Future climate change-related decreases in food quality may affect juvenile Chinook salmon growth and survival

Jessica Garzke[1]*, Ian Forster[2], Caroline Graham[1], David Costalago[1], Brian P.V. Hunt[1,3,4]

[1] Institute for the Oceans and Fisheries, University of British Columbia, AERL, 2202 Main Mall, Vancouver, BC, Canada, V6T 1Z4

[2] Pacific Science Enterprise Center, Fisheries and Oceans Canada, 4160 Marine Dr., West Vancouver BC, Canada V7V 1N6

[3] Department of Earth, Ocean and Atmospheric Sciences, University of British Columbia, 2020 – 2207 Main Mall, Vancouver, BC, Canada, V6T 1Z4

[4] Hakai Institute, PO Box 309, Heriot Bay, BC, Canada, V0P 1H0

* Corresponding author: Jessica Garzke (garzke@zoology.ubc.ca)

Keywords: Zooplankton, essential fatty acids, RNA:DNA ratio, fish condition, mortality, growth, DHA, EPA, DHA:EPA ratios

Highlights

- Climate change may decrease the quality of salmon prey through changes in the fatty acid composition.
- Low dietary essential fatty acid levels reduce growth and condition and increase mortality rates in juvenile Chinook salmon.
- Food quality changes within zooplankton species but also by changes between species.
- Results suggest potential cascading effects on higher trophic levels when zooplankton species composition shifts to lower quality species.
- Higher food intake cannot compensate for low food quality.

Abstract

The global temperature increase due to global change is predicted to be between 3.3 – 5.7°C by 2100 leading to changes at the base of the marine food web in species composition, abundance, and quality at the base of the marine food web leading to flow-on effects of higher trophic levels such as fish and humans. Changes in marine prey availability and nutritional quality can affect juvenile salmon conditions (i.e., growth, condition, and mortality) during the early marine phase. There is limited knowledge of the interplay between prey availability and prey quality and the importance of food quality under food-satiated conditions. Here, a three-phase feeding experiment measured the effects of nutritional quality (fatty acid composition and ratios) on juvenile Chinook salmon (*Oncorhynchus tshawytscha*) condition. Experimental diets represented the present three different climate scenarios with a present-day diet (*Euphausia pacifica*), a control diet (commercial aquaculture diet), and a predicted IPCC worst-case scenario diet with low essential fatty acid concentrations (IPCC SSP5-8.5). We tested how potential future low quality food affects growth rates, body condition, fatty acid composition and mortality rates in juvenile Chinook salmon compared to present-quality prey. Fatty acids were incorporated into the salmon muscle at varying rates but, on average, reflected dietary concentrations. High dietary concentrations of DHA, EPA and high DHA:EPA ratios resulted in increased fish growth and condition. In contrast, low concentrations of DHA and EPA and low DHA:EPA ratios in the diets were not compensated for by increased food quantity. This result highlights the importance of considering food quality when assessing fish response to changing ocean conditions.

Introduction

Zooplankton play a fundamental role in aquatic food webs by consuming phytoplankton and bacteria and being the prey of higher trophic levels. At different life stages, many fish species derive their nutrition predominantly from zooplankton, which provide energy and essential biomolecules such as fatty and amino acids (Copeman and Laurel, 2010; Litz et al., 2017; Paulsen et al., 2014; Reitan et al., 1997). The nutritional quality of zooplankton prey directly affects fish condition, survival, and growth (Ahlgren et al., 2005; Daly et al., 2010; Olsen, 1999). Zooplankton nutritional quality varies significantly among species, and changes in the composition may therefore play an essential role in the feeding success, growth, and mortality of fish (Burke et al., 2013; Miller et al., 2014, 2013), particularly during vulnerable phases (Beamish et al., 2004; Pearcy and McKinnell, 2007). Understanding how prey composition impacts consumers has become a necessary step for estimating the survival of commercially important fish species.

Environmental variability in aquatic ecosystems is a key determinant of phenology, abundance, and species composition of zooplankton prey for foraging fish (Mackas et al., 2013; Mahara et al., 2019). This environmental variability occurs in space (e.g., bathymetry, tidal mixing) and time across a range of scales imposed by both natural cycles (e.g., seasonality, climate oscillations) and anthropogenic forcing (e.g., warming, acidification)(Vasseur and McCann, 2007). Over the past decades, the global mean surface temperature increased by ~0.2°C per decade, reaching 1.0°C above the pre-industrial period in 2017 (Hoegh-Guldberg et al., 2019) and the newest predictions indicate a potential temperature increase of between 2.01 – 4.07°C of the global mean sea surface temperature (worst-case scenario; SSP5-8.5)

by 2100 (IPCC, 2021). These temperature changes can lead to notable changes at the base of aquatic food webs through species-range shifts (Benedetti et al., 2021; McGinty et al., 2021; Poloczanska et al., 2013), shifts in phenology (Garzke et al., 2015; Parmesan and Yohe, 2003; Poloczanska et al., 2013; Walther et al., 2002), and reduced abundance and body size (Garzke et al., 2015).

Marine species have extended their range at an average rate of 72 km/decade towards the poles and into deeper waters responding to warmer environments (Lotze et al., 2010; Poloczanska et al., 2013). Specifically for marine zooplankton, time series analyses in the North Pacific and Atlantic Ocean showed that smaller bodied warmer-water species appeared and larger-bodied cold-water species disappeared (Beaugrand et al., 2002; Mackas et al., 2013, 2007; Preikshot et al., 2010)(Preikshot et al., 2010). Cold-water zooplankton species tend to have different life histories compared to their warmer-water species counterparts, as they have to diapause for overwintering and, therefore, store wax esters, triacylglycerols, phospholipids or diacylglycerol, such as euphausiids, *Calanus*, *Calanoides*, *Eucalanus*, *Neocalanus*, and *Rhinocalanus* whereas warm-water species have no energy-storage strategies and present a lower quality as a food item (Colombo et al., 2017; Hiltunen et al., 2021; Lee et al., 2006; Record et al., 2018). It is predicted that globally more non-native species will occur in new habitats. More native species will be extinct by 2050, and species turnovers of over 60% of the present biodiversity, implying direct effects on energetic pathways in marine food webs (Cheung et al., 2009; Heneghan et al., 2023). Indirect temperature-related changes, such as prey species composition, increased stratification, and corresponding changes in nutrient supply, may affect the production of essential biomolecules, such as fatty acids (FAs). These changes

could alter the overall availability of fatty acids and the modification of an individual's fatty acid profile (Garzke et al., 2016; Sahota et al., 2022; Stevens et al., 2022), ultimately affecting the efficiency of energy and biomass transfer through food webs (Pontavice et al., 2020; Pontavice, 2019; Ullah et al., 2018).

FA's play a fundamental role in physiological processes in organisms from microorganisms to top predators, including humans, for energy storage, growth, and development, and trophic interactions in aquatic food webs (Dalsgaard et al., 2003; Parrish, 2009; Parrish et al., 2012). The precise composition of FAs is critical for the structure and function of all organisms and is thus considered an essential driver of organismal and ecosystem health and stability (Arts and Kohler, 2009; Parrish, 2009). Long-chain (i.e., >= 20 carbons) polyunsaturated fatty acids (LC-PUFA) and especially those of the n3 series, namely eicosapentaenoic acid (EPA; 20:5n-3) and docosahexaenoic acid (DHA; 22:6n-3), are key for brain and neural tissue growth, and cardiovascular health (Copeman et al., 2002). Despite their importance to animals, these n3 LC-PUFAs cannot be biosynthesized in sufficient quantity to meet their needs, and they must be obtained from their prey (Arts and Kohler, 2009; Hixson and Arts, 2016). LC-PUFAs are mainly synthesized by phytoplankton in aquatic food webs, especially by diatoms, cryptophytes, and dinoflagellates (Brett and Müller-Navarra, 1997). As primary consumers of phytoplankton, zooplankton act as conduits controlling the quantity and quality of LC-PUFA to higher trophic levels such as juvenile salmon. Accordingly, changes in phytoplankton FAs, zooplankton fatty acids, or the transfer between these two organisms will have cascading effects through the marine food web (Dalsgaard et al., 2003). Zooplankton consumers, such as juvenile fish, will be affected by the direct temperature-related impact on the organism's metabolism, affecting their growth, reproduction, and survival, and the

indirect temperature-related effects by changed bottom-up effects on the zooplanktons prey source, phytoplankton (Azani et al., 2021).

Global climate change could significantly affect the availability of long-chain omega-3 PUFAs in two not mutually exclusive ways: 1) by decreasing overall plankton biomass and growth rates and 2) by reducing the content of PUFAs, due to changes in zooplankton species composition with varying FA accumulation rates. For example, Hixson and Arts (2016) predicted that, in response to a 2.5°C increase in surface water temperature, phytoplankton could reduce DHA synthesis by up to 28% globally. Holm et al. (2022) predicted that EPA content in marine phyto- and zooplankton would substantially decline in the next century globally by 2%, ranging from 0.7% to 28.5%, depending on the geographic location. Since phytoplankton are the principal suppliers of DHA and EPA to higher trophic levels (e.g., fish), a reduction in the synthesis of DHA and EPA by phytoplankton as water temperatures rise has the potential to translate into a profound impact on the total supply of DHA and EPA to aquatic ecosystems, and hence the availability of these FAs to humans. Holm et al. (2022) even predicted that such a decline in essential FAs could lead to deleterious effects on fisheries.

In this study, we tested the climate-driven changes in zooplankton FA composition on fish condition, growth, mortality, and FA profiles. We performed a feeding experiment where juvenile Chinook salmon were fed either *Artemia* sp., which has an extremely low DHA and EPA concentration and low DHA:EPA ratio, a nutritionally enhanced control diet (commercial aquaculture), and *Euphausia pacifica* (a present-day diet representative).

2.1 Materials & Methods

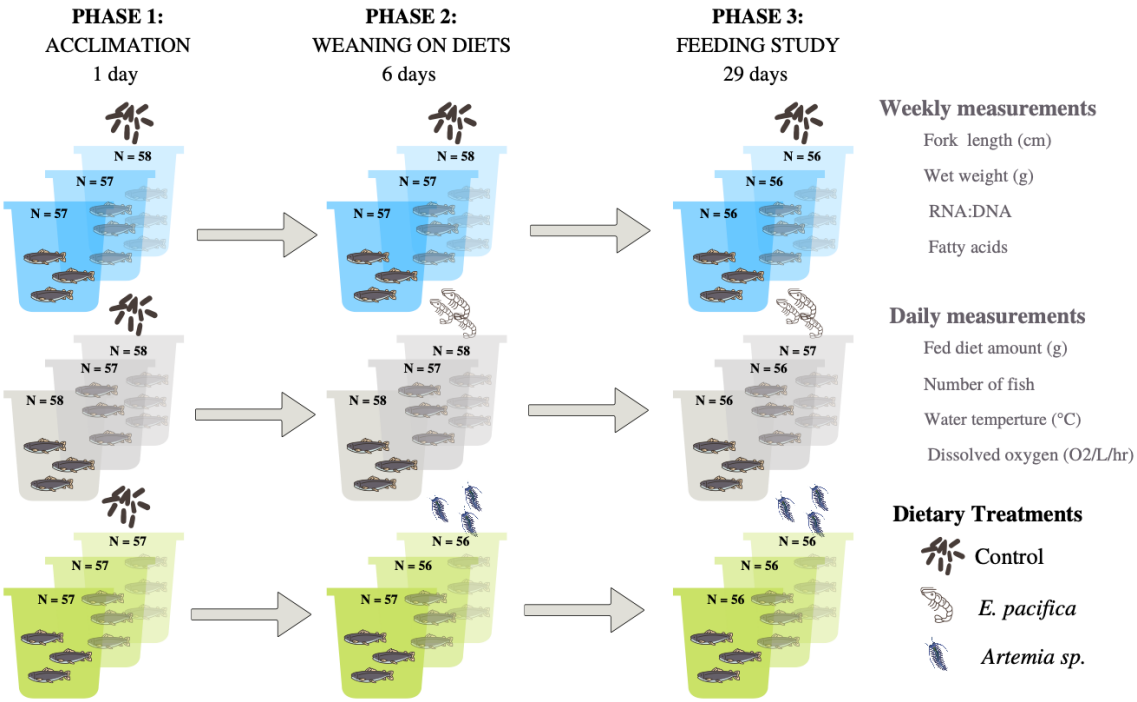
2.1 Ethics Statement

All animals and procedures used in this experiment were approved by DFO Pacific Region - Animal Care Committee (AUP # 2017-008).

2.2 Experimental design

This study was conducted on the premises of Fisheries and Oceans Canada at the Pacific Science Enterprise Centre in West Vancouver, British Columbia, Canada. Juvenile Chinook salmon (ocean type, Harrison River strain) from the brood year 2018 (4.14 g ± 0.72 body weight; 73.6 mm ± 4.67 standard length) were obtained from Capilano River Hatchery (49.35º N 123.11º W) and transported to the research facilities. Before starting the trial, the fish were fed commercial salmon feed (Bio-Oregon, BioVita, Skretting Canada). The juvenile fish were acclimated in freshwater tanks for one week before transitioning to salt water, and only one individual died during the transition. Fish were not fed on the day of the transfer. The remaining 517 fish were used in the experiment (Fig. 1). The rearing system consisted of nine 200 L oval fibreglass tanks, each supplied with flow-through seawater (drawn from Burrard Inlet and passed through a sand filter) at 9 L min^{-1} and provided with supplemental air through an air stone. The overhead lights supplied lighting, and the photoperiod followed the natural day length. The fish were randomly assigned to the tanks, and each tank was assigned to three treatments in triplicate (n=3, Fig. 1). Fish were maintained in the experimental set-up for 36 days. The experimental phase was split into three individual phases (Fig. 1): (1) the acclimation phase when the fish were transferred to the experimental tanks (Phase 1); (2) weaning fish from control diets to

182 their respective treatment diets (6 days; Phase 2); (3) feeding phase on experimental
183 diets (29 days; Phase 3). Over the experimental period, the fish were held at an
184 average temperature of 11.6ºC (± 0.61ºC).
185
186



187

188 **Fig. 1.** Three communities received a control diet (commercial aquaculture diet),
189 three received *Euphausia pacifica* (present-day diet species), and three received
190 *Artemia* sp. (climate change diet). Phase 1 lasted one day to acclimate fish to
191 smaller tanks, and fish received the control diet; Phase 2 was six days to wean fish
192 onto their respective diet treatments, and Phase 3 was the feeding study. Juveniles
193 received their treatment diets until satiation.
194

195 *2.3 Feeding*

196 The three diet treatments were: 1) a commercial control (Bio-Oregon, BioVita,
197 Skretting Canada), frozen *Euphausia pacifica* (Euphausia pacifica Canada) as

198 present-day feeding condition representative, and frozen *Artemia* sp. as low-quality,
199 climate change feeding condition representative (San Fransisco Bat Brand® Sally's
200 Frozen Spirulina Brine Shrimp™, Table 1). The commercial control had the lowest
201 moisture content of the three diets (avg. = 5.79% ± 0.007), while *E. pacifica* and
202 *Artemia* sp. had much higher moisture content (*E. pacifica*: avg. = 82.09% ± 0.24;
203 *Artemia* sp.: 91.63% ± 0.32; Table 1, Fig. 1). The commercial control feed pellets,
204 *Artemia* sp. and *E. pacifica* were thawed before feeding.
205
206 **Table 1.** Nutritional information of dietary treatments used in the experiment, for total
207 fatty acids (TFA), saturated fatty acids (SFA), mono-unsaturated fatty acids (MUFA),
208 polyunsaturated fatty acids (PUFA), docosahexaenoic acid (DHA), eicosatetraenoic
209 acid (EPA), and DHA-to-EPA ratio (DHA:EPA). (DW: dry weight)

Ingredients	Control	*Euphausia pacifica*	*Artemia* sp.
kJ g^{-1} (DW)	23.67	19.56	20.44
TFA (g g^{-1} DW)	161.483 (± 11.112)	24.069 (± 13.638)	7.503 (± 4.118)
SFA (%)	32.10 (± 0.10)	28.47 (± 0.11)	20.63 (± 0.10)
MUFA (%)	29.32 (± 0.68)	24.49 (± 0.22)	30.34 (± 0.51)
PUFA (%)	38.36 (± 1.85)	46.81 (± 0.13)	38.38 (± 0.42)
DHA (%)	11.02 (± 0.58)	16.80 (± 0.24)	0.68 (± 0.94)
EPA (%)	14.06 (± 0.28)	16.11 (± 0.15)	2.55 (± 0.15)
DHA:EPA	0.78 (± 0.02)	1.04 (± 0.02)	0.28 (± 0.40)

210
211
212 All diets were offered four times daily (7:30 – 8:00 am; 11:00 – 11:30 am; 12:30 –
213 1:00 pm; 2:00 – 2:30 pm) in smaller portions until the fish stopped actively feeding

214 (*satiation*) depending on the individual needs of the treatment fish (Yu and
215 Sinnhuber, 1979). The daily provided feed amounts were recorded. The tanks were
216 cleaned three times a week after the last feeding by stopping the water flow and
217 brushing the tank walls and bottom while the drain was plugged with a stopper. The
218 sediment was then siphoned and cleaned. After the cleaning, the water flow was
219 restarted, and the drain was unplugged.
220
221 *2.4 Sampling and laboratory analyses*
222 Throughout the experiment, four fish were randomly sampled from each tank weekly.
223 Fish were removed from the tanks using nets and then euthanized in TMS™ (0.4 g L^{-}
224 ^{1}). All fish were weighed to the nearest 0.1 g (Mettler Toledo EXCELLENCE Plus
225 balance, model XP16001L), and fork length (FL) was measured (mm). Individual fish
226 were placed into separate Whirlpak™ bags, labelled with a unique identifier, and
227 frozen at -80 ºC. The time between the mortality of the fish and the fish being placed
228 in the -80 ºC freezer was less than two minutes. Tissue was sampled from each fish
229 (n=336) for RNA:DNA ratios and fatty acids analysis using sterilized dissection tools.
230 Before dissection, the dissection area was cleaned first with 95% ethanol and then
231 with EZ-Zyme (Integra™ Miltex™ EZ-Zyme™ All-Purpose Enzyme Cleaner, Fisher
232 Scientific, Cat No 12-460-427). White muscle tissue (400 – 600 mg) was collected
233 close to the dorsal fin for RNA:DNA and fatty acid analysis. Tissue samples were
234 then returned to a -80ºC freezer until further analysis.
235
236 *2.4.1 RNA:DNA analysis*
237 The ratio of RNA and DNA of juvenile Chinook was determined according to Garzke
238 et al. (2022) using the cyanine base fluorescence dye RiboGreen® (Thermo Fisher

239 Scientific). Nucleic acids were extracted from freeze-dried white muscle tissues,
240 which had been homogenized in 1% N-lauroylsarcosine extraction buffer (VWR, Cat.
241 No. VWRV0719-500G), the supernatant was diluted to match the RiboGreen
242 saturation range (1:10, 1:20, 1:30, 1:50, 1:100). In short, we used a DNA standard
243 (ranging from 0-3 µg mL^{-1}; calf thymus, VWR, Supp. No. MB-102-01100) and RNA
244 (ranging from 0-3 µg L^{-1}; Escherichia *coli* 16S and 23S rRNA, Quant-iT RiboGreen
245 RNA assay Kit Thermo Fisher Scientific Cat. No. R11490). Triplicate technical
246 replicates from the diluted sample were added to a black, flat-bottom 96-well
247 microtiter plate. RiboGreen® reagent was added to the homogenized muscle tissue
248 samples and incubated in the dark for 5min; initial fluorescence (F_1) readings were
249 taken using a VarioSkan Flash Microplate Reader (ThermoFisher Scientific). RNase
250 A (bovine pancreas, Alpha Aesar, Cat. No. CAAAJ62232-EX3) was added to each
251 well and incubated at 37 ºC for 30min in the dark, followed by a second fluorescence
252 reading of the DNA only (F_2). Finally, we calculated the RNA concentration using F_1 -
253 F_2. RNA and DNA concentrations were calculated based on the standard curves,
254 followed by calculations of RNA:DNA.
255
256 *2.4.2 Fulton K, growth rates, and mortality rates*
257 For each sampling date and each diet treatment, we calculated the average body
258 condition factor as Fulton's K:
259 $$K = \frac{W}{L^3} \times 10^5 \quad (1)$$
260 Where *W* is the wet body mass (g), and *L* is length (mm) (Fulton, 1904). Specific
261 growth rates (SGR) for the weight (% W d^{-1}) were measured for experimental
262 $$SGR = \left(\frac{\left(ln(x_f) - ln(x_i)\right) \times 100}{t}\right) \quad (2)$$

263 Where X is the weight of the last sampling day of Phase 3, x_i is the weight of the first
264 sampling day of Phase 3, and t is the time (days) between observations.
265
266 We calculated the instantaneous mortality rate (Z) by comparing the numbers of fish
267 from one sampling day to the next for the duration of the experiment using (Sundby
268 et al., 1989):

269 $$Z = \left(\frac{ln(N_0) - ln(N_t)}{t} \times 100\right) \quad (3)$$

270 N_0 is the initial number of fish, N_t is the number of remaining fish between sampling
271 days, and t is the number of days between samplings.
272
273 *2.4.3 Fatty Acid analysis*
274 FAs were analyzed using a modified protocol following Puttick et al. (2009) in a one-
275 step fatty acid methyl ester (FAME) method. White muscle tissue samples were
276 weighed (wet weight), freeze-dried, and the respective dry weights measured for
277 calculations of moisture content. FAs were methylated with 3 N HCl in CH_3OH
278 (Sigma-Aldrich cat. #90964-500ML) with hexane (containing nonadecanoic acid
279 C19:0 as an internal standard) at 80 C for 18 h. FAMEs were analyzed with a gas
280 chromatograph (Scion 436-GC, Scion Instruments Canada, Edmonton, Alberta,
281 Canada) using a 50 m column (CP Sil-88, Agilent Technologies, Santa Clara,
282 California, USA) and a flame ionization detector (FID). Peaks were identified against
283 external standards (GLC 455 and GLC 37 Nu-chek Prep, Inc., Elysian, Minnesota,
284 USA) and measured before and after each sample measurement session.
285 All fatty acids were measured and reported as relative contents of total fatty acid
286 methyl esters. Concentrations of 28 fatty acids were calculated using calibration

curves of the known standard (C19:0). We calculated bioaccumulation factors (Kainz et al., 2006) of DHA, EPA, and ALA as

$$Bioaccumulation\ factor\ = \frac{FA\ [\%]\ in\ tissue\ of\ an\ individual}{FA\ [\%]\ in\ diet} \quad (4)$$

The bioaccumulation factor indicates consumers' internal synthesis or retention of FAs. Specifically, the retention or internal synthesis of a FA increases with the increasing value of the bioaccumulation factor, and it is positive if the factor is >1.

2.5 Statistical analysis

We used linear mixed models repeated measures analysis to examine the effects of the three diet treatments and sampling date on fish condition (RNA:DNA, Fulton's K) and individual fatty acid composition in fish muscle tissues across experimental Phase 3 to examine the effects of the three different prey qualities (using RStudio Version 1.1.456, lme4 R package (Bolker et al., 2009)). The tank number was set as a random factor.

Non-parametric PERMANOVA was used to test for differences in fish fatty acid composition between the three diet treatments. Subsequently, similarity percentages (SIMPER) were analyzed to identify the FA species that contributed most to the Bray-Curtis dissimilarities among the diet treatments. All multivariate tests were performed using the R package vegan (Oksanen et al., 2017).

Kolmogorov-Smirnov analysis was performed to test for normal distribution (α = 0.05). Analysis of Variance (ANOVA) was performed to identify differences between SGR between diet treatment, followed by a Tukey honest significance difference post hoc test. For non-normally distributed data, a Kruskal-Wallis test (Kassambara, 2020), followed by analyzing the effect size (H^2) and a pairwise comparison using the

Dunn's test were used to identify differences in instantaneous mortality rate (Z) and diet fatty acid data. The error rate for all tests was held at a 5% probability. All statistical tests were run at a significance threshold of α=0.05.

3. Results

Throughout the experiment, higher quantities of *Artemia* sp. were voluntarily eaten by fish compared to the control and *E. pacifica*. Percentage body weight (BW) consumed per day was: *Artemia* sp., 2.13% BW d^{-1} (of dry matter) ± 0.42 (range: 1.43% BW d^{-1} – 2.86% BW d^{-1}); control, 1.49% BW d^{-1} (of dry weight) ± 0.34% BW (range: 0.99% BW d^{-1} – 2.35% BW d^{-1}); *E. pacifica,* 1.68% BW d^{-1} (of dry matter) ± 0.66% (range: 1.04% BW d^{-1} – 2.50% BW d^{-1}), Fig 2). Fish consuming *Artemia* sp. during Phase 3 had an average mortality rate of 1.96 ind d^{-1} (± 0.59) which was significantly higher than for fish consuming *E. pacifica* (1.21 ind d^{-1} ± 0.05; p = 0.04), and the control feed (1.23 ind d^{-1} ± 0.07; p = 0.04).

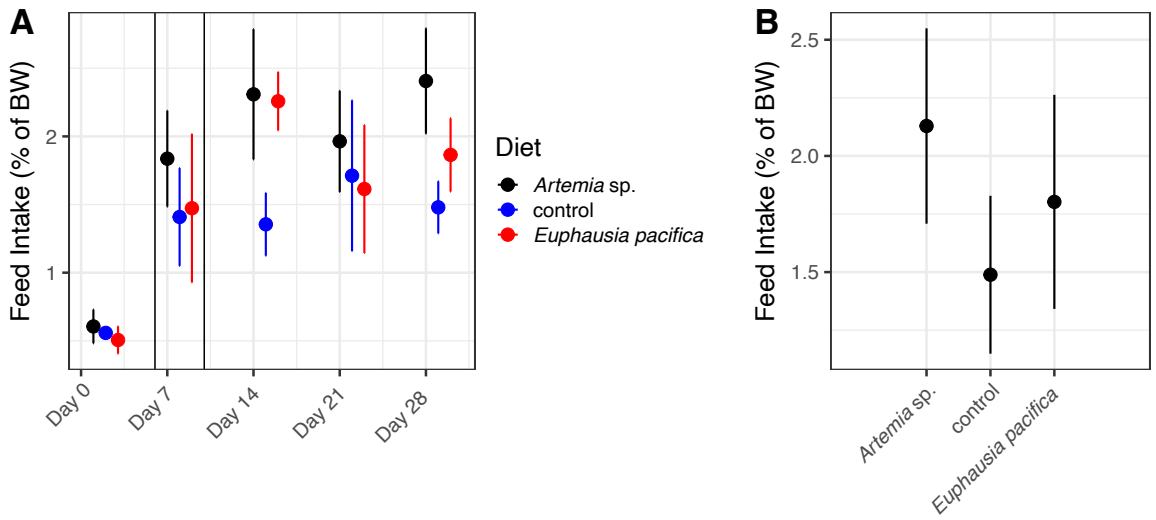
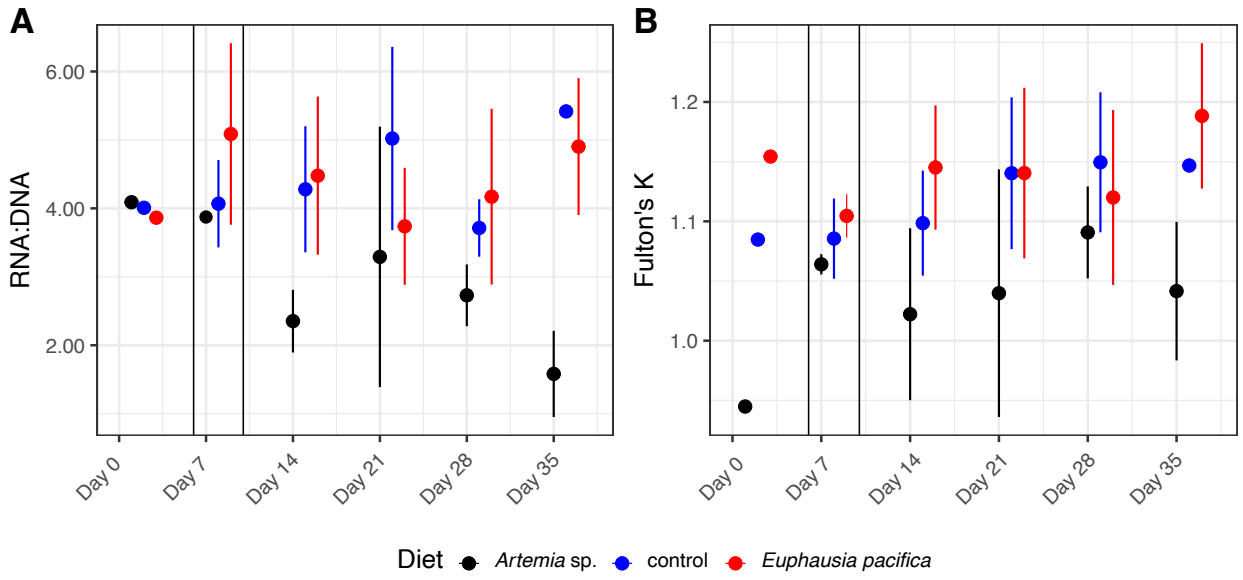


Fig. 2. A) Voluntary feed intake (mean values (± Std Dev) by juvenile Chinook salmon as A) % of body weight dry matter over time, and B) average voluntary feed intake for each diet treatment during Phase 3.

3.1 Fish condition

Chinook salmon RNA:DNA ratios did not differ significantly among the three diet treatments in Phase 1 and Phase 2 (weaning) (Fig 3A, Table 2, Table S2). When fish transitioned from Phase 2 into Phase 3 (feeding study), within the first week, RNA:DNA of *E. pacifica* and control treatments increased by 7.47% and 21.21%, respectively, and *Artemia* sp. treatments RNA:DNA decreased by 47.38% (Fig 3A). Throughout Phase 3, *Artemia* sp. treatments had a significantly lower RNA:DNA ratio (2.68 ± 1.1) than *E. pacifica* treatments (4.43 ± 0.94) and control treatments (4.78 ± 1.11). The RNA:DNA of the *Artemia* sp. treatment declined throughout Phase 3. The *E. pacifica* and control treatments had the highest RNA:DNA ratios and were not significantly different from each other (Table 2, Table S2).



345 **Fig. 3**. Mean values (± Std Dev) of A) RNA:DNA ratios and B) Fulton's K condition
346 index across the experimental period.
347
348
349 **Table 2.** Analysis of deviance of linear mixed models based on full models testing
350 effects of diet treatments over time on juvenile Chinook with the tank as random
351 effects (full linear mixed model results see Table S2. TFA = Total fatty acids, SFA =
352 saturated fatty acids, MUFA = monounsaturated fatty acids, PUFA = polyunsaturated
353 fatty acids, EFA = essential fatty acids, DHA = docosahexaenoic acid, EPA =
354 eicosapentaenoic acid, ALA = alpha-linolenic acid.
355

	Predictor	Chi^2	Df	P value
RNA:DNA	Treatment	77.194	2	<0.001
	Sampling date	10.989	1	<0.001
Fulton K	Treatment	16.959	2	<0.001
	Sampling date	24.902	1	<0.001
TFA	Treatment	5.168	2	0.075
	Sampling date	14.318	1	<0.001
SFA	Treatment	37.997	2	<0.001
	Sampling date	84.812	1	<0.001

MUFA	Treatment	15.099	2	<0.001
	Sampling date	0.642	1	0.422
PUFA	Treatment	28.114	2	<0.001
	Sampling date	15.279	1	<0.001
EFA	Treatment	30.242	2	<0.001
	Sampling date	18.258	1	<0.001
DHA	Treatment	53.184	2	<0.001
	Sampling date	23.060	1	<0.001
EPA	Treatment	29.836	2	<0.001
	Sampling date	58.886	1	<0.001
DHA:EPA	Treatment	37.184	2	<0.001
	Sampling date	40.481	1	<0.001
ALA	Treatment	244.25	2	<0.001
	Sampling date	2.84	1	0.0919

Fulton's K body condition index increased significantly after Phase 2 (Fig. 3B, Table 2). However, this increase was dependent on diet treatment. Fulton's K during Phases 1 and 2 did not differ within or among diet treatments (Fig 3B). When the fish

361 transitioned from Phase 1 into Phase 2, only minor changes in Fulton K were
362 measured (*Artemia sp.*: -1.86%; *E. pacifica*: +1.88%; control: -3.6%). After the
363 transition to Phase 3, after the first week, Fulton's K increased by 5.61% and 2.78%
364 in the *E. pacifica* and control treatments, respectively. Throughout Phase 3, Fulton's
365 K increased significantly in the *E. pacifica* and control treatments (start: control 1.11
366 ± 0.04, *E. pacifica* 1.12 ± 0.04; end: control 1.15 ± 0.05, *E. pacifica* 1.14 ± 0.05).
367 Fulton's K of *Artemia* sp. treatments was significantly lower than *E. pacifica* and
368 control treatments.
369
370 Specific growth rates across Phase 3 were not significantly different among the three
371 diet treatments (Table 3). The average weight increase of fish from the end of Phase
372 2 and the end of Phase 3 was 1.29% d^{-1} ± 0.40 for *Artemia sp.*, 1.35% d^{-1} ± 0.29 for
373 control, and 1.91% d^{-1} ± 0.58 for *E. pacifica* (Fig 4A). Instantaneous mortality rate (Z)
374 did not differ between treatments within experimental feeding Phase 3 ($p = 0.57$) but
375 was highest for *Artemia* sp. treatments (~1.7 vs. ~1.2 for *E. pacifica* and control
376 treatments; Fig 4B). The cumulative mortality was higher in Artemia treatments
377 compared to *E. pacifica* and control treatments (Fig. S1)
378
379 **Table 3.** Analysis of Variance (ANOVA) results of specific growth rates (SGR) of fish
380 from last day Phase 2 to last day of Phase 3; Df = degrees of freedom, Sum Sq =
381 Sum of Squares, Mean Sq = Mean squares).

		df	Sum Sq	Mean Sq	F value	p

SGR weight	Treatment	2	0.8914	0.4457	3.202	0.113
	Residuals	6	0.351	0.1392		

382

383 *3.2 Fatty Acids (FA)*

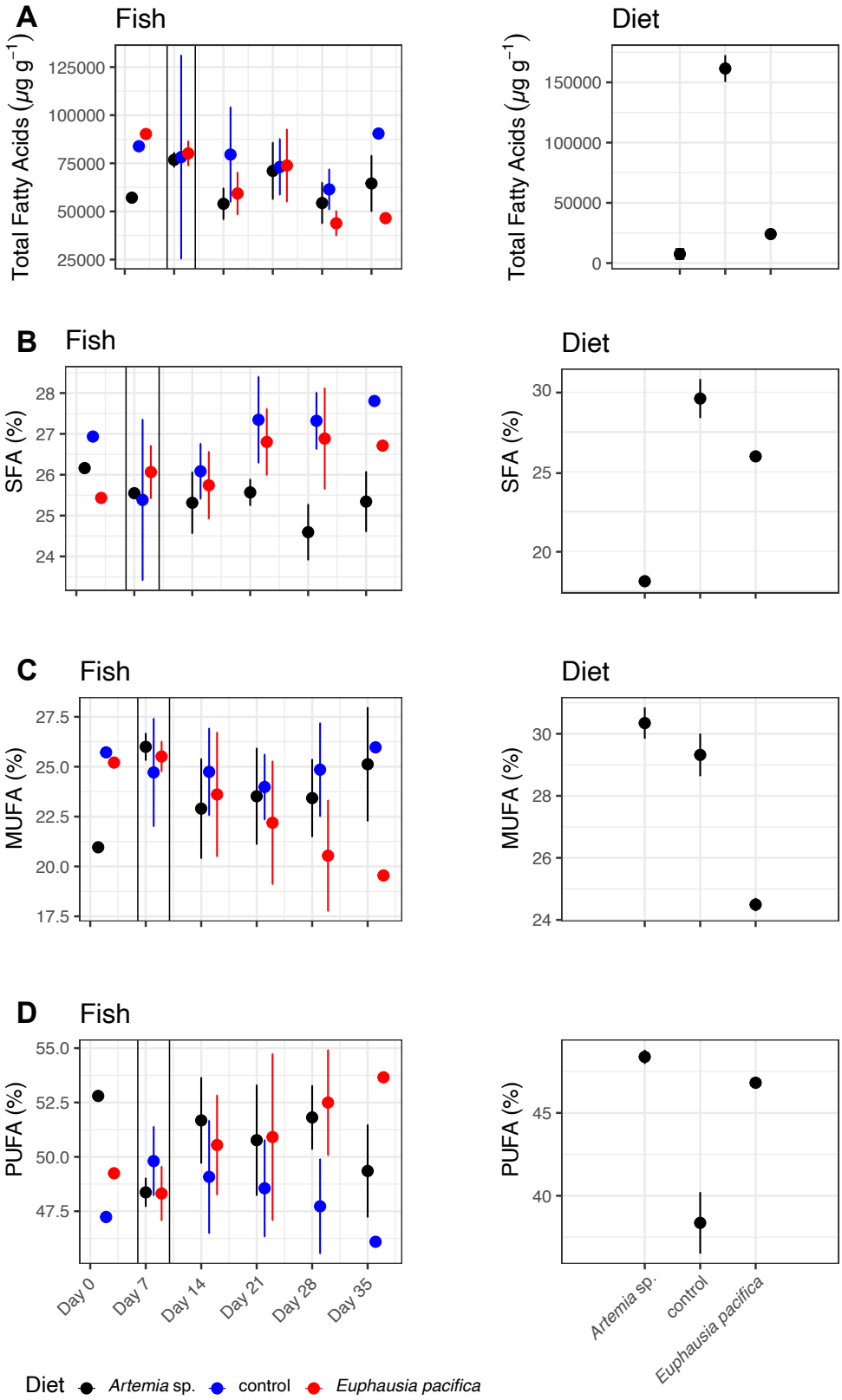
384 The PERMANOVA showed that fish FA profiles were significantly affected by diet
385 during experimental Phase 3 ($p < 0.001$; Table S3). DHA contributed the most
386 (>20%) to the dissimilarity between all treatment pairs and was most remarkable for
387 *E. pacifica* vs control (32%; Table S4). The proportion of ALA in *Artemia sp.* differed
388 by 18% and 18.8% from *E. pacifica* and control, respectively, while oleic acid
389 (C18:1n-9) differed by >12% between *E. pacifica* and control (Table S4). C16:0 was
390 also detected as a contributor to the dissimilarity between control and *E. pacifica* and
391 *Artemia* sp. We excluded this from our further analysis as C16:0 is a typical trophic
392 marker for a fish meal, the main ingredient in the control feed. The total fatty acid
393 concentration in fish did not differ significantly between the three diet treatments (Fig
394 4A, Table 2, Table S2).

395

396 *3.2.1 Fatty acid groups*

397 Fish SFA percentages differed among the three diet treatments (Table 2, Table S2).
398 SFA percentages were significantly higher in *E. pacifica* and control treatments
399 compared to *Artemia* sp. treatments (Fig 4B, Table 2, Table S2). SFA percentage
400 increased significantly from the first day of Phase 3 to the end of Phase 3 in *E.*
401 *pacifica* and control treatments (*E. pacifica*: start 26.75% ± 0.92, end 26.82% ± 0.78;
402 control: start 26.39% ± 0.95, end 27.46% ± 0.77 Fig 4B; Table 2, Table S2). The SFA

403 percentage of fish in *Artemia* sp. treatments decreased in the same phase from
404 26.02% (± 0.81) to 25.47% (± 0.36) (Fig 4C). No significant difference was detected
405 in the MUFA percentage between the different diet treatments (Fig 4C, Table 2,
406 Table S2). PUFA contribution was significantly lower in control treatments (49.79% ±
407 2.11) compared to *Artemia* sp. (51.37% ± 2.52) and *E. pacifica treatments* (50.67% ±
408 3.24; Table 3). PUFA composition remained constant in *Artemia* sp., and *E. pacifica*
409 treatments provided fish during Phase 3 (*E. pacifica*: start - 50.19% ± 3.03, end -
410 50.16% ± 3.08; *Artemia*: start - 50.92 ± 1.85, end - 51.50% ± 3.00; Fig 4D). PUFAs in
411 the control treatment fish decreased slightly from the beginning to the end of Phase
412 3 (49.61% ± 2.01 - 48.60% ± 2.62).
413
414



415

416 **Fig. 4.** Fatty acid composition (mean values ± Std Dev) of Chinook white muscle

417 tissue and diets in each treatment. A) Total fatty acids (TFA), B) saturated fatty acids

418 (SFA), C) monounsaturated fatty acids (MUFA), and D) polyunsaturated fatty acids
419 (PUFA). Mean values.
420
421
422 DHA was significantly higher in *E. pacifica* treatments compared to control and
423 *Artemia* sp. treatments (Table 2, Table S2), whereas DHA in control and *Artemia* sp.
424 treatments were similar (Fig 5A). All fish had a comparable inter-treatment DHA
425 percentage in Phase 2 (*Artemia sp.*: 30.68% ± 2.59; control: 29.25% ± 3.24; *E.*
426 *pacifica*: 30.27% ± 1.87) before the fish were switched to their experimental diets.
427 One week after the diet switch, all treatments slightly increased their DHA (*Artemia*
428 *sp.*: +0.74%; control: 0.94%; *E. pacifica*: +1.59%). Over Phase 3, DHA slightly
429 increased in the *E. pacifica* treatment (start 31.86% ± 4.07, end 32.42% ± 4.14),
430 whereas DHA in control and *Artemia sp.* treatments slightly decreased over time
431 (control: start 30.19% ± 2.24, end 29.37% ± 3.91; *Artemia sp.*: start 31.42% ± 2.97,
432 end 27.95%± 4.4; Fig 5A).
433
434 During Phase 3, EPA decreased in *Artemia* sp. treatments (0.81%), control
435 treatments (0.18%), and *E. pacifica* treatments (0.47%) within one week (Fig. 5B).
436 The EPA percentage in *Artemia* sp. treatments decreased slightly over Phase 3,
437 from 5.89% ± 0.31 to 4.93% ± 0.30, whereas in the *E. pacifica* and control
438 treatments, EPA was constant (*E. pacifica*: start 6.40% ± 0.51, end 6.64% ±0.63;
439 control: start 6.94% ±0.60, end 7.10% ±0.61; Fig. 5B).
440
441 ALA percentage was significantly higher in the *Artemia* sp. treatment compared to *E.*
442 *pacifica* and control treatments (Table 2, Table S2; Fig. 5C). During Phase 2, ALA in

all treatments was similar (*Artemia sp.*: 1.27% ± 0.07; control: 0.99% ± 0.15; *E. pacifica*: 0.99% ± 0.07). ALA increased in *Artemia* sp. after the complete diet switch at the beginning of Phase 3 (2.27%), but *E. pacifica* and control treatments remained relatively constant. ALA percentage increased over time during Phase 3 in *Artemia* sp. treatments (start 2.27% ± 0.65, end 6.72% ± 1.46). In *E. pacifica* treatments, the ALA percentage increased slightly over time (start 1.03% ± 0.10, end 1.25% ± 0.15), and in control treatments, the ALA percentage remained constant over time (start 0.91% ± 0.81, end 0.91% ± 0.11; Fig 5C).

DHA:EPA ratios were significantly affected by time (Table 2, Table S2). During Phase 2, DHA:EPA ratios were comparable among treatments (*Artemia sp.*: 4.64 ± 0.74; control: 4.18 ± 0.83; *E. pacifica*: 4.42 ± 0.41; Fig. 5D). After transitioning into Phase 3, DHA:EPA ratios increased in *Artemia* sp. and *E. pacifica* treatments (+0.61) (Fig 5D). The DHA:EPA ratio in the *Artemia* sp. and *E. pacifica* treatments increased slightly over experimental Phase 3 (Fig 5D). DHA:EPA ratios in the control treatment were significantly lower than *Artemia* sp. and *E. pacifica* and decreased slightly over Phase 3 (4.37 ± 0.47 - 4.19 ± 0.85). DHA:EPA ratios were highest in *Artemia*-fed Chinook in Phase 3 and slightly increased over time (start 5.34 ± 0.61, end 5.72 ± 1.21). *E. pacifica* treatments had a slightly lower DHA:EPA ratio than the *Artemia* sp. treatment (start 5.04 ± 0.98, end 4.96 ± 1.01).

We found that fish did not bioaccumulate ALA (all prey treatments) and EPA (control and *E. pacifica*), as the percentage of these fatty acids was lower in the tissues of the fish than in their diets (Table 4). In contrast, fish in all treatments bioaccumulated DHA, and the bioaccumulation factor was significantly higher in DHA deficient

468 *Artemia* sp. diet than in the control or *E. pacifica* treatments. Despite the higher
469 bioaccumulation, the relative concentration of DHA was significantly lower in muscle
470 tissue of DHA-deprived *Artemia* sp. treatments than in the control and *E. pacifica*
471 treatments.

472

473 **Table 4.** Mean bioaccumulation (± SD) of DHA, EPA and ALA across all three diet
474 treatments over the experimental phase Phase 3.

475

	***Artemia* sp.**	**Control**	***Euphausia pacifica***
DHA	**40.894 (**± 7.460)	**2.874 (**± 0.356)	**1.945 (**±0.255)
EPA	**1.956 (**± 0.122)	**0.486 (**± 0.031)	**0.412 (**± 0.039)
ALA	**0.212 (**±0.053)	**0.296 (**± 0.039)	**0.432 (**± 0.062)

476

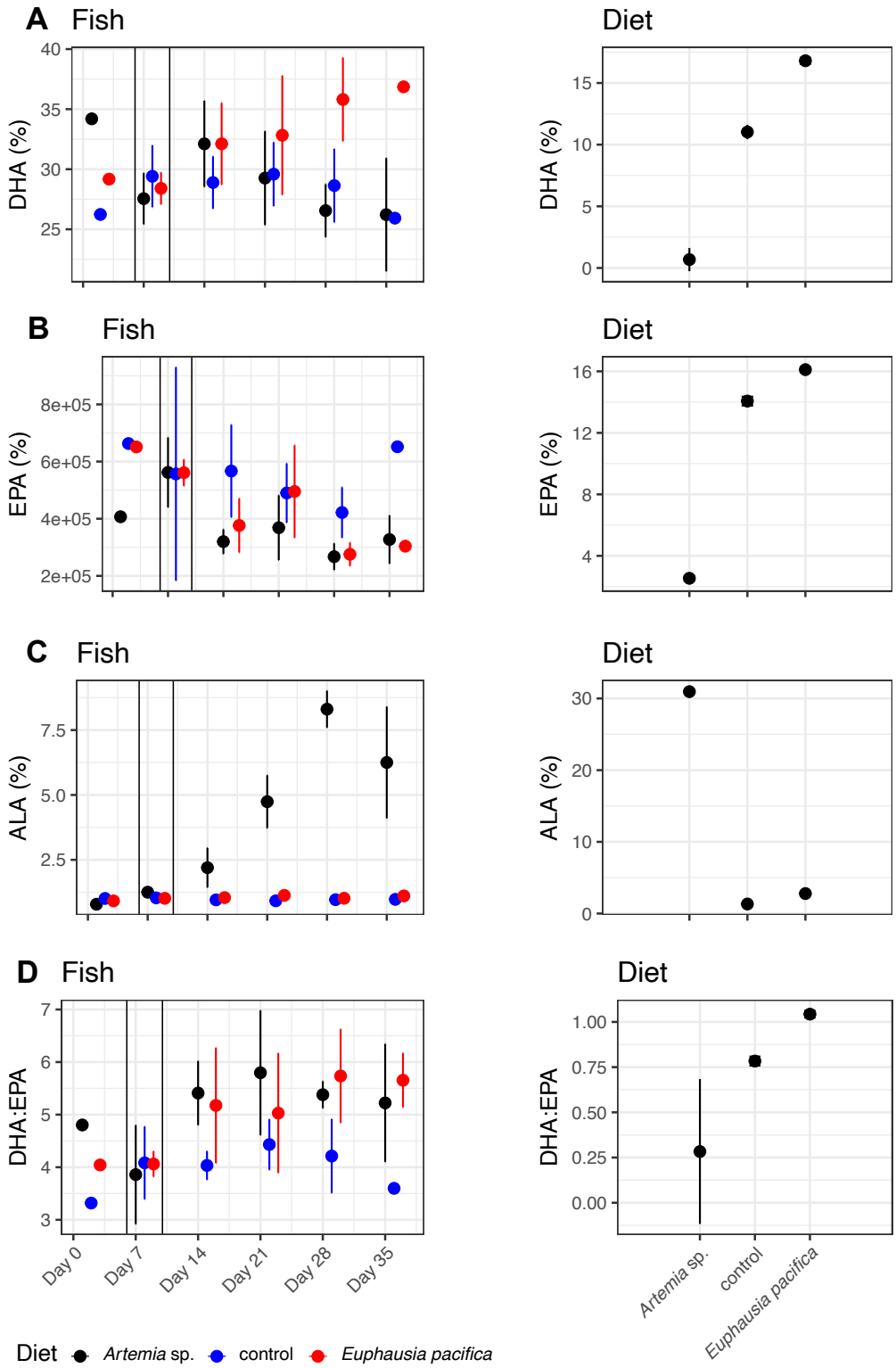


Fig. 5. Mean fatty acid percentual composition (± Std Dev) compared to TFA in Chinook white muscle tissue and provided diet treatments. A) DHA, B) EPA, C), ALA, D) DHA:EPA.

481 **4. Discussion**

482

483 Our results show that low dietary concentrations of DHA and EPA and low DHA:EPA
484 ratios induced by experimental treatment resulted in reduced growth and condition
485 compared to a control diet and the present dominant prey species, *Euphausia*
486 *pacifica* (Duguid, 2021). Furthermore, low concentrations of DHA and EPA and low
487 DHA:EPA ratios were not compensated for by higher food quantity intake. This
488 indicates that food quality needs to be considered when assessing fish response to
489 changing ocean conditions. Below we evaluate the response of juvenile Chinook
490 salmon to individual fatty composition as measured by condition factor, RNA:DNA
491 ratio and growth rate. Finally, we assess the implications for the response of fish to
492 climate-driven changes in zooplankton communities.

493

494 Dietary DHA:EPA ratios >2 are considered optimal for larval and juvenile marine fish
495 (Bell et al., 1995; Watanabe, 1993), and ratios of ~1 are sufficient in many species
496 (Copeman and Laurel, 2010; Rodriguez et al., 1997). *E. pacifica* was ~1, the control
497 diet ~0.75, and *Artemia* sp. below with ~0.25 (representing diets such as crab larvae
498 and polychaetes; Hiltunen et al. (2021)). Based on data for chum, coho, and Atlantic
499 salmon, SFA and MUFA in diets should be approximately 33% (Turchini et al.,
500 2009). All diets in the present study were below 33% in SFA and MUFA,
501 respectively. Additionally, essential fatty acids, the sum of DHA, EPA, ALA, and
502 linolenic acid, should contribute >1-2% of the diets' dry weight (Tocher, 2010). In the
503 present study, the contribution of EFA was highest in *Artemia* sp. and lowest in *E.*
504 *pacifica* and the commercial control. Still, the individual FA composition varied
505 significantly among feeds. *Artemia* sp. had the highest ALA concentration,

contributing to the significantly higher amount of EFAs than *E. pacifica* and control diets, which had lower ALA concentrations. SFA was significantly lower in *Artemia* sp. than in both *E. pacifica* and control diets. These differences were related to specific growth and FA composition responses in the experimental fish.

The bioaccumulation factor of DHA in all diet treatments was higher than one and about 40 times higher in salmon fed with DHA-deprived *Artemia* sp. than the control and the *E. pacifica* fish. In contrast, differences in EPA and ALA bioaccumulation factors between the diet treatments were not retained in the salmon tissues; only the bioaccumulation factor for EPA fed with *Artemia* sp. was ~1.9. This suggests that dietary EPA and ALA were not retained in the salmon tissues but instead likely used for cell energy synthesis by fish (Kainz et al., 2006; Závorka et al., 2021). Despite the high bioaccumulation factor, DHA was low in salmon of *Artemia* treatments, associated with lower growth rates, low body condition (RNA:DNA ratios, Fulton's K), and high mortality rates. This finding corresponds with previous studies and demonstrates that maintaining a similar DHA content as the control group is costly (Závorka et al., 2021; Taipale et al., 2018; Murray et al., 2014). Závorka et al. (2021) showed that juvenile salmon fed under n-3 PUFA-deprived conditions had lower mitochondrial efficiencies of ATP production in muscle tissues. The fastest metabolic pathway synthesizing adenosine triphosphate (ATP) is from SFA, followed by MUFA, and PUFA by β-oxidation in the peroxisome (Turchini and Francis, 2009). In salmon, EPA is extensively used for β-oxidation when supplied in dietary surplus (Codabaccus et al., 2011; Stubhaug et al., 2005), and DHA is more conserved, irrespective of its dietary concentration (Tocher, 2010). Chinook SFAs in the *Artemia* treatments decreased over time, suggesting that this was related to the low

nutritional SFA requiring ATP synthesis from stored SFA. Conversely, SFAs in *E. pacifica* and control treatments increased over the same experimental phase, indicating a sufficient supply of ATP for growth. We observed that Artemia-fed fish had higher intake rates than the control and *E. pacifica*-fed fish, which might have been a compensation mechanism leading to a higher energy intake to cover higher energy expenses.

The effects of food quality on fish growth demonstrated here and in other studies (Brett and Müller-Navarra, 1997; Malzahn et al., 2007b) highlight the important role of prey quality in fish condition and how changes in zooplankton due to climate change could affect zooplanktivorous consumers. In the case of zooplanktivorous fish, the quality of zooplankton prey can be determined by 1) the species composition of the zooplankton themselves or 2) by shifts within the zooplankton species due to changes in their phytoplankton prey. The fatty acid composition of zooplankton in the Salish Sea varies substantially among species (Costalago et al., 2020; Hiltunen et al., 2021). The relative proportion of these prey taxa can vary with ocean conditions, including whole community shifts from lipid-rich boreal species during cold conditions to lipid-poor species, including more gelatinous plankton, during warm conditions (El-Sabaawi et al., 2009a; Mackas et al., 2013). Such environmentally-driven shifts have been demonstrated to occur over decadal scales and in rapid response to recent marine heatwaves (Fisher et al., 2015; Miller et al., 2017; Peterson et al., 2017; Young et al., 2019). While distributional shifts in zooplankton are expected to be important, changes in their phytoplankton food base have also been demonstrated to be an important factor in their FA composition (El-Sabaawi et al., 2009a, 2009b). EFAs are primarily synthesized by phytoplankton and

transferred through the food web via zooplankton to higher trophic levels. Phytoplankton have taxon-specific FA profiles; for example, diatoms have higher proportions of EPA than DHA, while dinoflagellates have higher proportions of DHA than EPA (Dalsgaard et al., 2003).

Fatty acid composition within species can also be affected by environmental conditions. For example, experimental studies showed that nitrogen and phosphorus limitation could increase TFA, SFA and n-3 and n-6 FA concentrations in phytoplankton (Bi et al., 2018; Malzahn et al., 2007a; Schoo et al., 2014); warming can increase the SFA to PUFA ratio (Hixson and Arts, 2016; Jin et al., 2020); and ocean acidification leads to reduced concentrations of TFA and PUFA (Meyers et al., 2019), but also that changes are species-specific and change with community species composition (Dörner et al., 2020; Leu et al., 2013). Such changes in nutritional quality at the base of the food web have been demonstrated to transfer to higher trophic levels, affecting consumers, e.g., when phytoplankton has higher concentrations of DHA and EPA, the same can be measured in zooplankton and secondary consumers show increased nutritional condition (RNA/DNA) (Malzahn et al., 2007a).

5. Conclusions

This study found that high dietary concentrations of DHA and EPA and high DHA:EPA ratios promoted the growth and condition of juvenile Chinook salmon. Furthermore, poor food quality, characterized by low concentrations of DHA and EPA and low DHA:EPA ratios, was not compensated for by increased food quantity. Such changes in food quality are potentially an important mechanism behind the

response of zooplanktivorous fish to climate change. This highlights the importance of considering food quality when assessing fish response to changing ocean conditions. We recommend that future studies consider food quality when determining foraging conditions for juvenile salmon. Furthermore, species-specific data on zooplankton nutrition could be used to qualify zooplankton time series retrospectively to assess how foraging conditions have changed over time. Additionally, testing only various DHA:EPA ratios or concentrations with comparable base feed would be beneficial to understand the effect on juvenile Chinook growth and condition.

Acknowledgements

We gratefully acknowledge the contributions of Lauren Porter and Katarina Doughty. L. Porter provided technical assistance and supported the experiment during sampling, feeding, and dissections. K. Doughty assisted with the fatty acid analyses, the experimental setup, and fish acclimation phase. Their work was essential to the quality of the results presented in this paper.

Author contributions

Conceptualization: J.G, I.F, D.C., B.P.V.H.; Methodology: J.G., C.G.; Validation: J.G.; Data curation: J.G, C.G.; Writing – original draft: J.G.; Writing- review & editing J.G, I.F., C.G, D.C., B.P.V.H.; Supervision: B.P.V.H., I.F.; Project administration: B.P.V.H., I.F.; Funding acquisition: B.V.P.H., I.F.

Funding

This study was supported by a Fisheries and Oceans Canada Strategic Program for Ecosystem-Based Research and Advice (SPERA) grant. The Tula-Mitacs Canada Grants IT09911 and IT13677 supported J. Garzke.

References

Ahlgren, G., Nieuwerburgh, L.V., Wänstrand, I., Pedersén, M., Boberg, M., Snoeijs, P., 2005. Can. J. Fish. Aquat. Sci. 62, 2240–2253.

Arts, M.T., Kohler, C.C., 2009. Health and condition in fish: the influence of lipids on membrane competency and immune response, in: Lipids in Aquatic Ecosystems, Lipids in Aquatic Ecosystems. Lipids in Aquatic Ecosystems, pp. 237–256.

Azani, N., Ghaffar, M.A., Suhaimi, H., Azra, M.N., Hassan, M.M., Jung, L.H., Rasdi, N.W., 2021. Iop Conf Ser Earth Environ Sci 869, 012005.

Beamish, R.J., Mahnken, C., Neville, C.M., 2004. T. Am. Fish. Soc. 133, 26–33.

Beaugrand, G., C., R., P., Ibanez, F., Lindley, J.A., Edwards, M., 2002. Science 296, 1692–1694.

Bell, M.V., Batty, R.S., Dick, J.R., Fretwell, K., Navarro, J.C., Sargent, J.R., 1995. Lipids 30, 443–449.

Benedetti, F., Vogt, M., Elizondo, U.H., Righetti, D., Zimmermann, N.E., Gruber, N., 2021. Nat Commun 12, 5226.

Bi, R., Ismar, S.M.H., Sommer, U., Zhao, M., 2018. Biogeosciences 15, 1029–1045.

Bolker, B.M., Brooks, M.E., Clark, C.J., Geange, S.W., Poulsen, J.R., Stevens, M.H.H., White, J.-S.S., 2009. Trends Ecol. Evol. 24, 127–135.

Brett, M.T., Müller-Navarra, D.C., 1997. Freshwater Biol. 38, 483–499.

Burke, B.J., Peterson, W.T., Beckman, B.R., Morgan, C., Daly, E.A., Litz, M., 2013. PLoS One 8, e54134.

Cheung, W.W.L., Lam, V.W.Y., Sarmiento, J.L., Kearney, K., Watson, R., Pauly, D., 2009. Fish Fish 10, 235–251.

Codabaccus, M.B., Bridle, A.R., Nichols, P.D., Carter, C.G., 2011. British Journal of Nutrition 105, 1772–1782.

Colombo, S.M., Wacker, A., Parrish, C.C., Kainz, M.J., Arts, M.T., 2017. Environ Rev 25, 163–174.

Copeman, L.A., Laurel, B.J., 2010. Mar. Ecol. Prog. Ser. 412, 259–272.

Copeman, L.A., Parrish, C.C., Brown, J.A., Harel, M., 2002. Aquaculture 210, 285–304.

Costalago, D., Forster, I., Nemcek, N., Neville, C., Perry, R.I., Young, K., Hunt, B.P.V., 2020. Sci. Rep. 10, 1–12.

Dalsgaard, J., John, M.S., Kattner, G., Müller-Navarra, D., Hagen, W., 2003. Adv. Mar. Biol. 46, 225–340.

Daly, E.A., Benkwitt, C.E., Brodeur, R.D., Litz, M.N.C., Copeman, L.A., 2010. Mar. Biol. 157, 1975–1987.

Dörner, I., Hauss, H., Aberle, N., Lohbeck, K., Spisla, C., Riebesell, U., Ismar-Rebitz, S.M.H., 2020. Mar. Ecol. Prog. Ser. 647, 49–64.

Duguid, W., 2021. Fine-scale structure in the ecology of juvenile Chinook Salmon at sea. University of Victoria, Canada.

El-Sabaawi, R., Dower, J., Kainz, M., Mazumder, A., 2009a. Mar. Biol. 156, 225–237.

El-Sabaawi, R., Dower, J.F., Kainz, M., Mazumder, A., 2009b. Mar. Ecol. Prog. Ser. 382, 151–161.

Fisher, J.L., Peterson, W.T., Rykaczewski, R.R., 2015. Global Change Biology 21, 4401–4414.

Fulton, T.W., 1904. Fisheries Board of Scotland 141–241.

Garzke, J., Forster, I., Godwin, S.C., Johnson, B.T., Krkošek, M., Mahara, N., Pakhomov, E.A., Rogers, L.A., Hunt, B.P.V., 2022. Facets 7, 1328–1347.

Garzke, J., Hansen, T., Ismar, S.M.H., Sommer, U., 2016. PLoS One 11, e0155952 EP-.

Garzke, J., Ismar, S.M.H., Sommer, U., 2015. Oecologia 177, 849–860.

Heneghan, R.F., Everett, J.D., Blanchard, J.L., Sykes, P., Richardson, A.J., 2023. Nat Clim Change 1–8.

Hiltunen, M., Strandberg, U., Brett, M.T., Winans, A.K., Beauchamp, D.A., Kotila, M., Keister, J.E., 2021. Estuaries and Coasts 1–15.

Hixson, S.M., Arts, M.T., 2016. Global Change Biology 22, 2744–2755.

Hoegh-Guldberg, O., Jacob, D., Taylor, M., Bolaños, T.G., Bindi, M., Brown, S., Camilloni, I.A., Diedhiou, A., Djalante, R., Ebi, K., Engelbrecht, F., Guiot, J., Hijioka, Y., Mehrotra, S., Hope, C.W., Payne, A.J., Pörtner, H.-O., Seneviratne, S.I., Thomas, A., Warren, R., Zhou, G., 2019. Science 365.

Holm, H.C., Fredricks, H.F., Bent, S.M., Lowenstein, D.P., Ossolinski, J.E., Becker, K.W., Johnson, W.M., Schrage, K., Mooy, B.A.S.V., 2022. Science 376, 1487–1491.

IPCC, 2021. 2021: Climate Change 2021: The Physical Science Basis. Contribution of Working Group I to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press.

Jin, P., Gonzalez, G., Agusti, S., 2020. Evol. Appl. 13, 2497–2506.

Kassambara, A., 2020. rstatix: Pipe-Friendly Framework for Basic Statistical Tests, R. R.

Lee, R., Hagen, W., Kattner, G., 2006. Mar Ecol Prog Ser 307, 273–306.

Leu, E., Daase, M., Schulz, K.G., Stuhr, A., Riebesell, U., 2013. Biogeosciences 10, 1143–1153.

Litz, M.N.C., Miller, J.A., Copeman, L.A., Hurst, T.P., 2017. J. Exp. Mar. Biol. Ecol. 494, 20–31.

Lotze, H.K., Coll, M., Dunne, J.A., 2010. Ecosystems 14, 198–222.

Mackas, D., Galbraith, M., Faust, D., Masson, D., Young, K., Shaw, W., Romaine, S., Trudel, M., Dower, J., Campbell, R., Sastri, A., Pechter, E.A.B., Pakhomov, E., El-Sabaawi, R., 2013. Prog. Oceanogr. 115, 129–159.

Mackas, D.L., Batten, S., Trudel, M., 2007. Prog. Oceanogr. 75, 223–252.

Mahara, N., Pakhomov, E.A., Jackson, J.M., Hunt, B.P., 2019. J. Plankton Res. 41, 309–328.

Malzahn, A.M., Aberle, N., Clemmesen, C., Boersma, M., 2007a. Limnol Oceanogr 52, 2062–2071.

Malzahn, A.M., Clemmesen, C., Wiltshire, K.H., Laakmann, S., Boersma, M., 2007b. Mar. Ecol. Prog. Ser. 334, 205–212.

McGinty, N., Barton, A.D., Record, N.R., Finkel, Z.V., Johns, D.G., Stock, C.A., Irwin, A.J., 2021. Global Change Biol 27, 1431–1442.

Meyers, M.T., Cochlan, W.P., Carpenter, E.J., Kimmerer, W.J., 2019. PLoS One 14.

Miller, J.A., Peterson, W.T., Copeman, L.A., Du, X., Morgan, C.A., Litz, M.N.C., 2017. Prog. Oceanogr. 155, 1–12.

Miller, J.A., Teel, D.J., Baptista, A., Morgan, C.A., 2013. Can. J. Fish. Aquat. Sci. 70, 617–629.

Miller, J.A., Teel, D.J., Peterson, W.T., Baptista, A.M., 2014. PLoS One 9, e99814.

Oksanen, J., Blanchet, F.G., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin, P.R., O'Hara, R.B., Simpson, G.L., Solymos, P., Stevens, H.H., Szoecs,

E., Wagner, H., 2017. Vegan: Community Ecology Package. R package Version 2.4-3., R Program. R Program.

Olsen, Y., 1999. Lipids and Essential Fatty Acids in Aquatic Food Webs: What Can Freshwater Ecologists Learn from Mariculture?, in: Lipids in Freshwater Ecosystems, Lipids in Freshwater Ecosystems. Lipids in Freshwater Ecosystems, pp. 161–202.

Parmesan, C., Yohe, G., 2003. Nature 421, 37–42.

Parrish, C.C., 2009. Essential fatty acids in aquatic food webs, in: Lipids in Aquatic Ecosystems, Lipids in Aquatic Ecosystems. Lipids in Aquatic Ecosystems, pp. 309–326.

Parrish, C.C., French, V.M., Whiticar, M.J., 2012. J. Plankton Res. 34, 356–375.

Paulsen, M., Clemmesen, C., Malzahn, A.M., 2014. Mar Biol 161, 239–244.

Pearcy, W.G., McKinnell, S.M., 2007. Am. Fish. S. S. 57.

Peterson, W.T., Fisher, J.L., Strub, P.T., Du, X., Risien, C., Peterson, J., Shaw, C.T., 2017. J. Geophys. Res.-Oceans 122, 7267–7290.

Poloczanska, E.S., Brown, C.J., Sydeman, W.J., Kiessling, W., Schoeman, D.S., Moore, P.J., Brander, K., Bruno, J.F., Buckley, L.B., Burrows, M.T., Duarte, C.M., Halpern, B.S., Holding, J., Kappel, C.V., O'Connor, M.I., Pandolfi, J.M., Parmesan, C., Schwing, F., Thompson, S.A., Richardson, A.J., 2013. Nat Clim Change 3, 919–925.

Pontavice, H. du, 2019. 121–128.

Pontavice, H., Gascuel, D., Reygondeau, G., Maureaud, A., Cheung, W.W.L., 2020. Global Change Biol 26, 1306–1318.

Preikshot, D., Beamish, R.J., Sweeting, R.M., 2010. Changes in the diet composition of juvenile sockeye salmon in the Strait of Georgia from the 1960s to the present, North Pacific Anadromous Fish Comission. NPAFC.

Puttick, D., Dauk, M., Lozinsky, S., Smith, M.A., 2009. Lipids 44, 753–757.

Record, N.R., Ji, R., Maps, F., Varpe, Ø., Runge, J.A., Petrik, C.M., Johns, D., 2018. J Biogeogr 45, 2238–2251.

Reitan, K.I., Rainuzzo, J.R., Oie, G., Olsen, Y., 1997. Aquaculture 155, 207–221.

Rodriguez, C., Pérez, J.A., Diaz, M., Izquierdo, M.S., Fernandez-Palacios, H., Lorenzo, A., 1997. Aquaculture 150, 77–89.

Sahota, R., Boyen, J., Semmouri, I., Bodé, S., Troch, M.D., 2022. Mar. Biol. 169, 133.

Schoo, K.L., Aberle, N., Malzahn, A.M., Schmalenbach, I., Boersma, M., 2014. Oecologia 174, 581–594.

Stevens, C., Sahota, R., Galbraith, M., Venello, T., Bazinet, A., Hennekes, M., Yongblah, K., Juniper, S., 2022. Mar Ecol Prog Ser 687, 43–64.

Stubhaug, I., Frøyland, L., Torstensen, B.E., 2005. Lipids 40, 39–47.

Sundby, S., Bjørke, H., Soldal, A.V., Olsen, S., 1989. Rapports et procès-verbaux des Réunions. Conseil permanent international pour l'Exploration de la Mer 191, 351–358.

Tocher, D.R., 2010. Aquaculture Research 41, 717–732.

Turchini, G.M., Francis, D.S., 2009. British Journal of Nutrition 102, 69–81.

Turchini, G.M., Torstensen, B.E., Ng, W.K., 2009. Reviews in Aquaculture 1, 10–57.

Ullah, H., Nagelkerken, I., Goldenberg, S.U., Fordham, D.A., 2018. Plos Biol 16, e2003446.

Vasseur, D.A., McCann, K.S., 2007. The Impact of Environmental Variability on Ecological Systems, The Peter Yodzis Fundamental Ecology Series. Springer.

Walther, G.R., Post, E., Convey, P., Menzel, A., Parmesan, C., Beebee, T.J.C., Fromentin, J.M., Hoegh-Guldberg, O., Bairlein, F., 2002. Nature 416, 389–395.

Watanabe, T., 1993. Journal of the World Aquaculture Society 24, 152–161.

Young, K., Galbraith, M., Perry, I., 2019. State of the Physical, Biological and selected Fishery Resources of Pacific Canadian marine Ecosystems in 2018, Canadian Technical Report of Fisheries and Aquatic Sciences. Can. Tech. Rep. Fish. Aquat. Sci.

Yu, T.C., Sinnhuber, R.O., 1979. Aquaculture 16, 31–38.

765 **Supplementary material**

766 **Table S1.** Feed composition (producer)

767

	Ingredients	Guaranteed Analysis from producer		Our analysis
Artemia	Brine shrimp, spirulina, carrageenan	Crude Protein (min) Crude Fat (min) Crude Fiber (max) Moisture (max) Ash (max) Phosphorus (min)	3.7% 1.2% 0.2% 94.8% 1.6% 0.1%	91.63 % water 20.440 kJ/g 4.881 kcal/g
Bio-Oregon BioVita	Fish meal, fish oil, vitamins, minerals, natural astaxanthin, beta-glucans	Protein (min) Oil (min) Fiber (max) Moisture (max) Ash (max) DE (MJ/kg)	50% 22% 1.0% 8.5% 13% 19.6	5.79 % water 23.676 kJ/g 5.654 kcal/g
Krill Canada	Krill (*E. superba*)	Protein (min) Fat (min) Crude Fiber (max) Moisture (max) Ash (max) KJ Calories Carbohydrates	58% (55.06 g/100g) 18% (13.6 g/100g) 6% (3.1g/100g) 10% (7.6 g/100g) 13% (8.5 g/100g) 1692 (per 100g) 404 (per 100g) 15.6 (g/100g)	82% water 19.557 kJ/g 4.671 kcal/g

768

Table S2 Full statistical analysis of linear mixed models. TFA = Total fatty acids, SFA = saturated fatty acids, MUFA = monounsaturated fatty acids, PUFA = polyunsaturated fatty acids, EFA = essential fatty acids, DHA = docosahexaenoic acid, EPA = eicosapentaenoic acid, ALA = alpha-linolenic acid

		Predictor				Random Effects			N	Marginal R^2
		Intercept	Control	Krill	Sampling date	σ^2	t_{Tank}	N_{Tank}		
RNA/DNA	Estimates	261.77	1.27	1.21	-0.01	1.19	0.00	9	254	0.268
	CI	145.08 – 378.46	0.86 – 1.68	0.79 – 1.62	-0.02 – -0.01					
	p-value	**<0.001**	**<0.001**	**<0.001**	**<0.001**					
	df	249.21	5.99	5.72	249.22					
Fulton K	Estimates	-19.06	0.05	0.03	0.00	0.00	0.00	9	204	0.168
	CI	-27.08 – -11.03	0.02 – 0.08	0.00 – 0.06	0.00 – 0.00					
	p-value	**<0.001**	**0.007**	**0.041**	**<0.001**					
	df	200.00	5.98	5.09	200.00					
TFA	Estimates	5483.10	5.63	-4.07	-0.31	731.01	2.26	9	2.64	0.072
	CI	2652.09 – 8314.10	-4.86 – 16.11	-14.57 – 6.44	-0.47 – -0.15					
	p-value	**<0.001**	0.238	0.376	**<0.001**					
	df	258.58	6.10	5.64	258.59					
SFA	Estimates	-414.27	0.97	0.65	0.02	0.81	0.01	9	264	0.340
	CI	-508.71 – -319.82	0.58 – 1.36	0.26 – 1.05	0.02 – 0.03					
	p-value	**<0.001**	**0.001**	**0.007**	**<0.001**					
	df	257.96	6.09	5.74	257.97					

MUFA	Estimates	-86.86	0.96	-0.55	0.01	6.76	0.00	9	264	0.056
	CI	-359.05 – 185.27	-0.00 – 1.93	-1.52 – 0.41	-0.01 – 0.02					
	p-value	0.530	0.051	0.208	0.426					
	df	258.80	6.11	5.60	258.81					
PUFA	Estimates	605.93	-1.92	-0.08	-0.03	7.21	0.00	9	264	0.139
	CI	324.95 – 886.92	-2.92 – -0.92	-1.08 – 0.92	-0.05 – -0.02					
	p-value	**<0.001**	**0.003**	0.856	**<0.001**					
	df	258.80	6.11	5.60	258.81					
EFA	Estimates	685.73	-1.82	0.41	-0.04	803	0.00	9	264	0.152
	CI	389.06 – 982.40	-2.87 – -0.77	-0.64 – 1.46	-0.05 – -0.02					
	p-value	**<0.001**	**0.005**	0.375	**<0.001**					
	df	258.80	6.11	5.60	258.81					
DHA	Estimates	1004.45	0.09	3.67	-0.06	14.73	0.00	9	264	0.220
	CI	602.68 – 1406.21	-1.33 – 1.52	2.24 – 5.10	-0.08 – -0.03					
	p-value	**<0.001**	0.880	**0.001**	**<0.001**					
	df	258.80	6.11	5.60	258.81					
EPA	Estimates	-375.81	0.71	0.05	0.02	0.89	0.00	9	2.64	0.248
	CI	-474.33 – -277.29	0.36 – 1.05	-0.30 – 0.40	0.02 – 0.03					
	p-value	**<0.001**	**0.003**	0.715	**<0.001**					
	df	258.80	6.11	5.60	258.81					

DHA/EPA	Estimates	341.14	-0.62	0.29	-0.02	1.00	0.00	9	264	0.222
	CI	236.57 – 445.71	-0.99 – -0.25	-0.08 – 0.66	-0.02 – -0.01					
	p-value	**<0.001**	**0.006**	0.104	**<0.001**					
	df	258.80	6.11	5.60	258.81					
ALA	Estimates	-102.60	-2.54	-2.29	0.01	1.41	0.00	9	264	0.484
	CI	-227.03 – 21.84	-2.98 – -2.09	-2.74 – -1.85	-0.00 – 0.01					
	p-value	0.106	**<0.001**	**<0.001**	0.094					
	df	258.80	6.11	5.60	258.81					

772

Table S3. PERMANOVA of fatty acids of PHASE (experimental feeding Phase) of fish permutations = 999, method = Bray Curtis

	Df	Sum of Sqs	R2	F	p-value
Treatment	2	0.0765	0.4333	45.112	0.001
Residual	118	0.1001	0.5667		
Total	120	0.1766	1.0000		

779 **Table S4.** SIMPER Cumulative contributions of most influential fatty acids.

780

Artemia vs E. pacifica	C22:6-n3 (DHA)	C18:3-n3 (ALA)	C16:0	C18:1-n9	C16:1	
Cumulative %	0.2785	0.4579	0.5521	0.6456	0.7072	
% dissimilarity	27.85%	17.94%	10.12%	9.35%	6.16%	
Artemia vs. control	C22:6-n3 (DHA)	C18:3-n3 (ALA)	C16:1	C16:0	C18:1-n9	C20:5-n3 (EPA)
	0.2019	0.3903	0.4876	0.5774	0.6649	0.7452
	20.19%	18.84%	9.73%	8.81%	8.75%	8.03%
E. pacifica vs. control	C22:6-n3 (DHA)	C18:1-n9	C16:1	C16:0	C18:2-n6	
	0.3224	0.4487	0.5712	0.6373	0.7005	
	32.24%	12.63%	12.25%	6.61%	6.32%	

781

782

783

784

785

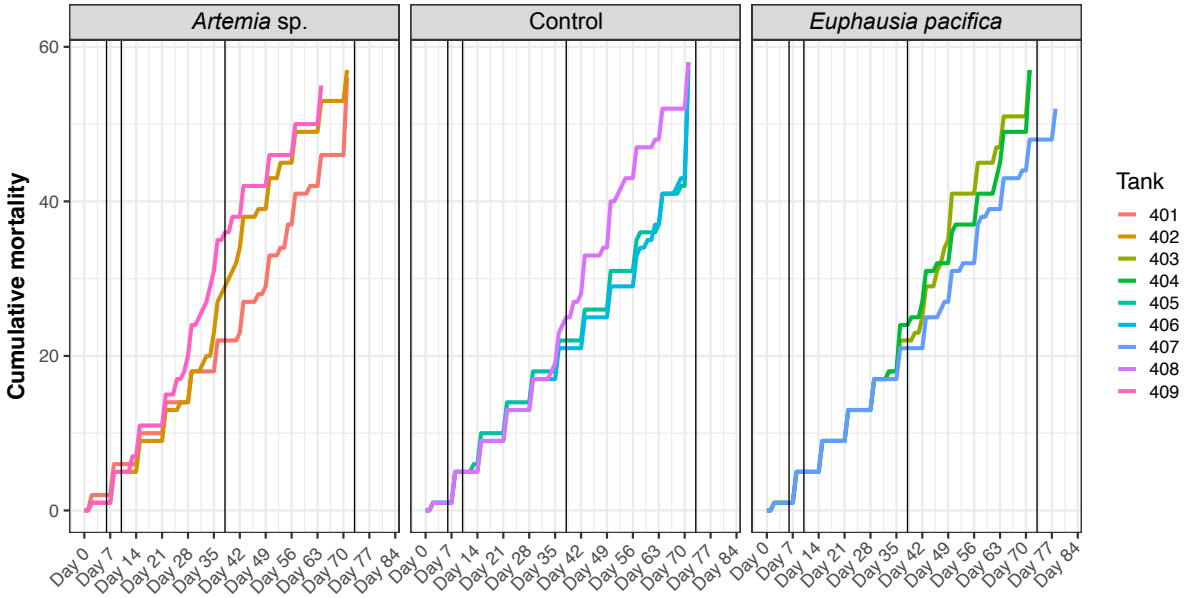



Figure S1. Cumulative mortality of fish in the different treatments (*Artemia* sp., control, *Euphausia pacifica*) over time.