

Photographs (see page 2) and morphometrics should be collected for all animals
 Chain of custody procedures should be followed for ALL samples, photographs, and datasheets
 If the animal is visibly oiled please call 877-433-8299 immediately

3/30/11

Field # LA 495/MCT-20110329-
LA001

**indicates priority sample

PAH/Chemistry:
 Code 2 & 3, 10-20g or 5 ml
 Freeze -20, I-chem vial or foil
 Blubber
 Liver
 Lung
 Kidney
 Freeze -20, I-chem or glass vial:
 **Bile
 **Urine
 Whole blood
 Aqueous humor

PAH/Chemistry con't:
 Freeze -20, foil, dull side in
 All conditions:
 **Swab
 **Tongue depressor (visibly oiled)

Biotoxins:
 Freeze -20, codes 2-4, 1 ml or 5g:
 **Feces
 **Urine (if enough after PAH)
 Liver

CYP 1A:
 Code 2 & 3, 1 cm thick section:
 Freeze, cryovial, -80 if possible
 Skin+full-thick blubber section
Formalin, 10:1 ratio
 Skin+full-thick blubber section

Parasites (codes 2 - 4):
 In 70% Isopropyl alcohol
 Representative sample from each species and each location
 External
 Internal

Viral:
 Code 2 and early 3: 2 sets - 1 in RNA later vial and 1 in cryovial, both frozen at -80
 Code late 3 and 4 - 1 set of priority samples (**) in RNA later if possible
 **Brain RNA later -80 Grossly affected Lymph nodes and other grossly affected tissues (list):
 Lung assoc LN- RNA later -80
 **Lung RNA later -80
 **Spleen RNA later -80
 **Adrenal gland RNA later -80
 Kidney RNA later -80
 **Liver RNA later -80

Life History (Codes 2 - 5):
 Freeze, -20:
 **Teeth (4 or whole mandible)
 **Skin (isotope)
 **Stomach whole (prey, biotoxin, & PAH)
 (see stomach instructions page 2)
 In DMSO or frozen:
 **Skin (genetics) LDWF

Bacterial Culture Swabs or tissues:
 Code 2 Ship to lab immediately on ice packs
 lung assoc lymph node
 **Brain Liver
 Spleen **Lung
 Other at vet's discretion:

Histology (code 2 & 3):
 10% neutral buffered formalin, 10:1 formalin:tissue ratio
 Samples fixed in formalin should be no more than 1 cm thick
 **Lung
 **Trachea
 **Heart (all 4 chambers)
 Aorta
 Pulmonary Artery
 Thymus
 **Thyroid
 **Laryngeal Assoc. Lymph. Tissue
 Pharynx
 Tongue
 Esophagus
 Small Intestine (proximal, mid, distal)
 Colon
 Pancreas
 **Spleen
 Spinal Cord (specify location)
 **Brain (cerebrum, cerebellum, stem, pituitary)
 **Liver
 **Kidney
 **Adrenal Gland
 Urinary bladder
 Blubber
 Skin
 Muscle (specify location)
 Eye (L/R)
 Gonad
 Mammary Gland
 **Uterus/Uterine horn
 Vagina & Cervix
 Prostate & Penis
 Lymph nodes (list):
 **Thoracic
 **Abdominal/Genital
Sublumbar

Live animals:
Blood:
 Collect the following and please see preparation information on page 2
 1 - 3ml purple top K₂EDTA
 5 - 10 ml red/gray tiger top SST tubes
 1 green top sodium heparin tube
Freeze -80 in Teflon for PAH:
 Whole blood
Swab:
 in viral transport media
 Blowhole Swab (ship immediately on ice packs if possible. If not, freeze at -80)

Pregnant or postparturient females (code 2 & 3):
 Uterine horn: Culture swab Formalin/histo -80 RNA later
 **Placenta: Culture swab Formalin/histo -80 RNA later
 **Amniotic fluid: Culture swab -80 RNA later
 Genital LN: Culture swab Formalin/histo -80 RNA later
 Fetus: follow fetal/neonate (<115 cm) protocol

Tursiops <115 cm (see also pg 2):
 Code 2/early 3 or visibly oiled:
 necropsy ASAP by network vet
 **Radiograph (before necropsy - dorso-ventral view skull & body)
 **Fetus/neonate datasheet
 **Skull and skeleton, freeze intact
 **Umbilical stump & body wall (formalin)
 Serum, from heart, freeze at -20, cryovial
Code Late 3 - 5 (no visible oil):
 **Freeze whole (photos, length, PAH swab first)

(E)

Other Samples Collected:		
<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
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Standardized external photographs - As feasible, please collect the following from all animals:

Whole body: left side, right side, dorsal, ventral Close ups: Dorsal fin, head, genitals Neonate add: umbilicus, tongue/mouth

Tursiops <115 cm additional details:

Code 2 OR early code 3 OR visibly oiled - any condition: These animals should be necropsied as soon as possible by a stranding network veterinarian. If your vet is unavailable, call the stranding pager and we will help find you an alternative.

Please collect dorso-ventral view radiographs of the skull & body before necropsy. Please save the entire skull and skeleton.

When collecting the urinary bladder for histo be sure to collect it whole or collect the tip and include the urachus.

Late code 3 - 5 (no visible oil): These should be frozen whole. Please collect your external PAH swabs at the stranding site.

Please take external photographs and the genetics skin sample in DMSO before freezing. If time allows, please score the external neonate characteristics from the neonatal characters data sheet

Blood preparation instructions - Blood from live animals:

The following blood tubes are for CBC, serum chemistry and serum electrophoresis:

Send 1st samples from intake or beach to Cornell and a pre-release sample to Cornell, other diagnostics may be done locally

Send on ice packs the night of collection, if not possible, send the next morning:

1 - 3ml purple top K3EDTA - refrigerate, send whole on ice packs

1 - 10 ml red/gray tiger top SST tube - spin and refrigerate, send on ice packs

The following blood tubes are for hormones and can be shipped to Cornell in batches:

(hormones include: cortisol, progesterone, testosterone, aldosterone and thyroid hormones - total T4, free T4 and total T3)

2 - 10 ml red/gray tiger top SST tubes - spin and remove the serum from both into a 5 ml cryovial, freeze at -80, ship on dry ice

Serum for archive:

1 - 10 ml red/gray tiger top SST tube - spin, remove serum into a cryovial and archive at -80

Serum for biotoxin:

1 - 10 ml red/gray tiger top SST tube - spin, remove serum into a cryovial, freeze at -20 until shipment on dry ice

Buffy coat (if possible):

1 green top tube, spin, carefully remove buffy coat into a cryovial and freeze at -80

Blood preparation instructions - Blood from dead animals:

Tursiops calves <115 cm - code 2 : Use red/gray tiger top SST tubes, collect as much blood as possible from a heart stick - spin, remove serum into a cryovial, freeze at -20 for future shipment, will be used for electrophoresis

Code 2 or 3 - please collect whole blood in an i-chem or glass vial and freeze at -20 for PAH

Stomach special instructions:

Tie off the stomach at both ends. Remove whole stomach and freeze in plastic. Do not open it!

A person will be identified who will receive the stomachs, examine them for internal lesions,

collect histology samples of lesions as needed, identify the prey and subsample for biotoxins

PAH/Chemistry:

If you have questions regarding collection methodology, please see attached Appendices 6 & 7 from the Marine Mammal Oil Spill Response Guidelines

Uia copy
Northern Gulf UME Sample Collection Checklist (Franklin County, FL to LA/TX Border) March 11, 2011

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<input type="checkbox"/> **Thyroid	<input checked="" type="checkbox"/> Muscle (specify location)
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Appendix 9. Oiled Marine Mammal Necropsy Form

Oiled Marine Mammal

Gross Necropsy Report

Spill Name: BP Deepwater Horizon	
Animal Log # MCT20110321-LA001 / LA 495	
Level A Field #	Tag #
Species: <i>Tursiops truncatus</i>	
Sex: M	Age: adult
Weight: _____ kg. (estimate/actual)	
SON: emaciated 1 2 3 4 5 6 7 obese	
Measurements: Blubber depth (bw) mm	
SL <input type="checkbox"/> Partial ear	AG cm UG cm XG cm

Form completed by: Cara Field		Date: 3/30/11
Enforcement Officer:		
Strand/Capture location:		
Collectors Name:		
Intake date:		Euthanasia
Death date: unknown		<input type="checkbox"/> yes
Post mortem date: 3/30/11		<input checked="" type="checkbox"/> no
Carcass Classification:		Frozen: <input type="checkbox"/> yes <input type="checkbox"/> no
2- fresh, 3- fair (organs intact) 4- poor 5- mummified		
Photographs: <input checked="" type="checkbox"/>		
Radiographs: <input type="checkbox"/> no		

Clinical signs/diagnosis:
Antibiotics given:
Pertinent lab results:

Ø

GROSS NECROPSY ABNORMALITIES: Blubber depth
Dorsal: 16mm Lateral: 15mm Ventral: 13mm

External: Penis erected, skin has multiple large bubbles underneath (separation of dermis), and is peeling off of dorsal fin, flukes + other areas. Carcass extremely bloated. Tongue severely bloated + black/green. Numerous rake and other assorted scratches over dorsum.

Teeth: all present and appear to be in good shape (little wear)

Eyes protruding + dried.

Internal: All tissues severely autolyzed. Large # gas released when incision made into cranial abdomen (explosive w/ intestinal protrusion).

Collected minimal samples for Code 4 (viral + life hx) + partial histo.

HYDROCARBON ANALYSIS SAMPLES		MICROBIOLOGY		SWABS: other	
bile	kidney	Lung	<input type="checkbox"/>		
blood	muscle	Liver	<input type="checkbox"/>		
urine	blubber/fat	other	<input type="checkbox"/>		
liver	other:				

HISTOLOGY SAMPLES				LYMPH NODES:	
lung	thyroid	ileum	kidney	muscle	colonic
trachea	tonsil	colon	ureter	gonad	gastric
heart	tongue	pancreas	urinary bladder	prostate	hepatic
aorta	esophagus	spleen	urethra	uterus	mediastinal
pulmonary artery	stomach	liver	blubber	vagina	inguinal
thymus	duodenum	gall bladder	bone marrow	cervix	axillary
salivary gland	jejunum	brain	spinal cord	penis	mesenteric
other:					tracheobronchial

Cause of death (preliminary diagnosis): open
 Examiner: Cara Field Examiner's signature: C. Field Date: 3/30/11

E