event (WGMMUME).

**<u>Rescue</u>**: Check this box if the animal was part of a rescue during an oil spill, disentanglement, out of habitat, or other such event.

**Biopsy**: Check this box if the sample was collected using remote biopsy (e.g., dart) or using a punch pole.

**<u>Repeat Event</u>**: Check this box if the animal stranded during a morbidity or mortality event that has been designated as a repeat event by the WGMMUME.

<u>Live Capture Release</u>: Check this box if the animal was part of a live-capture release research effort and samples were collected (e.g., blood, surgical biopsy)

<u>Other (specify)</u>: Check this box if the animal was part of an event that is not listed in this section and specify what the event is on the line provided next to the check box (e.g. research).

Incidental Take: Check if the animal was killed incidental to any human activities.

Fisheries: Check this box if the animal was obtained incidental to fishery activities.

**Other (specify)**: Check this box if the animal was obtained incidental to other human activities (e.g., incidental to permitted research activities, ship strike) and fill in the circumstances in the comment field.

<u>Subsistence</u>: Check this box if the animal was killed for subsistence use by Native Americans under comanagement agreements. List the means by which the legal take occurred:

<u>**Clubbed**</u>: Check this box if the cause of death was a striking with a cudgel, stick, bat, baton, nightstick, staff, shillelagh, or billy club during a hunt.

**Bow/Arrow**: The animal cause of death was due to the shooting of a bow and arrow during a hunt.

**Ballistics or Gunshot**: Check this box if the cause of death was from a projectile fired from a gun. When possible, please enter the caliber of the projectile used.

**Other (specify)**: Check this box if the cause of death was from a weapon other than the one listed on the form such as, a knife, a machete, etc. and specify weapon.

ndition:	() Ali	ve 🛛 🔘 Fresh Dea	d (Code 2)	Euthanized	Was anim	nal in rehabilitation	? 🖲 Yes 🖲 No
<i>oose one)</i> If e	uthanized:	With what:			If yes:	Where:	1987 - 32
		How much:			From:	dd / mm / yy	To: dd / mm / yy
		Where:			(pleas	e attach cllinical/n	nedical records)

<u>Condition</u> indicates the physical state of the animal (carcass or live animal) on the date when the samples were removed from the animal. The codes used follow the guidelines used in the Level A data sheet.

Code 1 – alive	Code 4 – Advanced decomposition
Code 2 – fresh dead	Code 5 – Mummified/Skeletal
Code 3 – moderate decomposition	Code 6 – Unknown

Alive (code 1): Check this box if the animal was alive during the removal of the specimen.

**Fresh Dead (Code 2)**: Check this box if the carcass was in good condition (fresh/edible). Normal appearance, usually with little scavenger damage; fresh smell; minimal drying and wrinkling of skin, eyes and mucous membranes; eyes clear; carcass not bloated, tongue and penis not protruded; blubber firm and white; muscles firm, dark red, well-defined; blood cells intact, able to settle in a sample tube; serum unhemolyzed; viscera intact and well-defined, gut contains little or no gas; brain firm with no discoloration, surface features distinct, easily removed and intact. If animal is early Code 3, moderate decomposition, note this in the Additional Comments section on Page1 of the Tissue Bank Form.

**Euthanized**: Check this box if the animal was found alive and was euthanized by an authorized entity for humane welfare, or medical reasons.

**If euthanized** indicates if the animal was euthanized by an authorized entity, please provide the following information about the method used:

With what Chemical: Indicate the drug(s) used to chemically euthanize the animal

**How much**: Indicate the dosage amount of the drug (total dose in milliliters) and include the concentration (mg/ml) of the drug as well as dose given.

<u>Where</u>: Indicate how and where the drug was administered to the animal by drug (e.g., IM acepromazine along left peduncle).

#### Was animal in rehabilitation?

<u>Yes</u>: Check this box if the animal was in an authorized rehabilitation facility when the samples were removed.

No: Check this box if the animal was not in an authorized rehabilitation facility.

If the animal was in a rehabilitation facility, please provide the following information about the animal medical history:

Where: Indicate the name of the authorized rehabilitation facility.

**From**: *DD/MM/YYYY* **To**: *DD/MM/YYYY* - Indicate the date that the animal was admitted into the rehabilitation facility and the date that the animal was removed from the rehabilitation facility upon the animal's death following the described date format. Also, attach the animal's medical history. If you are completing the form using the NMMTB Database, you may submit the medical history electronically.

	cean/Bay/Sea:	County:	10	City/Island/Community:	
Lo	ocality Details:	20			
Latitude:			N (dec degrees)	Longitude:	W (dec degrees)

Animal location is the geographic location of animal when the samples were collected.

**<u>State</u>**: The standard two letter abbreviation of a state in the United States.

<u>County or Parish</u>: The name of the county or parish in the state, if known.

<u>**City/Island/Community**</u>: The names for the city, island or community of the location. This should include boroughs, parishes, provinces, commonwealths, and territories.

**<u>Ocean/Bay/Sea</u>**: The name of the ocean, sea, gulf, bay, inlet or estuary that the animal was found on the shore of a body of water where the tissue was removed.

**Locality Details**: Using known landmarks, describe the precise locality where the animal was found. Compass bearings and relative distances are useful.

**Latitude**: These are the GPS coordinates of the geographic location of where the animal was when the tissues were collected. The units of measurements should be decimal degrees.

**Longitude**: These are the GPS coordinates of the geographic location of where the animal was when the tissues were collected. The units of measurements should be decimal degrees.

Time of death dd / (Zulu)	m / yy _ hr	Place of Death: Internal body temp. of animal:
If transported before tissue re	emoval:	Vehicle Type: Length of Transport:
		Ambient weather condition:
		Remarks:
Time of tissue removal	dd / mm/yy	hr Place of tissue removal:
(Zulu)	P	Internal body temp. of animal just before tissue removal:
If transported before process	sing:	Transportation storage: Dry ice Wet ice Other: Ambient weather condition: Interim storage of tissue: Teflon bag Teflon jar Other: Remarks:
Time of tissue processing	dd / mm / yy	hr Place of tissue processing:
(Zulu)		Ambient temperature at processing:
Time of interim freezing (Zulu)	dd / mm / yy	hr Freezer type: LN2 -80° C -30° C Other:
Time shipped to NIST	dd / mm / yy	hr
<i>(Zulu)</i> Time received at NIST <i>(Zulu)</i>	dd / mm / yy	hr

If samples were collected from a dead animal:

<u>**Time of Death**</u>: Indicate the date (*DD/MM/YYYY*) of the animal's death and time using Zulu which pertains to the hour in military time (i.e. 2 pm equals 1400).

<u>Place of Death</u>: Indicate the place of the animal's death. This may be different from the location of the animal during sample collection.

Internal Body Temp. of Animal: Indicate the animal's internal body temperature at the time of death.

<u>Celsius (C)</u>: Check this box if the unit of measurement is Celsius.

**Fahrenheit** (F): Check this box if the unit of measurement is Fahrenheit.

### <u>Rigor?</u>

<u>Yes</u>: Check this box if *rigor mortis*, the stiffening of the body muscles, was observed during the time of tissue collection.

**No**: Check this box if *rigor mortis* did not occur during the time the carcass was being observed prior to sample collection.

<u>If transported before tissue removal</u>: If the animal was transported before sample removal, please provide the following information describing the method of the animal relocation from initial site.

**<u>Vehicle Type</u>**: Indicate the type of transportation provided to relocate the animal from the initial site, such as, pick-up truck, open truck, closed truck, van, etc.

**Length of Transport**: Indicate the length of time the animal was in transport.

<u>Ambient Weather Conditions</u>: Indicate the temperature of the environment during transport. Provide units of measurements as Celsius (C) or Fahrenheit (F).

**<u>Remarks</u>**: Provide additional comments regarding the animal relocation.

<u>**Time of Tissue Removal**</u>: Indicate the date (*DD/MM/YYYY*) of the removal of samples from the animal and time using Zulu which pertains to the hour in military time (i.e. 2pm equals 1400).

<u>Place of Tissue Removal</u>: Indicate the location of the animal when the samples were removed. (e.g., necropsy laboratory, beach, landfill, etc).

**Internal Body Temperature Before Tissue Removal**: Indicate the animal's internal body temperature at the time of tissue removal. Check the units of measurement as Celsius (C) or Fahrenheit (F).

<u>If transported before processing</u>: Please provide information in this section about the storage of the tissue(s) that are transported prior to processing.

<u>**Transportation Storage</u>**: Check whether the storage unit used dry ice, wet ice or other to preserve the tissue while being transported before processing to NIST. If select "other", please indicate other storage method used (i.e. portable freezer, refrigerator/freezer, etc.)</u>

<u>Ambient Weather Conditions</u>: Indicate the environmental parameters for the transport event. Include temperature and if in an open area (outside) include precipitation and wind factor. Provide units of measurements as Celsius (C) or Fahrenheit (F).

**Interim Storage Container**: Check one of the boxes to indicate whether the tissue was in a Teflon bag, Teflon jar or other. If select "other", please describe the interim storage container.

**<u>Remarks</u>**: Provide additional comments regarding the storage of the tissue.

<u>**Time of Tissue Processing**</u>: Indicate the date (*DD/MM/YYYY*) and time (using Zulu which pertains to the hour in military time (i.e. 2pm equals 1400) that the sample collected was processed for long term storage.

Place of Tissue Processing: Indicate the place where the tissue was processed.

<u>Ambient Temperatures at Processing</u>: Indicate the temperature of the environment where the samples are being processed and if processing occurs outside include wind and precipitation. Provide units of measurements as Celsius (C) or Fahrenheit (F).

**<u>Time of Interim Freezing</u>**: Indicate the date (*DD/MM/YYYY*) and time (using Zulu which pertains to the hour in military time (i.e. 2pm equals 1400) of the initial freezing of the samples collected.

<u>Freezer Type and temperature</u>: Check the freezer type temperature as  $LN_2$ , -80 °C, -30 °C or other. If select "other", please provide the units of measurements for the freezing temperature as Celsius (C) or Fahrenheit (F).

**<u>Time Shipped to NIST</u>**: Indicate the date (*DD/MM/YYYY*) and time (using Zulu which pertains to the hour in military time (i.e. 2pm equals 1400) the sample was shipped to NIST from the site using the format provide on the form in this section

<u>**Time Received at NIST</u>**: Indicate the date (*DD/MM/YYYY*) and time (using Zulu which pertains to the hour in military time (i.e. 2pm equals 1400) the sample was received at NIST from the site using the format provide on the form in this section.</u>

Additional comments:							**
Sample weights:	Blubber (q):	Liver (q):	Kidney (q):	Whole Blood (mL):	Plasma (mL)	Serum (mL):	Other:
A B							

<u>Additional Comments</u>: Provide additional remarks regarding the tissues being processed. For instance, if animal is early Code 3, note it here.

<u>Sample Weights</u>: Indicate the sample weight for tissues classified, A or B. The weight should be measured in grams for the following tissues: blubber, liver, kidney. The measure for whole blood, plasma and serum should be in milliliters. If other, specify the tissue and provide the unit of measurement.

# ANIMAL INFORMATION

ield IC			Genus species:	
Sex:	🖲 Female 🔘 Male	Total length:	● cm ● in	Actual Estimated
	Unknown	Total weight:	🖲 kg 🔘 lb	Actual Estimated

**Field ID** is the unique identifier assigned to the animal from which the sample(s) were obtained. Format is open to each agency's requirements; however, please remain consistent within your agency. It is carried over from NIST Sample Processing – Page 1.

<u>Genus Species</u> is the Latin name for the animal in standard binomial nomenclature. It is carried over from NIST Sample Processing – Page 1.

Sex is the sexual classification of the animal.

**Female**: Check this box if the animal is confirmed as female.

Male: Check this box if the animal is confirmed as male.

<u>Unknown</u>: Check this box if the animal's sex cannot be confirmed by examination. It is expected that all unknown will be genetically identified as to 'sex'.

<u>Total Length</u> is the straight length (not contoured) measurement of the animal on the date of the initial examination. Please note in the comment field if the animal was towed or otherwise stretched prior to measurement.

<u>Cm</u>: Check this box if the measurement is taken in centimeters (metric system is preferred).

In: Check this box if the measurement is taken in inches.

<u>Actual</u>: Check this box if this is the actual physical measurement using standard measuring device (e.g., tape measure).

**Estimated**: Check this box if this is a visual measurement or if the animal's carcass is not intact (e.g. flukes degraded or severed).

**Total Weight** is the weight of the entire animal on the date of the initial examination.

**<u>Kg</u>**: Check this box if the weight of the animal is measured in kilograms (metric system is preferred).

**<u>Lb</u>**: Check this box if the weight of the animal is measured in pounds.

Actual: Check this box if this is the actual physical weight obtained using a scale.

**Estimated**: Check this box if this is an estimate based on length/growth charts or visual observation using an item of known length because the carcass was not intact or could not be measured. (e.g. flukes degraded or severed or floating).

Age Class: (choose one)	Adult Subadult	Actual	Age: By whom:	_ Date aged	dd /mm / yy
(L'IDOSE ONE)	<ul> <li>Pup/calf</li> <li>Yearling</li> <li>Unknown</li> </ul>	Estimated	Method Used:  Teeth (GLG's)  Baleen (choose one) Other:	Bone	Ear Plugs
Epiphysis:	Open Oclosed fused	Eused invis	Disposition of specimen: Photo Slide (Please attach copy of photo)	eter en elide l	

<u>Age Class</u> is the classification of the animal into one of six age classes using morphometric data, aging techniques on specific specimen (e.g., teeth, baleen, claw, bone, etc.) or having known age animals.

<u>Adult</u>: Check this box if the age class for an animal is determined to be physically or sexually mature.

**Subadult**: Check this box if the age class for an animal is determined to be greater than one year old, but not yet mature physically or sexually.

**<u>Pup/calf</u>**: Check this box if the age class for an animal is determined to be younger than one year old.

<u>**Yearling**</u>: Check this box if the age class for an animal is determined to be approximately one year old, using length or time of year or age determination.

**Unknown**: Check this box if the age class of an animal is unable to be determined.

Actual: – Check this box if the age can be verified.

**Estimated**: Check this box if the age is estimated.

**Epiphysis** is an area at the end of a long bone and other bones (e.g., vertebrae) that is the point of bone growth in length. The line of growth is called the physeal line and is made of cartilage in young immature animals. Over time and with physical maturity the physeal line reduces as the cartilage becomes ossified and the bones fuse. The timing of that fusion or ossification is species dependent and bone dependent, therefore for most species only gives approximate age classification not exact age. *Ossification* is the process of bone formation, in which connective tissues, such as cartilage are turned to bone or bone-like tissue. Measures of epiphyseal closure indicate physical maturity (classification as adult).

**<u>Open</u>**: Check this box if the epiphyses are open with the physeal line of cartilage apparent and in many cases the bone ends may separate from the main portion of the bone.

<u>**Closed fused**</u>: Check this box if the epiphyses are partially closed (ossified) but the physeal line is still quite visible.

**<u>Fused Invis</u>**: Check this box if the epiphysis is fused (ossified) to the rest of the bone and the physeal line is invisible.

Age is the approximate number of years determined for the animal based on aging techniques.

**By Whom** is the name of the qualified individual who determined the age classification of the animal.

**Date Aged**: The date (*DD/MM/YYYY*) that the age classification was determined.

<u>Method Used</u>: If the age was determined using a standard aging technique, please indicate the type of technique used to estimate the age of the animal.

**Teeth (GLG's)**: Check this box if the age estimation is determined by counting the growth layers (GLGs) or tooth layers of the animal's teeth, e.g. odontocetes' age classification is determined by counting the GLG's of the mid-mandibular teeth; pinnipeds' age classification is determined by counting the GLG's of the canines and first 3-4 postcanines (incisors and premolars); walruses' lower canines; and sea otters' lower first premolars.

**Baleen**: Check this box if the age estimation is determined using baleen plates.

**Bone**: Check this box if the age estimation is determined by counting the growth plate on the bone end; e.g., remove the tympanic bullae for a count of the layers or rings.

**Ear plugs**: (also known as "*wax plug*" in some mysticetes) Check this box if the age estimation is determined by removing the ear plug at the proximal end on the auditory canal and counting the ear plug layers.

<u>**Other</u>**: Check this box if other method was used, please indicate in the comment field the method used with a brief description of the method.</u>

**Disposition of Specimen**: Use this line to indicate where the photos or slides of the sample taken to determine the age classification of the animal are housed.

**Photo**: Check this box if you have pictures of the specimens used determining the age classification of the animal. If possible, attach copies of the photographs to the NMMTB form.

<u>Slide</u>: Check this box if you have slides of the specimens used determining the age classification of the animal. If possible, attach copies of the slides to the NMMTB form.

Reproductive condition:	Testis/Ovaries:	Length: Left:	Mid-Width:	Mid-depth:	Weigh	t: 
Pregnant Lactating	(circle one)	Right:			() in	og ⊚oz
Fetus length:	Corpora lutea #:	Corpora al	bicantia #:	Corpo	ora hemmorghagic	um #:

**<u>Reproductive Condition</u>** is the state of the animal's reproductive cycle at the time of the sample collection.

<u>Sexually Mature</u>: Check this box if the male or female has characteristics of reproductive maturity such as pregnancy, ovulation, corpora lutea or albicans, presence of milk, size of the uterus, sperm apparent on smear or histological examination.

**<u>Pregnant</u>**: Check this box if the animal is carrying one or more embryos or fetuses. If pregnant, please indicate fetal length.

**Lactating**: Check this box if the animal shows evidence of increased mammary development and milk can be expressed from the nipple or on cut surface of the mammary gland.

<u>Fetus Length</u> is the measurement of the length in centimeters of the fetus or embryo as a total straight length (tip of nose to notch in fluke or tip of tail).

**<u>cm</u>**: Check this box if the measurement is taken in centimeters (the metric system is preferred).

in: Check this box if the measurement is taken in inches.

<u>**Testies/Ovaries**</u>: Gonadal measurements are used to identify and quantify reproductive maturity and are evaluated for evidence of reproductive status. In this section, please provide the measurements of the gonadal length and weight.

Length: The full length measurement of the gonads (left and right separately).

Mid-Width: The arc girder length from the center of the gonad to the end.

**<u>Mid-Depth</u>**: The depth format the center of the gonad.

**<u>cm</u>**: Check this box if the measurement is taken in centimeters (the metric system is preferred.)

in: Check this box if the measurement is taken in inches.

Weight: The total weight amount of gonad. For testis, weigh without epididymus attached.

**g**: Check this box if the weight of the animal is measured in grams or kilograms (metric system is preferred).

oz: Check this box if the weight of the animal is measured in ounces.

<u>Corpora lutea #</u>: The Corpora lutea is the mass of tissue that forms when the granulosa cells move into the Graafian follicle after ovulation. The presence of a CL is indicative of recent ovulation and the uterus should be examined closely for the presence of a blastocyst.

**<u>Corpora albicantia #</u>**: The mass of white scar tissue formed in an ovary replacing the corpus luteum. Because corpora albicantia persist in cetaceans, the number of corpora albicantia can be used to approximate the maximum number of pregnancies.

**Follicle # and maximum diameter**: The presence of a large follicle indicates imminent ovulation and is a sign of sexual maturity. Maximum follicle diameter is measured as a mechanism to understand seasonality of ovulation and whether an immature female is close to maturity.

## Specify Units of Measurements:

Cetaceans:		2	
Snout to ant. ins. of flipper:	Girth:	Axillary:	
Snout to center of genital aperture:		Max:	a water
Snout to center of anus:		Anal:	(Location)
Flipper length:	Blubber thickness:	Thoracic:	
Fluke width:		Dorsal:	
Fluke notch to anus:		Lateral:	
Total counts: UL/LL:	UR/LR:	Ventral:	

<u>Specify units of measurements</u> – indicate using centimeters (cm) (metric system is preferred) or inches (in)

For measurement of all species, use straight length, not contoured. Refer to Geraci, J.R. and V.J. Lounsbury. 2005. Marine Mammals Ashore: A Field Guide for Strandings, Second Edition. National Aquarium in Baltimore, Baltimore, MD.

## Cetaceans:

**Snout to ant.(anterior) ins.(insertion) of flipper**: This is measured from snout to anterior insertion of flipper while the animal is lying flat, dorsal side up if possible.

**Snout to center of genital aperture**: This is measured from snout to center of genital aperture.

Snout to center of anus: This is measured from snout to center of anus.

**<u>Flipper length</u>**: This is measured from axilla (caudal attachment of the flipper to the body) to the tip of the flipper.

**<u>Fluke width</u>**: This is measured from tip to tip across the widest part of the fluke while the animal is lying flat, dorsal side up if possible.

Fluke notch to anus: This is measured from fluke notch to center of anus.

<u>Total Counts</u>: These are the total tooth counts of the animal.

<u>**UL/LL**</u>: Upper left/Lower left.

**<u>UR/LR</u>**: Upper right/Lower right.

Girth: The measurement around the body of the cetacean.

Axillary: This is the circumference of the animal measured at the caudal insertion of the flippers.

<u>Max</u>: This is the maximum circumference of the animal measured. Specify location next to the measurement.

Anal: This is the circumference of the animal measured at the level of the anus.

**Blubber Thickness**: For small cetaceans, such as the beluga, this is the sternal blubber thickness. For large cetaceans, such as the bowhead whale, the measurement might have to be made midway along the side depending on how the animal is lying.

**<u>Thoracic</u>**: This is the measurement of blubber thickness along the body over the thorax with three levels being measured.

**Dorsal**: This is the measurement of the blubber thickness along the dorsal surface (midline) over the thorax.

**Lateral**: This is the measurement of the blubber thickness at the mid-lateral position (halfway between the dorsal midline and the ventral midline) over the thorax.

**Ventral**: This is the measurement of the blubber thickness at the ventral midline over the thorax.

<u>Pinnipeds:</u>		
Nose to tail length:	Ant. length of hind flipper:	
Ant. length of foreflipper:	Blubber thickness over post. end of sternum:	
Axillary girth:	Other blubber thickness:	
Bacculum length:		(Location)

# Pinnipeds:

Nose to tail length: This measurement is the total length of the animal, from tip of snout to tip of tail.

<u>Ant.(anterior). length of foreflipper</u>: This is the measurement from the base of the foreflipper anteriororly to the tip of the foreflipper.

**<u>Axillary girth</u>**: This is the circumference of the animal measured just posterior of the front flippers.

**Bacculum length**: The length of the bacculum from tip to tip after excision from the penis.

<u>Ant.(anterior) length of hind flipper</u>: This is the measurement of base of the hind flipper anteriorly to the tip of the hind flipper.

<u>Blubber thickness over post.(posterior) end of sternum</u>: This is the thickness of the blubber at the posterior end of the sternum.

**Other blubber thickness**: If additional blubber thickness measurements are taken, note here the thickness and location.

<u>Polar Bears:</u>		
Girth of neck of axis:	Skull length:	
Girth of neck at shoulders:		9 <b>7</b>

# Polar Bears:

<u>Girth of neck of axis</u>: This measurement is the circumference at the top of the neck, just below the head and behind the ears.

Girth of neck at shoulder: This is the circumference at the base of the neck, just above the shoulders.

<u>Skull length</u>: (condylo basal length) Measure using calipers with bear in lateral recumbent position. Using your knee, bend the head to near 90 degrees at the neck to facilitate feeling the back of the skull. Measure distance between upper dental plate and bony process at the base of the skull.

Snout to angle of mouth: 	Right forepaw v Skull width:	vidth:			•	_
Axillary girth:	Tooth Wear:	Heav	γ	Med.	🖲 Light	None
Extimate of body fat stores:		None:	Little:	Average:	Excessive:	
	Subcutaneous:	۲	۲	۲	۲	
	Groin:cm	۲	۲	۲	۲	
	Kidneys:		۲		۲	
	Mesenteric:			۲	۲	

## Sea Otters:

<u>Snout to angle of mouth</u>: Open the sea otter's mouth and measure from the tip of the snout to the angle of the mouth.

**Skull length**: Cut away tissues so that the base of the skull is exposed where the neck vertebrae joins the skull. Place the large calipers at the base of the skull and extend it to the gum line of the upper incisors. Measure the distance between the jaws of the calipers in centimeters (cm) on the plastic ruler or measuring tape to the nearest 0.1 cm.

<u>Axillary girth</u>: On a flat surface, with the animal in ventral position, place the measuring tape beneath it. Wrap the tape around the otter at the xiphoid process (the tape should be snug but not pulled tight). The xiphoid process is located at the point where the last rib connects at the bottom of the sternum (breast bone). Record the circumference to the nearest 0.5 cm.

**<u>Right forepaw width</u>**: Measure the width of the right forepaw across the widest portion of the foot at the level with the pad using a small plastic calipers. Record the measurement to the nearest 0.1 cm.

**Skull width**: Cut away tissues at the zygomatic arch to expose the bone. The zygomatic arch is the "cheek bone" and it is an arch of bone that extends along the side of the skull beneath the eye. Place the large calipers on the outermost point of each zygomatic arch. Measure the distance between the jaws of the calipers in centimeters on the plastic ruler or measuring tape to the nearest 0.1 cm.

**Tooth Wear**: Measure base of upper right canine tooth with plastic calipers from front to back at gum line. Measure widest part visible without pushing gums down. Record to the nearest 0.1 cm.

**<u>Heavy</u>**: Check this box if the incisors and molars are worn to or near gumline, tips of canines are rounded, and molars are flattened with heavy pitting.

<u>Med. (Medium)</u>: Check this box if the incisors and molars show wear but the wear does not extend as far as the gumline. Moderate pitting in the molars may be observed.

**Light**: Check this box if there is slight rounding of the cusps of the molars and incisors, little or no pitting in the molars.

<u>None</u>: Check this box if the fresh pointed canines, incisors, and cusps of molars show no flattening or pitting on them. This is usually observed in animals less than or equal to one year of age. Check to see if the molars are in the process of replacement with adult teeth.

### **Estimate of body fat stores**:

Body fat stores in sea otters are evaluated at four levels: <u>None</u>: no visible fat on tissue. <u>Little</u>: some fat visually detected on tissue. <u>Average</u>: fat is easily detected on tissue. <u>Abundan</u>t: fat is simple and covers much of the surface of the tissue.

Fat is evaluated at four distinct sites on the animals.

**Subcutaneous**: Fat that is located between the skin and muscle layers that is readily visible after the animal has been skinned.

<u>**Groin</u>**: Fat in the area on the inside of the thigh where the leg meets the body. This is measured by slicing the fat layer with the scalpel and inserting a plastic ruler, measuring to the nearest 0.1cm.</u>

<u>**Kidneys</u>**: Fat located on the top of the kidney and the fat occurring between the reniculi of the kidney itself. Whole right kidneys are being collected to attempt to develop a quantitative measure for this fat store.</u>

<u>Mesenteric</u>: Fat associated with the supportive membranes which supply the intestines with blood.

# ADDITIONAL SAMPLES LIST

Field ID Number:				Genus s	pecies:	
Was animal necropsied	? 🛞 Yes	No No				
Necropsied by:				dd / mr	n/yy	
(Plea	ase attach necrop	nsy report)		Dat	te	
Samples collected:						
Histological samples:						
Individual/Organiza	ation:			Final destination	n:	
Tissues sampled:	Liver	Kidney	Blubber	Stomach	Heart	Intestine
(Choose all that apply)	🔲 Lung	Pancreas	Adrenals	🔲 Brain	Muscle	Skin
	Trachea	Spleen	Thymus	Colon	Thyroid	Esophagus
01						
Other: (Pleas	2000-0 m-10					
(11005						
	-					
Lymph Nodes:	Cubm	andibular 🔲 Pres	scapular 🔲 Axilli	ary 🔲 Hilar	Mesenteric	
Lymph Nodes.			capulai 🗀 Axili	ary 🗀 Allar	La mesenteric	
	Other I.	ı.: <sub></sub>				

**Field ID Number** is the unique identifier assigned to the animal from which the tissue(s) were obtained. Format is open to each agency's requirements; however, please remain consistent within your agency. This is automatically populated from Page 1.

<u>Genus species</u> is the Latin name for the animal in standard binomial nomenclature. It is carried over from NIST Sample Processing – Page 1.

#### Was animal necropsied?

<u>Yes</u>: Check this box if a full necropsy was undertaken to examine the carcass determining the cause of death as well as obtaining tissues samples for submission.

<u>No</u>: Check this box if a necropsy was not completely performed but tissues samples were obtained for submission.

<u>Necropsy by</u>: List the name and contact information of the primary person/facility who conducted the necropsy. In addition, attach a copy of the necropsy report to the tissue bank form.

Necropsy Date: Provide the date (DD/MM/YYYY) when the necropsy was done.

**Samples collected**: Indicate the tissue samples collected other than the NMMTB samples from the same animal for other research or diagnostic or archival purposes from the checklist provided in this section of the form.

<u>**Histological samples**</u>: If samples are collected for histolopathological examinations, these are recorded here. This indicates that specimens taken from the same animal for histological analysis are used for the direct assessment of the animal's cause of death.

**Individual/Organization**: List the person or facility that conducted the removal of these tissue samples.

**Final Destination**: This is the final location of where the histological samples/slides will be sent for analysis.

### **Tissue Samples**:

<u>**Tissue List</u>**: Check the box next to each tissue collected for histological analysis. These tissues are liver, kidney, blubber, stomach, heart, intestine, lung, pancreas, adrenals, brain, muscle, skin, trachea, spleen, thymus, colon, thyroid, and esophagus (i.e. check the box next to the liver if the liver was collected).</u>

<u>**Other**</u>: List additional tissues that were collected for histological analysis that were not included in the tissue sample list above.

**Lymph Nodes**: Check the box next to each lymph node if that lymph node was collected for histological analysis (i.e. check submandibular if the submandibular lymph node was collected...).

**<u>Other l.n.</u>**(lymph nodes): List additional lymph node samples that were collected for histological analysis that were not included in the lymph node list above.

Other samples collected:	Type of storage:	Where located (Ind./Org.):
	(Z-frozen, F-formalin, DMSO, ETOH)	
Teeth:		
Genetics (skin):		
Skull:		
Reproductive tract:		· · · · · · · · · · · · · · · · · · ·
Mammary tissue:		
Ovaries:		
Gonads/testes:		
Parasites:	<u>2</u>	(A
List type and location:	······	7
Stomach:	101 101 101	
List contents if applicable	۲	08
	43	
Other contaminant samples:		
(List tissue type, storage type and where located)	-	
Additional samples:		
(List tissue type, purpose of collection, storage type and where located)		
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	*	

<u>Other Samples Collected</u>: Samples collected from the same animal for other research purposes are recorded in this section. This information includes the kind of sample (s): Teeth, Genetics (skin), Skull, Reproductive tract, Mammary tissue, Ovaries and Gonads/testes.

**<u>Type of Storage</u>**: (Z-frozen, F-Formalin, DMSO, ETOH-ethyl alcohol, other...)

<u>Where located (Ind./Org.)</u>: Indicate the name and organization of the person (s) where the tissues are located.

**<u>Parasites (List type and location)</u>**: In addition to type of storage and where located (Ind./Org.), also list the type of parasite (s) collected and where in the cavity the parasite came from and indicate the number of parasites (0-20, 21-100, 100+).

<u>Stomach (List contents if applicable)</u>: In addition to type of storage and where located (Ind./Org.), identify food items if known and record some impression of degree of fullness. Note if internal parasites are present as well.

<u>Other contaminants/biotoxin samples</u>: (list the tissue type, storage type and where located). If other tissues are collected for contaminants or biotoxin analysis, list the tissue type sampled, storage type and where the tissue is being stored (Ind./Org.).

<u>Additional samples</u>: (list tissue type, purpose of collection, storage type and where located) If additional samples are collected that were not listed above, list tissue type, purpose of collection, storage type and where the sample is located (Ind./Org.).

Field ID Numbe		-		Genus species:	
Photos taken:	🖲 Yes 🏾 🔘 No	💿 Digital 💿 Film	If yes, how many?		
Video taken:	🖲 Yes 🍥 No			. (send copy with samples for NIST archive)	
Disposition:					
(primary locatio	in				<u>.</u>

**Field ID Number** is the identifier assigned to the animal from which the tissue(s) were obtained. Format is open to each agency's requirements; however, please remain consistent within your agency. It is carried over from NIST Sample Processing – Page 1.

<u>Genus species</u> is the Latin name for the animal in standard binomial nomenclature. It is carried over from NIST Sample Processing – Page 1.

### Photos taken:

Yes: Check this box if photographs were taken of the tissue sample or animal.

**<u>Digital</u>**: Check this box if photographs were taken with a digital camera.

**Film**: Check this box if photographs were taken with a film camera.

**If yes, how many?**: Indicate the number of photographs taken of the tissue specimen or animal. In addition, send copy of images with the samples for NIST archive.

<u>No</u>: Check this box if there were no photographs taken.

### Video taken:

Yes: Check this box if video was taken of the tissue sample or animal.

No: Check this box if there was no video taken.

**Disposition** (**Primary location of photos and/or video**): If photos and/or video were taken, list where the photos and/or video are located and the individual and organization responsible for the photos and/or video.

General comments:	-	
(Field notes)		18
25 E		
Consultant and the distribution		3
General appearance of individual:	P	
General appearance of organs: -		
	-	3
	04	
NMMTB Protocol:   Standard	Modified	
]		E
Please note any modifications:	2	_ [
	·	÷
×.		

<u>General comments</u> (Field notes): Any additional comments which were obtained from the field that were not indicated elsewhere on the data sheet.

<u>General appearance of individual</u>: Any comments, which describe the healthy or unhealthy appearance of the animal, external parasites, evidence of trauma, or body condition of the animal.

<u>General appearance of organs</u>: Any unusual appearances of any of the internal organs, particularly those to be sampled. If they all appear normal, note this also.

### **NMMTB Protocol**:

**<u>Standard</u>**: Check this box if there were no changes made to the protocol to obtain the tissues for the NMMTB.

**Modified**: Check this box if changes were made to the protocol to obtain the tissues for the NMMTB and indicate modifications below.

<u>Please note any modifications</u>: List the modifications made to the NMMTB protocol here.

Form prepared by:		A copy of this form and Level A Data Form should be shipped with samples to:
	Name	ATTN: Rebecca Pugh National Institute of Standards and Technology Hollings Marine Laboratory
	Affiliation	331 Fort Johnson Rd Charleston, SC 29412 (843) 762-8952

**Form prepared by**: Indicate the Name and Affiliation of the person (s) recording the data for the form.

Field ID Number:		
Other ID Number:		
VMMTB Reference/Storage ID Numbers:		
( <del></del>		
		dd / mm / y
Collector's signature	Method of transfer to processing stage	Date
		dd / mm / y
Processor's signature	Method of transfer to shipping stage	Date
		dd / mm / y
Shipper to NMMTB's signature	Method of transfer to MESB	Date
		_dd / mm / y
Receiver's signature		Date

**Field ID**: This is the identifier assigned to the animal from which the tissue(s) were obtained. Format is open to each agency's requirements; however, please remain consistent within your agency.

<u>Other ID Number</u>: This is the identifier related to the Field ID or identification of the animal. Examples include: previous field ID numbers if this animal previously stranded; ID numbers assigned by other organizations (including authorized rehabilitation facilities to which the animal is transferred), former identification numbers from scientific research projects, etc.

<u>NMMTB/Reference/Storage ID Numbers</u>: These numbers will be assigned by NIST personnel when the samples are received at the NMMTB in Charleston, SC.

<u>Collector's Signature</u>: This is the signature of the person that removed the tissue (collected) from the animal prior to processing the tissue.

<u>Method of Transfer to Processing Stage</u>: This is the method that was used to transfer the tissues from the collection site to the processing site, if applicable. (i.e. by hand, by truck, by cooler, Federal Express, DHL, UPS, USPS or indicate other if needed).

**<u>Date</u>**: This is the date (*DD/MM/YYYY*) that the samples were transferred from the collection site to the processing site.

<u>**Processor's Signature**</u>: This is the signature of the person that processed the tissue for eventual liquid nitrogen freezing and shipment to the NMMTB in Charleston, SC

<u>Method of Transfer to Shipping Stage</u>: This is the method that was used to transfer the tissues from the processing site to the shipping site, if applicable. (i.e. by hand, by truck, by cooler, Federal Express, DHL, UPS, USPS or indicate other if needed).

**<u>Date</u>**: This is the date (*DD/MM/YYYY*) that the samples were transferred from the processing site to the shipping site.

**Shipper to NMMTB's Signature**: This is the signature of the person that shipped the samples to the NMMTB in Charleston, SC.

<u>Method of Transfer to MESB</u>: This is the method that was used to transfer the tissues from the shipping site to the NMMTB in Charleston, SC. (i.e. by hand, by truck, by cooler, Federal Express, DHL, UPS, USPS or indicate other if needed).

**<u>Date</u>**: This is the date (*DD/MM/YYYY*) that the samples were transferred from the shipping site to the NMMTB in Charleston, SC.

**<u>Receiver's Signature</u>**: This is the signature of NIST personnel at the NMMTB in Charleston, SC that received the samples for archival.

**<u>Date</u>**: This is the date (*DD/MM/YYYY*) that the samples were received at the NMMTB in Charleston, SC.

### References

- Becker, P.R., S.A. Wise, B.J. Koster, R. Zeisler. 1988. Alaskan Marine Mammal Tissue Archival Project: A Description Including Collection Protocols. U.S. Dep. Commer., National Bureau of Standards, NBSIR 88-3750. Gaithersburg, Maryland.
- Becker, P.R., S.A. Wise, B.J. Koster, R. Zeisler. 1991. Alaska Marine Mammal Tissue Archival Project: Revised Collection Protocol. USDOC, National Institute of Standards and Technology, NISTIR 4529. Gaithersburg, MD.
- Becker, P.R., Porter, B.J., Mackey, E.A., Schantz, M.M., Demiralp, R., and Wise, S.A. 1999. National Marine Mammal Tissue Bank and Quality Assurance Program: Protocols, Inventory, and Analytical Results. U.S. Department of Commerce, National Institute of Standards and Technology, NISTIR 6279. Gaithersburg, MD. 183 pp.
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- Geraci, J.R. and V.J. Lounsbury. 2005. Marine Mammals Ashore: A Field Guide for Strandings, Second Edition. National Aquarium in Baltimore, Baltimore, MD.