SECTION 2
METHODS

2.1 MARINE RESOURCES STUDY SAMPLING STRATEGY

Tetra Tech conducted a sampling program to provide data to evaluate the incremental risks to human health from eating fish and shellfish collected from the muliwai downstream of MMR and in the nearshore waters and limu collected in nearshore waters. This study was designed to compare analytical results of fish and shellfish samples collected in the vicinity of MMR with analytical results for fish and shellfish samples collected from background locations. The data were collected in two phases. Fish and limu samples were collected between August 2 and August 24, 2006, while shellfish samples were collected between September 29 and October 10, 2008. Shellfish were not collected during the first phase due to concerns about the impact that collecting large numbers could have on their population. Limu samples were collected only from the MMR nearshore environment, so these samples were evaluated independently. Furthermore, the ecological risks to several taxonomic groups identified in the muliwai were evaluated. A complete description of the sampling program strategy and objectives is provided in the SAP.

2.2 SITE SELECTION AND CHEMICAL TRANSPORT PATHWAYS

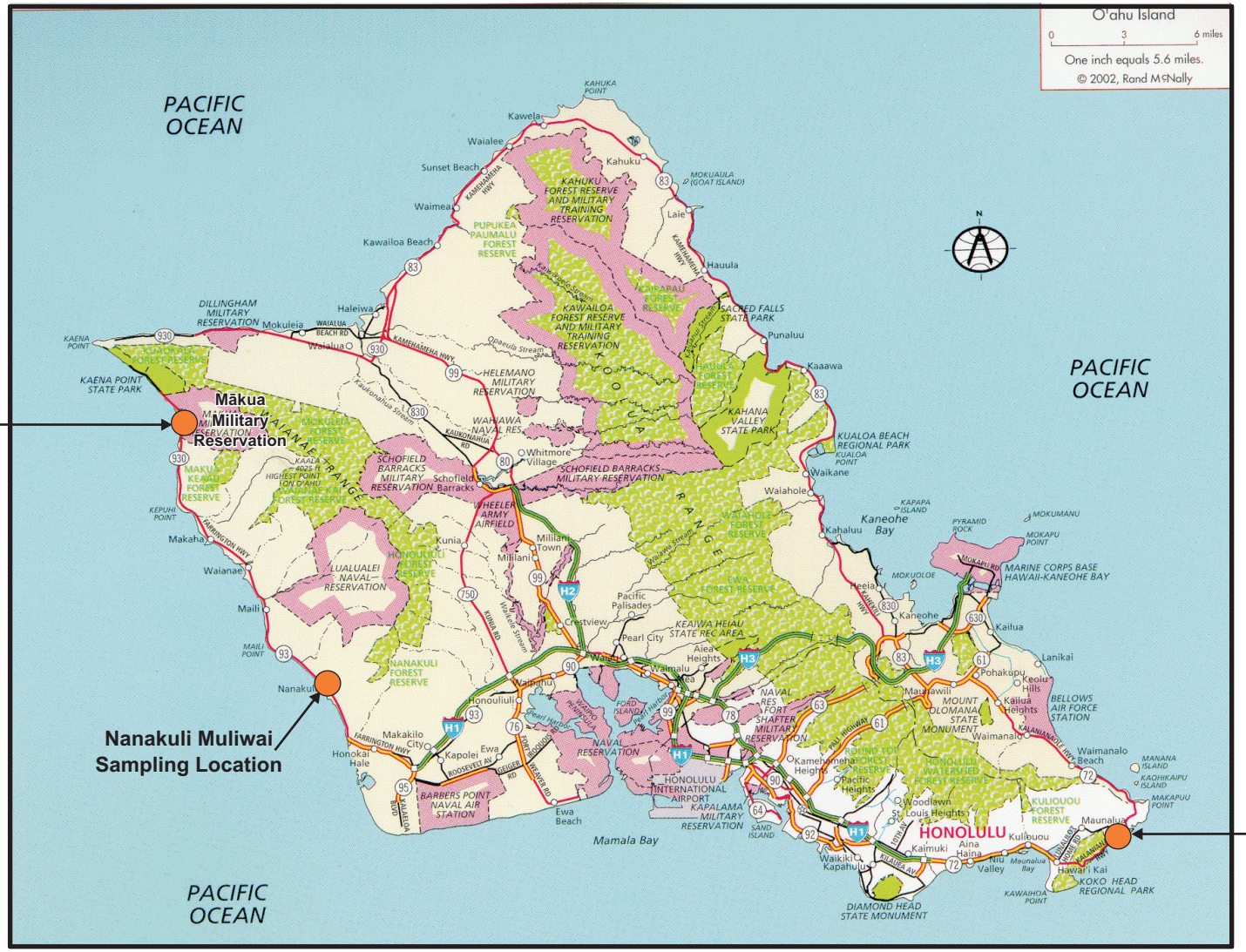
To meet the objectives identified in the SA, the preparers of this study addressed whether chemicals of potential concern have transported beyond the boundaries of MMR. In general, chemicals are transported by air, surface water, and groundwater. Chemical emissions generated from various activities, including military training, could enter the air and be transported downwind. They can be dissolved in water or adsorbed to particles that are transported by permanent or intermittent streams (streams that flow infrequently) or surface water runoff. Furthermore, water underlying the surface (groundwater) can transport chemicals that originate from the earth's crust or infiltrate from the earth's surface. Previous studies have evaluated the presence of chemicals of potential concern in air, soil, streambed sediment, surface water, groundwater, and muliwai sediment at Mākua in the vicinity of MMR. For this marine resources study, sites were selected that could link compounds originating at MMR to marine resources in the Mākua muliwai and nearshore waters. Background sites were selected to evaluate whether chemicals found in the vicinity of MMR were different from chemicals found in other parts of O'ahu.

The north and south muliwai, in the vicinity of the MMR, as shown on Figure 2-1, were selected for sampling. The north muliwai is the pond that results from runoff from Mākua Stream, the principal stream that runs through the center of MMR. The south muliwai is the pond that results from runoff from Kaiahi Gulch, which runs along the south side of MMR. Based on the topography and the drainage ditches along the MMR access road, runoff from most of the live-fire areas and the disposal areas at MMR drains to the south muliwai. Nearshore sampling at MMR was conducted adjacent to the north and south muliwai, as shown on Figure 2-1.

Background locations for the Marine Resources Study were selected after careful consideration of a wide variety of factors. One of the objectives of the Marine Resources Study was to identify if Army activities at MMR have potentially impacted Mākua Valley resources. Using an uncontaminated, pristine, or minimally developed watershed for a background location would hinder distinguishing between the Army and other sources on O'ahu. This is a significant concern as there are many potential sources of contamination to the muliwai and nearshore environments other than the MMR. To adequately address the Army's impact alone on Mākua Valley resources, an appropriate control site i.e., background location would be a valley where biotic and abiotic variables are as similar as possible to Mākua. Since inter-watershed transport of contaminants is facilitated by wind and rain, the control valley should have similar wind and rain patterns as that of Mākua. Biogeochemical processes affecting contaminants are a function of temperature and substrate, and these attributes also should be as similar as possible to Mākua Valley in order to identify impacts that can be potentially attributed to Army activity. There are distinct differences in the substrate (mineralogy and age) as well as in the human population and accompanying anthropogenic impacts among the Hawaiian Islands, making the selection of background locations on another island inappropriate.

Because of this, the most appropriate control watersheds are on the leeward (Waianae) coast of O'ahu. As long as the background sites selected are representative of ambient conditions for the general Mākua vicinity and have not received contamination from the MMR, they are considered acceptable, according to the USEPA (1989, 2002a) risk assessment guidance. It is for this reason that the SAP states "Background muliwai will be located on the Waianae Coast within watersheds that are not subjected to military activity" (Section 2.2). The SAP further states that "Samples will be collected from locations distant enough from Mākua Valley that biota would be unlikely to be affected by target chemicals (explosives, by-products of explosives, and metals) originating from MMR. Background muliwai will be located in watersheds that are not subject to military activity" (Section 2.2.3).

The background muliwai selected for this study was the Nanakuli muliwai, which is located on the Waianae Coast, south of the town of Nanakuli, approximately 15 miles south of MMR. One of the sources of freshwater to the Nanakuli muliwai is runoff from Nanakuli Stream; another likely source of freshwater to the muliwai is groundwater discharge. Other muliwai located on the Waianae Coast were evaluated as potential background sampling locations, but none of these other muliwai contained water during the summer, when the sampling program was implemented.



Source: Rand McNally 2002

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Fish, Shellfish, and Limu Sample Location Map

Mākua Military Reservation
O´ahu, Hawai´i

Tetra Tech, Inc.

Figure 2-1

The background nearshore area was located at Sandy Beach, on the southeast side of O'ahu, as shown on Figure 2-1. Sandy Beach is considered to be similar to the Mākua nearshore area because both support rocky areas and sandy beaches, with very low rainfall. There is much greater movement of water and fish in nearshore areas than in a muliwai, so there is much less need for the background nearshore area to be next to a watershed that is similar to Mākua than the need for the background muliwai to be located in a watershed that is similar to Mākua.

2.3 SPECIES OF INTEREST

One of the goals of the Marine Resources Study was to sample a representative range of species that may be consumed by subsistence and recreational fishermen on the Waianae Coast. Species of interest for this study were identified through discussions with regional commercial fishermen, local recreational fishermen, area divers and spear fishermen, and local residents from the Waianae coast. These discussions indicated that local fishermen are typically opportunists who consume most of the fish they are able to catch and are not selective of species. In addition, a preliminary shellfish survey was conducted to identify species that inhabit the muliwai and nearshore areas of Mākua in sufficient quantities to meet the laboratory analytical requirements. Although the SA stated that the Army should complete one or more studies to determine if fish, shellfish, limu, and other marine resources are contaminated, it is not possible to collect samples of all available marine resources. A substantial effort was made to select and collect marine resources that were representative of and readily available in the habitats of the Mākua muliwai and nearshore waters and similar watersheds where military training exercises have not occurred in the recent past. Since trophic level influences the potential uptake and concentration of contaminants, species from a range of trophic levels (primary producer, herbivore, omnivore and carnivore) were targeted in the study.

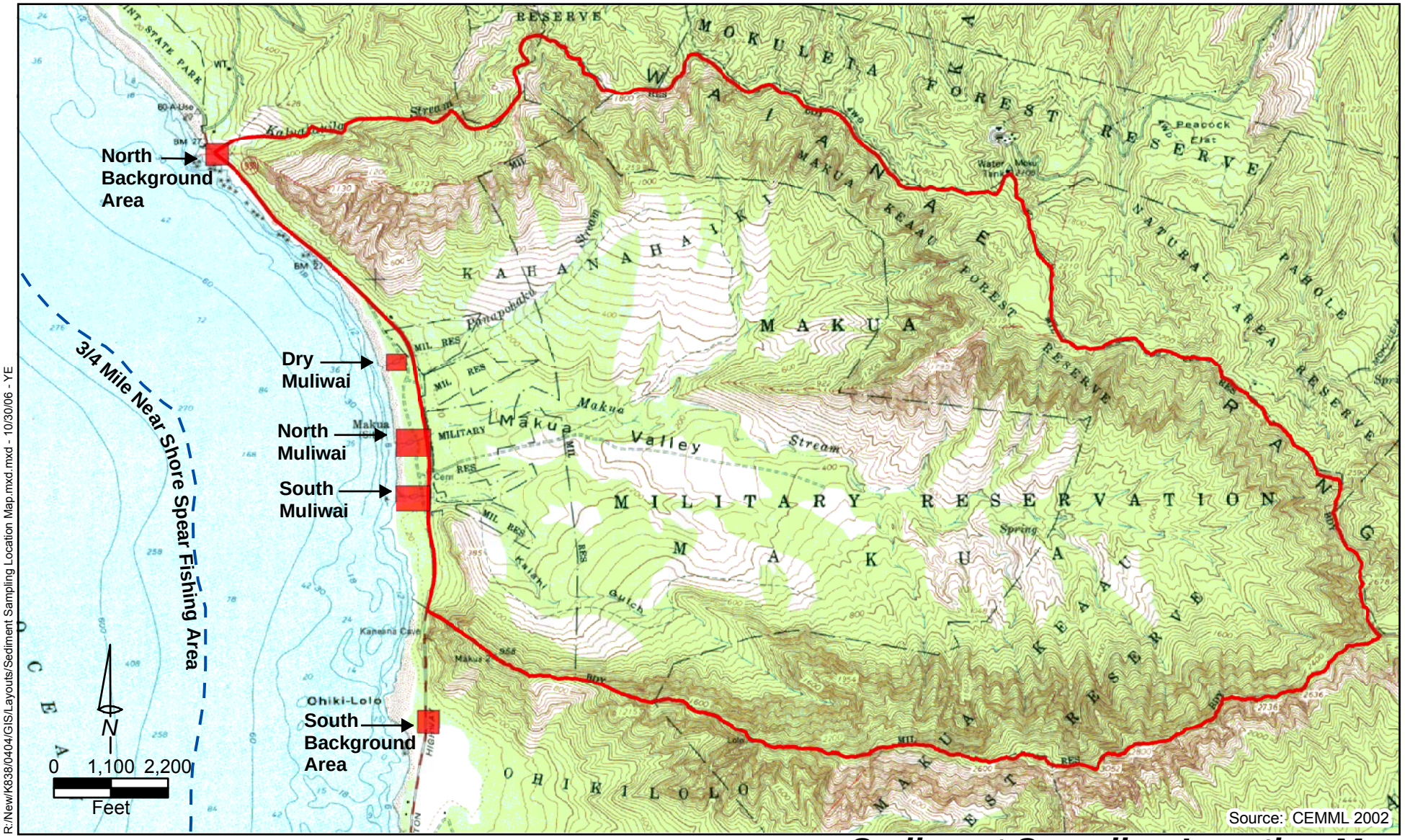
Target species were prioritized by the following criteria:

- Serve as a food source for humans;
- Spend part of their life cycle in or near brackish or freshwater (e.g., muliwai); and
- Represent a variety of trophic levels and feeding niches.

Table 2-1 identifies the target species that were collected in each habitat. Every effort was made to collect similar species in all sampling locations, but natural differences in species composition and abundance dictated which species could be collected.

2.4 FIELD SAMPLING METHODS

Field sampling of fish and limu for the marine resources study occurred between August 2 and August 24, 2006, while field sampling of shellfish occurred between September 29 and October 10, 2008. Field data sheets are provided in Appendix A. Multiple sampling methods were employed to accommodate the irregular shapes and rocky bottoms of the muliwai and the surf conditions in the nearshore waters. Hook and line sampling was used in the nearshore waters, while sampling methods used in the muliwai included seine nets, gill nets,



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Sediment Sampling Location Map

Legend

Mākua Military Reservation

Mākua Military Reservation
Oʻahu, Hawaiʻi

Table 2-1
Marine Resources Sampling Locations and Species of Interest at MMR

Common Name	***Scientific Name***
Muliwai Target Species	
Hawaiian flagtail (aholehole)	*Kuhlia sandvicensis*
Striped mullet (`ama`ama)	*Mugil cephalus*
Medaka	*Poeciliidae* sp.
Tilapia	*Talapia zillii*, *T. rendalii*, *Oreochromis macrochir*, *O. mossambicus*, *Sarotherdon melanotheron melanotheron*
Samoan crab	*Scylla serrata*
Hawaiian prawn	*Macrobrachium grandimanus*
Malaysian snail	*Thiaridae sp.*
Rock crab	*Pachygrapsus minutus*
Red rock crab	*Plagusia depressa tuberculata*
Nearshore Target Species	
Picasso triggerfish (humuhumu nukunuku a puaa)	*Rhinecanthus rectangulus*
Blackspot sergeant (kupipi)	*Abudefduf sordidus*
Christmas wrasse (hinalea)	*Thalassoma trilobatum*
Saddle wrasse (hinalea lau-wili)	*T. duperry*
Manybar goatfish (moano)	*Parupeneus multifasciatus*
(Limu wawae`iole)	*Codium edule*
(Limu manauea)	*Gracilaria coronopifolia*
Kona crab	*Ranina ranina*
Slipper lobster	*Parribacus antarcticus*
Helmet urchin	*Colobocentrotus atratus*
Oblong urchin	*Echinometra oblonga*
Thin-shelled rock crab	*Grapsus tenuicrustas*
Black purse shell	*Isognomon californicum*
Dotted periwinkle	*Littoraria pintado*
Black nerite	*Nerita picea*
Rock-boring urchin	*Echinometra mathaei*
False `opihi	*Siphonaria normalis*
Purple rock barnacle	*Nesochthamalus interextus*
Pleated rock crab	*Pachygrapsus plicatus*
Snakedhead cowry	*Cypraea caputserpentis*
Blue-back urchin	*Echinotrhix diadema*
Black-foot `opihi	*Cellana exarata*

hook and line, crab and minnow traps. Limu was handpicked from the nearshore area. Each of these field methods is described below. A variety of other methods used by local fishermen, including spear fishing, were not used in the study because of the potential to introduce metals and other types of contamination into the fish.

2.4.1 Seine Nets

The seine nets used in this study ranged from 10 to 30 feet long and three feet in depth, with 1/4 inch mesh. The top of the nets were lined with floats and the bottoms of the nets were lined with lead. Each end was tied to a 4 foot pole, which was pulled by two team members through the water, dragging the leaded net bottom across the muliwai bottom and floating the top across the water surface, in effect creating a netted wall which was used to corral fish up to the banks of the muliwai. While two team members pulled the net across the bottom, a third person followed behind to free the net from an assortment of rocks or debris. Seining continued until sufficient biomass was obtained for each sample and no new species were collected.

2.4.2 Gill Nets

Gill nets used in this study were 20 feet long and 5 feet in width, with ½ or ¾ inch mesh. The top length of the net was lined with floats and the bottom length of the net was lined with lead. Gill nets were deployed across the width of the muliwai. The gill nets were used independently and in conjunction with the seine nets. Gill nets were stretched across the muliwai to compartmentalize the muliwai and facilitate seining within a smaller area restricting fish from escaping into the inaccessible areas of the muliwai. Gill nets were deployed for periods ranging from 30 minutes to 1 hour; Tetra Tech personnel remained on site whenever gill nets were deployed. Gill net sampling continued until sufficient biomass was collected for each sample and no new species were recorded.

2.4.3 Hook and Line

The hook and line method was used in the muliwai and in the nearshore fishing areas. Appropriate weight fishing lines and hook size for each target species were used for hook and line sampling in the muliwai and nearshore waters. Bread, limu, aku belly, shrimp, squid, and in some cases live fish were used for bait. Sampling continued until sufficient biomass was collected for each sample and no new species were recorded.

2.4.4 Crab Nets

Crab nets with a two-foot diameter frame and containing either nylon or cotton two-inch stretched mesh, were baited with Kona Kampachi and deployed in both the nearshore and muliwai during daylight hours. Nets were checked every one to two hours.

2.4.5 Lobster Traps

Lobster traps, three feet by two and a half feet by two feet, with rigid two-inch by two-inch rigid mesh and two eight-inch by four-inch funnel openings, were baited with Kona Kampachi, deployed in the early evening, and retrieved the next morning.

2.4.6 Crab and Minnow Traps

Several minnow trap designs with ¼ inch mesh were employed. These traps were baited with aku belly, chicken parts, squid, shrimp, and canned tuna. Traps were placed in a variety of habitats throughout the day. At the end of the day, traps were redeployed and remained in place throughout the night and collected the next morning.

2.4.7 Hand-Picked Limu

Limu was hand picked using clippers and was cut at the stipe above the holdfast and placed in a netted bag or a bucket of water during collection. Before being weighed and identified to species, the limu samples were checked for any accidental removal of holdfasts. Any holdfast that may have been removed was returned to the reef. Scientists at the Bishop Museum identified the limu species.

2.5 SAMPLE HANDLING

Samples were handled in accordance with procedures outlined in the SAP. Once caught, the fish were placed in buckets and brought to the sample station where they were identified, measured, and labeled. Fish and limu samples were wrapped in foil and a plastic bag and placed on ice until delivery to the laboratory. Information describing the individual fish was recorded on field data sheets, including the time, date and approximate location of collection, length and weight of the fish, and method of collection. Samples were shipped to the analytical laboratories on dry ice and under chain-of-custody. Shellfish samples were placed in one-liter glass jars and stored on wet ice until being shipped on wet ice to the laboratory for sample preparation and analysis. The time, date, and approximate location of collection, full length, carapace length, and width and mass of the shellfish and method of collection were recorded on field data sheets.

2.6 LABORATORY ANALYSIS

Chemical parameters and analytes for sample analysis were identified in the SAP and are presented in Table 2-3. Primary fish and limu samples collected in 2006 were submitted to two laboratories, Columbia Analytical Services and Agricultural and Priority Pollutants, Inc. (APPL). Columbia Analytical Services analyzed the samples for the analytes listed in Table 2-2, and APPL analyzed the samples for explosives. APPL, Battelle Marine Sciences Laboratory, and Severn Trent Laboratories analyzed the quality control (QC) samples. Shellfish samples collected in 2008 were submitted to APPL, where they were composited, and QC samples were forwarded to Test America, Inc. The laboratory sample analysis scheme is presented in Table 2-3.

2.7 NATURAL AND ANTHROPOGENIC SOURCES OF CHEMICAL ANALYTICAL GROUPS

The chemicals of particular concern for the Marine Resources Study were explosives compounds (RDX, nitroglycerin, 2,4-dinitrotoluene, and perchlorate) and several metals. These chemicals are associated with past and proposed training at MMR. The following additional analytical groups were included in this study after public comments were received on the SAP:

Table 2-2
Summary of Fish, Shellfish, and Limu Samples, Makua Military Reservation Marine Resources Study

Site	Sample ID	Matrix	Species	Sample Type
Makua North Muliwai	1	Fish	Striped mullet	Primary
Makua North Muliwai	3	Fish	Hawaiian flagtail	Primary
Makua North Muliwai	4	Fish	Tilapia	Primary
Makua North Muliwai	1b	Fish	Tilapia	Primary
Makua North Muliwai	5	Fish	Tilapia	Primary
Makua North Muliwai	MNM-04	Shellfish	Samoan crab	Primary
Makua South Muliwai	6	Fish	Striped mullet	Primary
Makua South Muliwai	2fd	Fish	Striped mullet	QC
Makua South Muliwai	7	Fish	Striped mullet	Primary
Makua South Muliwai	Comp 8,8a	Fish	Medaka	Primary
Makua South Muliwai	9	Fish	Tilapia	Primary
Makua South Muliwai	Comp 9fd, 10a	Fish	Tilapia	QC
Makua South Muliwai	10	Fish	Tilapia	Primary
Makua South Muliwai	MSM-01	Shellfish	Rock crab	Primary
Makua South Muliwai	MSM-02	Shellfish	Hawaiian prawn	Primary
Nanakuli Muliwai	12	Fish	Tilapia	Primary
Nanakuli Muliwai	13	Fish	Tilapia	Primary
Nanakuli Muliwai	14	Fish	Tilapia	Primary
Nanakuli Muliwai	NM-01	Shellfish	Hawaiian prawn	Primary
Nanakuli Muliwai	NM-02	Shellfish	Rock crab	Primary
Nanakuli Muliwai	NM-01	Shellfish	Hawaiian prawn	QC
Nearshore waters at Makua	NW2	Fish	Picasso triggerfish	Primary
Nearshore waters at Makua	NW3	Fish	Blackspot sergeant	Primary
Nearshore waters at Makua	NW4	Fish	Manybar goatfish	Primary
Nearshore waters at Makua	NW1fd	Fish	Manybar goatfish	QC
Nearshore waters at Makua	NW5	Fish	Christmas wrasse	Primary
Nearshore waters at Makua	MNS-03	Shellfish	Helmet urchin	Primary
Nearshore waters at Makua	NMS-03	Shellfish	Helmet urchin	QC
Nearshore waters at Makua	MNS-05	Shellfish	Kona crab	Primary
Nearshore waters at Makua	NW1SW3-1	Limu	All four samples are composites of *Acanthophora spicifera, Sargassum muticum*, and *Sargassum polyphyllum*	Primary
Nearshore waters at Makua	NW1SW1-1	Limu		Primary
Nearshore waters at Makua	NW1SW2-2	Limu		Primary
Nearshore waters at Makua	NW1SW1-1fd	Limu		QC
Nearshore waters at Sandy Beach	NW2fd	Fish	Blackspot sergeant	QC
Nearshore waters at Sandy Beach	NW9	Fish	Picasso triggerfish	Primary
Nearshore waters at Sandy Beach	NW10	Fish	Manybar goatfish	Primary
Nearshore waters at Sandy Beach	SBNS-01A	Shellfish	Helmet urchin	Primary
Nearshore waters at Sandy Beach	SBNS-01B	Shellfish	Helmet urchin	Primary
Nearshore waters at Sandy Beach	SBNS-01A	Shellfish	Helmet urchin	QC

Table 2-3
Sample Analytes and Analytical Methods

Analyte	Analytical Procedure	Primary Sample Analysis	QC Sample Analysis
Dioxins/Furans (17 congeners of concern)		**Columbia[1] APPL[2]**	**STL[1] TestAmerica[2]**
HpCDD	USEPA Method 8290		
HpCDF	USEPA Method 8290		
HxCDF	USEPA Method 8290		
OCDD	USEPA Method 8290		
OCDF	USEPA Method 8290		
TCDD	USEPA Method 8290		
Gasoline (Purgeable Organics)		**Columbia[1] APPL[2]**	**APPL[1] TestAmerica[2]**
Ethylbenzene	USEPA Method 8260B		
m-Xylene	USEPA Method 8260B		
p-Xylene	USEPA Method 8260B		
o-Xylene	USEPA Method 8260B		
Toluene	USEPA Method 8260B		
Metals		**Columbia[1] APPL[2]**	**Battelle[1] TestAmerica[2]**
Aluminum	USEPA Method 200.8		
Antimony	USEPA Method 200.8		
Arsenic	USEPA Method 200.8		
Barium	USEPA Method 200.8		
Beryllium	USEPA Method 200.8		
Cadmium	USEPA Method 200.8		
Chromium	USEPA Method 6010B		
Cobalt	USEPA Method 200.8		
Copper	USEPA Method 200.8		
Iron	USEPA Method 6010B		
Lead	USEPA Method 200.8		
Manganese	USEPA Method 200.8		
Mercury	USEPA 7471A (USEPA Method 245.6)		
Methyl Mercury	USEPA Method 1630 modified		
Selenium	USEPA Method 7740		
Silver	USEPA Method 200.8		
Thallium	USEPA Method 200.8		
Vanadium	USEPA Method 6010B		
Zinc	USEPA Method 200.8		
Explosives (Nitroaromatics/Nitramines)		**APPL[1, 2]**	**STL[1] TestAmerica[2]**
2,4-DNT	USEPA Method 8330		
RDX (Cyclonite)	USEPA Method 8330		
Nitroglycerine	USEPA Method 8330 modified		
Perchlorate	USEPA Method 314		

Table 2-3
Sample Analytes and Analytical Methods

Analyte	Analytical Procedure	Primary Sample Analysis	QC Sample Analysis
Organochlorine Pesticides		**Columbia[1] APPL[2]**	**APPL[1] TestAmerica[2]**
4,4'-DDT	USEPA 8081A		
Aldrin	USEPA 8081A		
alpha BHC	USEPA 8081A		
beta BHC	USEPA 8081A		
delta BHC	USEPA 8081A		
gamma BHC (lindane)	USEPA 8081A		
Heptachlor	USEPA 8081A		
Heptachlor epoxide	USEPA 8081A		
VOCs/SVOCs		**Columbia[1] APPL[2]**	**APPL[1] TestAmerica[2]**
Styrene	USEPA Method 8260B		
1,2,4-Trimethylbenzene	USEPA Method 8260B		
Pyrene	USEPA Method 8270C		
Phthalate Esters			
Bis(2-ethylhexyl) phthalate	USEPA Method 8270C		
Di-n-butyl phthalate	USEPA Method 8270C		
Diethyl phthalate	USEPA Method 8270C		
Dimethyl phthalate	USEPA Method 8270C		
Di-n-octyl phthalate	USEPA Method 8270C		

1 - Analyzed fish and limu samples
2 – Analyzed shellfish samples

- Dioxins/furans;
- Organochlorine pesticides;
- VOCs;
- SVOCs; and
- Additional metals.

The chemicals in these analytical groups have a variety of potential sources and, if detected in the fish, shellfish, and limu samples, would be difficult to attribute to activities at MMR. An extensive literature review was conducted to identify potential natural and man-made sources of each chemical or chemical analytical group. Furthermore, efforts were directed toward identifying anthropogenic sources that were unique to the military because these chemicals could indicate a chemical migration pathway.

2.8 HUMAN HEALTH AND ECOLOGICAL RISK ASSESSMENTS

Methods used to conduct the human health and ecological risk assessments are described in Sections 4 and 5, respectively.