Exxon Valdez Oil Spill
Restoration Project Final Report

Towards Sustainable Management in the Kenai River Watershed: Linking Human and Resource Development with Nutrient and Energy Pathways Across Terrestrial, Aquatic and Marine Systems

Restoration Project 030684

Final Report

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Towards Sustainable Management in the Kenai River Watershed: Linking Human and Resource Development with Nutrient and Energy Pathways Across Terrestrial, Aquatic and Marine Systems

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Study History: A group of individuals representing agencies and organizations with interest in the Kenai River watershed have met over the past five years to discuss and identify issues related to marine and terrestrial derived nutrients in the watershed. Three workshops and a number of smaller meetings were convened to bring together those interested in collaboration in a larger research initiative for the Kenai River watershed (Kenai RW). Two technical bulletins, a CD ROM, a Study plan (EVOS Final Report 02612) and this final report have been created to foster an understanding of watershed issues and stakeholder interest and input on present and future research in the Kenai RW. The report herein on Restoration Project 030684 presents a first phase proposal for submission to EVOS for funding an integrated and interdisciplinary field research project to examine nutrient and energy pathways and terrestrial-aquatic linkages in the Kenai RW.

Abstract: Kenai River Watershed (Kenai RW) is recognized as a national treasure for its abundant fish, wildlife and diversity of habitats. Extensive consultation among stakeholders, communities and agencies has led to the development a detailed research proposal on the role of marine-derived nutrients (MDN) in sustaining the productivity of Kenai RW. The proposed research plan indicates that in the first two years we develop robust methods and monitoring protocols to detect, understand and predict changes in MDN and its linkage to productivity and biological resources. During this period we intend to test the precision and validity of several distinct indicators or proxies (nutrients, stable isotopes, fatty acids, contaminants, foodwebs) of MDN across different ecosystem components of Kenai RW. In a 3rd research year, we will synthesize data, compare results with other complementary projects and produce a final GEM report. During the final year of the Kenai RW research study, we will begin to develop protocols to test the validity of these indicators to quantify the fate/transport of MDN linking various components of the watershed and their implications for the productivity of Kenai RW. The group of researchers we have assembled through this collaborator process will continue to participate in networking and communication among various research groups looking at watershed level changes in MDN and resource productivity.

Key Words: anadromous fish, energy, estuaries, Gulf Ecosystem Monitoring, habitat linkages, Kenai River, lakes, marine-derived nutrients, rivers, salmon, streams, watersheds, wetlands.

Project Data: *Description and format of data* – Data used in this study are stored electronically at University of Victoria (UVIC), and Alaska Department of Fish and Game (ADF&G), Commercial Fisheries Division, Soldotna. Data are formatted as follows: maps (JPEG – From ArcView 3.2), physical data and zooplankton information (Microsoft Excel), water chemistry, nutrient and chlorophyll concentrations (Microsoft Excel), and fisheries (adult spawning timing, adult abundance) data (Microsoft Excel). *Custodian* - Custodian of the data used in this project is: J.A. Edmundson, ADF&G, Division of Commercial Fisheries, 333 Raspberry Road, Anchorage, AK 99518-1599. TEL: 907/267-2123, EMAIL: jim_edmundson@fishgame.state.ak.us. A. Mazumder, Environmental Management of Water and Watersheds, Department of Biology, University of Victoria, Victoria, V8W 3N5BC, Canada. TEL: 250/472-4789, FAX: 250/472-4766, EMAIL: mazumder@uvic.ca. Website: www.uvic.ca/water. *Availability* – UVIC and ADF&G hold proprietary research rights to the data. Requested data will be made available under discretion of the data custodians.

Citation: Johannes, M.R.S., J.A. Edmundson and A. Mazumder. 2004. Towards sustainable management in the Kenai River watershed: linking human and resource development with nutrient and energy pathways across terrestrial, aquatic and marine systems *Exxon Valdez* Oil Spill Restoration Project Final Report (Restoration Project 030684), University of Victoria, Victoria, BC; Alaska Department of Fish and Game, Commercial Fisheries Division, Soldotna, Anchorage, Alaska.

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EXECUTIVE SUMMARY

The Kenai River Watershed (Kenai RW) is a unique site within Alaska and the Pacific Northwest. It is a productive, diverse system supporting a wide variety of anadromous and non-anadromous fish species, marine, freshwater, sport, commercial, subsistence and personal use fisheries, wildlife, and forest resources contained within a large scale terrain setting comprised of lakes, streams, wetlands, mountains and glaciers connected to the Cook Inlet and the greater Gulf of Alaska ecosystem. The resources of the Kenai RW have high economic and ecological value to the culture and socio-economic function of communities on the Kenai Peninsula and Alaska.

Over the past five years, a team of researchers and managers have developed a broad stakeholder consultation process which resulted in an actionable research plan including an extensive literature review on nutrients and productivity in the Kenai RW, coastal watersheds and salmon ecosystems and a meta-analysis showing information and knowledge gaps in the Kenai RW. The research plan will implement a long-term integrated program to examine the nutrient cycling and energy pathways that link freshwater habitats in the Kenai RW, their surrounding drainage basins including riparian areas, wetlands and terrestrial environments and downstream nearshore marine ecosystems. The conclusion drawn from an ongoing dialogue is that there is a critical need to sponsor research to develop tools, techniques and models for restoration and management of the Kenai RW. The science and integration developed from future project research projects can potentially be applied to other Alaskan, and international watersheds exposed to similar human and resource use and development towards sustainable management.

The Kenai RW research initiative has now evolved with a mission to integrate interdisciplinary knowledge on nutrients and energy and their sources and pathways within the watershed, and link the cycle of nutrients / energy with watershed and resource productivity that cuts and integrates across habitat types (terrestrial, freshwater of lakes and streams and marine systems) and watersheds within the larger Gulf of Alaska ecosystem. The Kenai RW comprises unique and diverse landscapes and networks of lakes, rivers, streams and wetlands with strikingly different geomorphology. Each sub-basin, wetland and tributary contributes different levels and compositions of nutrients to the Kenai River and the downstream nearshore marine ecosystems, where each component benefits differently from the input of marine-derived or nutrient sources. These observations are the basis of a research initiative to explore and examine the diversity of sources, sinks and pathways of nutrients and energy, how nutrients cycle and the sensitivity of the Kenai RW to changes in nutrient inputs in the context of regional and global climatic scenarios coupled with anthropogenic impacts. The first stage of the research plan is to develop robust methods and monitoring protocols to detect, understand and predict changes in marine derived nutrients (MDN) and its linkage to productivity and biological resources. During this period our research will test the robustness and validity of several distinct indicators or proxies (nutrients, stable isotopes, fatty acids, contaminants, foodwebs) of MDN across different ecosystem components of Kenai RW. In a 3rd research year, we will synthesize data, compare results with other complementary projects and produce a final GEM report. During the final year of the Kenai research study, we will begin to develop protocols to test the validity of these indicators to quantify the fate/transport of MDN linking various components of the watershed and their implications for the productivity of Kenai RW. The group of researchers we have assembled through this collaborator process will continue to participate in networking and communication among various research groups looking at watershed level changes in MDN and resource productivity.

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GEM PROPOSAL SUMMARY PAGE

Project Title: Marine-Derived Nutrients in the Kenai River Watershed: Methods for Detecting Change.

Project Period: October 1st 2003 to September 30th 2006 (FY 03- FY 06).

Proposer(s): Asit Mazumder, University of Victoria, P.P Box 3020, Stn. CSC, Victoria, BC, V8W 3N5, 250-472-4789, mazumder@uvic.ca; Jim A. Edmundson, ADF&G Commercial Fisheries Division, 43961 Kalifornsky Beach Rd., Soldotna, AK, 99669, 907-260-2917, jim_edmundson@fishgame.state.ak.us ; Bob Clark, ADF&G, Sport Fish Division, 333 Raspberry Road, Anchorage, AK, 907-267-2222, bob_clark@fishgame.state.ak.us; Mark Willette, ADF&G Commercial Fisheries Division, 43961 Kalifornsky Beach Rd., Soldotna, AK 99669, 907-260-2961, mark_willette@fishgame.state.ak.us

Study Location: Kenai River watershed

Abstract: Kenai River Watershed (Kenai RW) is recognized as a national treasure for its abundant fish, wildlife and diversity of habitats. Extensive consultation among stakeholders, communities and agencies has led to this research proposal on the role of marine-derived nutrients (MDN) in sustaining the productivity of Kenai RW. In the first two years, we propose to develop robust methods and monitoring protocols to detect, understand and predict changes in MDN and its linkage to productivity and biological resources. We will test the robustness and validity of **several <u>distinct</u>** indicators or proxies (nutrients, stable isotopes, fatty acids, contaminants, foodwebs) of MDN across different ecosystem components of Kenai RW. In the 3rd year, we will synthesize data, compare results with other complementary projects and produce a final GEM report, and begin to develop protocols to test the validity of these indicators to quantify the fate/transport of MDN linking various components of the watershed and their implications for the productivity of Kenai RW. We will also participate in networking and communication among various research groups looking at watershed level changes in MDN and resource productivity.

Funding:	EVOS Funding Requested:	FY 04	$ 164,617.50	
		FY 05	$ 154,277.50	
		FY 06	$ 151,955.00	TOTAL: $470,850
	Non-EVOS Funds to be Used:	FY 04	~$ 20,000	
		FY 05	~$ 20,000	
		FY 06	~$ 20,000	TOTAL: ~$60,000

Date: June 11, 2003 Date proposal prepared – REVISED Oct 28,03

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I. NEED FOR THE PROJECT

A. Statement of Problem

The Kenai River Watershed (Kenai RW) is recognized by many to be a precious resource for its abundant fish and wildlife and diversity of landscapes unique to Alaska and the Pacific Northwest. The Kenai RW is at a cross roads; now experiencing the cumulative stress of human activities such as resource use, urban and industrial development and the apparent influence of ocean-climate changes from the Gulf of Alaska (GOA) (c.f Beamish and Boullion 1993; Hare and Francis 1995; Mantua et al. 1997). To effectively sustain productive resources and the biological diversity of the Kenai RW, there is a critical need to understand the linkages among the major biological, biogeochemical and hydrological processes in the watershed in the context of marine climate and nutrient influences from the GOA ecosystem and increasing anthropogenic pressures. Perhaps one of the most significant features of the Kenai RW is the abundance of anadromous fish stocks, in particular Pacific salmon (*Oncorhynchus sp*.). The role of anadromous fish in transferring nutrients from marine to freshwater environments has received considerable attention in recent years (Willson et al. 1998; Bilby et al. 2001; Gende et al. 2002). We recognize that anadromous fish transport large quantities of marine-derived nutrients (MDN) into freshwater through migration, spawning and subsequent carcass deposition to their natal spawning streams and lakes. While decomposing salmon carcasses are an important seasonal food resource for an array of invertebrate and vertebrate wildlife, it has also been suggested that releases of MDN are vital to the productivity, diversity and overall ecosystem health of Pacific Northwest watersheds (Cederholm et al. 1999; Naiman et al. 2002). The influence of salmon carcasses on the availability of MDN has been demonstrated in multiple small order streams (i.e. Bibly et al. 1996, Wipfli et al. 1998, 1999, Wold and Hershey 1999). For example, enhanced salmon carcass densities lead to increased lower trophic production (i.e. biofilm, macro-invertebrates) and juvenile salmonid growth (i.e. coho, rainbow trout). This evidence suggests that coho salmon and rainbow trout growth increments level off or decline at carcass densities above 1 to 2 pink salmon per square meter (Bilby et al. 2001, Wipfli et al. 2003). However, robust methods and tools have not been used to detect changes in MDN inputs under variable climate conditions, variable salmon run sizes and altered freshwater habitats. Furthermore, the role and dynamics of MDN in sustaining the productivity in whole watersheds is yet to be fully demonstrated and characterized at the level of an entire watershed and its component ecosystems.

We know that millions of salmon and other anadromous fish enter the Kenai RW and other watersheds in the region (i.e. Johannes et al. 2002), however we have a poor understanding of the ecological consequences of this widespread marine input. Similarly, the effects of increasing human activities within the watershed and impacts on fish habitats, while noticeable, have not always been chronicled or evaluated. In addition, short-term climatic events (e.g., El Nino, La Nina), Pacific decadal oscillations in oceanographic conditions, and long-term ocean-climate changes pose new questions about how and to what extent GOA ecosystem processes influence coastal watersheds such as the Kenai RW. The converse of this is that we cannot ignore the impact of MDN output from larger watersheds on the structure, function and productivity of the nearshore coastal environments, Alaska Coastal Current (ACC) and GOA. Yet, we have little direct information on the nature and function of these important physical, chemical and biological processes and their related links. Following two years of consultation with local, state and federal agencies, stakeholders, community groups and academic institutions, we developed an interdisciplinary research plan to examine the role of MDN through anadromous fish, climate, their cycling and pathways within the Kenai RW and its ecosystem components of lakes, streams, wetlands, terrestrial and nearshore environments (Johannes et al. 2002). To move forward with this plan, which is a priority mandate of GEM under the watershed theme, we first need to develop, explore and validate methods, techniques and models to detect changes in the occurrence and magnitude of MDN inputs to the Kenai RW and determine the effects of the inputs on the productivity of the watershed ecosystem. Detecting, understanding and modeling the effects of MDN on ecosystem productivity at the level of a whole watershed is a challenging task and cannot be achieved by examining single ecosystem components of the watershed. The following are some of the critical knowledge gaps associated with detecting, understanding and modeling marine, freshwater and terrestrial linkages through MDN including:

- Use, development and validation of single or combinations of indicators to detect the occurrence and magnitude of changes in MDN;
- The validation of the use $\delta^{15}N$ signals in freshwater as indicators of variation in MDN, nutrient loading and anadromous fish run strength;
- The influence of large and often cyclical anadromous fish runs (i.e. pink salmon, eulachon) on MDN input into watersheds;
- Ability to detect and differentiate the relative contributions of nutrients derived from MDN and climate inputs.
- Understanding the fate and transformation of MDN among different components within a watershed and their relevance to ecosystem level productivity;

To address these knowledge gaps, we developed an interdisciplinary team of researchers and built strong partnerships for cost sharing and communication.

B. Relevance to GEM Program Goals and Scientific Priorities

The goals for development of the Kenai RW research initiative and the research and monitoring proposed here, are consistent with the priority research needs and schedule of the Gulf Ecosystem Monitoring (GEM) program Science Plan (May 1, 2003). The mission of the GEM program is "to sustain a healthy and biologically diverse marine ecosystem in the northern Gulf of Alaska (GOA) and the human use of the marine resources in that ecosystem through greater understanding of how its productivity is influenced by natural changes and human activities". The broad goal of our proposed project is to identify, detect, monitor and better understand the dynamics of MDN in the Kenai RW ecosystem to provide information about how its productivity is influenced by natural changes in MDN under altered and variable climate and human activities. Consistent with the GEM perspective of research needs, we plan to:

- Identify the most sensitive and suitable indicators of marine related sources of nutrients;

- Differentiate isotopic forms in the different components of the foodweb and their utility to detect MDN and nutrient loading;
- Identify and calibrate other potential proxies or indicators of MDN (i.e. fatty acids, ammonium, persistent organic pollutants);
- Quantify the distribution and dynamics of MDN within and among ecosystem components of a entire watershed and its nearshore and estuary habitats;
- Differentiate the sources of nutrients derived from atmospheric, anadromous fish and anthropogenic inputs;
- Help create a network of researchers focusing on MDN in watersheds to communicate methods, results and establish collaborations.

II. PROJECT DESIGN

A. Objectives

Our approach for the Kenai RW research plan for FY 04-06 is to develop and validate a suite of robust methods for monitoring annual changes in MDN input into watersheds. During the first 2 years (FY 04-05), we will identify the best proxy indicators of MDN using established methods such as water nutrient concentrations of phosphorus (P), nitrogen (N) and carbon (C); stable isotopes of N, C; and sulfur (S) and marine-derived fatty acids, ammonium levels and persistent contaminants (i.e. persistent organic pollutants). During the 3rd year (FY06), we will determine a more rigorous proposal to determine MDN fate and transport within the aquatic and terrestrial foodwebs and their role in regulating resource productivity and diversity in the context of changing climatic and anthropogenic scenarios. Our research has been designed to be cost effective by complimenting monitoring and assessment activities for fish and water quality conducted by agencies, NGO's and community based citizen groups. While we propose to address specific short term objectives and goals within a 3-year plan, we recognize that a much longer-term research and monitoring program will be needed to understand effects of MDN on resource productivity in an entire watershed, like the Kenai RW, and to detect decadal climate changes and variation. Another objective of our work includes assistance with development of a watershed network and collaboration among other existing watershed-based projects and researchers in the Cook Inlet and south-central Alaska regions.

Our research plan is driven by the following specific testable hypotheses that:

A. inputs of MDN to watersheds can be detected in food webs at selected trophic levels as an indicator signature (isotope, chemical, biochemical) rather than increased trophic level biomass or productivity;
B. the occurrence and magnitude of MDN input, as a function of anadromous fish run strength, will be dependent on the habitat type of the sub basin (water type and hydrology) (i.e. clear, glacial, stained) and ecosystem types (i.e. stream, lake, estuary);
C. inputs of MDN to watersheds are proportional to the run strength (biomass) of anadromous fish entering these watersheds or sub basins and independent of climatic and anthropogenic inputs;
D. MDN uptake in aquatic foodwebs is primarily through direct consumption of fish carcasses and eggs rather than through bottom-up decomposition and microbial uptake; and,

E. the isotopic signatures of $\delta^{15}N$ at any trophic level is a consistent indicator of MDN input.

The specific objectives of this proposal are:

1. FY04-05 – collect field samples and develop methods and proxies for detecting MDN, and differentiating isotopic, chemical and biochemical forms of MDN and validate these indicators associated with different salmon run strength by sub basin and water type (Table 1);
2. FY 04-05 - compile data, synthesize results and develop predictive models to quantify total MDN input and output within different aquatic ecosystem components of the Kenai RW;
3. FY04-06 collaborate with local stakeholders within the Kenai River Watershed and among other watershed studies and researchers.

B. Procedural and Scientific Methods

Our methods involve field sampling across stream, lake and estuary watershed components at sites within 7 sub basins of the Kenai RW (Table 1). Sample collections will be made in the following 3 target habitats (ecosystem components): (a) stream/rivers, (b) lakes, and (c) lower Kenai River estuary. Sampling locations and sites selection will be consistent with existing monitoring programs and established to maximize the use of infrastructure and monitoring stations and historic data collected by citizen groups (Kenai Watershed Forum, Cook Inlet Keepers) and federal (USGS, USFS and USFWS) and state (ADF&G) agencies. The locations of the specific watersheds and study sites have been chosen to represent the different typology and sources of running and standing waters (i.e., clear, stained, and glacial), gradient of tidal (salinity) influence upriver, freshwater and major upland and lowland riparian and vegetation zones. This design is intended to test whether a given indicator of MDN is equally robust under variable target habitats and associated physical and chemical characteristics. We will sample a total of 10 river/streams and 6 lakes locations in the Kenai RW. The first full field sampling season will begin in May 2004 and the final year of field sampling will be 2006. Field sampling will be conducted during the period before salmon begin returning to spawn (early May), continue though peak spawning activity (July-August) and carry on into late fall (November) when it is assumed that nutrients released from the decomposition of spawned-out salmon is nearly complete (Table 2). This sampling schedule is also meant to capture the seasonal hydrograph for the rivers and streams in the different watersheds. Following each survey, all water and biological samples will be returned to the ADF&G Central Region Limnology laboratory in Soldotna, Cook Inlet Keepers new laboratory facilities and University of Victoria, where they will be preserved and readied for processing. Detailed field and laboratory methods are outlined below.

Field Methods

Streams - Rivers

Each stream and river site (Table 1) will be sampled twice per month for nutrients beginning in May and continuing through December; weather and ice conditions permitting. Approximately one liter of unfiltered water will be collected by submerging a pre-cleaned polybottle upstream from where anyone has crossed or waded. Samples will be stored in a cooler and transported to the laboratory for processing. During 3 specific spring, summer and fall sample periods, periphyton, benthic macroinvertebrates (grazing insect and midges: c.f. Table 2) will be collected by standard methods (surber samplers, D-frame nets). Organisms will be rinsed into sample jars and frozen at -80°C until processed for stable isotope composition and fatty acids (potentially other indicators). In addition, seines, small mesh nets and minnow traps will be used to collect samples of resident fish (sculpins and rainbow trout) and juvenile salmon (coho, chinook). Fish samples will be frozen at -80°C for analysis.

Table 1: Sampling matrix for sites within the Kenai RW for proposed FY04-06, including sampling at specific freshwater habitats including streams/rivers and lakes. For example, sample site selection is based on water type, hydrograph, water clarity and relative salmon run density. Presence of anadromous eulachon / smelt and pink salmon in watersheds and specific sites within the Kenai RW was provided as a potential variable for examining marine-derived nutrient inputs into watersheds.

Replicate Watersheds / Sub basins	Watershed Component Sampling	(1) Water Type / Clarity	(2) Recent Average Salmon Escapement (#'s)	(3) Salmon Density (#/m^3/sec)	Index of Relative Salmon Density	Eulachon / Smelt Presence	Pink Salmon Presence	(4) Anadromous Salmon Species Presence
Kenai Mainstem - Kenai*	Stream / Lake / Estuary	Glacial	~2,500,000	~19,000	Medium	Present	Present	so, co, ch
Hidden - Kenai	Stream / Lake	Clear	~40,000	~138,000	High			so
Quartz - Kenai	Stream	Clear	~17,000	NA	Medium			so, co
Snow - Kenai	Stream	Glacial	~6,000	~220	Low			so, co
Russian - Kenai	Stream / Lake	Clear	~150,000	~46,000	Medium-High			so, co
Grant - Kenai	Stream / Lake	Glacial	~100	~20	Low-Zero			so, ch
Moose - Kenai	Stream	Stained	~5000	~700	Low	Present		so, co, ch

(1) Water Type / Clarity (Typology): Clear Visibility, Glacial Turbidity, Tannin Stained

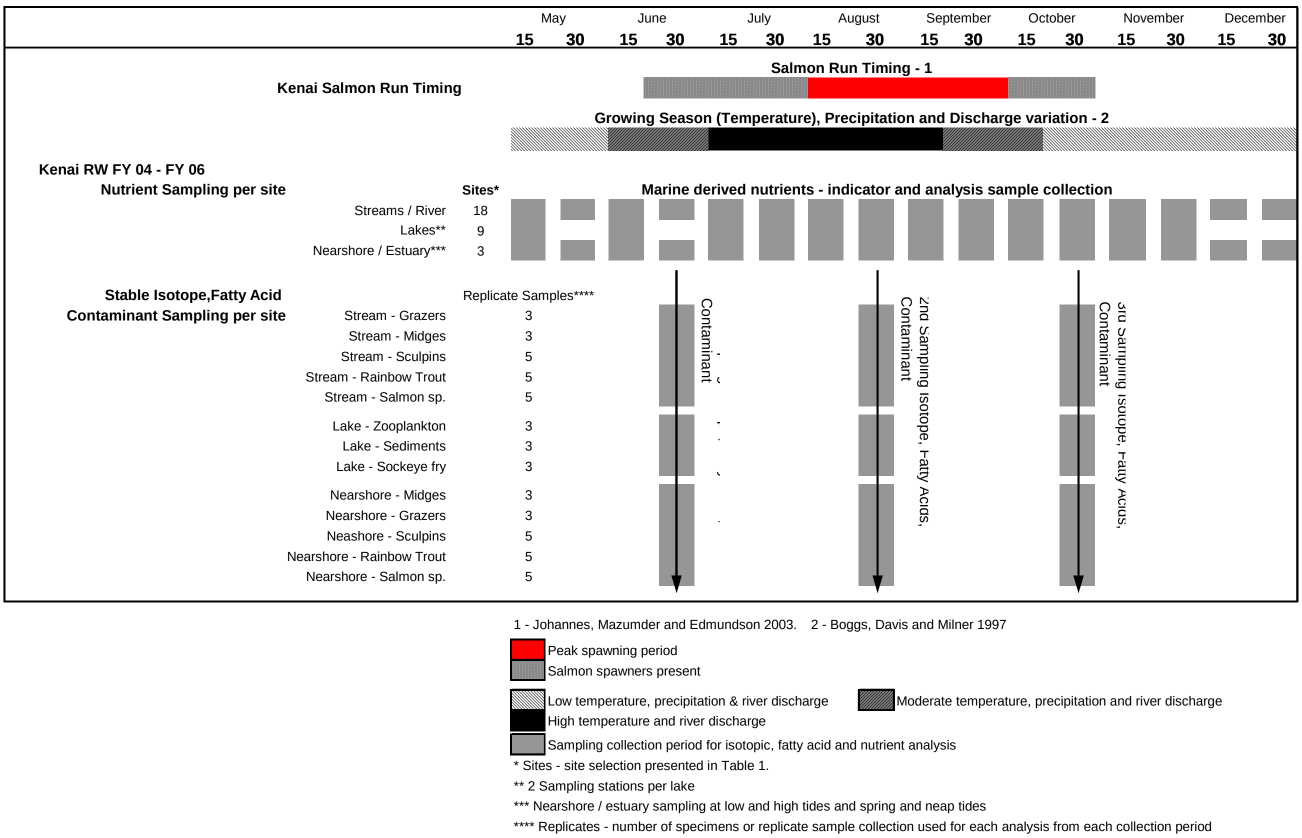
(2) Recent Average Salmon Escapement = 10 year average
(3) Salmon Density = Escapement per unit river discharge (mean annual flow)
(4) Anadromous Salmon Species so = sockeye, co = coho, ch = chinook
* Skilak and Kenai Lakes to be sampled in the Kenai Mainstem

Lakes

We will collect samples for nutrients from 2 pelagic sites per lake, once a month in May and June and twice per month until November. Samples will be collected with an integrated epilimnetic tube sampler (Mazumder et al. 1988) and stored in acid washed carboys and returned to the lab for processing. During 3 specific sampling periods, spring (May-June), summer (July-August) and fall (September-October), zooplankton will be collected from 50m to the surface vertical hauls (153-µm-mesh net, 0.5 diameter).

In addition, we will capture sockeye fry using tow-nets. Both zooplankton and fry samples will be frozen at -80°C until processed for stable isotope composition and fatty acids (potentially other indicators). We will install sediment traps and sample them once per month during the ice free season, to collect sinking material (eplimnetic plankton, organic material) in each lake. The accumulated sediment material will be concentrated to 100 ml (water and sedimented material) and frozen at -80°C for isotope and nutrient analysis.

Table 2: Proposed sampling protocol using nutrient, isotopic, fatty acid, and contaminant indicators of MDN relative to know salmon run timing and trends in growing summarized from climate data and the Kenai RW hydrograph.



Estuary

In the lower the Kenai River estuary, nutrient samples will be collected from a range of fresh to saline waters. Collected samples are to be collected and preserved as described above for the lakes. During 3 specific spring, summer and fall sample periods, benthic macroinvertebrates (grazing insect and midges: c.f. Table 2) will be collected by standard methods (epibenthic sled). Organisms will be rinsed into sample jars and frozen at -80°C until processed for stable isotope composition and fatty acids (potentially other indicators). In addition, seines, small mesh nets and minnow traps will be used to collect samples of resident fish (sculpins and rainbow trout) and juvenile salmon (sockeye, pink, coho, chinook). Invertebrates and fish will be frozen at -80°C for analysis. We will conduct a preliminary survey to determine suitable benthic invertebrates to use taxonomic or species groups as indicator of MDN signals.

Laboratory Methods

For the analysis of nutrients, portions of the bulk water samples from the field will be (1) refrigerated until analyzed for turbidity, (2) frozen for the determination of total phosphorus (P) and Kjeldahl nitrogen (N); and (3) filtered through a Whatman GFF grade filter and then frozen for the analysis of nitrate + nitrite, ammonium and true color. Turbidity will be measured with a HF model 00B turbidimeter and true color is to be determined on a filtered (GFF) sample by measuring the spectrophotometric absorbance at 400 nm and converting to equivalent platinum-cobalt units (Koenings et al. 1987). Total-P will use molybdenum blue method following acid-persulfate digestion. Nitrate + nitrite will be measured as nitrite following cadmium reduction and determinations of ammonia will follow the phenyl-hypochlorite procedure using colorimetric analyses. Kjeldahl-N is to be measured as ammonia following acid-block digestion. We will estimate total-N concentration as the sum of Kjeldahl-N and nitrate+nitrite. All procedures for water chemistry and nutrient analysis are detailed in Koenings et al. (1987).

For determination of carbon-13, nitrogen-15 and sulfur-34, samples of tissue 0.5-5mg dry weights (dependent on N and C contents) are placed in tin metal capsules and combusted (1100°C). Stable isotope ratio of carbon (^{13}C), nitrogen (^{15}N) and sulfur (^{34}S) ratio in the gas from the combusted bulk material compared to a standard will be determined using a mass spectrometer (Thermo Finnigan Delta Plus Advantage – Conflo to CHNSO analyzer). Results are expressed as deviation values (δ) in parts per thousand (‰) difference between sample and standard ratios according to the equation: $\delta^{13}C$, or $\delta^{15}N$, or $\delta^{34}S = [(R_{\text{sample}} - R_{\text{standard}})/R_{\text{standard}}] \times 1000$, where $R = {}^{13}C/{}^{12}C$ or ${}^{15}N/{}^{14}N$ or ${}^{34}S/{}^{33}S$.

For fatty acid analysis, samples will be freeze dried, homogenized and extracted in a chloroform / methanol mix (Parish 1999). The extraction mixture will be sonicated, vortexed and centrifuged (Kainz et al. 2002). The lower chloroform layer will be removed and stored below 0^0C under nitrogen. The extraction will be washed in chloroform to purify. Lipids plus classification will be determined using Chromarod-Iatroscan thin-layer chromatography with flame ionization detection. For detailed fatty acid analysis, extracted samples will be transformed into fatty acid methyl ester (FAME). FAME will be analyzed for individual fatty types using gas chromatograph with mass spectrometer (GCMS). An Omegawax 320-column, specially designed for the separation of polyunsaturated fatty acids will be used (Parrish 1999, Arts and Wainman 1998).

Table 3: Total number of samples and replicate for analysis.

Kenai RW FY 04-05, Kenai RW FY06		FY04-06	Total Water Samples Collected for Analysis			Total Annual Sampling Periods
Nutrient Indicator Sampling per site		**Sites**	**FY 04**	**FY 05**	**FY 06**	**FY04-06**
(i.e. TP, TN, Ammonia)	Streams / River	18	288	288	72	16
	Lakes	9	108	108	30	12
	Nearshore / Estuary*	3	95	57	30	19
	Total Samples		**491**	**453**	**132**	

		Biological Indicator Replicate	Total Biological Samples Collected			Total Annual Sampling Periods
Biological Indicator Sampling per site		**FY04-05**	**FY 04**	**FY 05**	**FY 06**	**FY04-06**
(i.e. Isotopes, fatty acids etc.)	Stream - Grazers	3	162	162	81	3
	Stream - Midges	3	162	162	81	3
	Stream - Sculpins	5	270	270	135	3
	Stream - Rainbow Trout	5	270	270	135	3
	Stream - Salmon sp.	5	270	270	135	3
	Lake - Zooplankton	3	81	81	54	3
	Lake - Sediments	3	81	81	54	3
	Lake - Sockeye fry	3	81	81	54	3
	Estuary - Midges	3	27	27	18	3
	Estuary - Grazers	3	27	27	18	3
	Estuary - Sculpins	5	45	45	30	3
	Estuary - Rainbow Trout	5	45	45	30	3
	Estuary - Salmon sp.	5	45	45	30	3
	Total Samples		**1566**	**1566**	**855**	

* Nearshore / estuary sampling at low and high tides and spring and neap tides

C. Data Analysis and Statistical Methods

We will develop relational databases in MS Access for storing inventory and monitoring data (c.f. GEM Data Management Standards). The database will be constructed to input and link spatial data to a geographic information system (GIS). The core database structure will have the following components linked to sampling site and date of collection: water nutrients ($\mu g\ L^{-1}$), taxonomic group, stable isotopes of carbon, nitrogen, sulfur (‰), fatty acids designation and density attributes for biological characteristics being sampled (density, biomass, species diversity etc). Because watersheds are affected by many different factors, we will organize databases relative to the spatial / geographic relationship between measurements at various sites and the physical and biological characteristics specific to that site and the project experiment / empirical design.

Since the main purpose of our study is to develop a reliable indicator of MDN, we will use <u>linear and non-linear</u> regression techniques to test the functional relationships between salmon abundance and our suite of MDN indicators. Our sample design considers individual sub basin sites in the watershed as the unit of observation with independent abundance and timing of salmon escapement and carcass deposition. In addition, we will use analysis of covariance (ANCOVA) to test for differences in slopes and intercepts among watershed sub basin types (clear, stained and glacial) using a significance level of $\alpha = 0.05$. For a variable considered functionally dependent on more than one other variable, we will use a multiple regression approach, where the significance level for retaining variables will be $\alpha = 0.1$. However, because we do not know whether the underlying form of this relationship is linear, curvilinear or exhibits some asymptotic behavior, we will also employ non-linear and non parametric statistical techniques. The advantage of the non-linear approach is that it allows a broad range of functions that can be fit. All statistical tests will be conducted using SAS or Systat software. To determine the best indicator of MDN, we will select among a set of models providing multiple views of the data (Hilborn and Mangel 1997) relating salmon abundance to the various proxy indicators of MDN. Assuming a set of *a prior* candidate models, we will employ Akaiki's Information Criterion (AIC) model selection process (Burnham and Anderson 1998). AIC allows models to be ranked and scaled with AIC values interpreted as the likelihood that a given model is the best model of the alternatives, given the data. We will seek additional consultative advice and appropriate analysis techniques in conjunction with our collaborators from other watershed groups.

D. Description of Study Area

Numerous river systems drain into the Cook Inlet basin covering an area of 101,473 km^2 (Fig. 1). The habitat formed by rivers, sloughs, lakes, wetlands, estuaries and nearshore environments provide corridors for fish migration, and important spawning and rearing habitat for all five species of Pacific salmon, anadromous euchalon and resident salmonids and other fish. Three water types can be identified in the streams, river and lakes of the Kenai RW (Table 1). In addition to clear water systems, many of the rivers and streams are fed by glaciers and contain large amounts of suspended silt particles imparting turbidity, while others originate from peaty soils, have large concentrations of dissolved organic compounds and are highly stained (Milner et al. 1997; Edmundson and Carlson 1998). The Kenai RW provides a unique setting for interdisciplinary research on a coastal watershed with a mixture of clear, glacial, wetland and salmon based ecosystems. Very few other Alaskan watersheds of this size and scale exhibit such varied terrestrial, freshwater and estuarine landscapes. The watershed is 5,054 km^2 in size, with a diversity of landscapes and habitats; six important species of salmonids; six abundant mammal species; and large forested and natural areas (Johannes et al. 2002). Average annual rainfall is about 45 cm in the Kenai area. July temperatures for this area average about 12°C compared to -12°C in January. Table 1 summarizes the various sub basin study sites.

Nutrients such as phosphorus and nitrogen enter the watershed from a variety of marine, terrestrial and atmospheric sources. Meltwater from headwater glaciers also contributes nutrients and large amounts of rock flour and silt (Edmundson and Carlson 1998) to many areas of the drainage. The largest lakes in the system (Kenai and Skilak) function as important buffers to variations in river discharge, silt and nutrient loading from the upper watershed (Dorava and Milner 2000). These lakes are considered the main nursery habitat for juvenile sockeye salmon, the most abundant salmon stock in the watershed (Edmundson et al. 2003). The river channel below the largest lake (Skilak) is lower gradient, meanders through forested areas and wetland bogs, and has numerous side channels and sloughs. The wetlands, and the tributaries which drain them, are nutrient rich relative to portions of the upper watershed, and provide complex habitats in support of chinook, coho, chum and pink salmon, and a diversity of wildlife species including moose, bear and wolves. On average over one million salmon, smelt and other anadromous fish enter the drainage annually to spawn and die, leaving marine-derived nutrients (phosphorus and nitrogen), lipids and proteins from carcasses throughout the watershed.

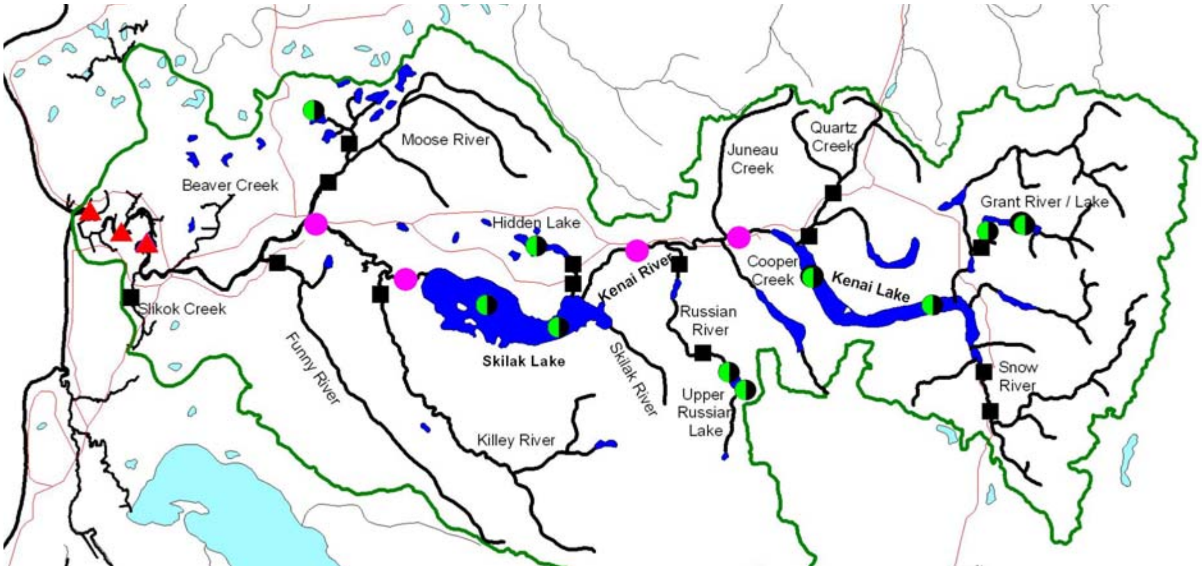


Figure 1: Sampling sites in the Kenai RW. Red triangle – estuary site; black square – stream site; purple circle – Kenai River mainstem site; green-black circle – lake site.

E. Coordination and Collaboration with Other Efforts

The principal investigator (PI) and the study team have considerable experience in conducting and coordinating large-scale studies. The PI will provide the leadership to maintain focus, facilitate communication and promote collaborations, research initiatives and partnerships. Through discussion with all the participating researchers, collaborators, agencies and stakeholders, we have developed a framework for coordination, collaboration and management. Our project will be managed by the PI, the three Co-PI's and staff members. The PI and Co-PIs will serve leadership functions to ensure collaboration, effective networking and the integration of specific research activities into comprehensive management strategies for Kenai RW.

A Public Advisory Group (composed of the PI, one an invited member from each of GEM, ADG&G, KRSFA, UCIDA, KRWAF, USGS, USEPA, USWFS) will work with and advise the scientific team on ongoing stakeholder issues to help support research and stakeholder collaboration and ongoing interactions with the Kenai community at large. Existing community stakeholders like the Kenai Watershed Forum and Kenai River Sportfishing Association are integral components of our research strategy and will assist in support of sample collection. An administrative office, including the PI, a program manager and an administrative assistant will manage the business and communication activities. This office will attend to the accounting of funds, management of information, internal and external communications, maintenance of the Kenai RW web site, liaison, promotion of science to stakeholders, and co-ordination of the periodic workshops, and other necessary meetings and discussions. The research administration and Accounting

Department at the host institution will manage the financial administration and auditing of funds, and will provide annual audited financial statements.

This Kenai River Watershed MDN Research Initiative is the result of two years of communication, coordination and discussion with researchers from several academic institutions and agencies, stakeholders and community groups. To be successful in its most important goals, we must address the complex issue of detecting MDN and the sensitivity of Kenai RW to MDN input and its processing at the scale watershed and its component ecosystems, which will require integration of interdisciplinary teams covering terrestrial/wetland, river/stream, lake, and near-shore ecosystems. To achieve these, we have put together a team of excellent researchers from these diverse disciplines. We will work off our existing working relationship among all the researchers, collaborators and stakeholders in the Kenai RW. The Kenai RW proposal will not only foster the integration of various perspectives and expertise, but will also assist in the development of expertise, thereby increasing scientific and knowledge transfer capacities in critical regions of Alaska. We propose to help facilitate regular meetings, workshops and symposia to transfer our results to other research groups in the region.

III. SCHEDULE

A. Project Milestones

- Finalize study design and initiate field work: April 1, 2004
- Develop partnerships and additional funding opportunities to support long term monitoring and research linking Kenai RW to the GOA ecosystems 2004-2006.
- Develop indicators of MDN: December 15, 2005
- Compare and contrast (statistical analysis) Kenai sub basins as a functions salmon run strength and indicators of MDN – December 15, 2005
- Estimate MDN input by analyzing nutrient flow patterns to the Kenai RW: September 30, 2006
- Complete draft final report: April 1, 2006
- Submit peer-reviewed manuscript and final report: September 30, 2006

We realize that processing of nutrient and biological samples from Year 3 (May 2006 – November 2006) along with the accompanying data compilation and analysis will not be completed by the end of FY 06 (30 September 2006). In Year 3, we plan to prepare a final report based largely on our research findings from the first two years of field and laboratory work; for example, establishing methods and determining robust indicators of MDN. In our third year, we will begin our synthesis of MDN indicators and comparative input to the Kenai RW and examine the feasibility of various methods to quantify productivity responses to MDN. However, it is our intent to develop a subsequent proposal to examine both the full ecosystem component and watershed responses to MDN at different scales and their implications for the productivity of the Kenai RW. The general focus of such a proposal will include the following objectives: (1) quantify total MDN input and output within different ecosystem components of the Kenai RW, (2) quantify and model the relationship between MDN input and the productivity of foodweb components and/or indicator species, (3) identify the relationship and potential influence of MDN input on fish and wildlife survival, growth and productivity under variable climate and land management conditions, and (4) determine the relative importance of

MDN constituents (C, N, S, P and Fatty acids) in limiting productivity of different trophic levels in the Kenai RW. This proposal would be submitted with a heightened awareness and understanding of MDN effects in the Kenai RW, the dual linkages between coastal watersheds and GOA, and the needs of GEM.

B. Measurable Project Tasks

<u>FY 04-06</u>

By Quarter

- Quarterly progress update for distribution to Kenai RW contact list;
- Kenai RW Website to act as repository for existing research ideas and update;
- Develop partnerships and additional funding opportunities to support long term research and monitoring linking Kenai RW to the GOA ecosystems 2004-2006;
- Conference call – researchers on progress update and planning;

<u>FY04</u>

1st quarter (October 1, 2003-December 31, 2003)

- Assign project team members and recruit staff and graduate students
- Finalize study plan, purchase supplies and equipment

2nd quarter (January 1, 2004-March 31, 2004)

- Make field sampling preparations
- Attend annual EVOS/GEM meeting (January)
- Convene 1st Alaska watersheds workshop (February);
- Convene 1st Scientific and Public Advisory Workshop (March)

3rd quarter (April 1, 2004-June 1, 2004)

- Implement watershed sampling and monitoring program
- Begin analytical work on chemical and biological samples

4th quarter (July 1, 2004-September 30, 2004)

- Continue watershed sampling and monitoring program
- Continue analytical work on chemical and biological samples
- Prepare annual report

<u>FY 05</u>

1st quarter (October 1, 2004-December 31, 2004)

- Complete field work for Year 1
- Continue analytical work on chemical and biological samples
- Data compilation and analysis

2nd quarter (January 1, 2005-March 31, 2005)

- Complete analytical work for Year 1
- Data compilation and analysis
- Attend annual EVOS/GEM meeting (January)
- Convene 2nd Alaska watersheds workshop (February);
- Convene 2nd Scientific and Public Advisory Workshop (March)

3rd quarter (April 1, 2005-June 1, 2005)

- Implement Year 2 watershed sampling and monitoring program

- Begin Year 2 analytical work on chemical and biological samples

4th quarter (July 1, 2005-September 30, 2005)

- Continue watershed sampling and monitoring program
- Continue analytical work on chemical and biological samples
- Prepare annual report

<u>FY 06</u>

1st quarter (October 1, 2005-December 31, 2005)

- Complete field work for Year 2
- Continue analytical work on chemical and biological samples
- Data compilation and analysis

2nd quarter (January 1, 2006-March 31, 2006)

- Initiate development of proposal for longer term research relating MDN with Kenai RW productivity
- Complete analytical work from Year 2
- Develop indicators of MDN
- Attend annual EVOS/GEM meeting (January)
- Convene 3rd Alaska watersheds workshop (February);
- Convene 3rd Scientific and Public Advisory Workshop (March)

3rd quarter (April 1, 2006-June 1, 2006)

- Implement Kenai River watershed sampling and monitoring program
- Begin Year 3 analytical work on chemical and biological samples
- 2 Graduate will defend thesis, submission of peer reviewed papers

4th quarter (July 1, 2006-September 30, 2006)

- Complete Year 3 watershed sampling and monitoring program
- Complete Year 3 analytical work on chemical and biological samples
- Estimate total MDN input to the Kenai RW
- Prepare final report.

IV. RESPONSIVENESS TO KEY TRUSTEE COUNCIL STRATEGIES

A. Community Involvement and Traditional Ecological Knowledge (TEK)

At present our Kenai RW contact list incorporates 75 individual contacts from 39 different stakeholder groups and agencies. Our project is sensitive to ongoing information dissemination and communication and will endeavor to maintain ongoing and establish new stakeholder interest and support. We recognize that involving local communities in Kenai RW research planning is extremely important we will make this a top priority throughout the delivery of this proposal. We have also partnered with Kenai Watershed Forum and Cook Inlet Keepers to support water nutrient sample collections in the Kenai RW. We intend to expand our collaborations with both groups and other community and Native groups in the Kenai RW and other watersheds of interest to support ongoing field sample collection and through communication and awareness of activities. For the Kenai RW research initiative to be successful, we will support collaboration with local agencies, government, NGO's and community groups to support informed decision making processes toward sustainable ecosystem based management to key salmon resources and their nutrients returning to the Kenai RW.

B. Resource Management Applications

This research proposal and the final research initiative in the watershed is not intended to explicitly deliver management solutions for terrestrial, freshwater, fisheries and marine resource and human use issues. However, the intent of this research is to allow perspective management agencies and stakeholders to support informed decision making towards sustainable ecosystem management in the watershed and marine areas of influence to the Kenai River.

V. PUBLICATIONS AND REPORTS

The following reporting will be delivered through this proposal:

- Technical bulletins – Scientific and Public advisory (i.e. Johannes et al. 2002a, b)
- White paper for broad distribution
- Communication poster and conference poster
- Published and peer reviewed primary literature publications
- Cook Inlet and South-central Alaska Watershed Researchers Network web site

VI. PROFESSIONAL CONFERENCES

- Will participate in annual (FY 06) 2005 EVOS / GEM meeting
- Contribute to development of special symposium on MDN in coastal watersheds at: GEM, ASLO, AFS and NABS meetings

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RESUMES

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