

The importance of dietary phosphorus and highly unsaturated fatty acids for sockeye (*Oncorhynchus nerka*) growth in Lake Washington — a bioenergetics approach

Ashley P. Ballantyne, Michael T. Brett, and Daniel E. Schindler

Abstract: Juvenile sockeye salmon (*Oncorhynchus nerka*) in Lake Washington experience very high growth rates, but the mechanisms regulating their growth rates are poorly understood. The potential for food quality limitation of growth was tested using a coupled bioenergetics and mass balance model. Sockeye and zooplankton prey species were analyzed for their fatty acid composition. Ratios of phosphorus to carbon (P:C) for both sockeye and zooplankton were obtained from the literature. Based on sockeye stomach contents and zooplankton weights, diets of zooplankton biomass were calculated and monthly values of specific fatty acids and P:C ratios in the diet were derived. An increase in highly unsaturated fatty acids, especially in the liver, was observed with increased fork length up to smoltification size, with a subsequent decrease in smolts and kokanee. Measurements of docosahexaenoic (DHA) in daphnids were significantly lower than in cyclopoids. Model predictions suggest that sockeye are not limited by the availability of phosphorus or eicosapentaenoic acid (EPA) in their diet but may experience limited growth because of DHA deficiencies. Thus, the ability and efficiency of sockeye at elongating 18:3 ω 3, 18:4 ω 3, and EPA to DHA may be of critical importance to lacustrine growth and production.

Résumé : Les jeunes saumons rouges (*Oncorhynchus nerka*) du lac Washington ont des taux de croissance très élevés, mais les mécanismes qui contrôlent ces taux restent mal connus. Nous avons évalué la possibilité d'une limitation de la croissance par la qualité de la nourriture en couplant une étude énergétique et un modèle de bilan de masse. La composition en acides gras a été déterminée chez les saumons et leurs proies dans le zooplancton. Les rapports phosphore:carbone (P:C) du saumon et du zooplancton ont été tirés de la littérature. L'étude des contenus stomacaux des saumons et de la masse du zooplancton nous a permis de calculer la biomasse du zooplancton dans le régime alimentaire et d'obtenir les mesures mensuelles des acides gras spécifiques et du rapport P:C dans le régime. On observe une augmentation des acides gras hautement insaturés, particulièrement dans le foie, à mesure de l'accroissement de la longueur à la fourche jusqu'à la taille de la transformation en saumoneaux; ces acides gras diminuent ensuite chez les saumoneaux et les kokanis. Les concentrations d'acide docosahexanoïque (DHA) sont significativement plus basses chez les daphnies que chez les cyclopoïdes. Les prédictions du modèle font croire que les saumons rouges ne sont pas limités par la disponibilité du phosphore ou de l'acide eicosapentanoïque (EPA) dans leur régime, mais que leur croissance peut être restreinte par manque de DHA. Ainsi, la capacité et l'efficacité du saumon rouge à allonger les acides gras 18:3 ω 3, 18:4 ω 3 et l'EPA en DHA peuvent être des facteurs déterminants de la croissance et de la production dans le lac.

[Traduit par la Rédaction]

Introduction

The constraints imposed on the growth of organisms by the availability of resources have long been the focus of study in ecology (Tilman 1982). Researchers investigating factors governing consumer growth (Gulati and DeMott 1997) have re-

vised this focus of research in the aquatic sciences. However, almost all research in ecology that has been conducted on food quality has focused on the phytoplankton–zooplankton interface. Few studies have attempted to apply principles describing lower-trophic level processes to subsequent higher-trophic level consumers, such as fish (Nakashima and Leggett

Received 8 August 2002. Accepted 30 September 2002. Published on the NRC Research Press Web site at <http://cjfas.nrc.ca> on 4 February 2003.
J17036

A.P. Ballantyne^{1,2} and **M.T. Brett**. Department of Civil and Environmental Engineering, University of Washington, Seattle, WA 98195, U.S.A.

D.E. Schindler. Department of Zoology, University of Washington, Seattle, WA 98195, U.S.A.

¹Corresponding author (e-mail: apb14@duke.edu).

²Present address: Division of Earth and Ocean Sciences, Duke University, Durham, NC 27708-0229, U.S.A.

1980; Kraft 1992). Results from bioenergetics analyses of several freshwater fish populations feeding on a variety of prey revealed that all fish considered grew below optimal levels, but more interestingly, their growth was never predicted to be limited by nitrogen (N) and seldom limited by phosphorus (P) (Schindler and Eby 1997). A study of fatty acid composition among various fish species from lakes of differing trophic state found higher ω 3 fatty acid content in fish sampled from relatively oligotrophic lakes with phytoplankton assemblages dominated by diatoms rich in ω 3 fatty acids (Ahlgren et al. 1996). However, the potential for growth limitation because of essential fatty acid content has never been explored using bioenergetics models.

There is a wealth of knowledge on nutritional physiology of fish with respect to fatty acids in the aquaculture literature. For example, Yu and Sinnhuber (1979) fed coho salmon (*Oncorhynchus kisutch*) artificial feeds with varying fatty acid content and found that the optimal dietary ω 3 fatty acid content for maximum growth was between 1 and 2.5% dry weight. Coho consuming diets containing a percentage of ω 3 fatty acids beyond this range experienced reduced growth. Higher growth rates have been observed in rainbow trout (*Oncorhynchus mykiss*) and chum salmon (*Oncorhynchus keta*) when supplied with diets rich in the long-chained ω 3 fatty acids eicosapentaenoic acid (20:5 ω 3 or EPA) and docosahexaenoic acid (22:6 ω 3 or DHA) (Takeguchi and Watanabe 1982). EPA and DHA, collectively referred to as highly unsaturated fatty acids (HUFA), are important nutrients for salmonid growth. Physiological studies have revealed that ω 3 fatty acids are required for proper development of eye and brain tissue within developing rainbow trout (Bell and Tocher 1989). However, few studies have examined wild salmonid populations feeding on natural diets.

Sockeye salmon (*Oncorhynchus nerka*) were selected as the subject of this study for several reasons. First, they have high concentrations of ω 3 fatty acids (Higgs et al. 1995) and it has been suggested that they may in some systems experience phosphorus-limited growth (Schindler and Eby 1997). Second, they have evolved a unique "adfluvial" life history, whereby young fry remain in nursery lakes to develop for 1–2 years before seaward migration, which greatly facilitates studies of their early development. However, the growth rates and residence times in these lakes vary tremendously; individuals that grow quickly leave the lake after only 1 year, whereas others grow more slowly and remain in the lake for 2 years (Weatherley and Gill 1995). Finally, according to Eggers (1982), sockeye in Lake Washington are obligate planktivores and feed almost exclusively on zooplankton throughout their lacustrine development, which greatly simplifies the analysis of prey items in their diet.

Extensive research has been conducted on the sockeye populations of Lake Washington (Woodey 1972; Beauchamp et al. 1989). Sockeye were first introduced to the Lake Washington system in 1935 and have proliferated since the 1960s (Edmondson 1994). Lake Washington is renowned for producing large sockeye smolts relative to other nursery lakes and the early departure of these smolts after only 1 year (Eggers 1978). The explanation provided by Eggers (1978) is that "the large size achieved by Lake Washington sockeye is due to a long growing season as well as a high rate of

growth". Although this describes the proximate mechanisms, it fails to describe the ultimate nutritional and physiological factors that may enhance or restrict sockeye growth in Lake Washington.

The purpose of this research is to test the hypothesis that sockeye salmon in Lake Washington experience growth limitation because of content of phosphorus or specific essential fatty acids in their diet. A permutation of the basic mass balance model as proposed by Nakashima and Leggett (1980) was coupled with a bioenergetics model for sockeye (Beauchamp et al. 1989) to explore the possibility that sockeye experience limited growth resulting from dietary constraints. This analysis will assess the degree to which food quality, in terms of phosphorus, EPA, and DHA, limit sockeye growth in Lake Washington.

Materials and methods

Field sampling

Samples for both zooplankton and sockeye were collected concurrently from Lake Washington throughout the April 1999 to September 1999 growth season. Zooplankton were sampled from 0- to 20-m depth using a 250- μ m zooplankton net. Zooplankton taxa were identified to genus and enumerated. Lake Washington has five *Daphnia* species (*D. ambigua*, *D. galeata*, *D. thorata*, *D. schodleri*, and *D. pulicaria*), of which *D. pulicaria* is the most abundant. *Daphnia* spp. were only distinguished to genus as they vary little in size and have similar feeding behaviors and elemental composition (Hessen and Lyche 1991; Brett et al. 2000). *Daphnia* spp. were, however, separated based on the presence or absence of eggs and were analyzed separately. The other prey taxa collected were *Cyclops* spp., which consisted of two species (*C. bicuspidatus* and *C. vernalis*) with the former being the dominant, as well as *Epischura nevadensis* and *Diaptomus ashlandi*. Other prey taxa such as the pelagic *Bosmina longirostris* and the benthic *Neomysis mercedis* were present at such low levels in zooplankton tows that they probably constitute a small portion of the sockeye diet in Lake Washington. Furthermore, of 688 juvenile sockeye salmon stomach samples collected, only three contained *Neomysis mercedis*, thus *Neomysis mercedis* constitute less than 1% of the sockeye diet (Jennifer Anson, Department of Zoology, University of Washington, Seattle, WA 98195, U.S.A., unpublished data). Eggers (1982) identified six zooplankton taxa in the diet of Lake Washington sockeye: *Cyclops*, *Diaptomus*, *Bosmina*, *Diaphanosoma*, *Epischura*, and *Daphnia*. Ivlev electivity indices

$$E_i = (r_i - p_i)/(r_i + p_i)$$

were calculated based on Eggers' (1982) data, where r_i is the percent of prey in the diet and p_i is the percent of prey in the environment. The proliferation of *Daphnia* in Lake Washington, which occurred in 1976 (Edmondson 1994), coincided with a decline in *Diaphanosoma*. Thus, *Diaphanosoma* is probably less important in the diet of sockeye today and was excluded from further analysis. Subsequently, *Daphnia* constitute an even greater proportion of the sockeye diet today as inferred from stomach content data. Sockeye were collected at night using a beach seine. Fork length was measured and flesh samples were taken from dorsal muscle and liver tissue.

All samples were freeze-dried and weighed, and an internal standard of 21:0 was added, as this fatty acid is not commonly found in nature. They were then sonicated, extracted three consecutive times, and methylated (Kattner and Fricke 1986) before injection into a gas chromatograph (HP6890) equipped with a programmable temperature vaporizer-injector (PTV) for measuring samples of low concentration. A fused silica DB-WAX capillary column, manufactured by J&W Scientific (Folsom, Calif.), with a length of 30 m, a diameter of 320 μm , and a thickness of 0.25 μm was installed. Injected samples were subject to the following oven temperature regime: 5 min at 40°C, then heated to 150°C at a rate of 10°C/min, then kept at 150°C for 5 min, further heated to 220°C at a rate of 2°C/min, and finally kept at 220°C for 20 min. Sample peaks were detected using a flame ionization detector. Retention times were verified with a quantitative mixture of fatty acids (No. 625024, Alltech, Australia) and area curves compared between standards and samples for 18:3 ω 3, EPA, and DHA showed less than 5% variability.

An analysis of variance (ANOVA) was conducted on fatty acid concentrations among the prey genera followed by a Tukey test to identify significant differences between prey types.

Bioenergetics modeling

The basic mass balance model as it applies to food quality includes parameters describing growth (G_Q), consumption (C_Q), egestion (E_Q), and excretion (U_Q), where Q represents the food quality parameter (i.e., phosphorus or fatty acids) and is expressed accordingly:

$$(1) \quad G_Q = C_Q - E_Q - U_Q$$

However, as fatty acids can be readily converted to triacylglycerols and stored as a major fuel source (Stryer 1981), they are probably not excreted but rather metabolized. Thus, essential fatty acids are first incorporated into growing tissues and then, if supplied in excess, are utilized as an energy source. The mass of fatty acids that can be metabolized (M_Q) in fish can then be written as

$$(2) \quad M_Q = C_Q - G_Q - E_Q$$

The mass of consumed fatty acids or phosphorus that are assimilated can be written as $C_Q \cdot \text{AE}_Q$, where AE represents the assimilation efficiency. The assimilation efficiency of 0.93 (Bell and Sargent 1992) for polyunsaturated fatty acids in a similar salmonid (*O. keta*) was used as an estimate of AE for sockeye. The average assimilation efficiency for phosphorus of 0.74 (± 0.06) (Nakashima and Leggett 1980) was used because it represents the potentially limiting element to sockeye growth, based on the stoichiometric relationship between carbon and phosphorus in their diet. Thus, eq. 2 can be rewritten as

$$(3) \quad M_Q = (\text{AE}_Q \cdot C_Q) - G_Q$$

Therefore by solving eq. 3 for M_Q we can calculate the amount of surplus fatty acids that may be allocated to energy stores and predict whether growth limitation is experienced when feeding on an actual diet of varying prey types. Likewise, eq. 3 can be evaluated for the possibility of growth limitation resulting from the relative availability of phospho-

rus to carbon (P:C). Thus, a fish with a high P:C ratio could experience phosphorus-limited growth on a diet with a relatively low P:C ratio. This suggests that as the term for food that can be metabolized (M_Q) approaches zero with increased specific growth rate, surplus fatty acids or phosphorus are reduced, resulting in growth limitation. The main assumption of this model is the conservation of mass, meaning that fatty acids and phosphorus do not change following consumption. Although ω 3 fatty acids cannot be synthesized by fish de novo and are thus considered to be essential, there is evidence that certain species can elongate 18 carbon chain ω 3 fatty acids after consumption (Buzzi et al. 1996; Ruyter et al. 2000a). However, the actual rates and metabolic costs of elongation have not yet been measured in salmonids.

Because of the allometric (Peters 1983) and thermal scaling of metabolism (Weatherley and Gill 1995), the mass balance model (eq. 3) must be coupled to a bioenergetics model to evaluate changes in consumption and metabolism at various growth rates and thermal regimes. The original bioenergetics model refined by Hewett and Johnson (1991) and later used in concert with mass balance equations (Kraft 1992) is as follows:

$$(4) \quad \text{Consump} = \text{Growth} + \text{Resp}_{\text{standard}} + \text{Resp}_{\text{activity}} \\ + \text{Resp}_{\text{digestion}} + \text{Egestion}_{\text{fecal}} + \text{Excretion}_{\text{urine}}$$

This bioenergetics model can be distinguished from the original mass balance equation (eq. 1) by the presence of the respiration terms for standard, activity, and digestion, which are strongly influenced by temperature. The other important distinction is that this model functions in units of energy (J) as opposed to the mass balance equations (eqs. 1–3), which function in units of mass ($\text{g} \cdot \text{g wet weight (wet wt.)}^{-1}$). Specific equations and parameter values for the sockeye bioenergetics model can be found in Table 1.

Energy densities ($\text{J} \cdot \text{g wet wt.}^{-1}$) for all prey species and sockeye were also required as inputs to the model, along with measurements of fatty acid content and P:C ratios for each of the prey species (Table 2). Stomach contents for Lake Washington sockeye were collected during the day and night at monthly intervals for the period 1994–1998 (Jennifer Anson, Department of Zoology, University of Washington, Seattle, WA 98195, U.S.A., unpublished data). Stomach contents were identified to genus and enumerated. From these data, monthly averages of zooplankton consumed were converted to biomass, based on zooplankton weights averaged over the sockeye growing season. Finally, monthly weighted averages for P:C ratios, EPA, DHA, and energy density were calculated and input to the model. It is also important to mention that the model, through a series of algorithms, fits a growth curve based on the energy density of the prey. The trajectory of this growth curve is reflected in P_{max} , or the proportion of maximum consumption, based on temperature and the initial and final wet weights of the fish. Thus, by maximizing consumption (i.e., $P_{\text{max}} = 1.0$), maximum growth rates were predicted for comparison with observed growth rates. Moreover, all dietary parameter inputs to the model are in terms of wet weight because measurements of fish biomass are in wet weight.

Data on sockeye growth in Lake Washington were obtained from Woodey (1972) and these data were corroborated by

Table 1. Equations and parameter values for the bioenergetics model obtained from Beauchamp et al. (1989) and references therein.

Equations and parameters	Description and units	Values
Consumption (C)		
$C = C_{\max} P_{\max} f(T)$		
$f(T) = K_A - K_B$		
where		
$K_A = (CK1 \cdot L1) / (1 + CK1(L1 - 1))$		
$L1 = e^{(G1(T - CQ))}$		
$G1 = (1 / (CTO - CQ)) \cdot \ln((0.98(1 - CK1)) / (CK1 \cdot 0.02))$		
$K_B = (CK4 \cdot L2) / (1 + CK4(L2 - 1))$		
$L2 = e^{(G2(CTL - T))}$		
$G2 = (1 / (CTL - CTO)) \cdot \ln((0.98(1 - CK4)) / (CK4 \cdot 0.02))$		
CA	Intercept: mass dependence of maximum consumption (C_{\max}) ($\text{g} \cdot \text{g}^{-1} \cdot \text{day}^{-1}$)	0.303
CB	Coefficient: mass dependence of C_{\max}	-0.275
CQ	Temperature for CK1 ($^{\circ}\text{C}$)	3
CTO	Optimum temperature for C_{\max} ($^{\circ}\text{C}$)	20
CTL	Temperature for CK4 ($^{\circ}\text{C}$)	24
CK1	Proportion of C_{\max} at temperature CQ	0.58
CK4	Proportion of C_{\max} at temperature CTL	0.5
Respiration (R)		
$R = RA \cdot W^{RB*} \cdot f(T) \cdot \text{ACT}$		
$f(T) = e^{(RQ \cdot T)}$ and $\text{ACT} = e^{(RTO \cdot \text{VEL})}$		
$\text{VEL} = W^{RK4}$		
$S = \text{SDA}(C - E)$		
RA	Intercept: R ($\text{g O}_2 \cdot \text{day}^{-1}$)	0.00143
RB	Coefficient: mass dependence of R	-0.209
RQ	Coefficient: R with respect to temperature	0.086
RTO	Coefficient: R with respect to swimming speed (U)	0.0234
ACT	Intercept: U ($\text{cm} \cdot \text{s}^{-1}$)	9.9
RK4	Coefficient: mass dependence of U	0.13
BACT	Coefficient: U with respect to temperature	0.0405
SDA	Specific dynamic action	0.172
S	Proportion of energy lost to SDA	
W	Fish mass (g)	Monthly values (see Fig. 1)
Egestion (E)		
$E = \text{PF} \cdot C$		
$\text{PF} = ((\text{PE} - 0.1) / 0.9)$		
$\text{PE} = \text{EA} \cdot T^{\text{EB}} \cdot e^{\text{EG}}$		
EA	Intercept: proportion egested	0.212
EB	Coefficient: E with respect to temperature	-0.222
EG	Coefficient: proportion of maximum consumption (P_{\max}) with respect to E	0.631
Excretion (U)		
$U = \text{UA} \cdot T^{\text{UB}} \cdot e^{\text{UG}}(C - E)$		
UA	Intercept: proportion excreted	0.0314
UB	Coefficient: U with respect to temperature	0.58
UG	Coefficient: P_{\max} with respect to U	-0.299

more recent observations (T. Quinn, School of Aquatic and Fishery Sciences, University of Washington, Seattle, WA 98195, U.S.A., unpublished data) (Fig. 1). Initial and final weights for each month were then used as inputs to the model. Specific growth rates (g) were also calculated for each month interval according to eq. 5 for comparison with predicted growth rates:

$$(5) \quad g = [\ln(W_f) - \ln(W_i)] / t$$

where W_i represents the initial weight, W_f the final weight, and t is the length of each month in days.

Temperature regimes were obtained for 20-m depth (D.E. Schindler, Department of Zoology, University of Washington, Seattle, WA 98195, U.S.A., unpublished data) and input to the model. The depth of 20 m was decided on as an intermediate depth between sockeye seasonal distribution estimates by Beauchamp et al. (1999) of 15–18 m and Eggers (1978) of 20–40 m. In the winter months, sockeye tend to aggregate in the shallower littoral zones of the lake; however, during this time of year Lake Washington is isothermal and there is little difference between surface temperatures and temperatures at 20 m. Thus, the annual temperatures at 20 m are reasonably representative of temperatures experi-

Table 2. Inputs to the bioenergetics and mass balance models.

	Fatty acids ^a (\pm SD)		Bioenergetics parameters	
	EPA content (mg·g wet wt. ⁻¹)	DHA content (mg·g wet wt. ⁻¹)	P:C (by weight)	Energy density (J·g wet wt. ⁻¹)
<i>Daphnia</i>	0.70 (\pm 0.04)	0.02 (\pm 0.03)	0.018 ^b	2281 ^c
<i>Daphnia</i> with eggs	0.84 (\pm 0.31)	0.21 (\pm 0.31)	—	2281 ^c
<i>Epischura</i>	1.04 (\pm 0.24)	1.40 (\pm 0.27)	0.023 ^d	3004 ^c
<i>Cyclops</i>	0.60 (\pm 0.70)	3.05 (\pm 2.87)	0.013 ^e	3028 ^c
<i>Diaptomus</i>	1.08 (\pm 0.97)	0.63 (\pm 0.48)	0.007 ^e	3075 ^c
<i>Oncorhynchus nerka</i> (liver)	3.67 (\pm 1.76)	5.45 (\pm 2.20)	—	= 5.234 + 7.75W ^f
<i>Oncorhynchus nerka</i> (dorsal)	1.22 (\pm 0.21)	3.20 (\pm 0.83)	0.024 ^g	= 5.234 + 7.75W ^f

Note: Values of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are reported as wet weight and standard deviations (SD). P:C = ratio of phosphorus to carbon; wet wt. = wet weight.

^aAll fatty acid values obtained in present study.

^bBrett et al. (2000).

^cEnergy densities are averages from representative genera from Cummins and Wuycheck (1971).

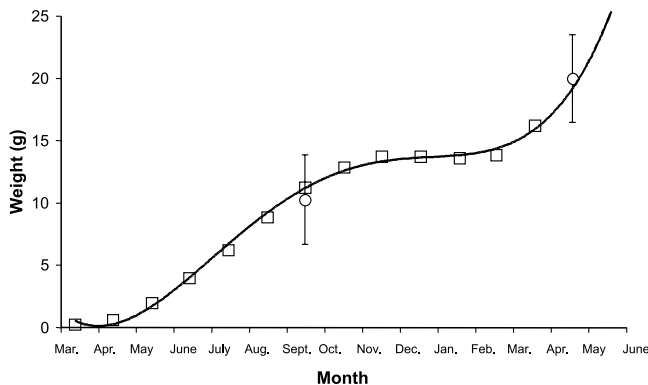
^dElser et al. (2000).

^eHessen and Lyche (1991), Fig. 1.

^fEquation for sockeye of ≤ 196 g wet weight, where W represents wet weight (Beauchamp et al. 1989).

^gValue calculated from 0.3648% P, wet weight (Donaldson 1967), and 14.69% C, wet weight (present study).

Fig. 1. Historical weight data from Woodey (1972) for sockeye (*Oncorhynchus nerka*) in Lake Washington compared with more recent data. The data were fit using a 4th-order polynomial equation.



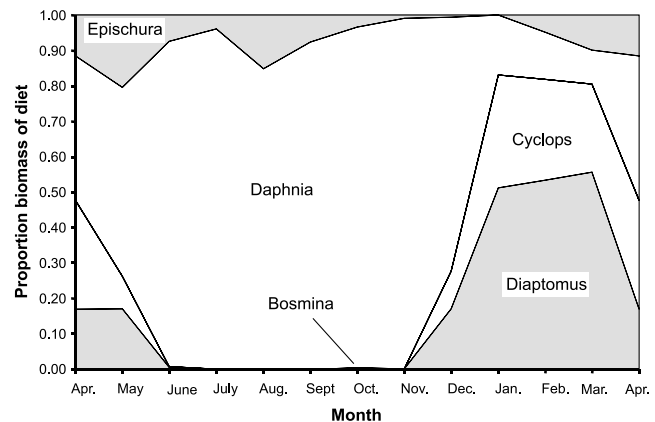
enced by sockeye throughout their lacustrine growth. A fourth-order polynomial was fit to the temperature data at 20-m depth to predict daily temperatures for input to the model. Simulations were also run at optimal temperature (15°C), which is the temperature of maximum growth, provided a maximum ration of food (Brett et al. 1969) and optimal lake temperatures ($\leq 15^\circ\text{C}$) for comparison. Model simulations were run for the 13-month lacustrine growth period from March to the following May, and the potential for growth limitation resulting from dietary availability of the fatty acids EPA and DHA as well as phosphorus was evaluated.

Results

Field sampling

By comparing the biomass estimates of prey taxa in the sockeye diet (Fig. 2) with the Ivlev electivity indices (Fig. 3), it is apparent that although *Epischura* may be selected for, their contribution in terms of biomass to the total diet is comparatively small. Furthermore, the copepods *Cyclops* and *Diaptomus* constitute a large portion of the diet biomass, especially in the winter months, despite their negative

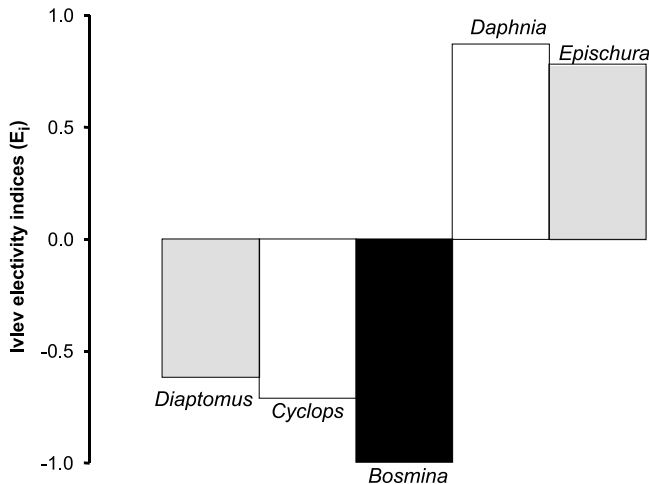
Fig. 2. Composition of sockeye (*Oncorhynchus nerka*) diet in Lake Washington. Based on stomach content data that were transformed into monthly averages and multiplied by the following individual weights (\pm standard deviation) in μg to obtain percent biomass of each prey genus in the diet: *Epischura*, 36.6 (± 10.10); *Daphnia*, 35.3 (± 12.80); *Bosmina*, 1.0 (± 1.56); *Cyclops*, 4.8 (± 0.04); *Diaptomus*, 4.7 (± 1.33).



electivity indices. More importantly, however, *Daphnia* constitute approximately 90% of the biomass in the diet of Lake Washington sockeye throughout their growing season (June–November).

There was very little variability in the amount of α -linolenic acid (ALA, 18:3 ω 3) among all prey types (Fig. 4a) and the EPA content of all prey types measured was also very similar (Fig. 4b). The highest variability in measurements of EPA was observed in *Diaptomus* and *Cyclops*. The highest total ω 3 content was observed in *Cyclops* (Fig. 4d). According to the ANOVA, there were no significant differences in EPA, ALA, or total ω 3 contents among the different prey types. Zooplankton DHA content was more variable among prey types compared with the other fatty acids measured, with the highest intrageneric variability observed within *Cyclops* (Fig. 4c). High variability of DHA concentrations in *Cyclops* was probably the result of a small sample mass

Fig. 3. Ivlev electivity indices calculated for the five dominant genera in Lake Washington sockeye (*Oncorhynchus nerka*) diet. Positive indices suggest that a prey item is selected for relative to its ambient abundance and negative indices suggest that a prey item is selected against. Calculations based on data collected from 1974–1975 (Eggers 1982).



(210–304 μg) analyzed for fatty acids. Significant differences in DHA content among prey groups were substantiated by the ANOVA and the subsequent Tukey test ($p = 0.05$), which were attributed to the disparate levels of DHA in *Daphnia* and *Cyclops*. *Daphnia* were practically devoid of DHA, whereas the highest observed concentrations of DHA were in *Cyclops*. Because of the shift in prey taxa consumed during the sockeye growth season, pronounced fluctuations in the relative amounts of dietary DHA and P:C ratios are experienced (Fig. 5). The diet of sockeye during winter is dominated by copepods and characterized by low P:C ratios and high DHA, whereas *Daphnia* are more prevalent in the diet during summer months resulting in high P:C ratios and low DHA content. Because there is relatively low variability among prey in EPA content, the content of EPA in the diet remains fairly constant throughout the year.

Sockeye dorsal muscle contained approximately half the amount of fatty acids as the liver tissue (Fig. 6a). The content of EPA and DHA were higher in sockeye liver tissue (Fig. 6b) than contents found in most of the prey types, except *Cyclops*, which had very high levels of DHA. There was also an increasing trend in all of the fatty acids until a fork length of about 110 mm, after which the fatty acid content declined. This trend was the most pronounced in the liver tissue. Resident kokanee liver and dorsal tissue samples had relatively low essential fatty acid content.

Bioenergetics modeling

Observed growth was consistently below the predicted maximum growth rate for all simulations. However, observed growth did approach maximum potential growth rates in the month of June. Predicted maximum growth greatly exceeded observed growth in the first April of lacustrine development. This could be an artifact of field sampling bias that may have undersampled fish <50 mm in fork length; thus, resulting in an underestimate of newly emergent fry growth rates (Woodey 1972). Growth limitation resulting from dietary EPA was not predicted during any month (Fig. 7a). This is

evidenced by the fact that observed growth was always less than predicted EPA-limited growth. Moreover, the predicted EPA-limited growth always exceeded maximum growth, suggesting growth limitation resulting from EPA content is highly unlikely.

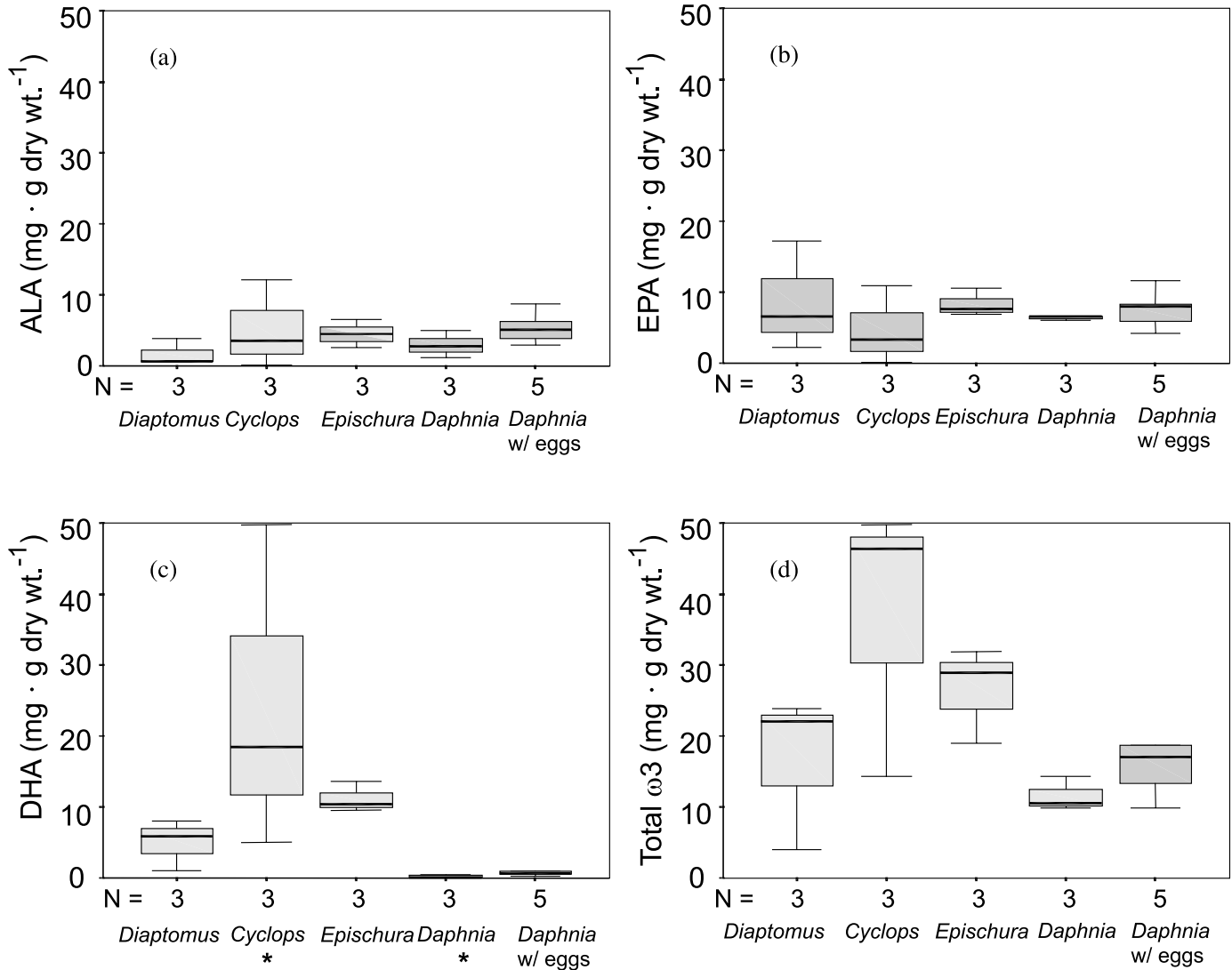
In contrast, the results for the other essential fatty acid DHA suggest that sockeye in Lake Washington may experience limited growth resulting from the DHA content of their diet much of the year (Fig. 7b). Based on these simulations growth appears to be limited by dietary DHA in all months except April, when DHA-limited growth coincides with optimal growth, and in the winter months, when observed growth is negligible. To test this assertion, we conducted a sensitivity analysis of the bioenergetics model by rerunning the simulations using the maximum observed measurements of DHA in *Daphnia* spp. ($0.76 \text{ mg}\cdot\text{g wet wt.}^{-1}$) and maintaining all other variables constant. These simulations were run for the months of greatest reported growth limitation (May–August). The results were consistent with our initial findings and indicate that even if we consider the scenario in which sockeye are feeding on the most nutritious *Daphnia* spp. available (i.e., individuals with the highest DHA content) they are still predicted to experience growth limitation because of insufficient DHA in their diet. Therefore we may conclude that sockeye in Lake Washington are predicted to experience DHA-limited growth, even if we consider the uncertainty of our parameters.

Model predictions of growth based on ratios of P:C in the diet of sockeye were similar to those for EPA and revealed no apparent limitation (Fig. 7c). At no point during the year did sockeye growth in Lake Washington appear to be limited by the amount of phosphorus in their diets. Suboptimal temperatures (i.e., temperatures <15°C) experienced by sockeye appeared to have little effect on their growth. Differences in predicted growth for the three temperature scenarios, ambient (at 20 m), optimal lake temperature ($\leq 15^\circ\text{C}$), and optimal temperature (15°C), were negligible. Therefore, if we partition out the limitation effects of lake temperature, food quantity, and food quality, in terms of DHA content, we can see how much each one of these factors potentially inhibits the growth of sockeye in Lake Washington during different months of the year (Table 3). Based on this comparison, it is evident that food quality, in terms of DHA, is potentially limiting to sockeye growth from May to November and that during the winter months suppressed growth may be due to insufficient food quantity.

Discussion

The results of this study suggest that sockeye growth rates in Lake Washington may be limited by food quality, especially the DHA content, of their diet. This is due to the disproportionate amount of DHA in sockeye compared with their prey. Sockeye are rich in DHA and their diet is composed mostly of *Daphnia*, which are deficient in DHA. Taxa considered to be lacking in phosphorus and of low nutritional value, such as *Cyclops*, had an abundance of EPA and the highest levels of DHA and yet are not heavily preyed upon by sockeye. Conversely, *Daphnia*, which are relatively rich in phosphorus, contained little EPA and were practically devoid of DHA. This is especially true for *Daphnia* without

Fig. 4. Box-and-whisker plots of (a) 18:3 ω 3 (ALA, alpha-linolenic acid), (b) EPA (eicosapentaenoic acid), (c) DHA (docosahexaenoic acid), and (d) total ω 3 content of sockeye (*Oncorhynchus nerka*) prey types with sample sizes listed on the x axis (w/ eggs = with eggs). The lines through the boxes show the median, the outer edges correspond with the 25th and 75th percentiles, and the whiskers represent the 10th and 90th percentiles. Asterisks (*) identify prey types with significantly different contents of DHA. The sample size (N) appears on the x axis. Each sample consisted of 50–100 individual zooplankton.



eggs. This disparity in the relative amounts of phosphorus with respect to essential fatty acids in copepods and cladocerans may have implications for the growth of other freshwater planktivorous fish, particularly taxa with high somatic DHA content. Furthermore, fatty acids appear to play a metabolic role in the physiological changes associated with smoltification as accumulations of fatty acids increase markedly up to smolt size (~120 mm), especially in the liver, and then subsequently decline.

Based on the Ivlev electivity indices, sockeye select for the phosphorus-rich *Daphnia* spp., which contain very little DHA, especially those without eggs, and avoid the relatively phosphorus-deficient copepods (*Cyclops* spp. and *Diaptomus*), which have a higher DHA content. One may infer from this result that phosphorus is an important constraint on sockeye growth and they avoid this constraint by selecting the most phosphorus-rich zooplankton available. However, there

are many confounding behavioral and physiological factors affecting this predator–prey interaction. For instance, *Daphnia* spp. have a slower swimming response, rendering them much more vulnerable to sockeye predation than copepods, which are more adept at evading predators (Vinyard et al. 1982). In addition, egg-laden *Daphnia* may be more conspicuous to predators and less adept at evading predation; further, *Daphnia* bearing eggs appear to have slightly higher levels of DHA. Thus for behavioral reasons it is logical that sockeye would select for *Daphnia* irrespective of their phosphorus or DHA content. *Daphnia* are also the most abundant prey taxa present in the lake during the spring and summer and their spatial distributions coincide with sockeye; therefore, they are frequently encountered.

In the analysis conducted by Schindler and Eby (1997), one of the few fish species predicted to experience phosphorus-limited growth was sockeye feeding exclusively on cope-

Fig. 5. Seasonal amounts of P:C (triangles), EPA (eicosapentaenoic acid; squares), and DHA (docosahexaenoic acid; diamonds) in the sockeye (*Oncorhynchus nerka*) diet based on weighted averages of zooplankton from the diet.

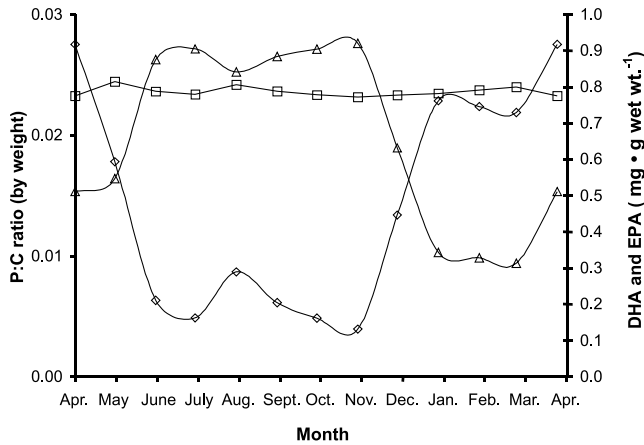
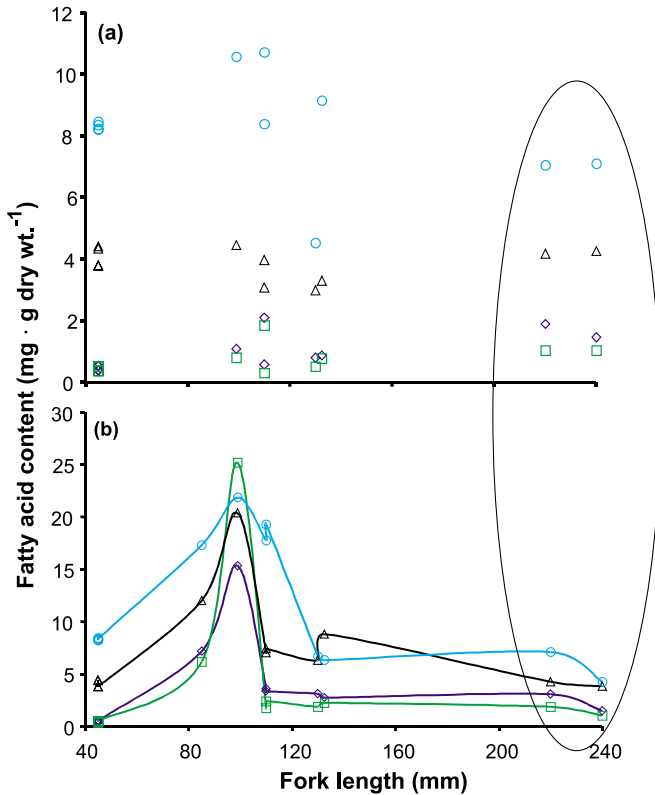
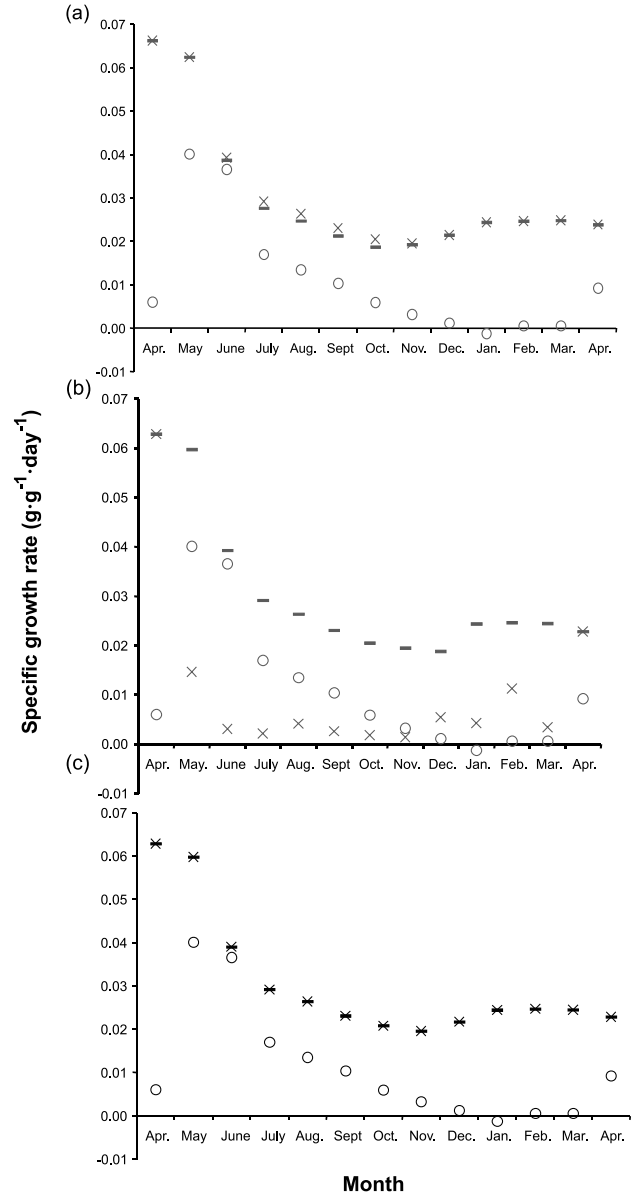


Fig. 6. Content of specific fatty acids in (a) the dorsal muscle and (b) liver tissue of sockeye salmon (*Oncorhynchus nerka*) from Lake Washington. Specific fatty acids are 18:3 ω3 (diamonds), 18:4 ω3 (squares), EPA (eicosapentaenoic acid; triangles), and DHA (docosahexaenoic acid; circles). Kokanee are the nonanadromous form of sockeye.



pod. According to their analysis, Lake Washington sockeye were not growth limited because their diet was dominated by phosphorus-rich *Daphnia* spp. Their results are corroborated by those of the present analysis, which was conducted at a greater resolution (i.e., monthly intervals), with much

Fig. 7. Specific growth rate scenarios plotted for actual observed growth, maximum growth at optimal consumption, and (a) EPA (eicosapentaenoic acid) limitation, (b) DHA (docosahexaenoic acid) limitation, and (c) phosphorus limitation (P:C); respective limitation indicated with an × on each panel; open circles = observed; dashes = maximum. All simulations were conducted for the 13-month period of lacustrine development of sockeye salmon (*Oncorhynchus nerka*) in Lake Washington.



more descriptive diet data and using prey ratios of P:C. Although the diet of sockeye in Lake Washington consists primarily of the phosphorus-poor *Diaptomus* for 5 months during their lacustrine development, this is during winter months when growth is suppressed by low prey abundance and phosphorus demands are low.

The assimilation efficiency of polyunsaturated fatty acids in salmon is quite high (0.93) relative to the assimilation efficiency of other elements, such as nitrogen (0.80; Brett and Groves 1979) and phosphorus (0.74; Nakashima and Leggett 1980). This marked difference could be interpreted as a

Table 3. Percentage of limitation resulting from lake temperature, food quantity, and food quality (docosahexaenoic acid, DHA) to growth of sockeye in Lake Washington.

	Temperature, %	Quantity, %	DHA, %
April	6	94	0
May	12	0	88
June	0	26	74
July	0	0	100
August	0	0	100
September	0	0	100
October	0	0	100
November	0	0	100
December	13	21	66
January	0	21	79
February	0	44	56
March	2	11	87
April	7	93	0

greater evolved affinity for polyunsaturated fatty acids in the diet compared with nitrogen or phosphorus or that polyunsaturated fatty acid molecules are more easily metabolized. However, as polyunsaturated fatty acids are utilized to form cell membranes, as precursors for important metabolites, and as an energy source, this high assimilation efficiency is not surprising. Moreover, as many polyunsaturated fatty acids are unable to be synthesized endogenously, their high assimilation efficiency would be expected.

The growth curve of sockeye is quite unusual. Unlike most vertebrates that experience their greatest exponential growth immediately after birth, sockeye experience their highest growth rates after smoltification upon reaching the ocean (Weatherley and Gill 1995). The a priori explanation of this is that there is a greater abundance of food in the marine environment. However, if we compare the content of EPA and DHA in the freshwater prey of sockeye with that of their marine prey (Table 4), a clear disparity emerges. As evidenced here, freshwater prey species have substantially less highly unsaturated fatty acids than their marine counterparts, especially the freshwater insects, which have less EPA and virtually no DHA. In contrast, marine species, such as euphausiids, mysids, and molluscs, are extremely rich in EPA and DHA. This disparity between the essential fatty acid content of marine and freshwater prey may explain the observation that freshwater fish are more adept at elongating precursors to form EPA and DHA than are marine fish (Watanabe 1982).

It is also thought that sockeye have a lower food conversion efficiency in fresh water (20.6%) compared with the food conversion efficiency (27.0%) during their marine growth (Brett 1995). When sockeye are growing at their greatest specific growth rate in Lake Washington, they are feeding almost exclusively on *Daphnia*; however, the essential fatty acid content of *Daphnia* is relatively low compared with other freshwater prey and very low compared with marine prey taxa. These two lines of evidence, in conjunction with the bioenergetics modeling results, suggest an alternative hypothesis to limited growth of sockeye in the freshwater environment, perhaps the allocation of carbon to growth is governed by how much carbon is available in the form of essential fatty acids.

According to the model simulations presented, sockeye are DHA limited to such an extent that they must be provided with an alternative source of DHA to grow at the observed growth rates. Sockeye can avoid DHA limitation by either complimenting their diet with other zooplankton species that are richer in DHA or through the elongation of the shorter length fatty acids linolenic (18:3 ω 3) acid and EPA (20:5 ω 3). The former scenario is not likely in Lake Washington as sockeye are feeding almost exclusively on *Daphnia* in the spring and early summer. It is generally believed that freshwater fish species are more adept at elongating shorter chain essential fatty acids than marine fish species, which may lack the necessary enzymes and desaturases for elongation (Watanabe 1982). This is true of the marine species *Scophthalmus maximus* (turbot; Ghioni et al. 1999), which is much less efficient at elongating ω 3 fatty acids than Atlantic salmon (*Salmo salar*), although both of these species were limited in their ability to biosynthesize DHA from shorter carbon chain precursors. The conversion of EPA and linolenic acid to longer chain DHA in rainbow trout (*O. mykiss*) has been definitively confirmed by using radiotracer (^{14}C) studies (Buzzi et al. 1996). It is quite possible that luxury uptake and subsequent elongation of EPA, which was not found to be limiting to sockeye growth, may account for the DHA required to sustain observed growth rates. However, trout fed on diets rich in ω 3 fatty acids had 20 times more DHA in their hepatocytes at the conclusion of the experiment, implying that trout had much more DHA that could be metabolized when it was supplied in their diet than when dependent on elongation. The inefficiency in the conversion of shorter essential fatty acids has been verified in Atlantic salmon, in which it is thought that the rates of elongation may be insufficient to meet the metabolic needs of growth. Moreover, Atlantic salmon raised on diets deficient in essential fatty acids often suffer from pale swollen livers and fin erosion (Ruyter et al. 2000b). Although elongation is known to occur in salmonids, there appear to be metabolic costs associated with the elongation of existing fatty acids, and higher growth rates in salmonids are still observed when these HUFAs are supplied directly in the diet (Takeguchi and Watanabe 1982; Ruyter et al. 2000b). Further efforts should be made to quantify the elongation efficiency of these essential fatty acids with respect to salmonids.

The gradual increase in essential fatty acids in the liver of sockeye throughout their development leading up to smoltification and the subsequent decline is indicative of the physiological demands associated with smoltification. The relatively low concentrations of essential fatty acids among kokanee may be indicative of fat stores being utilized by fish overwintering in the lake. Among the essential fatty acids, stearidonic acid (18:4 ω 3) was observed to fluctuate the most. As this fatty acid is one of the precursors for elongation to form EPA and DHA, as well as an intermediate in the oxidation of HUFAs to lesser fatty acids yielding energy, it is not surprising that this particular fatty acid fluctuates most with sockeye size.

The catabolism of lipids yields 9 kcal·g⁻¹ compared with carbohydrates and proteins, which yield only 6 kcal·g⁻¹, thus lipids constitute the most efficient means of storing energy (Stryer 1981). For this reason lipids provide the energy for

Table 4. Eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) contents (% total fatty acids) of various freshwater and marine prey taxa of salmonids (revised from Higgs et al. 1995 and references therein).

Freshwater prey taxa	EPA	DHA	Marine prey taxa	EPA	DHA
Insecta			Crustacea		
Diptera	7.7	0.0	<i>Calanus helgolandicus</i>	14.3	15.4
Trichoptera	6.5	0.0	<i>Palandus jordani</i>	18.0	10.1
Coleoptera	4.3	0.0	<i>Eugommarus confervicolus</i>	14.5	6.8
Ephemeroptera	12.5	0.0	<i>Corophium volutator</i>	25.8	7.1
Plecoptera	13.9	0.0	<i>Euphausia pacifica</i>	20.5	12.3
Hymenoptera	0.0	0.0	<i>E. pacifica</i> (California)	30.9	6.4
Odonata	9.4	0.0	<i>E. pacifica</i> (Washington)	27.9	16.1
Crustacea			<i>E. gibboides</i>	10.3	21.0
Cladocera	9.2	1.5	<i>E. brevis</i>	14.9	24.4
<i>Daphnia cucullata</i>	11.1	1.4	<i>E. americana</i>	13.5	19.5
<i>Daphnia magna</i>	13.9	0.0	<i>E. krohnii</i>	14.0	21.9
<i>Daphnia</i> spp.†	13.7	1.5	<i>E. superba</i>	25.3	14.6
<i>Cyclops strenuus</i>	8.8	16.8	<i>Thysanoessa inermis</i>	21.4	10.9
<i>Cyclops</i> spp.†	5.4	20.0	<i>Neomysis integer</i>	20.9	11.0
<i>Epischura nevadensis</i> †	14.9	20.2	<i>N. integer</i> (with eggs)	26.9	12.6
<i>Eudiaptomous gracilis</i>	8.8	8.8	<i>N. spinosa</i>	24.0	9.0
Mean	9.3	4.7	<i>N. awatschensis</i>	17.4	11.3
Standard deviation	4.2	7.8	<i>N. nakazawai</i>	21.1	10.5
			Mollusca		
			<i>Loligo opalensis</i>	12.8	29.6
			Squid (whole)	17.7	31.1
			Mean	19.6	15.1
			Standard deviation	5.8	7.3

Note: Taxa denoted by † are values from this study.

many metabolic functions and should be considered one of the major currencies of bioenergetics. The total lipid contained in the liver of some fishes has been used to accurately predict their recruitment (Marshall et al. 2000). More extensive research should focus on the importance of lipids in the physiological ontogeny of fish species with an emphasis on essential fatty acids.

Based on the results of this study, we can prioritize the factors most likely limiting sockeye growth. We may conclude that food quality, especially the DHA content of the diet, may limit sockeye growth for a good portion of their lacustrine development, depending on their ability to elongate shorter chain length unsaturated fatty acids. Furthermore, a high P:C ratio in the diet of sockeye makes it improbable that growth is ever limited by phosphorus. The second most important factor in the limitation of sockeye growth in Lake Washington is food availability. During the winter months, sockeye may be growing at suboptimal rates because they are not feeding near maximum consumption (i.e., low food abundance). Finally, we may conclude that even though sockeye aggregate at depths where they experience less than ideal temperatures for growth, these temperatures do not substantially limit their growth. Thus, the avoidance of piscivorous predators such as northern pikeminnow (*Ptychocheilus oregonensis*) and cutthroat trout (*Oncorhynchus clarki*) in addition to prey distribution are probably more important in controlling the vertical distribution of sockeye in Lake Washington than is temperature.

Acknowledgements

We would like to thank Doerthe Müller-Navarra for her assistance with the fatty acid analysis and Anne Liston and Sudeep Chandra at UC Davis for their laboratory assistance. The past and present students of Tom Quinn's fish ecology class deserve recognition for their help with data collection. Finally, this project would not have been possible without Sally Abella and Arni Litt's assistance with zooplankton sampling and Jennifer Anson's help with the identification and enumeration of sockeye prey species. This research was funded by an NSF grant (DEB 9615888) awarded to M.T.B.

References

- Ahlgren, G., Sonesten, L., Boberg, M., and Gustafsson, I.B. 1996. Fatty acid content of some freshwater fish in lakes of different trophic levels—a bottom-up effect? *Ecol. Freshw. Fish.* **5**: 15–27.
- Beauchamp, D.A., Stewart, D.J., and Thomas, G.L. 1989. Corroboration of a bioenergetics model for sockeye salmon. *Trans. Am. Fish. Soc.* **118**: 597–607.
- Beauchamp, D.A., Baldwin, C.M., Vogel, J.L., and Gubala, C.P. 1999. Estimating diel, depth-specific foraging with a visual encounter rate model for pelagic piscivores. *Can. J. Fish. Aquat. Sci.* **56**(Suppl. 1): 128–139.
- Bell, J.G., and Sargent, J.R. 1992. The incorporation and metabolism of polyunsaturated fatty acids in phospholipids of cultured cell from chum salmon (*Oncorhynchus keta*). *Fish Physiol. Biochem.* **10**: 99–109.

- Bell, M.V., and Tocher, D.R. 1989. Molecular species compositions of the major phospholipids in brain and retina from rainbow trout (*Salmo gairdneri*). *Biochem. J.* **264**: 909–915.
- Brett, J.R. 1995. Energetics. In *Physiological ecology of Pacific salmon*. Edited by C. Groot, L. Margolis, and W.C. Clarke. UBC Press, Vancouver. pp. 50–66.
- Brett, J.R., Shelbourn, J.E., and Shoop, C.T. 1969. Growth and body composition of fingerling sockeye salmon, *Oncorhynchus nerka*, in relation to temperature and ration size. *J. Fish. Res. Board Can.* **26**: 2363–2394.
- Brett, J.R., and Groves, T.D.D. 1979. Physiological energetics. In *Fish physiology*. Vol. VIII. Bioenergetics and growth. Edited by W.S. Hoar, D.J. Randall, and J.R. Brett. Academic Press, New York. pp. 279–352.
- Brett, M.T., Müller-Navarra, D.C., and Park, S.K. 2000. Empirical analysis of the effect of phosphorus limitation on algal food quality for freshwater zooplankton. *Limnol. Oceanogr.* **45**: 1564–1575.
- Buzzi, M., Henderson, R.J., and Sargent, J.R. 1996. The desaturation and elongation of linolenic acid and eicosapentaenoic acid by hepatocytes and liver microsomes from rainbow trout (*Oncorhynchus mykiss*) fed diets containing fish oil or olive oil. *Biochim. Biophys. Acta*, **1299**: 235–244.
- Cummins, K.W., and Wuycheck, J.C. 1971. Caloric equivalents for investigations in ecological energetics. *Mitt. Internat. Verein. Limnol.* **18**: 1–158.
- Donaldson, J.R. 1967. The phosphorus budget of Iliamna Lake, Alaska, as related to the cyclic abundance of sockeye salmon. Ph.D. dissertation, University of Washington, Seattle, Wash.
- Edmondson, W.T. 1994. Sixty years of Lake Washington: a curriculum vitae. *J. Lake Res. Manag.* **10**: 75–84.
- Eggers, D.M. 1978. Limnetic feeding behavior of juvenile sockeye salmon in Lake Washington and predator avoidance. *Limnol. Oceanogr.* **23**: 1114–1125.
- Eggers, D.M. 1982. Planktivore preference by prey size. *Ecology*, **63**: 381–390.
- Elser, J.J., Fagan, W.F., Denno, R.F., Dobberfuhl, D.R., Folarin, A., Huberty, A., Interlandi, S., Kilham, S.S., McCauley, E., Shulz, K.L., Siemann, E.H., and Sterner, R.W. 2000. Nutritional constraints in terrestrial and freshwater food webs. *Nature (London)*, **408**: 578–580.
- Ghioni, C., Tocher, D.R., Bell, M.V., Dick, J.R., and Sargent, J.R. 1999. Low C18 to C20 fatty acid elongase activity and limited conversion of stearidonic acid, 18:4(*n*-3), to eicosapentaenoic acid, 20:5(*n*-3), in a cell line from the turbot, *Scophthalmus maximus*. *Biochim. Biophys. Acta*, **1437**: 170–181.
- Gulati, R.D., and Demott, W. (Guest Editors). 1997. The role of food quality for zooplankton. *Freshw. Biol.* **38**.
- Hessen, D.O., and Lyche, A. 1991. Inter- and intraspecific variations in zooplankton element composition. *Arch. Hydrobiol.* **121**: 343–353.
- Hewett, S.W., and Johnson, B.L. 1991. Fish bioenergetics model 3.0. University of Wisconsin Sea Grant Institute, Madison, Wis., WIS-SG-92-250.
- Higgs, D.A., Macdonald, J.S., Levings, C.D., and Dosanjh, B.S. 1995. Nutrition and feeding habits in relation to life history stage. In *Physiological ecology of Pacific salmon*. Edited by C. Groot, L. Margolis, and W.C. Clarke. UBC Press, Vancouver. pp. 200–280.
- Kattner, G., and Fricke, H.S.G. 1986. Simple gas–liquid chromatographic method for the simultaneous determination of fatty acids and alcohols in wax esters of marine organisms. *J. Chromatogr.* **361**: 263–268.
- Kraft, C.E. 1992. Estimates of phosphorus and nitrogen cycling by fish using a bioenergetics approach. *Can. J. Fish. Aquat. Sci.* **49**: 2596–2604.
- Marshall, T.C., Yaragina, N.A., Lambert Y., and Kjesbu, O.S. 2000. Total lipid energy as a proxy for total egg production by fish stocks. *Nature (London)*, **402**: 288–290.
- Nakashima, B.S., and Leggett, W.C. 1980. Natural sources and requirements of phosphorus for fishes. *Can. J. Fish. Aquat. Sci.* **37**: 1540–1549.
- Peters, R.H. 1983. The ecological implications of body size. Cambridge University Press, Cambridge.
- Ruyter, B., Roesjoe, C., Maesoeval, K., Einen, O., and Thomassen, M.S. 2000a. Influence of dietary *n*-3 fatty acids on the desaturation and elongation of ¹⁴C 18:2*n*-6 and ¹⁴C 18:3*n*-3 in Atlantic salmon hepatocytes. *Fish Physiol. Biochem.* **2**(23): 151–158.
- Ruyter, B., Rosjo, C., Einen, O., and Thomassen, M.S. 2000b. Essential fatty acids in Atlantic salmon: effects of increasing dietary doses of *n*-6 and *n*-3 fatty acids on growth, survival and fatty acid composition of liver, blood and carcass. *Aquacult. Nutr.* **6**: 119–127.
- Schindler, D.E., and Eby, L.A. 1997. Stoichiometry of fishes and their prey: implications for nutrient recycling. *Ecology*, **78**: 1816–1831.
- Stryer, L. 1981. *Biochemistry*. 2nd ed. W.H. Freeman and Co., New York. pp. 383–386.
- Takeguchi, T., and Watanabe, T. 1982. Effects of various polyunsaturated fatty acids on growth and fatty acid compositions of rainbow trout (*Salmo gairdneri*), coho salmon (*Oncorhynchus kisutch*), and chum salmon (*Oncorhynchus keta*). *Bull. Appl. Soc. Sci. Fish.* **48**: 1745–1752.
- Tilman, D. 1982. Resource competition and community structure. Princeton University Press, Princeton, N.J.
- Vinyard, G.L., Drenner, R.W., and Hanzel, D.A. 1982. Feeding success of hatchery-reared kokanee salmon when presented with zooplankton prey. *Prog. Fish Cult.* **44**: 37–39.
- Watanabe, T. 1982. Lipid nutrition in fish. *Comp. Biochem. Physiol. B*, **73**: 3–15.
- Weatherley, A.H., and Gill, H.S. 1995. Growth. In *Physiological ecology of Pacific salmon*. Edited by C. Groot, L. Margolis, and W.C. Clarke. UBC Press, Vancouver. pp. 114–153.
- Woodey, J.C. 1972. Distribution, feeding and growth of juvenile sockeye salmon in Lake Washington. Ph.D. thesis, University of Washington, Seattle.
- Yu, T.C., and Sinnhuber, R.O. 1979. Effect of dietary (ω 3 and ω 6) fatty acids on growth and feed conversion efficiency of coho salmon (*Oncorhynchus kisutch*). *Aquaculture*, **16**: 31–38.