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Project No: ________________
Date Received: ________________

GEM PROPOSAL SUMMARY PAGE

(To be filled in by proposer)

Project Title:	The Influence of Adult Salmon Carcasses on Energy Allocation in Juvenile Salmonids
Project Period:	"FY 04-FY 06"
Proposer(s):	Ron Heintz and Lawrence Schaufler NOAA Fisheries Auke Bay Laboratory
Study Location:	Kenai Peninsula
Abstract:	A brief (150 words or less) summary of the project. Include what question(s) the project will address, what products the project will produce, and where and when the work will be done. The abstract may be edited for clarity, brevity, and readability by Trustee Council staff.

This project examines the effect of adult salmon carcasses on the energy allocation in juvenile salmon. Juveniles must allocate energy between the competing demands of growth and energy storage to minimize exposure to predation while forestalling starvation over winter. However, little is known about how the energy offered by carcasses helps juveniles resolve these competing demands. We propose contrasting annual energy dynamics in age-0 Dolly Varden from Kenai peninsula streams with and without carcasses present. Fatty acid analysis will be used to identify marine signal strength and persistence in the lipids of the juveniles. We will combine proximate and lipid class analyses to determine the proportions of their total energy allocated to storage versus structure, and examine how seasonal variation in allocation differs among streams and carcass densities. Finally, we examine the influence of carcasses on growth rate and the relation between growth and energy allocation.

Funding:	EVOS Funding Requested:	FY 04	$ 48.4	
		FY 05	$ 42.3	
		FY 06	$ 14.1	TOTAL:104.8
	Non-EVOS Funds to be Used:	FY 04	$15.3	
		FY 05	$27.6	
		FY 06	$16.5	TOTAL:59.4

Date: June 12, 2003

(NOT TO EXCEED ONE PAGE)

The Influence of Adult Salmon Carcasses on Energy Allocation in Juvenile Salmonids

Ron Heintz and Lawrence Schaufler

I. NEED FOR THE PROJECT

A. Statement of Problem

The presence of salmon carcasses in the rearing habitat of juvenile salmonids represents an important energetic resource, but little is known about how juvenile salmonids exploit that energy. Juvenile salmonids rearing in lotic systems must balance their need for growth against the need to store energy for winter (Shuter and Post, 1990) and decisions regarding the allocation of energy have long-term effects on life history decisions (Thorpe, 1990). These issues are particularly important for age-0 salmonids, whose small size makes them susceptible to a greater range of predators and relatively high mass-specific metabolic rates make them more susceptible to starvation (Schmidt-Nielsen, 1984). The amount of energy offered by salmon carcasses may therefore make them an important feature of juvenile salmon habitats. The presence of carcasses has been shown to both increase juvenile coho growth (Wipfli et al. In press) and energy reserves (Heintz et al., in press) under controlled conditions. Increased size and energy reserves at the end of the growing season in juvenile fish leads to increased over-winter survival (Conover and Schultz, 1997), consequently the energy availability in juvenile-rearing habitats has long-term implications for the future productivity of those habitats.

Understanding the relationship between carcass density and energy allocation in juvenile salmonids provides an integrated measure of the influence of marine nutrients on watersheds. Juvenile salmonids occupy a pivotal position in the foodwebs of Alaskan watersheds. Growth and energy storage in juvenile salmon integrates abiotic features as well as the availability of prey and the relative predation risk (Post and Parkinson, 2001). The presence of carcasses increases the availability of prey by increasing stream production (Wipfli et al., 1999) or by providing tissue that can be consumed (Gende et al., 2002). The increased availability of prey and tissue may also potentially reduce predation risk. Thus, by measuring the growth and energy storage of age-0 salmonids exposed to various levels of marine nutrients the overall impact of the nutrients on the population can be characterized.

This study will examine how the presence of carcasses influences energy allocation in juvenile salmonids, by contrasting cycles of growth and energy storage in wild populations that are exposed or not exposed to carcasses. Salmonids exposed to large energy sources during the growing seasons are likely to alter their allocation strategies relative to those of salmonids that depend on energetic resources derived entirely from the watershed. These shifts in energy allocation are likely to have long-term implications for the life histories of juveniles rearing in lotic systems. Currently our understanding of the importance of carcasses in the life history of juvenile salmonids is limited to a few phenomenological studies that hint at the importance of carcasses in the life history of anadromous populations, but provide little direct demonstration of the benefit. Understanding how the presence of carcasses influences annual cycles of growth and energy storage in juvenile salmonids will ultimately lead to an understanding of the best periods for monitoring the impacts of marine-derived nutrients on freshwater habitats.

There are no published data documenting the effect of carcasses on the annual lipid cycle in juvenile salmonids, but unpublished data suggest that the presence of carcasses modifies the way juvenile salmonids typically process lipids. For example, Sashin Creek in southeastern Alaska has a barrier waterfall which prevents the immigration of adult salmon beyond the first 2 kilometers of stream. A population of steelhead was transplanted from below the barrier to the section above approximately 60 years ago. Between April and September, the lipid content of juvenile steelhead rearing in the lower section decreased (Figure 1). In contrast, juvenile steelhead rearing in the upper section increased their lipid content during the same period. The pattern observed among fish in the upper section is consistent with patterns typically reported in published work (Gardiner and Gettes, 1980). However, the pattern observed for the lower section is consistent with observations we have made in other anadromous streams near Juneau (Figure 2). Fluctuations in lipid content generally follow fluctuations in the amount of lipid stored as triacylglycerols, suggesting that these data are evidence for the effect of carcasses on energy allocation during the year. However, there are no data describing how carcasses might influence energy allocation towards growth throughout the year.

B. Relevance to GEM Program Goals and Scientific Priorities

The GEM plan identifies a need for indicators that show how and when to measure marine-related biological production in wathersheds. A useful index for monitoring the effects of marine nutrients on watersheds should integrate effects across trophic levels, correlate closely with variation in economically important features of the watershed, and be relatively inexpensive to measure. Such an index would relate the magnitude of the marine subsidy to its ultimate value in terms that have direct utility to resource managers. Recently, analysis of the lipids of juvenile salmonids has shown promise as such an index. Changes in the fatty acid composition of juvenile salmon exposed to various densities of carcasses suggested consumption of adult salmon tissues were accompanied by significant increases in growth and stored energy (Heintz et al., In press; Wipfli et al., In press). The changes in fatty acid composition represent a type of marine signal that was easily detected in coho tissues. The marine signal was characterized by elevated levels of $\omega3$, $\omega11$ and some $\omega9$ fatty acids, along with diminished levels of $\omega6$, mono-unsaturated and saturated fatty acids. Thus, an identifiable marine signal was found to correlate closely with the energetic status of economically important species. It is important that the signal and correlated response are found in salmonids, because salmonids are the ultimate agents for importing marine nutrients into watersheds. Streams that produce a large number of well-nourished smolts are more likely to produce a large number of adults (Bradshaw and Heintz, 2000). Consequently, monitoring the effect of carcasses on the energetic status of juvenile salmon provides a direct measure of the effect on an important component of the local foodweb, but also provides an index to the future subsidies of the system.

While the presence of carcasses produces marine signals in the tissues of juvenile salmon, the persistence of the signal may be a better indicator of its importance to juvenile salmonids. Isotopic studies performed in conjunction with adult salmon runs have demonstrated juvenile salmon take up marine-derived nitrogen and carbon (Kline et al., 1990). In addition, the presence of carcasses results in increased juvenile growth (Wipfli et al., 1998), lipid reserves and altered fatty acid composition (Heintz et al., in press). These data demonstrate that identifiable marine signals are taken up and reveal the immediate fate of marine-derived carbon, but they do little to convey the overall impact of this carbon on the well-being of populations residing in anadromous streams. For example, it is not known how long these signals and lipid reserves persist in the tissues of juvenile salmonids. Atlantic salmon parr may derive as much as 50% of their energy from lipid reserves during winter (Berg and Bremset, 1998), indicating juvenile salmonids require a relatively large lipid reserve prior to winter. Thus, variations in carcass availability may directly influence the over-winter survival of juvenile salmon. Consequently, changes in the intensity of the marine signal and lipid reserves between fall and spring may be the most sensitive tool for monitoring impacts of marine nutrients on the production and survival of juvenile salmonids.

II. PROJECT DESIGN

A. Objectives

This project is a component of the project proposed by Walker et al., so field sampling conforms to the plan outlined in that proposal. The aim of this project is to understand how the presence of carcasses influences the allocation of energy between growth and storage in age-0 Dolly Varden charr. This age class has been selected because its survival is most tightly linked to the availability of energy and therefore it is likely to be the best indicator for monitoring marine influences. Fish will be sampled periodically from the Anchor River, a stream with adult salmon runs, and Happy Valley Creek, a stream with no adult salmon runs. Dolly Varden reside in both of these systems located on the Kenai peninsula, making comparisons between systems possible. On each sampling occasion fish will be collected to determine their energy density (Kjoules/gram), proximate, lipid class, and fatty acid compositions. Energy density will provide a basis for contrasting the energetic content of different populations, while the lipid class and proximate composition data will indicate how the energy is partitioned between energy storage (triacylglycerols) and structure (protein and phospholipids). Fatty acid analysis will be used to verify the presence of the marine signal. In addition, growth rates will be determined by examining the RNA/DNA ratio in samples of muscle. From these data we will determine how the presence of marine subsidies delivered by adult salmon influence the allocation of energy in juvenile salmonids.

The project has the following objectives:

I. Characterize the persistence of a marine signal in the lipids of age-0 Dolly Varden charr.

<u>Rationale</u>

Fatty acid analysis will be used to identify and characterize the persistence of the marine signal in the lipids of age-0 Dolly Varden. The marine signal will be measured by sampling the fatty acid composition of returning adult salmon and characterized with a principal component model. Fatty acid composition of the triacylglycerols isolated from juvenile fish will be examined to determine if the marine signal is present in stored lipids and measure its strength. These data will be used to test the following hypotheses:

Ia) The presence of carcasses leads to the incorporation of marine-derived lipids in Dolly Varden

Ib) The persistence of this signal is a function of the original signal strength.

II. Characterize seasonal variation in energy allocation in populations of age-0 Dolly Varden exposed and not exposed to adult salmon carcasses.

Rationale

Energy allocation will be determined by calculating the proportion of total calories in each fish devoted to structure (protein, cholesterol, and phospholipid) or storage (triacylglycerols and wax esters) and related to presence or absence of carcasses to test the following hypotheses:

IIa) The presence of carcasses results in greater allocations of energy towards storage throughout the year.

IIb) Age-0 Dolly Varden in streams with carcasses enter winter with higher energy reserves than those in streams with no carcasses.

IIc) Age-0 Dolly Varden in streams with carcasses have higher energy stores after winter than those in streams with no carcasses.III. Characterize seasonal growth patterns in Dolly Varden exposed and not exposed to adult salmon carcasses.

Rationale

Growth rate, measured by RNA/DNA ratios in fish sampled for lipids will be related to the presence or absence of carcasses and the proportion of energy allocated to structure to test the following hypotheses:

IIIa) The presence of carcasses results in higher growth rates during the growing season in age-0 Dolly Varden.

IIIb) Age-0 Dolly Varden with high growth rates are able to allocate more energy towards storage than those with lower growth rates.

B. Procedural and Scientific Methods

Ideally, fish will be collected six times during the year, but collections will be limited by procedures outlined in the companion proposal submitted by Walker et al. Ideal sampling periods will begin following the 2003 growing season, immediately before the 2004 king and coho salmon runs begin in spring, twice during the runs, immediately after the 2004 run, and the following spring. Each collection will consist of 6 age-0 fish from each of the 5 stations on each river. Thus, 30 fish will be collected from each stream on each sampling date. In addition, 5 samples of dead male and female (without eggs) adult king and coho salmon will also be collected to provide a fatty acid fingerprint representing the marine sources. Thus, a total of 380 samples will be processed representing various locations in two different streams at four times of the year.

Fish will be collected, their lengths recorded, and then immediately frozen in liquid nitrogen. Frozen fish will be shipped to Auke Bay Lab and stored at -80 °C prior to processing. Prior to homogenizing each fish, the otoliths will be removed for aging, and a sample of muscle will be removed for RNA/DNA analysis using the method of Wang et al. (1993). The lipid will extracted using the Bligh and Dyer method (Bligh and Dyer, 1959) on a Dionex 200 Accelerated Solvent Extractor (ASE), followed by rotovapping and volumetric dilution to a final volume of 1 ml. Total lipid content will be determined gravimetrically from the lipid fraction after solvent evaporation. For quality control, duplicate samples will be analyzed per group of 15-20 fish, and a standard reference sample analyzed with each group. The Dumas method will be used to determine protein content with a Leco FP-528 nitrogen analyzer, in which homogenate samples are dried and subject to combustion at 850 °C, followed by total nitrogen gas quantitation (AOAC Methods, 1995). Nitrogen content is converted to % protein using 6.25 as the conversion factor (AOAC Methods, 1995). Protein measurements will be performed in duplicate, and samples reanalyzed if the duplicates vary significantly. Moisture and ash contents will be determined gravimetrically on a Leco TGA-601 ash analyzer. National Institute for Standards and Technology (NIST) standard reference materials and reference standard samples will be used for quality-control purposes in the protein and ash analyses. Carbohydrate content will be determined by subtraction of the contents of the other components from 100 %. Energy density will be calculated from proximate composition using the constants 39.3, 23.49 and 17.05 KJ/g for lipid, protein, and carbohydrate, respectively.

Lipid class composition will be determined by high performance liquid chromatography (HPLC) on a Hewlett Packard HP1050 equipped with a Sedex 55 Evaporative light scattering detector. Lipid classes will be separated with a 3 µm silica column using a solvent gradient and quantified against 3-point calibration curves normalized to an internal standard (PDME). Calibration standards include myristate, cholesterol, triolein, monolein, C21:0, choline, plant phosphatidylinositol, egg phosphatidylethanolamine, and bovine phosphtidylserine. A proportioning valve will be used to collect 40% of the eluting triacylglycerol volume for fatty acid analysis.

Fatty acids in the triacylglycerol fraction will be trans-esterified following the method of Christie (1982) and quantified on a Varian Saturn 2200 Gas Chromatograph with a Mass Spectrometric detector (GC/MS). Fatty acid methyl esters (FAMEs) will be separated with a 30 m Omegawax-250 fused silica column operating under a temperature program. FAME quantification will rely on 5-point calibration curves developed for 29 different FAMEs and normalized to an internal standard added prior to transesterification. Quality control is verified using the measured recovery of surrogate compounds added prior to injection. FAMEs identified by mass spectrometry, but not represented by a calibration standard will be quantified using the calibration curve developed for the most closely related isomer. Samples are processed in groups of 15-20, along with standard reference materials and duplicated samples to evaluate repeatability and as quality control measurements.

Analytical results will conform to QA standards set by a consortium of Federal and University labs actively engaged in lipid analysis in the Pacific Northwest and Alaska. Participants in the consortium include: University of Washington, National Marine Fisheries Service (NMFS) Northwest Fishery Science Center, NMFS Alaska

Fishery Science Center, University of Alaska Fairbanks Marine Technology Center, University of Alaska Anchorage, Alaska Department of Fish and Game, and Dalhousie University.

C. Data Analysis and Statistical Methods

<u>Objective I: Characterize the persistence of a marine signal in the lipids in age-0 salmonids</u>

The fatty acid data will be used to verify the presence of the marine signal in the lipids of the fish collected from the stream receiving marine subsidies in the form of an returning adult salmon. Initially, the mean fatty acid composition of adult fish returning to the Anchor River will be characterized by a principal components model. One such model will be used for each of the adult runs: chinook, coho, and pink salmon. Variables included in the model will be determined from stepwise linear discriminant function analysis. Once the model has been constructed, the fatty acid compositions of the age-0 Dolly Varden will be fit to the adult model and the residual standard deviation (RSD) will be calculated. The ratio of the RSD for the Dolly Varden fatty acids divided by the mean RSD for the carcass model is distributed as an F statistic and will be used the test the hypothesis that the fatty acid vectors of the carcass and Dolly Varden are equal (Wold and Sjonstrom, 1977).

The inverse of the RSD for each fish will be used as a measure of signal strength. The persistence of the signal will be examined by relating the change in signal strength for fish sampled from a given location as a function of sampling date. The slope of the resulting relation will measure persistence for each of the five locations sampled in Anchor River. The relation between signal persistence and initial strength will be determined by regressing the persistence measure for each location against average signal strength at the end of the growing season and testing if the resultant slope differs from 0. The mean signal strength for fish from a given location will also be regressed on values of the water chemistry proxies developed by Walker et al..

<u>Objectives II and III: Characterize seasonal variation in energy allocation and growth</u>

The hypotheses regarding energy allocation will be examined by an ANOVA and regression using the proportion of total energy allocated to storage as the response variable. A two-way factorial ANOVA will be used to examine differences between stream types and season. Replicates will be the means response for each of the 5 locations sampled in each stream on each sampling date. A significant interaction between stream type and sampling period will be interpreted as differences in the seasonal allocation strategies between streams. The absence of an interaction but differences in the main effects will be interpreted as differences in allocation strategy resulting from either seasonal or carcass effects. Mean growth rates of Dolly Varden from the two streams will be compared using a similar ANOVA as used for the analysis of energy allocation. Pairwise tests will be used to compare the proportion of energy allocated to storage between the subsidized and unsubsidized streams during the periods prior to and after winter, using growth rates as a covariate.

D. Description of Study Area

In the first year of the study, fish will be collected from Happy Valley Creek and the North Fork of the Anchor River on the Kenai Peninsula. A detailed description of the study area can be found in the companion proposal submitted by Walker, Wipfli and Stricker (Presence and Effects of Marine Derived Nutrients in Stream, Riparian and Nearshore Ecosystems on the Southern Kenai Peninsula, Alaska: Developing Monitoring Tools for GEM). The Happy Valley system is a 19 km stream that has no anadromous populations, but does have a resident population of Dolly Varden char. The Anchor River is 236 km long and also has a Dolly Varden population in addition to king, coho, and pink salmon. Salmon runs begin in May and continue through October in the Anchor River system. Thus, juvenile Dolly Varden will be exposed to carcasses throughout the growing season.

E. Coordination and Collaboration with Other Efforts

This study is a component of the larger project proposed by Walker et al. titled: Presence and Effects of Marine Derived Nutrients in Stream, Riparian and Nearshore Ecosystems on the Southern Kenai Peninsula, Alaska: Developing Monitoring Tools for Alaska. That project will provide all the onsite infrastructure and sample collection in addition to important data on trophic interactions. Samples collected will be shipped to Auke Bay Lab for analysis under this project. Consequently, data generated under this study will be correlated with data collected under the study proposed by Walker et al.. Thus, the effects of energy allocation identified under this study can be related to proxy measures of salmon escapements collected under the Walker et al. study. In addition, data collected under this study will be comparable to collections underway in Sashin Creek in southeastern Alaska.

III SCHEDULE

A. Project Milestones

Objectives I, II, III:

Sample collection will begin at the beginning of FY04 and completed early in the third quarter of FY05. Chemical analysis of the samples will be completed by the end of the fourth quarter of FY05. Statistical analysis will be completed at the end of the first quarter in FY06 and the final report will be issued immediately afterward.

B. Measurable Project Tasks

FY04

First two quarters (October 1, 2003 - March 31, 2004)

October:	Receive funding and collect initial samples
November	Develop RNA/DNA methods
January	Attend GEM workshop

Third quarter (April 1 - June 30, 2004)

May	Collect samples
June	Begin chemical analyses, collect samples

Fourth quarter (July 1 - September 30, 2004)
July Collect samples, continue chemical analyses

FY05

First two quarters (October 1, 2004 - March 31, 2005)
October: Collect samples
November Complete analyses on FY04 and October samples
January Attend GEM workshop, present 1 year's data

Third quarter (April 1 - June 30, 2005)
May Collect final samples
June Chemical analyses of last sample set

Fourth quarter (July 1 - September 30, 2005)
July Complete all chemical analyses
September Complete all statistical analyses

FY06

First quarter (October 1 to December 31, 2005)
October Produce final report

Second quarter (January 1, 2006 - April 30, 2006)
January Attend GEM workshop, present final report

IV. RESPONSIVENESS TO KEY TRUSTEE COUNCIL STRATEGIES

A. Community Involvement and Traditional Ecological Knowledge (TEK)

Community involvement includes the production of published reports and presentation of data in public forums. In addition, the work will be described to school-aged children during the SeaWeek program operated by ABL.

B. Resource Management Applications

This project is designed at providing salmon managers with a tools for fine-tuning escapement goals. If the relationships between energy acquisition and escapement of salmon into watersheds are defined, managers will have an additional criterion for setting escapement goals for systems in addition to egg deposition. These data will form the basis for understanding how many carcasses are necessary to ensure adequate nutrition of existing juveniles. Thus, future approaches to managing salmon escapement may attempt to balance the requirement for egg deposition and the nutritional benefit afforded to rearing juveniles.

V. PUBLICATIONS AND REPORTS

Proposed publications include:

Strength and persistence of a fatty acid signal reflecting marine subsidies in juvenile salmon. Transactions of the American Fisheries Society.

Effect of adult salmon carcasses on energy allocation in juvenile salmon. Journal of Fish Biology

VI. PROFESSIONAL CONFERENCES

Data are proposed to be presented at the annual meeting of the American Fisheries Society in 2005 and 2006

Literature Cited

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Heintz, RA, BD Nelson, ML Larsen, LG Holland, MS Wipfli and JP Hudson. In Press. Marine subsidies in freshwater: Effects of salmon on lipid class and fatty acid composition of juvenile coho salmon. Trans. Am. Fish. Soc. Accepted April 23, 2003.

Kline, TC, JJ Goering, OA Mathiesen, and PH Poe. 1990. Recycling of elements transported upstream by runs of Pacific salmon I: d15N and d13C evidence in Sashin Creek, southeastern Alaska. Can. J. Fish. Aquat. Sci. 47:136-144.

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Wipfli, MS, JP Hudson, JP Caouette, and DT Chaloner. In Press. Marine subsidies in freshwaters: Salmon carcasses increase the growth rates of stream-resident salmonids. Trans. Am. Fish. Soc.

Wipfli, MS, JP Hudson, DT Chaloner, et al. 1999. Influence of salmon spawner densities on stream productivity in Southeast Alaska. Can. J. Fish. Aquat. Sci. 56:1600-11.

Wipfli MS, JP Hudson, and J Caouette. 1998. Influence of salmon carcasses on stream productivity: response of biofilm and benthic macroinvertebrates in southeastern Alaska, USA. Can. J. Fish. Aquat. Sci. 55:1503-11.

Wold S. and M. Sjonstrom. 1977. SIMCA: A method for analyzing chemical data in terms of similarity and analogy, in Chemometrics: Theory and Application. BR Kowalski ed. American Chemical Society Symposium Series Number 52. American Chemical Society. Washington, DC. pp 243-282.

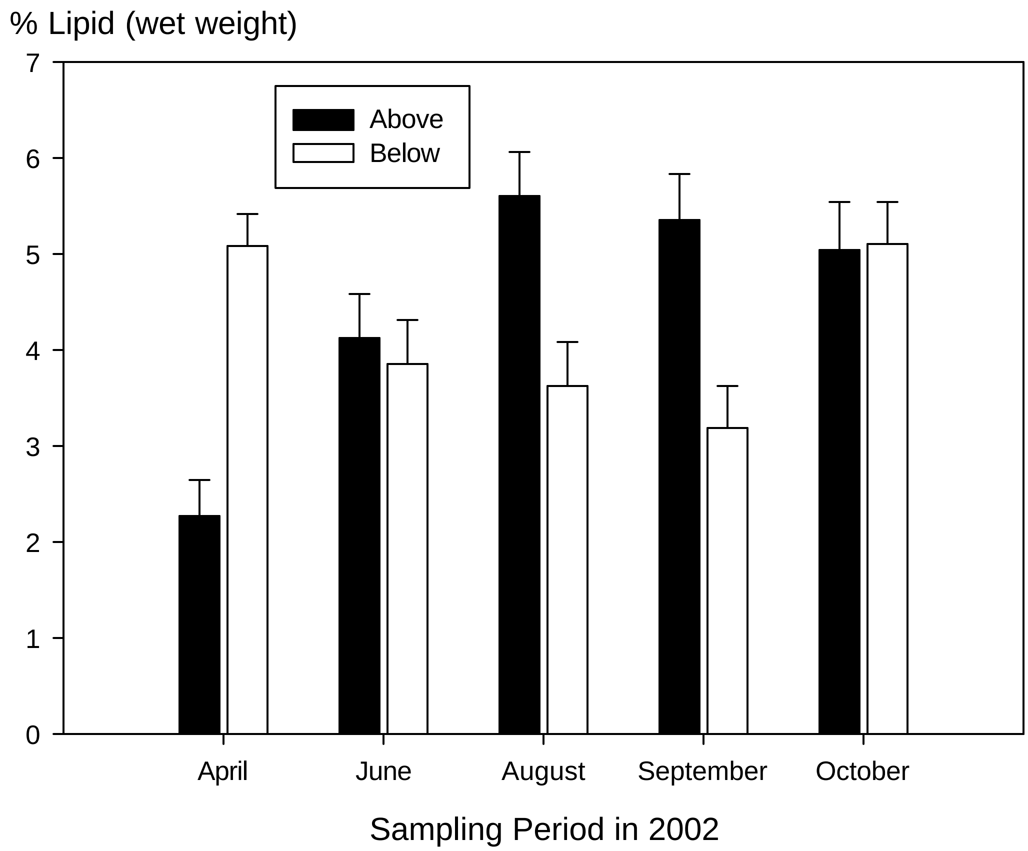

Figure 1. Seasonal changes in the mean (+/- 1 s.e.) lipid content in two populations of juvenile *Oncorhynchus mykiss* rearing in Sashin Creek, Alaska. The populations are separated by a barrier falls which prevents the juveniles rearing above the barrier from being exposed to adult pink salmon carcasses. Sampling in September coincided with the peak of the pink salmon run.

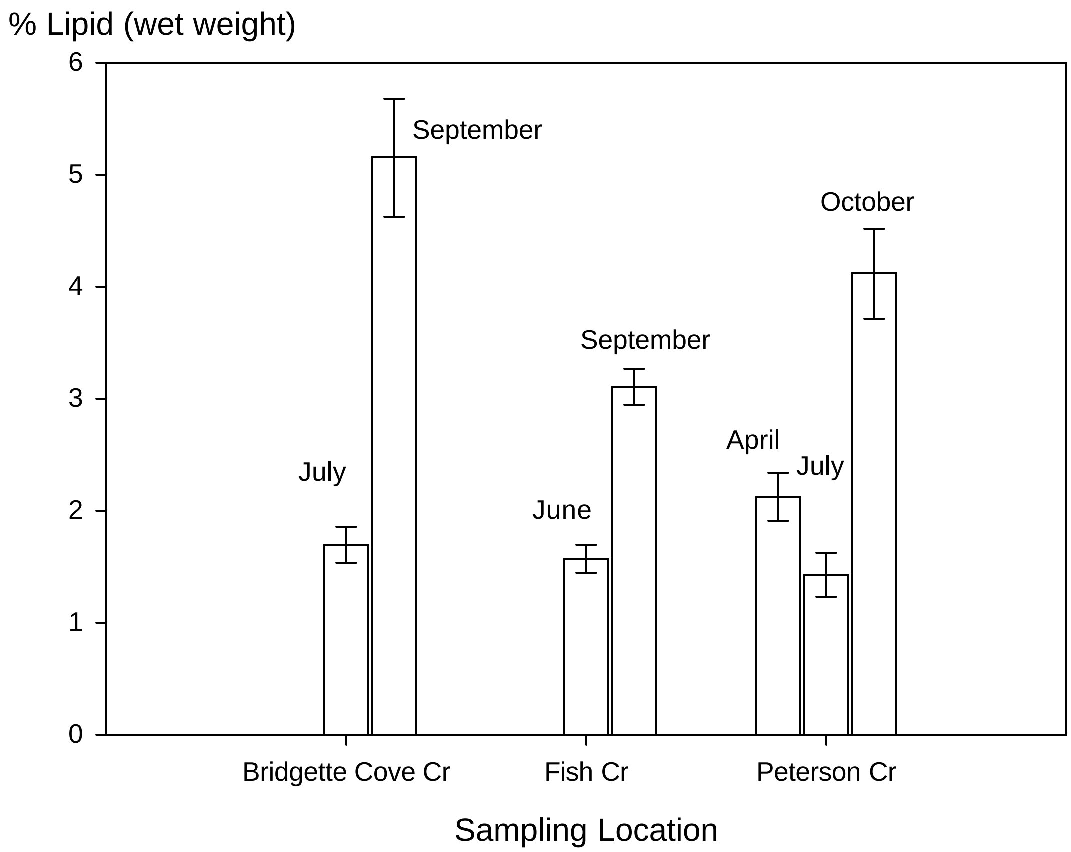

Figure 2. Seasonal variation in the mean (+/- 1 s.e.) lipid content of juvenile coho salmon rearing in streams located near Juneau, Alaska. Adult salmon return to spawn in each of these systems between August and September.

Curriculum Vitae of Ron Heintz

Contact Information

NOAA Fisheries
Auke Bay Laboratory
11305 Glacier Highway
Juneau, AK 99801

Office: (907) 789-6058
Home: (907) 789-7025
Fax: (907) 789-6094
Email: Ron.heintz@noaa.gov

Education

M.S.	*Fisheries*	University of Alaska	1987
B.A.	*Ecology*	University of Illinois Champaign	1979

Employment History

9/92-present	Fishery research biologist assigned to Habitat section of the Auke Bay Laboratory
12/85-9/92	Fishery research biologist assigned to NMFS Little Port Walter Research station

Current Responsibilities

Head of Nutritional Ecology Lab at NOAA Auke Bay Laboratory, which employs a biochemist, two biologists and 3 contract employees. Current lab projects include evaluation of the nutritional quality of Steller sea lion prey, development of models to describes Steller sea lion diets, use fatty acids to discriminate herring stocks, evaluate the role of contaminants in the decline of Steller sea lions.

Selected Publications Relevant to this Proposal

Heintz, RA, BD Nelson, ML Larsen, LG Holland, MS Wipfli and JP Hudson. In Press. Marine subsidies in freshwater: Effects of salmon on lipid class and fatty acid composition of juvenile coho salmon. Trans. Am. Fish. Soc. Accepted April 23, 2003.

Short, JP and **RA Heintz**. 1997. Identification of Exxon Valdez oil in sediments and tissues from Prince William Sound and the northwestern gulf of Alaska based on a PAH weathering model. Environ. Sci. Technol. 31:2375-2384.

Heintz, RA. 1987. The use of space and forage by Dolly Varden Char in Osprey Lake, Alaska. Masters Thesis. University of Alaska. 137 pp.

<u>Drafted Lipid Related Reports Currently in Final Preparation or Review:</u>

Gende, SM; Quinn, TP; Willson, MF; **Heintz RA**; Scott, TM. *Submitted*. Chemical composition of spawning salmon (*Oncorhynchus* ssp) with consequences for selective comsumption by bears. Submitted to Can. Jour. Zool.

Wipfli, MS; Hudson, JP; Caouette JP; **Heintz, RA**; Chaloner, DT; Larsen, ML; Holland LG. *Submitted*. Marine Subsidies in Fresh Water: Salmon Carcasses Increase Growth and Lipids of Stream Salmonids. Submitted to Trans. Am. Fish. Soc.

Heintz, RA; Vollenweider, JJ.. Estimation of Northern Fur Seal Diets Using Fatty Acid Analysis. *Submitted to NMML for review*

Otis, EO, **RA Heintz**, and KP Severin. Discriminating among Alaska's Pacific herring stocks using heart fatty acid profiles and otolith microchemistry. Trans. Am. Fish. Soc.

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 04 - FY 06

Budget Category:	Proposed FY 04	Proposed FY 05	Proposed FY 06	TOTAL PROPOSED
Personnel	$21,000.0	$21,000.0	$10,500.0	$52,500.0
Travel	$2,385.0	$2,460.0	$2,460.0	$7,305.0
Contractual	$10,000.0	$10,000.0	$0.0	$20,000.0
Commodities	$10,998.4	$5,392.0	$0.0	$16,390.4
Equipment	$0.0	$0.0	$0.0	$0.0
Subtotal	$44,383.4	$38,852.0	$12,960.0	$96,195.4
General Administration (9% of Subtotal)	$3,994.5	$3,496.7	$1,166.4	$8,657.6
Project Total	$48,377.9	$42,348.7	$14,126.4	$104,853.0

Cost-share Funds:
In this box, identify non-EVOS funds or in-kind contributions used as cost-share for the work in this proposal. List the amount of funds, the source of funds, and the purpose for which the funds will be used. Do not include funds that are not directly and specifically related to the work being proposed in this proposal.

NMFS/ABL will provide Bradshaw
1.5 months labor for proximate analysis (GS11/4) $10.8
k
Heintz
6.0 months labor for data analysis, report writing and project management (GS12/6) $ 48.6 k
3 months maintenance costs on all analytical instruments $ 1.5
k

FY 04-06

Date Prepared:

Project Number:
Project Title: influence of carcasses on energy allocation
Agency: NMFS

FORM 3A TRUSTEE AGENCY SUMMARY

EXXON VALDEZ OIL SPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 04 - FY 06

Personnel Costs:		GS/Range/	Months	Monthly		Personnel
Name	Description	Step	Budgeted	Costs	Overtime	Sum
Lawrence Schaufler	Biochemist	GS/12/1	3.0	7000.0		21,000.0
hplc/gcmsd operation and lab quality control						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
	Subtotal		3.0	7000.0	0.0	
					Personnel Total	$21,000.0

Travel Costs:	Ticket	Round	Total	Daily	Travel
Description	Price	Trips	Days	Per Diem	Sum
Annual Meeting of EVOS	450.0	1	4	165.0	1,110.0
					0.0
travel to homer to oversee site and sample collection	450.0	1	5	165.0	1,275.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
				Travel Total	$2,385.0

FY 04

Project Number:
Project Title: influence of carcasses on energy allocation
Agency: NMFS

FORM 3B
Personnel
& Travel
DETAIL

EXXON VALDEZ OIL SPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 04 - FY 06

Contractual Costs:		Contract Sum
Description		
Contract labor for preparing and esterifying samples		10,000.0
If a component of the project will be performed under contract, the 4A and 4B forms are required.	**Contractual Total**	$10,000.0

Commodities Costs:		Commodity Sum
Description		
sample vials/weighing pans/gases/solvents/ reagents/misc. supplies for sample prep., extraction and esterification, fatty acid and lipid class analysis	$10.27/sample x 250 sample	2,567.5
sample cups/solvents/reagents/gases/crucibles for proximate analysis	$11.17/sample x 250 sample	2,792.5
Liquid nitrogen and cylinder rental		1,500.0
dewar shipping		500.0
reagents and vials for RNA/DNA anlaysis	15.16/sample x 240 sample	3,638.4
	Commodities Total	$10,998.4

FY 04

Project Number:
Project Title: influence of carcasses on energy allocation
Agency: NMFS

FORM 3B
Contractual
&
Commoditie

EXXON VALDEZ OIL SPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 04 - FY 06

New Equipment Purchases: Description	Number of Units	Unit Price	Equipment Sum
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
		New Equipment Total	$0.0

Existing Equipment Usage: Description	Number of Units	Inventory Agency
Dionex Accelreated Solvent Extractor	1	1
Varian GC/MSD	1	
Hewlett Packard HPLC/ELSD	1	1
Leco Nitrogen Analyzer	1	1
Leco Thermogravimetric Analayzer	1	1

FY 04

Project Number:
Project Title: influence of carcasses on energy allocation
Agency:

FORM 3B
Equipment
DETAIL

EXXON VALDEZ **OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 04 - FY 06

Personnel Costs:		GS/Range/	Months	Monthly		Personnel
Name	Description	Step	Budgeted	Costs	Overtime	Sum
Lawrence Schaufler	Biochemist	GS/12/1	3.0	7000.0		21,000.0
hplc/gcmsd operation and lab quality control						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
	Subtotal		3.0	7000.0	0.0	
					Personnel Total	$21,000.0

Travel Costs:	Ticket	Round	Total	Daily	Travel
Description	Price	Trips	Days	Per Diem	Sum
Annual Meeting of EVOS	450.0	1	4	165.0	1,110.0
					0.0
Travelto meeting to present data	750.0	1	4	150.0	1,350.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
				Travel Total	$2,460.0

FY 05

Project Number:
Project Title: influence of carcasses on energy allocation
Agency: NMFS

FORM 3B
Personnel
& Travel
DETAIL

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 04 - FY 06

Contractual Costs:		Contract
Description		Sum
Contract labor for preparing and esterifying samples		10,000.0
If a component of the project will be performed under contract, the 4A and 4B forms are required.	**Contractual Total**	$10,000.0

Commodities Costs:		Commodity
Description		Sum
sample vials/weighing pans/gases/solvents/ reagents/misc. supplies for sample prep., extraction and esterification, fatty acid and lipid class analysis	$10.27/sample x 120 sample	1,232.4
sample cups/solvents/reagents/gases/crucibles for proximate analysis	$11.17/sample x 120 sample	1,340.4
Liquid nitrogen and cylinder rental		750.0
dewar shipping		250.0
	$15.16/sample x 120 sample	1,819.2
	Commodities Total	$5,392.0

FY 05

Project Number:
Project Title: influence of carcasses on energy allocation
Agency: NMFS

FORM 3B
Contractual
&
Commoditie

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 04 - FY 06

New Equipment Purchases: Description	Number of Units	Unit Price	Equipment Sum
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
		New Equipment Total	$0.0

Existing Equipment Usage: Description	Number of Units	Inventory Agency
Dionex Accelreated Solvent Extractor	1	1
Varian GC/MSD	1	
Hewlett Packard HPLC/ELSD	1	1
Leco Nitrogen Analyzer	1	1
Leco Thermogravimetric Analayzer	1	1

FY 05

Project Number:
Project Title: influence of carcasses on energy allocation
Agency: NMFS

FORM 3B
Equipment
DETAIL

EXXON VALDEZ OIL SPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 04 - FY 06

Personnel Costs:		GS/Range/	Months	Monthly		Personnel
Name	Description	Step	Budgeted	Costs	Overtime	Sum
Lawrence Schaufler	Biochemist	GS/12/1	1.5	7000.0		10,500.0
						0.0
Assist in report preparation						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
	Subtotal		1.5	7000.0	0.0	
					Personnel Total	$10,500.0

Travel Costs:	Ticket	Round	Total	Daily	Travel
Description	Price	Trips	Days	Per Diem	Sum
Annual Meeting of EVOS	450.0	1	4	165.0	1,110.0
					0.0
Travelto meeting to present data	750.0	1	4	150.0	1,350.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
				Travel Total	$2,460.0

FY 06

Project Number:
Project Title: influence of carcasses on energy allocation
Agency: NMFS

FORM 3B
Personnel
& Travel
DETAIL

Contractual Costs:	Contract
Description	Sum
Contractual Total	$0.0

Commodities Costs:	Commodity
Description	Sum
Commodities Total	$0.0

FY 06

Project Number:
Project Title: influence of carcasses on energy allocation
Agency: NMFS

FORM 3B
Contractual
&
Commoditie

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 04 - FY 06

New Equipment Purchases: Description	Number of Units	Unit Price	Equipment Sum
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
		New Equipment Total	$0.0

Existing Equipment Usage: Description	Number of Units	Inventory Agency

FY 06

Project Number:
Project Title: influence of carcasses on energy allocation
Agency: NMFS

FORM 3B
Equipment
DETAIL

Budget Justification

EVOS Funding Requested:	FY 04	$ 48.4	
	FY 05	$ 42.3	
	FY 06	$ 14.1	TOTAL:104.8
Non-EVOS Funds to be Used:	FY 04	$15.3	
	FY 05	$27.6	
	FY 06	$16.5	TOTAL:59.4

Costs for the FY04 and FY05 reflect costs associated with sample collection and analysis. Costs associated with FY06 are for closing out the project. Requested labor costs are for Schaufler who runs our lipid laboratory, but is not supported by agency funds. Labor costs for permanent NMFS personnel will be provided by the agency, these include Bradshaw who performs our proximate analyses and Heintz who will manage the project at ABL, perform statistical analyses and lead the report writing. Costs in FY04 include 1 month of Bradshaw for sample analysis and 1 month of Heintz for management. In-kind costs in FY05 include 0.5 months of Bradshaw and 3.0 months of Heintz. FY05 includes 2.0 months of Heintz. Costs in FY04 and FY05 for contracted labor are for homogenizing and esterifying samples prior to analysis. Travel costs in the FY04 include transportation to Homer to inspect the sampling locations and instruct staff at the Kachemak Bay Research Reserve on sample collection and handling procedures. Travel costs in outlying years are associated with the annual GEM workshop and presentation of data at professional meetings. Shipping costs are for transporting dewars of liquid nitrogen between Homer, Juneau and the supplier. Commodity costs are for the materials and reagents required for sample analysis.