NEARSHORE CETACEAN & SEA TURTLE PREY ITEM SAMPLING PLAN

Prepared by:

The Prey Species Information Subgroup
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For the Marine Mammals and Sea Turtles Working Group of the
Mississippi Canyon 252 Trustees

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Approval of this work plan is for the purposes of obtaining data for the Natural Resource Damage Assessment (NRDA). Each party reserves its rights to produce its own independent interpretations and analyses of any data collected pursuant to this work plan.

The trustees have developed a preliminary conceptual model of the DWH release, potential pathways and routes of exposure, and potential receptors. This preliminary model has informed the trustees' decision to pursue the studies outlined in the work plan. By signing this work plan and agreeing to fund the work outlined, BP is not endorsing the model articulated in the work plan.

This plan will be implemented consistent with existing trustee regulations and policies. All applicable state and federal permits will be obtained prior to conducting work.

APPROVED:

Tom Brosnan 7/6/11

NOAA Trustee Representative: **Date**

FOR ROLAND GUIDRY 7/11/11

Louisiana Trustee Representative: **Date**

Joyce Hulley 7-7-2011

BP Representative: **Date**

Summary

This document presents an assessment plan for prey items of nearshore cetaceans and sea turtles in Louisiana, where habitat has been impacted by Mississippi Canyon 252 (MC 252) oil and dispersants, (hereafter referred to as "MC 252 oil") , as confirmed by Louisiana Department of Wildlife and Fisheries (LDWF) field observations (LDWF, personal communication). The data collection described in this plan targets ephemeral data and can be reasonably expected to support a natural resource damage assessment under the Oil Pollution Act.

Objective

To obtain representative samples of prey species potentially consumed by nearshore cetaceans and sea turtles that are known to inhabit the geographic area of interest over a gradient of potential MC 252 oil impacts.

The Plan specifically addresses the following topics:

I. **Approach and rationale.** This section describes the overall purpose of and need for a prey sampling plan as a way to evaluate the potential for nearshore cetacean and sea turtle exposure to MC 252 oil via the trophic transfer exposure pathway. This plan describes monthly surveys to be conducted for approximately 10 months, beginning upon signature of this plan.

II. **Data needs and sources.** This section provides an overview of the types of data that will be collected to further the assessment of nearshore cetacean and sea turtle prey species in the northern Gulf of Mexico.

III. **Health and safety.** This section summarizes health and safety protocols that are relevant to this effort. It includes a number of procedures by reference, all of which should be carefully reviewed and adhered to by all field team members.

IV. **Site selection strategy.** This section describes the approach for identifying sites for evaluation.

V. **Site procedures.** This section provides guidance on how to handle the samples and data gathered in the field. This section makes frequent reference to protocols in Appendix I and also to NRDA-wide procedures that are incorporated by reference. Field team members should make sure they understand and adhere to procedures, whether included here directly or by reference.

Appendix I: Detailed Standard Operating Procedures (SOPs) and Field Data Sheets. This section will be compiled as SOPs and field data sheets become available. Preliminary draft SOPs will be shared with BP or its representative before this plan is signed.

Appendix II: Protected Species Mitigation Measures (Protected Resources, NOAA NMFS Southeast Regional Office)

I. Approach and Rationale

Consumption of prey contaminated with MC 252 oil (exposure via trophic transfer) could potentially cause injury to either or both nearshore cetaceans and sea turtles in the Gulf of Mexico (Peterson et al., 2003). More specifically, within the geographic scope of this plan, bottlenose dolphins (*Tursiops truncatus)*, Kemp's ridley (*Lepidochelys kempii)* and loggerhead (*Caretta caretta)* sea turtles consume prey items that may have been exposed to MC 252 oil. To help determine whether exposure has occurred, samples of the prey items will be collected for contaminant analyses, to be addressed in an addendum to this plan. The Trustees shall provide a draft of the cooperative analytical addendum to BP by August 15, 2011. BP and the Trustees agree to work together in good faith to cooperatively develop this addendum and agree that the addendum will include analyses for Polycyclic Aromatic Hydrocarbons (PAHs) and fingerprinting for MC252 oil, where technically practicable, and a schedule of delivery for data resulting from the proposed analyses within a reasonable timeframe. However, if BP and the Trustees are unable to reach consensus on any individual element(s) of the analytical addendum, the Trustees reserve the right to proceed independently on those elements on which no agreement was reached. If the Trustees and BP have not entered into a cooperative analytical addendum by October 1, 2011, upon request of BP, samples will be split if practicable and each Party may proceed independently.

This study will attempt to determine levels of prey exposure to MC 252 oil by sampling key prey items in inshore and nearshore Louisiana waters (no further out than the 10-meter isobath). Known prey items will be targeted for collection using trawls and gill nets. This study plan has been reviewed by NOAA protected resources staff in the NOAA NMFS Southeast Regional Office to ensure that appropriate mitigation measures are taken. This review resulted in the identification of preventive measures to help avoid marine mammal or sea turtle interactions during scheduled NDRA sampling. Based on this review and subsequent follow-up conference calls with NOAA NMFS staff, the sampling teams will undertake measures to help avoid marine mammal and sea turtle interactions as noted in the Protected Species Mitigation Measures document in Appendix II.

BP or its representative will be provided an opportunity to participate in all sampling events described in this plan.

II. Data needs and sources

Table 1 provides a list of typical prey items for bottlenose dolphins, Kemp's ridley and loggerhead sea turtles (Barros & Odell, 1990; Bjorndal, 1996; Barros & Wells, 1998; Maze-Foley & Mullin 2006; etc.). Field teams will attempt to collect up to 2 independent samples of any targeted species listed in Table 1 in each net haul or trawl at each sampling station (see Section V for site procedures). Each of these samples should weigh approximately 30 g to provide adequate material for future analyses. A spring scale will be utilized by field sampling teams to weigh samples. If individual organisms within a species weigh less than 30 g, field teams will collect enough individuals to obtain two samples of at least 30 g each. Each haul will be sorted to species, and individuals will be selected at random from the sorted species until two 30 gram samples have been collected for each species. If individual organisms weigh > 30 g, field sampling teams will collect 2 individuals and prepare each as a separate sample. Samples of muscle tissue (approximately 30 g each from 2 individuals) will be taken from fish that are > 600 mm in length (see Appendix 1 for detailed SOP).

Although not proposed as part of the current scope of the work in this work plan, sampling and potential analyses of liver and bile samples will be addressed separately by the Trustees.

Table 1: Typical prey items for bottlenose dolphins, Kemp's, and loggerhead sea turtles (Barros & Odell, 1990; Bjorndal, 1996; Barros & Wells, 1998; Maze-Foley & Mullin 2006; etc.).

Dolphin Prey		Sea Turtle Prey	
Common name	**Scientific name**	**Common name**	**Scientific name**
Atlantic croaker	*Micropogonias undulatus*	blue crab	*Callinectes sapidus*
bay anchovy	*Anchoa mitchilli*	lesser blue crab	*Callinectes similus*
Gulf menhaden	*Brevoortia patronus*	speckled swimming crab	*Arenaeus cribrarius*
pinfish	*Lagodon rhomboides*	iridescent swimming crab	*Portunus gibbesii*
spot	*Leiostomus xanthurus*	Florida lady crab	*Ovalipes floridanus*
striped mullet	*Mugil cephalus*	calico box crab	*Hepatus epheliticus*
brown shrimp	*Farfantepenaeus aztecus*	flame box crab	*Calappa flammea*
pink shrimp	*Farfantepenaeus duorarum*	yellow box crab	*Calappa sulcata*
white shrimp	*Litopenaeus setiferus*	mottled purse crab	*Persephona mediterranea*
Atlantic brief squid	*Lolliguncula brevis*	portly spider crab	*Libinia emarginata*
Gulf toadfish	*Opsanus beta*	incongruous ark	*Anadara brasiliana*
hardhead catfish	*Ariopsis felis*	blood ark	*Anadara ovalis*
sand seatrout	*Cynoscion arenarius*	Atlantic cutlassfish	*Trichiurus lepturus*
silver seatrout	*Cynoscion nothus*	horseshoe crab	*Limulus polyphemus*
spotted seatrout	*Cynoscion nebulosus*		
striped anchovy	*Anchoa hepsetus*		
black drum	*Pogonias cromis*		
gafftopsail *catfish*	*Bagre marinus*		
red drum	*Sciaenops ocellatus*		
southern flounder	*Paralichthys lethostigma*		

III. Health and Safety

- The team leader and field crew shall have completed all applicable health and safety training as directed by NOAA or state agency oil spill policy.

- All field team members must complete the NOAA safety training and documentation requirements as set forth in "Safety Requirements for All Personnel Working on NOAA led NRDA teams for MS Canyon 252 Incident" (NOAA Safety Documentation Requirements.doc).

- Exception: if a specific team's site collection activities do not include use of either a boat or an aircraft, then familiarity with the safety documents for these vehicles is not required.

- At least **one week prior** to the start of field sampling, the project leader will inform the NOAA NRDA Field Ops Team [redacted]) of intended operations. This email should outline specific information regarding the operation, including at a minimum:
    - Point of Contact, including cell phone number and email,
    - Number of field teams and number of samplers per field team, including contact information for federal representatives,
    - Start date,
    - Number of anticipated field days,
    - Whether assistance in securing vessels will be needed,
    - General description of planned activities,
    - General operational area(s), and
    - Whether sample/data intake will be needed.

- Before conducting fieldwork, the following will be completed:
    - **Team Tracking Field Plan** - Updated daily to provide Field Ops with information about your operations (staff, operational area, launch, contact information)
    - **Site Safety Plan** - The team will be expected to review and comply with the current Site Safety Plan accessed on noaanrda.org in the Documentation: Safety Protocols section. The project leader must determine what required safety equipment the team needs
    - **Field teams mu**st adhere to all procedures set forth in the "Deepwater Horizon NRDA Site Safety Plan Version 12/08/2010," ("NRDA_OPS_Safety_Plan_08DEC2010.docx").[8]

IV. Site Selection Strategy

Sampling is intended to encompass an area where a gradient from heavy to little or no oiling is anticipated to exist. Sampling will be conducted in waters between 0 m to 10 m in depth from approximately 20 km west of Terrebonne Bay around Pass a Loutre to a point on the northeastern coast of the Mississippi River Delta (see Figure 2). It is anticipated that the oiling level in Barataria Bay may be on the high end of the gradient, as this area is still actively being cleaned up, and that the oiling level may diminish moving westward.

Sampling within this area will be subdivided into 5 strata: (I) Barataria Bay inshore,(II) Barataria Bay offshore, (III) Pass a Loutre, (IV) Terrebonne/Timbalier Bay inshore, and (V) Terrebonne/Timbalier Bay offshore (see Figure 1). The locations of sampling stations within each strata were allocated using a generalized tessellation stratified sampling (GRTS) scheme (Stevens and Olsen 1999; Stevens and Olsen 2004). For planning purposes, a GRTS sample of 30 locations was selected in each stratum to receive field measurements. Alternate locations were also created in the GRTS procedure to be used in the event that one or more of these 30 locations are determined, at the discretion of field sampling teams, to be unsuitable for sampling. Sites that are unsuitable for sampling by one or more gear types will not be visited, but will be replaced by the next site selected in the GRTS procedure and the reason for any

[8] This file is available or[redacted]

changes will be noted on data collection sheets. This will ensure that gear can be deployed successfully at all sites.

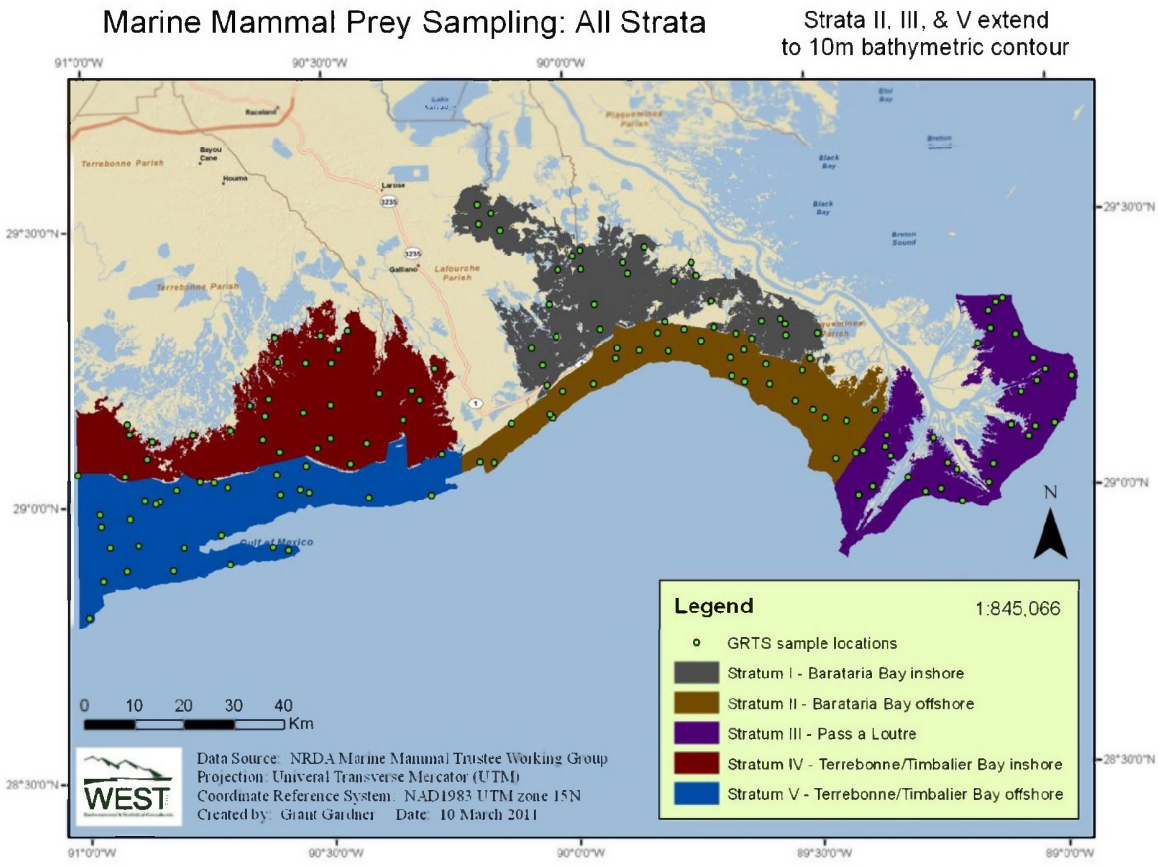

Figure 1: Sampling stations, within each stratum, that were allocated using a generalized tessellation stratified sampling (GRTS) scheme (Stevens and Olsen 1999; Stevens and Olsen 2004).

V. Site procedures

Targeted prey item sampling

The goal of the sampling effort under this Plan is to obtain whole samples and muscle tissue from nearshore cetacean, Kemp's ridley and loggerhead sea turtle prey items that will be archived per NRDA sample storage protocol (see Appendix 1) for contaminant analysis at a future date. A total of 150 sampling stations have been allocated over all 5 strata (5 strata containing 30 stations each). This is intended to ensure that an adequate number of sites are sampled in each stratum. Individual stations will be sampled once every 5 months for a total of 10 months (beginning upon signature of this plan), the stations within each of the 5 strata being sampled as a set. Field teams will thus rotate from one

stratum to the next in sequence, on a monthly basis, so that specimens from each stratum will be sampled during different seasons over the course of the study. It is anticipated that strata will be sampled in the following order (see Figure 2): Barataria Bay inshore (Stratum I), Barataria Bay offshore (Stratum II), Pass a Loutre (Stratum III), Terrebonne/Timbalier Bay inshore (Stratum IV), Terrebonne/Timbalier Bay offshore (Stratum V). Such sequencing may change based upon time of initiation, seasonal considerations, or preliminary results. Sampling will be adjusted to avoid sampling in the seasonal (late summer) hypoxic zone. This zone is not fixed from season to season, but is located primarily in Stratum II.

The Trustees and BP will meet (either in person or by phone) after seven months of sampling have been completed to discuss the need for an additional round of sampling. If BP decides it will no longer fund the study after 10 months, the Trustees may proceed independently. BP may also cease funding and withdraw from the study if 45 days pass without such a meeting despite BP's good faith efforts to schedule one. In either case, should the Trustees continue the study, BP agrees not to initiate field work that duplicates or overlaps with the Trustees' prey sampling efforts.

Field teams will use gill nets and trawls in initial sampling efforts. Where sampling stations are located at depths less than 8 feet, and with conditions appropriate for the use of gill nets (to be determined at the discretion of field sampling teams), field teams will utilize (strike) gill nets once and then trawl the study site once. Trawls will be towed for 10 minutes each (per LDWF 2010). Procedures for decontamination of these gears will be included in the SOPs in Appendix I. Effectiveness of these gears will be evaluated after each sampling event and field protocols will be amended if determined necessary by the Trustees and BP. Catch will be sorted and identified to species.

Any whole body fish < 600 mm in length will be preserved separately for analysis by wrapping in pre-cleaned aluminum foil (provided by Newfields lab) and storing on ice until it can be frozen at -20°C (within 24 hours of sample collection). For fish > 600 mm in length, muscle tissue samples (one sample) will be taken as described above in section II, and preserved separately. For species < 30 g, stored samples will consist of groups of one species taken from one site using one type of gear. For example, all white shrimp from one site caught with trawls will be wrapped in foil together. All samples will be collected by data intake teams who will meet field sampling teams at the dock at the end of each sampling day.

VI. Data Handling and Sharing

A. *Data Handling*

MC 252 NRDA chain-of-custody procedures will be observed for all samples. All samples will be transferred with appropriate chain-of-custody forms. Camera memory cards will be handled under Chain-of-Custody after a card is full or after the study is completed pursuant to the NRDA Field Sampler Data Management Protocol, 10-24-2010, which includes the protocol for transferring and uploading digital photos (most up-to-date versions of all NRDA protocols available at noaanrda.org).

All field and laboratory data will be collected, managed and stored in a secure facility under trustee control in accordance with written SOPs. The appropriate training on particular equipment or in the conduct of specific field studies for all personnel involved with the project shall be documented and those records shall be kept on file for the duration of this project.

All materials associated with the collection or analysis of samples under these protocols or pursuant to any approved work plan, except those consumed as a consequence of the applicable sampling or analytical process, must be retained unless and until approval is given for their disposal in accordance with the retention requirements set forth in paragraph 14 of Pretrial Order # 1 (issued August 10, 2010) and any other applicable Court Orders governing tangible items that are or may be issued in MDL No. 2179 IN RE: Oil Spill by the Oil Rig "DEEPWATER HORIZON" (E.D. LA 2010). Such approval to dispose must be given in writing and by a person authorized to direct such action on behalf of the state or federal agency whose employees or contractors are in possession or control of such materials.

B. *Data Sharing*

Copies of all non-analytical data collected in accordance with this plan, (including raw data, field sheets, and field notes, photos, photo logger forms and GPS files), will be transferred to the NOAA NRDA Sample Intake Team following NRDA data management protocols. An identical copy of all documentation will be provided to BP/Cardno-ENTRIX and the Louisiana Oil Spill Coordinator's Office (LOSCO) within a reasonable timeframe once data intake, QA analyses and data entry procedures are complete but no later than 45 days after the data are collected. All samples collected pursuant to this Plan will be submitted to laboratories that are operated in a manner that is consistent with the guidelines of the Analytical Quality Assurance Plan (AQAP) for the Mississippi Canyon (Deepwater Horizon) Natural Resource Damage Assessment (version 2.2).

This Plan does not include any laboratory analysis. The following provision will apply to any laboratory data generated pursuant to a cooperative addendum developed as described above in Section I:

Each laboratory shall simultaneously deliver raw data, including all necessary metadata, generated as part of this work plan as a Laboratory Analytical Data Package (LADP) to the trustee Data Management Team (DMT), the Louisiana Oil Spill Coordinator's Office (LOSCO) on behalf of the State of Louisiana and to BP (or ENTRIX on behalf of BP). The electronic data deliverable (EDD) spreadsheet with pre-validated analytical results, which is a component of the complete LADP, will also be delivered to the secure FTP drop box maintained by the trustees' Data Management Team (DMT). Any preliminary data distributed to the DMT shall also be distributed to LOSCO and to BP (or ENTRIX on behalf of BP). Thereafter, the DMT will validate and perform quality assurance/quality control (QA/QC) procedures on the LADP consistent with the authorized Analytical Quality Assurance Plan, after which time the validated/QA/QC data shall be made available simultaneously to all trustees and BP (or ENTRIX on behalf of BP). Any questions raised on the validated/QA/QC results shall be handled per the procedures in the Analytical Quality Assurance Plan and the issue and results shall be distributed to all parties. In the interest of maintaining one consistent data set for use by all parties, only the validated/QA/QC data set released by the DMT shall be considered the consensus data set. In order to ensure reliability of the consensus data and full review by the parties, no party shall publish consensus data until 7 days after such data has been made available to the parties. Also, the LADP shall not be released by the DMT, LOSCO, BP or ENTRIX prior to validation/QA/QC, absent a showing of critical operational need. Should any party show a critical operational need for data prior to validation/QA/QC, any released data will be clearly marked "preliminary/unvalidated" and will be made available equally to all trustees and to BP (or ENTRIX on behalf of BP).

Lead Investigators

Description of Duties for Lead Investigators

Name	Role
Mandy Tumlin Louisiana Department of Wildlife and Fisheries	Principal Investigator

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2011 BUDGET ESTIMATE

This budget assumes that only one stratum will be sampled at a time. Sampling at more than one location concurrently will increase costs; this budget does not include analytical costs.

DESCRIPTION	**AMOUNT**
INITIAL ONE-TIME EXPENDITURES:	
2 CHEST FREEZERS (-20°C)	$ 3,000
SAMPLING GEAR AND ACCESSORIES INCLUDING: GILL NETS, TRAWLS, WEIGHTS, FLOATS, ROPES, ETC.	$ 26,265
STAFF/SALARY	
6 LDWF STAFF MEMBERS AT A RATE OF 40 HOURS PER WEEK @ $22.99/HR (INCLUDES SAMPLING, PREP TIME, DATA ENTRY AND COMPILATION, SAMPLE STORAGE, CLEANING, ETC.)	$ 220,704
FRINGE BENEFITS (34%)	$ 75,040
VESSEL OPERATING EXPENDITURES	
FOR VESSELS UTILIZED DURING SAMPLING REGIME	$ 91,960
VEHICLE USEAGE	$ 7,500
TRAVEL	
LODGING (WHERE DEPARTMENT FACILITIES ARE NOT AVAILABLE)	$ 4,000
SAMPLING SUPPLIES	
MISCELLANEOUS SUPPLIES	$ 30,000
TOTAL FOR 10 MONTHS	**$458,469**

The Parties acknowledge that this budget is an *estimate* and that actual costs may prove to be higher. BP's commitment to fund the costs of this work includes any additional reasonable costs within the scope of this approved work plan that may arise. The trustees will make a good faith effort to notify BP in advance of any such increased costs.

Appendix 1:
Fish Collection SOP

Materials

1. Small cooler with ice (to hold samples until frozen)
2. Datasheets or notebooks
3. Scalpel handles (#3) and blades (#11)
4. Stainless steel forceps (small and large), scissors, hemostats
5. Stainless steel or Teflon cutting board
6. Pre-cleaned aluminum foil
7. Nitrile or other non-contaminating gloves
8. Isopropanol or 95% ethanol in a squirt bottle
9. Distilled water
10. Alconox soap for cleaning equipment and tools
11. Clear vial and spring inserts for low-volume composite samples (http://www.sun-sri.com; catalog numbers 200-774 and 200-778, respectively).
12. 20-mL scintillation vials for use with large fishes
13. 1-cc tuberculin syringes with needles
14. Marking pens with indelible ink for labeling tasks
15. Chain of Custody Forms and security tape
16. Clear packing tape
17. Sharps containers (for scalpel blade disposal) and garbage bags.

Sample Labeling and Packaging Guidelines

1. All parts of the animal must be retained.
2. A single fish or a sample of a group of fish of the same species will be assigned a single NRDA ID number.
3. Each component (carcass and/or tissue) will be labeled as "A" or "B" (e.g., LAAM24-A0502-T0102A, LAAM24-A0502-T0102B for the tissue and carcass).
4. Carcass and tissue samples will each be wrapped separately in pre-cleaned aluminum foil, placed in separate plastic bags with the NRDA ID number written on the bag and on a cloth tag contained within the bag.
5. Each sample (composite, tissue, etc.) should have its own bag with accompanying sample name and tag, as specified. Subsamples from a single fish can be bagged together for convenience, provided that all NRDA sample IDs and pertinent information are written on the bag and also on a cloth tag within the bag.
6. All samples should be iced as soon as practicable, and archived at -20°C for pending analyses.

Sample Processing Guidelines

1. Place fish into a covered storage container (polypropylene or stainless steel) temporarily, until each can be frozen or subsampled (samples will spend as little time as possible sitting out in the open air on the deck of the ship).

2. A subsample of muscle tissue should be taken from fish >600 mm in length (see 4-7 below)
3. Fish samples should be wrapped in pre-cleaned aluminum foil (provided by Newfields lab), placed in plastic freezer bag, labeled per NRDA sample collection protocol, and placed in freezer at -4°C or on ice for temporary storage until sample can be stored at -20°C pending analysis.
4. Clean table with Alconox solution and seawater
5. Put on a clean pair of nitrile gloves
6. Obtain a new disposable scalpel blade: muscle tissue sampler should change scalpel blades between subsamples of each fish.
7. Once fish is on the table, carefully clean the external area to be subsampled with diH_2O.
    a. After sacrificing each animal (by severing the spinal column just posterior to the fish's head), open the body cavity using scissors, forceps or new disposable scalpel blade. One set of tools will be used to cut open the animal and a separate set of tools will be used for cutting tissue sections inside the animal.
    b. Take approximately 30 g or more of muscle tissue from behind the head.
8. Wrap and label each sample independently in pre-cleaned aluminum foil immediately after sample is collected.
9. Place aluminum foil-wrapped tissue in plastic freezer bag; let as much air as possible out of bag and seal bag, label per NRDA sample collection protocol.
10. Place in freezer at -4°C or on ice for temporary storage.

<u>Recording protocols</u>

The following information should be recorded on the sampling sheets:

1. Collection date
2. Station number
3. Species and gender (if possible, per NMFS standard protocol)
4. Sequential specimen # (assigned by person collecting sample; start over at each station)
5. Any other information required by NRDA sample collection instructions
6. Length and weight of specimen

<u>Equipment cleaning procedures</u>

1. Rinse dissecting tools with isopropyl between each fish.
2. If tools are dirty with caked-on tissue, wash in Alconox solution, rinse with diH_2O, and then rinse with isopropyl alcohol.
3. Don't wash scalpel blades; just replace with a new scalpel blade.

Appendix 2:
Protected Species Mitigation Measures