Title

Antagonistic effects of temperature and dissolved organic carbon on fish growth in California mountain lakes

Permalink

Journal

ISSN

Authors

Symons, Celia C
Schulhof, Marika A
Cavalheri, Hamanda B
et al.

Publication Date

2019

DOI

Peer reviewed

GLOBAL CHANGE ECOLOGY – ORIGINAL RESEARCH

CrossMark

Antagonistic effects of temperature and dissolved organic carbon on fish growth in California mountain lakes

Celia C. Symons[1] · Marika A. Schulhof[1] · Hamanda B. Cavalheri[1] · Jonathan B. Shurin[1]

Abstract
Resources and temperature play major roles in determining biological production in lake ecosystems. Lakes have been warming and 'browning' over recent decades due to climate change and increased loading of terrestrial organic matter. Conflicting hypotheses and evidence have been presented about whether these changes will increase or decrease fish growth within lakes. Most studies have been conducted in low-elevation lakes where terrestrially derived carbon tends to dominate over carbon produced within lakes. Understanding how fish in high-elevation mountain lakes will respond to warming and browning is particularly needed as warming effects are magnified for mountain lakes and treeline is advancing to higher elevations. We sampled 21 trout populations in the Sierra Nevada Mountains of California to examine how body condition and individual growth rates, measured by otolith analysis, varied across independent elevational gradients in temperature and dissolved organic carbon (DOC). We found that fish grew faster at warmer temperatures and higher nitrogen (TN), but slower in high DOC lakes. Additionally, fish showed better body condition in lakes with higher TN, higher elevation and when they exhibited a more terrestrial $\delta^{13}C$ isotopic signature. The future warming and browning of lakes will likely have antagonistic impacts on fish growth, reducing the predicted independent impact of warming and browning alone.

Keywords Allochthony · Brook trout · Rainbow trout · Elevation · Bottom-up control

Introduction

A dominant paradigm in freshwater ecology is that primary and secondary production is largely determined by nutrients, primarily nitrogen and phosphorus (i.e., bottom-up control Hairston et al. 1960). In addition to N and P, dissolved organic carbon (DOC) has emerged as another major determinant of the structure and function of lake ecosystems (Williamson et al. 1999; Pace et al. 2004; Wilkinson et al. 2013; Tanentzap et al. 2017). It is important to understand the role of terrestrial carbon because climate warming is altering the distribution of vegetation and accelerating detrital decomposition, increasing the supply of organic matter to aquatic systems (Evans et al. 2006; Larsen et al. 2011; Monteith et al. 2007). Understanding how carbon subsidies to lakes will influence fish production is critical to predicting how climate change will alter this ecosystem service.

Dissolved organic carbon quantity can have positive or negative effects on secondary production in lakes (Jones et al. 2012; Karlsson et al. 2009). DOC can increase secondary production by blocking harmful UV rays and providing a source of carbon to aquatic micro-organisms that are resources for invertebrates and ultimately fish (Hessen and Andersen 1990; Stasko et al. 2012). However, colored DOC can decrease production by reducing the amount of photosynthetically active radiation (PAR) in the water column, resulting in light limited algae growth, decreasing energy available to higher trophic levels (Jones et al. 2012). A lake survey found that fish abundance was unimodally related to DOC in nutrient-poor lakes (Finstad et al. 2014). However, fish abundance may be a poor indicator of their productivity or growth rates. Two studies of fish growth along DOC gradients found a negative association between DOC

✉ Celia C. Symons
csymons@ucsc.edu

1 Ecology Behavior and Evolution Section, Department of Biological Sciences, University of California, San Diego, La Jolla, CA 92093, USA

concentration and fish growth (Benoît et al. 2016; Karlsson et al. 2009), another found a positive association (Tanentzap et al. 2014), while a whole-lake experiment found no association between fish growth and DOC (Koizumi et al. 2018). These conflicting results suggest that there are unknown interactions with DOC that need to be determined to understand when and how DOC will influence fish production in lakes.

DOC can come from different sources and have distinct effects on lake ecosystems. Algal- and terrestrial-derived DOC differs in chemical characteristics and impacts on food webs. Terrestrially derived DOC contains more recalcitrant, colored compounds which attenuate light, increase bacterial production and suppress phytoplankton production (reviewed in Williamson et al. 1999). Above treeline, atmospheric dust is a significant source of DOC that can stimulate bacterial production (Mladenov et al. 2011), a poor food source for zooplankton (Wetzel 1995). Comparatively, algal-derived DOC is not as colored, and therefore has a reduced shading impact on primary production (Williamson et al. 1999). Less shading allows more phytoplankton production, providing high quality fatty acids for higher trophic levels (Brett et al. 2009). Fish growth in a Canadian Boreal Shield lake was related to both DOC quantity and quality (Tanentzap et al. 2014), suggesting that quantity and quality may exert different effects on food webs (Tanentzap et al. 2017).

In addition to resource availability, fish growth depends on temperature (Elliott 1976). Within lakes, fish growth has been found to be both positively and negatively correlated to variation in temperature (Morrongiello et al. 2011; Guzzo et al. 2017). However, the effect of temperature among populations is shaped by the potential for local adaptive responses (Somero 2010), which can occur over a few generations in fishes (reviewed in Crozier and Hutchings 2014). Determining how fish growth varies over broad temperature gradients will help elucidate the impact of climate change on fish production.

The importance of studying DOC and temperature in mountain lakes

Our study is focused on oligotrophic mountain lakes of the Sierra Nevada (CA). Historically, these lakes were fishless; however, most lakes now contain trout (primarily brook trout *Salvelinus fontinalis* and rainbow trout *Oncorhynchus mykiss*) which were introduced approximately 100 years ago to support recreational fisheries (Knapp et al. 2001).

Mountain lakes tend to be oligotrophic and clear, making them especially sensitive to even small changes in DOC concentrations relative to darker lakes (Stasko et al. 2012), yet they remain underrepresented in the literature examining the effect of DOC on fish (but see Finstad et al. 2014). Similarly, most studies of fish along DOC gradients are in low-elevation lakes, where allochthonous carbon tends to dominate. In mountain systems, DOC quality varies with elevation: low-elevation lakes have relatively greater allochthonous DOC inputs due to the higher abundance of plants below treeline, and high-elevation lakes have relatively greater autochthonous DOC production (Rose et al. 2015; Sadro et al. 2012). High-elevation lakes may be particularly vulnerable to changing DOC because of (1) advancing treeline (increasing allochthonous DOC; Gottfried et al. 2012; Tanentzap et al. 2017), (2) increased precipitation to deliver terrestrial carbon (increasing allochthonous DOC; Sadro and Melack 2012) and (3) greater runoff from glaciers which can stimulate primary producers (increasing autochthonous DOC; Sommaruga 2015). Additionally, UV levels can be biologically damaging at high elevation, therefore the shading effect of DOC may be more beneficial to organisms in alpine lakes compared to low-elevation lakes (Rose et al. 2009). Alpine aquatic ecosystems are also particularly sensitive to climate warming (Hauer et al. 1997), as reduced snowpack and ice cover result in decreased albedo, magnifying the effect of higher air temperatures (Bradley et al. 2004). Determining independent effects of DOC and temperature in mountain lakes has been difficult as DOC and temperature are typically correlated, with both negatively covarying with elevation.

We chose lakes arrayed along independent gradients in temperature and DOC to examine how these variables, among others, influence individual fish growth rates and body condition. Both temperature and DOC are increasing in mountain lakes, yet we do not understand how they will independently or jointly influence fish production. We hypothesized that (1) in lakes with higher DOC concentration, fish growth rates would be higher because DOC can increase energy to higher trophic levels by providing a source of nutrients and reducing UV stress at these low concentrations of DOC and (2) growth rates would be higher in warmer lakes in accordance with the metabolic theory of ecology (Brown et al. 2004).

Materials and methods

We chose 21 lakes arrayed along broad environmental gradients from 2506 to 3337 m elevation in the Sierra Nevada, California, to test the effects of temperature and DOC on fish growth. The mountainous terrain provides a natural elevational gradient in temperature and DOC, while water chemistry shows less variation due to similar underlying geology (Sadro et al. 2012). We chose lakes to have independent gradients in temperature and DOC to determine their individual effects (temperature and DOC correlation $r^2 = 0.01$, $p = 0.29$). Lake fish assemblages are low in diversity because the lakes are naturally fishless and stocked with

trout (Knapp et al. 2001). Brook trout (*Salvelinus fontinalis*), rainbow trout (*Oncorhynchus mykiss*), brown trout (*Salmo trutta*), cutthroat trout (*Oncorhynchus clarkii*) and golden trout (*Oncorhynchus mykiss aguabonita*) were stocked starting in the late 1800s until cessation in the 1970s. Brook trout and rainbow trout are the most common species found in the lakes of the Sierra Nevada (Knapp et al. 2001). We sampled each lake for water chemistry, physical characteristics and fish between 01-Jul-2015 and 05-Sept-2015, and 06-Jun-2016 and 31-Aug-2016 between one and six times per lake.

Lake sampling methods

At the deepest point in each lake, in situ measurements of temperature, conductivity, dissolved oxygen (DO) and pH were taken using a YSI probe (YSI Incorporated, Yellow Springs, Ohio, USA). Surface water samples were filtered through 63-μm mesh to remove zooplankton and processed for chlorophyll-*a* (chl-*a*), particulate organic matter (POM), total nitrogen (TN), total phosphorus (TP) and dissolved organic carbon (DOC). For chl-*a* quantification, a known volume of water was filtered through 0.7 μm glass fiber filters (GF/F Fisher Scientific) and frozen. Chl-*a*, a proxy for phytoplankton biomass, was measured using a fluorometer after a 24 h cold methanol extraction. For POM isotope analysis, a known volume of water was filtered through pre-weighed pre-combusted (7 h, 500 °C) 0.7 μm glass fiber filters. Upon returning to the laboratory, filters were dried for 24 h at 60 °C, weighed and packaged in tin capsules for ^{13}C and ^{15}N isotope analysis. Total nitrogen and total phosphorus samples were collected in HDPE vials and preserved with H_2SO_4 to a pH < 2 and stored at ~ 4 °C until analysis. TN and TP were measured using an auto analyzer (LaChat QuikChem 8500, persulfate digestions). Leaves of several common plant species were collected from shoreline and frozen until processing for isotopic analysis. Leaves were sorted into broad functional groups (grasses, shrubs, pine) and dried at 60 °C for 2 days. A mortar and pestle was used to grind the leaf samples before packaging in tin capsules for isotope analysis. Based on a subset (10 lakes) of the plant data, we chose to process a grass and pine sample to capture the maximum variation in isotopes within the terrestrial organic matter entering lakes.

To quantify DOC, water samples were filtered through pre-combusted glass fiber filters (Whatman GF/F, pore size 0.7 μm) into triple-rinsed 20 mL glass vials and preserved with HCl to a pH < 2. DOC was measured using a total organic carbon analyzer (TOC-V CSN, Shimadzu Scientific Instruments, Japan). To characterize DOC quality, we used UV–Vis absorbance, spectrofluorometry and spectrophotometry, which reflect several aspects of the molecules comprising the light absorbing and fluorescing DOM pool, respectively. We used excitation emission matrices (EEMs) as a three-dimensional representation of fluorescence intensities scanned over a range of excitation/emission wavelengths (Coble 1996; Chen et al. 2003). EEMs were collected with a JY-Horiba Spex Fluoromax-3 spectrofluorometer (HORIBA, Japan) at room temperature using 5 nm excitation and emission slit widths and an integration time of 1.0 s. The Aqualog spectrophotometer simultaneously collects both fluorescence and absorbance spectra on a sample. All fluorescence spectra were collected in signal-to-reference (*S*:*R*) mode with instrumental bias correction. Instrument-specific corrections, Raman area normalization and Milli-Q blank subtraction were conducted with Matlab (2009). From the UV–Vis absorbance and EEMs data, we calculated two indices of DOC quality: the freshness index (FI) and specific UV absorption (SUVA). FI (β:α) is a ratio of emission intensity at 380 nm to that of the region between 420 and 435 nm at an excitation of 310 nm and is reflective of recently produced algal organic matter (Parlanti et al. 2000). SUVA is a DOC-normalized index of aromaticity calculated as UV absorbance at 254 nm/[(DOC (mg L^{-1}) × path length (0.01 m)] (Weishaar et al. 2003). FI increases with autochthonous carbon production, whereas SUVA increases with allochthonous carbon production.

All isotope samples were analyzed by the University of California, Davis Stable Isotope Facility for ^{13}C and ^{15}N, using an elemental analyzer interfaced to a continuous flow isotope ratio mass spectrometer.

Fish sampling

At each of the 21 lakes, we caught fish by angling. Each fish was identified to species, weighed, photographed and measured (TL; maximum length). We collected a dorsal muscle sample from each individual which was frozen until processing for stable isotope analysis. Upon returning to the laboratory, muscle samples were freeze dried for 24 h, ground with a mortar and pestle and packaged for ^{13}C and ^{15}N analysis. Otoliths were removed, cleaned, dried and stored in vials for age determination and growth rate analysis. We calculated catch per unit effort (CPUE) as the total catch divided by the number of person-hours spent angling at each lake. All applicable institutional guidelines for the use of animals were followed and approved by the Institutional Animal Care and Use Committee at the University of California, San Diego (Protocol #S14140).

Fish sample processing

Fish in temperate regions can be aged by examining calcified structures called otoliths, which form annuli—rings that correspond to low winter growth. The width of the annuli is an indicator of annual growth (Casselman 1990). To determine age and annual growth, the sagittal otoliths were mounted on

the edge of a microscope slide with the core positioned just within the microscope slide's edge and polished to section the otolith. The otolith was then flipped onto the transverse cross section and polished again until the core was exposed in the transverse section similar to Taylor and McIlwain (2010). Annuli were counted by two independent readers in the absence of information about fish size or lake. Ages were in agreement for 84% of the otoliths, and never differed by more than 1 year. For otoliths where the age determinations disagreed, the two readers examined the otoliths together and were able to reach consensus. The width of each annuli was measured using imaging software (Image J).

Calculations and statistical analysis

For body condition comparison, we calculated the condition using Eq. 1 (Fulton 1902):

$$\text{Condition} = (\text{wet body mass/total body length}^3) \times 100. \tag{1}$$

To quantify DOC quality, we conducted a PCA on the correlation matrix of two indices of DOC quality: freshness index and SUVA. Both SUVA and freshness loaded positively on PCA axis 1, and SUVA loaded positively on axis 2, while freshness loaded negatively (Online Resource 1, Figure S2). The site scores along axis 2 were used as a measure of DOC quality for further analysis where low values of this quality variable are indicative of autochthonous DOC and high values indicate allochthonous DOC.

Trophic level (TL) for each fish was calculated using Eq. 2:

$$\text{TL} = \frac{\delta^{15}N_{\text{Fish}} - (\delta^{15}N_{\text{Plant}} + \delta^{15}N_{\text{POM}}/2)}{3.4}. \tag{2}$$

We calculated fish trophic level as the number of trophic levels above the average plant and particulate organic matter (POM) $\delta^{15}N$, assuming a trophic enrichment of $\delta^{15}N$ of 3.4‰ per trophic level (post-2002).

To determine the best predictors of body condition, we fit a linear model with lake temperature, elevation, DOC, fish $\delta^{13}C$, fish trophic position, CPUE, chl-*a*, TN, date caught, species and DOC quality as predictors. TP was not included as a predictor because it consistently fell below detection limits. Variance inflation factors were calculated to determine the degree of multicollinearity and all VIFs were < 4 suggesting that these predictors are appropriate to use. DOC, Chl-*a* and CPUE were log transformed to improve normality (Online Resource 1, Figure S1). We then fit the same model, including all predictors, but allowed for a second-order polynomial relationship to DOC to account for the possibility that fish condition was unimodally related to DOC (Finstad et al. 2014). Models with linear and polynomial relationships between condition and DOC were compared using AIC. A

linear relationship between growth and DOC was the most parsimonious (ΔAIC = 1.8). We used a backward selection AIC-based procedure to fit the best models. In addition, we calculated η^2 values for predictors in the reduced model using the *etasq* function in the R package "heplots". This set of analyses allows us to (1) determine the predictors that explain the data in the simplest way (backward selection) and (2) report the relative importance of the covariates.

To examine individual-level growth rates, we restricted our analysis to fish that were at least 4 years old to generate time series of comparable lengths similar to Romo-Curiel et al. (2015). Growth was represented as the length of the radii from the core to each annulus as a function of age (Fig. 2a). To calculate a measure of individual growth rate, we fit a linear model to the annuli radius as a function of age from age 1 to 4. The slope of this line was then used as a measure of individual growth rate for further analyses. These slopes were highly correlated with the growth parameter of the Von Bertalanffy growth function (K), but required fewer assumptions (Online Resource 1, Figure S3; linear slope and K Pearson's correlation $r = 0.94$). We fit a linear model as above to predict growth. We removed current trophic position and date caught as predictors because these are not relevant to growth that occurred in the past years. We added an additional predictor because fish of different ages experienced early growth during different calendar years. To estimate temperature experienced by the fish during the years they were between 1 and 4 years old, we compiled a daily average temperature dataset from 13 NOAA stations arrayed along an elevation gradient in our study area (from −118.9617 to −119.9161 in longitude and 38.07–37.63 in latitude) from 1997 to 2017 (NOAA stations: USC00041697, USC00043939, USC00044881, USC00045280, USC00045400, USC00049063, USC00049855, USR0000C-CRE, USR0000CDPP, USR0000CTUO, USR0000CWWO, USS0019L13S, USW00053150). Air temperatures are highly correlated with surface water temperatures (Livingston et al. 1999), allowing us to estimate past water temperatures where direct measurements are not available. We regressed the June–September average daily temperatures with elevation, and used the linear relationship to predict the mean summer temperature at the elevation of our lakes. The average temperature over the 3-year period of interest (fish was age 1–4 years) was calculated for each fish and used as a predictor in the model of fish growth. Model selection and η^2 values were calculated as above. A linear relationship between growth and DOC was a better fit to these data than a polynomial fit, and the most parsimonious (ΔAIC = 2.3). Results were largely consistent when we calculated growth over different durations (e.g., growth from age 1 to 3, or age 1 to 5; Online Resource 1, Table S1).

Our study design reflects a trade-off between sampling many different sites and sampling fewer sites more
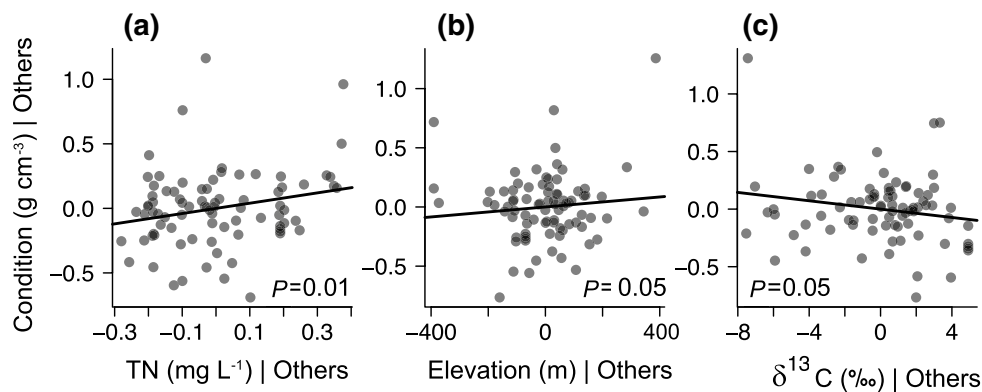
Fig. 1 Added-variable plots for significant predictors of body condition in the final model selected by backward AIC selection ($N = 87$) a) total nitrogen, b) elevation and c) δ^{13}C. Axes in added-variable plots are the residuals of the model excluding all other variables in the final model (e.g., in a) the y-axis represents the residuals of the relationship between condition and elevation and δ^{13}C)

extensively. Because regression designs are generally more powerful than ANOVA designs at detecting associations between the independent and dependent variables (Cottingham et al. 2005), we sampled many sites (21 lakes) and collected fewer fish per lake. To explore how our results may have been influenced by our sampling design, we used simulations to test how the probability of type I and II errors vary with the number of fish sampled per lake with the observed levels of variation among fish and between lakes. We focused on the relationship between

DOC and fish growth as an example of a significant finding (Fig. 2b). We simulated fish growth rates when there was no relationship between DOC and growth to explore how type I error rates vary with the number of fish sampled per lake (simulation 1). We then simulated datasets where we assume that DOC influenced growth rates according to the relationship we observed in our data ($\text{growth} = -0.25 \times \text{DOC} + 0.68$) to test how type II error rate vary with the number of fish sampled per lake (simulation 2; Online Resource 1).
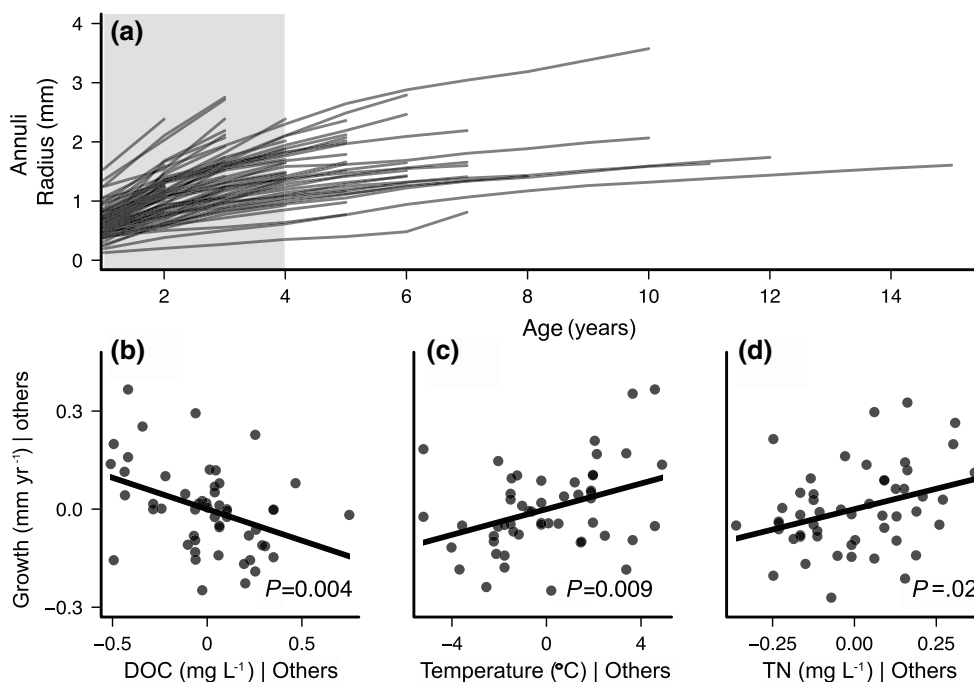

Fig. 2 **a** Growth trajectories for individual fish plotted as the otolith annuli radius as a function of age. Linear models were fit to the growth from age 1 to 4 years for individuals that were at least 4 years (shaded box) and used as the measure of growth for future analysis ($N=52$). **b–d** Added-variable plots for significant predictors of individual growth in the final model selected by backward AIC selection

For the condition and growth analysis, we plotted the data as added-variable plots (AV plots; Figs. 1, 2). These plots show the effect of x on y after controlling for variability due to other explanatory variables (i.e., marginal effects). AV or residual plots more accurately depict the relationships found by the model, since each regression coefficient represents the residuals of different models plotted against one another (Draper et al. 1966).

All analyses were done in R (R Core Team 2016).

Results

The lakes varied in many of their physical and chemical parameters (Table 1). Epilimnion temperature ranged from 8.7 to 20.3 °C and was negatively correlated with elevation (Pearson's correlation, $r = -0.71$, $p < 0.001$). Lakes sampled varied in DOC concentration from 0.83 to 3.6 mg L^{-1}, which was also negatively correlated with elevation (Pearson's correlation, $r = -0.28$, $p = 0.008$). Although temperature and DOC were both correlated with elevation, we specifically chose lakes to have independent DOC and temperature gradients, resulting in a set of lakes where DOC and temperature were unrelated (Pearson's correlation, $r = 0.16$, $p = 0.13$; Online Resource 1, Figure S1).

Fish body condition increased with TN and elevation, and decreased with $\delta^{13}C$ (Fig. 1; Table 2). We caught 87 fish across the 21 lakes (range 1–12, average 4), including 76 brook trout and 11 rainbow trout. The mean total length

Table 1 Median and range of physical and environmental variables from the 21 lakes sampled

	Median	Range
Elevation (m)	3147	2506–3337
Area (ha)	4.5	0.5–22
Depth (m)	6.9	3–4
DOC (mg L^{-1})	1.5	0.83–3.6
TN (mg L^{-1})	0.3	0.02–0.58
Lake temperature (°C)	15.9	8.7–20.3
SUVA	1.2	0.06–2.7
Freshness index	0.58	0.34–0.77
$\delta^{15}N$ of fish (‰)	6.6	3.9–9.7
$\delta^{13}C$ of fish (‰)	−19.9	−30.9 to −14.4
Chl-a (μg L^{-1})	0.52	0.18–1.23
CPUE (fish h^{-1})	2	0.04–4
Trophic position	3	1.9–3.7
Temperature (age 1–4, °C)	10.7	8.2–14.8
Calculated light attenuation coefficient (K_D, m^{-1})	0.1	0.07–0.23

The light attenuation coefficient K_D was calculated using the equation presented in Jones et al. (2012): $K_D = 0.213 + 0.0177\text{Chl }a + 0.0514\text{DOC}$

Table 2 Model estimates, p values and partial eta-squared values for predictors of fish condition (mg mm^{-3}) from a backward AIC selected linear model

Predictor	Estimate	p-value	η^2
Elevation (m)	**0.0005**	**0.05**	**0.05**
TN (mg L^{-1})	**0.51**	**0.01**	**0.09**
$\delta^{13}C$ (‰)	**−0.02**	**0.05**	**0.05**

Bold values represent predictors that were significant in the final model

of fish varied between lakes and ranged from 12 to 36.5 cm, and their weight varied from 0.03 to 0.62 kg. Because Fulton's body condition can be biased by body size (and our goal was to describe the weight of fish across equivalent lengths, thus removing length-related biases), we investigated if condition was correlated with length, which was not (Pearson's correlation, $r = -0.2$, $p = 0.07$). Additionally, condition did not vary among the two fish species of equivalent lengths ($F = 1.48$, $p = 0.09$ for effect of species in a two-way ANOVA that included species and length).

Otoliths

Early life growth was positively related to temperature and TN (Fig. 2b, c; Table 3) and negatively related to the quantity of DOC (Fig. 2d; Table 3). Chl-a was retained in the backward AIC selected model, but was not a significant predictor of growth (Table 3). DOC and chl-a were not correlated (Pearson's correlation, $r = -0.09$, $p = 0.36$). Fish ranged in age from 1 to 15 years, and averaged 4.8 years. Of the 21 sampled lakes, we had 18 lakes with fish > age 4 years to estimate fish growth rates. Growth curves varied among individuals (Fig. 2a). Growth rates did not differ between the two trout species (t test, $t = -1.38$, $p = 0.19$). Though few fish were caught per lake, simulations show that both the probability of type I error (false positive in the case of no significant relationship) and of type II error (false negative in the case of a significant relationship like the ones we found) decreased with the number of fish sampled per lake; however type I and type II errors remained low for any number of

Table 3 Model estimates, p values and partial eta-squared values for predictors of fish growth (mm otolith year^{-1}) from a backward AIC selected linear model

Predictor	Estimate	p value	η^2
Log DOC (mg L^{-1})	**−0.27**	**0.004**	**0.17**
Temperature (age 1–4, °C)	**0.02**	**0.009**	**0.14**
TN (mg L^{-1})	**0.24**	**0.02**	**0.11**
Log Chl a (μg L^{-1})	0.06	0.13	0.05

Bold values represent predictors that were significant in the final model

fish sampled (type I: 0.043–0.058, type II: 0–0.03; Online Resource 1). The precision of the estimate of slope increased with the number of fish (Online Resource 1).

Discussion

We found opposing effects of temperature and DOC on fish growth, with faster growth in warmer, higher TN lakes, and slower growth in higher DOC lakes. Although terrestrial organic matter inputs can provide additional energy at the base of the food web and stimulate bacterial production, our results suggest that DOC largely acted to limit fish growth, potentially through shading effects on primary producers. Additionally, TN and carbon with a more terrestrial isotopic signal were associated with higher fish body condition, suggesting that the resource base impacts food quality for fish. Overall, our results suggest that as lakes become warmer and browner, fish production will depend on the magnitude of the change in each of these stressors. A concurrent increase in temperature and DOC could buffer fish growth rates from predicted change in response to these individual stressors.

Two studies of European temperature lakes found that DOC was negatively related to fish abundance (Finstad et al. 2014; Karlsson et al. 2009). However, abundance is not a proxy for growth, and changes in the abundance may have the opposite effect on growth. For example, Benoît et al. (2016) hypothesized that a decrease in abundance with DOC may increase density-dependent fish growth rates. Despite this prediction, they found a negative impact of DOC on growth rates, suggesting DOC was functioning to reduce primary production through shading (Benoît et al. 2016). Our study expands this result by examining lower DOC lakes, which are not currently represented in the literature (Benoît et al. 2016: 2.6–9 mg L^{-1}, current study 0.83–2.2 mg L^{-1}). Oligotrophic and clear lakes are predicted to respond strongly to even small increases in DOC (Stasko et al. 2012). Therefore, determining if DOC will reduce or increase fish growth rates at these low concentrations is important to understanding how lake 'browning' will impact important ecosystem functions in the future.

We found a negative relationship between DOC and individual fish growth at these very low concentrations of DOC (Fig. 2b). The slower growth in lakes with higher DOC is potentially due to a negative effect of DOC on primary productivity via photon absorption (Carpenter et al. 1998; Jones et al. 2012; Thrane et al. 2014). DOC can absorb ten times as many photons in the PAR region as phytoplankton (as % of PAR entering the lake per concentration by mass), reducing light availability for primary production in phytoplankton and benthic algae (Thrane et al. 2014). Within the range of DOC and chl-a concentrations in our study, the calculated extinction coefficient of PAR (K_D)

is expected to increase ~ threefold from 0.07 to 0.23 m^{-1} (Table 1), reducing light available for photosynthesis and decreasing the euphotic zone. Thus, although Sierra Nevada lakes are very clear, even small changes in DOC can reduce the light energy available for in-lake production. Primary production in clearer lakes is often dominated by benthic periphyton, which is more sensitive to variation in water color than pelagic production (Hansson 1992; Vadeboncoeur et al. 2001; Seekell et al. 2015). Lower benthic productivity due to light limitation can have large effects on fish since the zoobenthos can account for the majority of their resources (Vander Zanden and Vadeboncoeur 2002), and zoobenthic prey is the mechanistic link between DOC and fish biomass (Karlsson et al. 2009). This explanation is consistent with the increase in individual growth rates with TN (Fig. 2d), where productivity, both benthic and pelagic, increases with TN (Seekell et al. 2015), though unfortunately we did not measure benthic production in this study. Our results suggest that fish growth is regulated through bottom-up processes, where primary production at the base of the food web determines the energy available for higher trophic levels.

DOC can also alter the visual environment in which fish must operate. Lower light intensity can decrease the visual acuity of planktivorous fish (Bramm et al. 2009; Vinyard and O'Brien 1976), decreasing their consumption rates (Carter et al. 2010; Weidel et al. 2017). Therefore, the negative relationship between growth and DOC could be mediated through changes in the efficiency of trout foraging in different light environments.

We measured DOC and TN concentrations during different calendar years than the growth of fish from age 1 to 4 years, implicitly assuming that measured DOC and TN represent differences among lakes that persist over the life span of fish. In Sierra Nevada lakes, DOC and TN show distinct seasonal patterns associated with snow melt (Piovia-Scott et al. 2016); however, on annual timescales DOC and nutrient load to lakes are largely driven by the characteristics of the terrestrial ecosystems such as primary production and vegetation structure which are likely stable over the inter-annual timescale of interest (Larsen et al. 2011). Among lake variation in DOC and water chemistry may therefore be maintained despite interannual variability driven by weather conditions.

The positive effect of temperature on growth was consistent with patterns observed for a wide range of ectotherms. Fish growth rates are related to temperature within populations (Elliott 1976) and this growth is captured by variation in otolith width (Black et al. 2005). Higher growth rates in warmer lakes likely represent faster metabolic processes that increase growth in these ectothermic animals. Indeed, in Norwegian alpine lakes, higher temperatures increased metabolic rate resulting in higher somatic growth in brown trout (*Salmo trutta* L.) until age

at maturity (Thaulow et al. 2017). Additionally, the seasonal duration of temperatures that permit physiological processing of food can limit growth in ectotherms (Sinervo et al. 2010). Colder, high-elevation lakes have shorter ice-free seasons, and likely a shorter window of time in which temperatures are optimal for growth. As temperatures warm, fish growth rates will likely increase.

We did not find that fish density, as measured by angling CPUE, predicted growth rates or body condition (Tables 2, 3). However, angling estimates of fish density are not as reliable as other estimates such as gill nets (Pierce and Tomcko 2003). Higher fish density can reduce individual growth rates through increased competition for food (Magnuson 1962), as has been documented in rainbow trout (Holm et al. 1990). It is possible that with a more accurate estimate of CPUE, fish density may impact individual growth rates.

Fish body condition was generally related to different lake variables than growth (Tables 2, 3). Body condition varies on shorter timescales than growth and indicates recent prey availability and quality (Blackwell et al. 2000). We found that TN, a variable associated with aquatic productivity (Elser et al. 2000), was related to better body condition likely representing a greater availability of prey (Fig. 1a). Body condition also increased with elevation (Fig. 1b). This may represent an unmeasured variable that covaries with elevation. A likely candidate is prey composition, as both benthic macroinvertebrate (Smith et al. 2016) and zooplankton communities (Symons and Shurin 2016) vary with elevation. Finally, more negative $\delta^{13}C$ values were associated with better body condition (Fig. 1c). In Sierra Nevada lakes, the $\delta^{13}C$ in benthic grazers is less negative than both terrestrial plants and POM which are similar to each other. The fish $\delta^{13}C$ we measured were less negative than our measured plant or POM signature even after accounting for trophic enrichment of ^{13}C (Post 2002). Thus, our stable isotopes suggest that fish have a major zoobenthic diet, which was supported by stomach content analysis (C. Banach, personal communication), and generally by studies of lake fish (Vander Zanden and Vadeboncoeur 2002). Therefore, our pattern suggests prey sources that are more negative in $\delta^{13}C$—either pelagic or terrestrial prey—increase condition more than the consumption of benthic prey. Greater body condition when consuming pelagic or terrestrial prey runs counter to expectations that benthic prey are generally larger and therefore more nutritionally valuable than zooplankton (Vander Zanden and Vadeboncoeur 2002). This counterintuitive pattern could be explained by terrestrial insect consumption. Terrestrial insects can be energetically preferable and represent a significant portion of trout diets (Marcarelli et al. 2011; C. Banach, personal communication). Therefore, better condition at more negative $\delta^{13}C$ values may represent the high quality or availability of terrestrially derived insects.

We hypothesized that DOC would have a positive effect on fish growth rates by protecting against damaging levels of UV and providing a source of N and P in these highly oligotrophic lakes. Though some positive effects may have occurred (e.g., UV protection), the predominant effect of DOC was to reduce growth across the entire DOC gradient sampled (Fig. 2b). Previously, Finstad et al. (2014) found a unimodal relationship between trout biomass and DOC, where fish biomass initially increased with DOC before declining. In Finstad et al. (2014), fish biomass was highly correlated with fish density ($r^2 = 0.79$). We suggest that at the lowest DOC concentrations, fish density is low due to bottom-up control on abundance, and because density is low individual growth rates are high (low competition). As DOC increases, resulting in lower fish biomass (density), the individual growth rates do not increase (Benoît et al. 2016), because DOC shades benthic and pelagic production reducing resources available for consumption and growth (Karlsson et al. 2009; Jones et al. 2012).

Conclusions

Understanding the role of temperature and DOC in structuring aquatic ecosystems is imperative as the climate warms and the influence of terrestrial production increases. This study adds to the body of evidence showing that terrestrial inputs of DOC depresses fish production through the shading of primary producers (Benoît et al. 2016; Finstad et al. 2014; Karlsson et al. 2009). Our study examined fish growth in high-elevation lakes, expanding the DOC concentration gradient to include lower DOC lakes. Even in these clear, low DOC lakes, there was a negative impact of DOC on fish growth and body condition, which was likely caused by bottom-up effects from DOC shading primary production. If space is a good proxy for time, then we expect that future browning will reduce growth rates in fish via shading effects on bottom-up processes and that future warming within the thermal tolerance of these fish will increase growth. Lakes will warm and brown at different rates depending on their geographical context, and thus the response of fish growth to climate change will be context dependent. In lakes of the Sierra Nevada where warming and browning will occur together, these changes will likely have antagonistic impacts on the growth rates of fish, reducing the negative impact of DOC on the productivity of an economically important recreational fishery.

References

Benoît P-O, Beisner BE, Solomon CT (2016) Growth rate and abundance of common fishes is negatively related to dissolved organic carbon concentration in lakes. Can J Fish Aquat Sci 73:1230–1236. https://doi.org/10.1139/cjfas-2015-0340

Black BA, Boehlert GW, Yoklavich MM (2005) Using tree-ring cross-dating techniques to validate annual growth increments in long-lived fishes. Can J Fish Aquat Sci 62:2277–2284. https://doi.org/10.1139/f05-142

Blackwell BG, Brown ML, Willis DW (2000) Relative weight (Wr) status and current use in fisheries assessment and management. Rev Fish Sci 8:1–44. https://doi.org/10.1080/10641260091129161

Bradley RS, Keimig FT, Diaz HF (2004) Projected temperature changes along the American cordillera and the planned GCOS network. Geophys Res Lett 31:L16210. https://doi.org/10.1029/2004gl020229

Bramm ME, Lassen MK, Liboriussen L, Richardson K, Ventura M, Jeppesen E (2009) The role of light for fish-zooplankton-phytoplankton interactions during winter in shallow lakes—a climate change perspective. Freshw Biol 54:1093–1109. https://doi.org/10.1111/j.1365-2427.2008.02156.x

Brett MT, Kainz MJ, Taipale SJ, Seshan H (2009) Phytoplankton, not allochthonous carbon, sustains herbivorous zooplankton production. Proc Natl Acad Sci 106:21197–21201. https://doi.org/10.1073/pnas.0904129106

Brown JH, Gillooly JF, Allen AP, Savage VM, West GB (2004) Toward a metabolic theory of ecology. Ecology 85:1771–1789. https://doi.org/10.1890/03-9000

Carpenter SR, Cole JJ, Kitchell JF, Pace ML (1998) Impact of dissolved organic carbon, phosphorus, and grazing on phytoplankton biomass and production in experimental lakes. Limnol Oceanogr 43:73–80. https://doi.org/10.4319/lo.1998.43.1.0073

Carter MW, Shoup DE, Dettmers JM, Wahl DH (2010) Effects of turbidity and cover on prey selectivity of adult smallmouth bass. Trans Am Fish Soc 139:353–361. https://doi.org/10.1577/T08-159.1

Casselman JM (1990) Growth and relative size of calcified structures of fish. Trans Am Fish Soc 119:673–688. https://doi.org/10.1577/1548-8659(1990)119%3c0673:GARSOC%3e2.3.CO;2

Chen W, Westerhoff P, Leenheer JA, Booksh K (2003) Fluorescence excitation–emission matrix regional integration to quantify spectra for dissolved organic matter. Environ Sci Technol 37:5701–5710. https://doi.org/10.1021/es034354c

Coble PG (1996) Characterization of marine and terrestrial DOM in seawater using excitation emission matrix spectroscopy. Mar Chem 51:325–346. https://doi.org/10.1016/0304-4203(95)00062-3

Cottingham KL, Lennon JT, Brown BL (2005) Knowing when to draw the line: designing more informative ecological experiments. Front Ecol Environ 3:145–152. https://doi.org/10.1890/1540-9295(2005)003%5b0145:KWTDTL%5d2.0.CO;2

Crozier LG, Hutchings JA (2014) Plastic and evolutionary responses to climate change in fish. Evol Appl 7:68–87. https://doi.org/10.1111/eva.12135

Draper NR, Smith H, Pownell E (1966) Applied regression analysis. Wiley, New York

Elliott JM (1976) The energetics of feeding, metabolism and growth of brown trout (*Salmo trutta* L.) in relation to body weight, water temperature and ration size. J Anim Ecol 45:923–948. https://doi.org/10.2307/3590

Elser JJ et al (2000) Nutritional constraints in terrestrial and freshwater food webs. Nature 408:578–580. https://doi.org/10.1038/35046058

Evans CD, Chapman PJ, Clark JM, Monteith DT, Cresser MS (2006) Alternative explanations for rising dissolved organic carbon export from organic soils. Glob Change Biol 12:2044–2053. https://doi.org/10.1111/j.1365-2486.2006.01241.x

Finstad AG, Helland IP, Ugedal O, Hesthagen T, Hessen DO (2014) Unimodal response of fish yield to dissolved organic carbon. Ecol Lett 17:36–43. https://doi.org/10.1111/ele.12201

Fulton TW (1902) The rate of growth of fishes. 20th Annual report of the Fishery Board of Scotland, vol 3, pp 326–446

Gottfried M et al (2012) Continent-wide response of mountain vegetation to climate change. Nat Clim Change 2:111–115. https://doi.org/10.1038/nclimate1329

Guzzo MM, Blanchfield PJ, Rennie MD (2017) Climate alters the diet and growth of fish. Proc Natl Acad Sci 114:9912–9917. https://doi.org/10.1073/pnas.1702584114

Hairston NG, Smith FE, Slobodkin LB (1960) Community structure, population control, and competition. Am Nat 94:421–425. https://doi.org/10.1086/282146

Hansson L-A (1992) Factors regulating periphytic algal biomass. Limnol Oceanogr 37:322–328. https://doi.org/10.4319/lo.1992.37.2.0322

Hauer FR et al (1997) Assessment of climate change and freshwater ecosystems of the Rocky Mountains, USA and Canada. Hydrol Process 11:903–924. https://doi.org/10.1002/(SICI)1099-1085(19970630)11:8%3c903:AID-HYP511%3e3.0.CO;2-7

Hessen DO, Andersen T (1990) Bacteria as a source of phosphorus for zooplankton. Hydrobiologia 206:217–223. https://doi.org/10.1007/bf00014087

Holm JC, Refstie T, Bø S (1990) The effect of fish density and feeding regimes on individual growth rate and mortality in rainbow trout (*Oncorhynchus mykiss*). Aquaculture 89:225–232. https://doi.org/10.1016/0044-8486(90)90128-A

Jones SE, Solomon CT, Weidel BC (2012) Subsidy or subtraction: how do terrestrial inputs influence consumer production in lakes? Freshw Rev 5:37–49. https://doi.org/10.1608/FRJ-5.1.475

Karlsson J, Bystrom P, Ask J, Ask P, Persson L, Jansson M (2009) Light limitation of nutrient-poor lake ecosystems. Nature 460:506–509. https://doi.org/10.1038/nature08179

Knapp RA, Matthews KR, Sarnelle O (2001) Resistance and resilience of alpine lake fauna to fish introductions. Ecol Monogr 71:401–421. https://doi.org/10.1890/0012-9615(2001)071%5b0401:RAROAL%5d2.0.CO;2

Koizumi S et al (2018) Experimental whole-lake dissolved organic carbon increase alters fish diet and density but not growth or productivity. Can J Fish Aquat Sci. https://doi.org/10.1139/cjfas-2017-0283

Larsen S, Andersen TOM, Hessen DO (2011) Climate change predicted to cause severe increase of organic carbon in lakes. Glob Change Biol 17:1186–1192. https://doi.org/10.1111/j.1365-2486.2010.02257.x

Livingston DM, Lotter AF, Walkery IR (1999) The decrease in summer surface water temperature with altitude in Swiss alpine lakes: a comparison with air temperature lapse rates. Arct Antarct Alp Res 31:341–352. https://doi.org/10.1080/15230430.1999.12003319

Magnuson JJ (1962) An analysis of aggressive behavior, growth, and competition for food and space in medaka (*Oryzias latipes* (Pisces, Cyprinodontidae)). Can J Zool 40:313–363. https://doi.org/10.1139/z62-029

Marcarelli AM, Baxter CV, Mineau MM, Hall RO (2011) Quantity and quality: unifying food web and ecosystem perspectives on the role of resource subsidies in freshwaters. Ecology 92:1215–1225. https://doi.org/10.1890/10-2240.1

Mladenov N et al (2011) Dust inputs and bacteria influence dissolved organic matter in clear alpine lakes. Nat Commun 2:405. https://doi.org/10.1038/ncomms1411

Monteith DT et al (2007) Dissolved organic carbon trends resulting from changes in atmospheric deposition chemistry. Nature 450:537–540. https://doi.org/10.1038/nature06316

Morrongiello JR, Crook DA, King AJ, Ramsey DSL, Brown P (2011) Impacts of drought and predicted effects of climate change on fish growth in temperate Australian lakes. Glob Change Biol 17:745–755. https://doi.org/10.1111/j.1365-2486.2010.02259.x

Pace ML et al (2004) Whole-lake carbon-13 additions reveal terrestrial support of aquatic food webs. Nature 427:240–243. https://doi.org/10.1038/nature02227

Parlanti E, Wörz K, Geoffroy L, Lamotte M (2000) Dissolved organic matter fluorescence spectroscopy as a tool to estimate biological activity in a coastal zone submitted to anthropogenic inputs. Org Geochem 31:1765–1781. https://doi.org/10.1016/S0146-6380(00)00124-8

Pierce RB, Tomcko CM (2003) Variation in gill-net and angling catchability with changing density of northern pike in a small Minnesota lake. Trans Am Fish Soc 132:771–779. https://doi.org/10.1577/T02-105

Piovia-Scott J, Sadro S, Knapp RA, Sickman J, Pope KL, Chandra S (2016) Variation in reciprocal subsidies between lakes and land: perspectives from the mountains of California. Can J Fish Aquat Sci 73:1691–1701. https://doi.org/10.1139/cjfas-2015-0549

Post DM (2002) Using stable isotopes to estimate trophic position: models, methods, and assumptions. Ecology 83:703–718. https://doi.org/10.2307/3071875

R Core Team (2016) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.r-project.org/

Romo-Curiel AE, Herzka SZ, Sosa-Nishizaki O, Sepulveda CA, Aalbers SA (2015) Otolith-based growth estimates and insights into population structure of White Seabass, *Atractoscion nobilis*, off the Pacific coast of North America. Fish Res 161:374–383. https://doi.org/10.1016/j.fishres.2014.09.004

Rose KC, Williamson CE, Schladow SG, Winder M, Oris JT (2009) Patterns of spatial and temporal variability of UV transparency in Lake Tahoe, California-Nevada. J Geophys Res Biogeosci 114:G00D03. https://doi.org/10.1029/2008JG000816

Rose KC, Williamson CE, Kissman CEH, Saros JE (2015) Does allochthony in lakes change across an elevation gradient? Ecology 96:3281–3291. https://doi.org/10.1890/14-1558.1

Sadro S, Melack JM (2012) The effect of an extreme rain event on the biogeochemistry and ecosystem metabolism of an oligotrophic high-elevation lake. Arct Antarct Alp Res 44:222–231. https://doi.org/10.1657/1938-4246-44.2.222

Sadro S, Nelson CE, Melack JM (2012) The influence of landscape position and catchment characteristics on aquatic biogeochemistry in high-elevation lake-chains. Ecosystems 15:363–386. https://doi.org/10.1007/s10021-011-9515-x

Seekell DA, Lapierre JF, Ask J, Bergström AK, Deininger A, Rodríguez P, Karlsson J (2015) The influence of dissolved organic carbon on primary production in northern lakes. Limnol Oceanogr 60:1276–1285. https://doi.org/10.1002/lno.10096

Sinervo B et al (2010) Erosion of lizard diversity by climate change and altered thermal niches. Science 328:894–899. https://doi.org/10.1126/science.1184695

Smith TC, Knapp RA, Briggs CJ (2016) Declines and extinctions of mountain yellow-legged frogs have small effects on benthic macroinvertebrate communities. Ecosphere 7(6):e01327. https://doi.org/10.1002/ecs2.1327

Somero GN (2010) The physiology of climate change: how potentials for acclimatization and genetic adaptation will determine 'winners' and 'losers'. J Exp Biol 213:912–920. https://doi.org/10.1242/jeb.037473

Sommaruga R (2015) When glaciers and ice sheets melt: consequences for planktonic organisms. J Plankton Res 37:509–518. https://doi.org/10.1093/plankt/fbv027

Stasko AD, Gunn JM, Johnston TA (2012) Role of ambient light in structuring north-temperate fish communities: potential effects of increasing dissolved organic carbon concentration with a changing climate. Environ Rev 20:173–190. https://doi.org/10.1139/a2012-010

Symons CC, Shurin JB (2016) Climate constrains lake community and ecosystem responses to introduced predators. Proc R Soc B 283:20160825. https://doi.org/10.1098/rspb.2016.0825

Tanentzap AJ, Szkokan-Emilson EJ, Kielstra BW, Arts MT, Yan ND, Gunn JM (2014) Forests fuel fish growth in freshwater deltas. Nat Commun 5:4077. https://doi.org/10.1038/ncomms5077

Tanentzap AJ et al (2017) Terrestrial support of lake food webs: synthesis reveals controls over cross-ecosystem resource use. Sci Adv 3:e1601765. https://doi.org/10.1126/sciadv.1601765

Taylor BM, McIlwain JL (2010) Beyond abundance and biomass: effects of marine protected areas on the demography of a highly exploited reef fish. Mar Ecol Process Ser 411:243–258. https://doi.org/10.3354/meps08672

Thaulow J, Haugen TO, Brogstrøm R (2017) Parallelism in thermal growth response in otoliths and scales of brown trout (*Salmo trutta* L.) from alpine lakes independent of genetic background. Ecol Freshw Fish 26:53–65. https://doi.org/10.1111/eff.12250

Thrane J-E, Hessen DO, Andersen T (2014) The absorption of light in lakes: negative impact of dissolved organic carbon on primary productivity. Ecosystems 17:1040–1052. https://doi.org/10.1007/s10021-014-9776-2

Vadeboncoeur Y, Lodge DM, Carpenter SR (2001) Whole-lake fertilization effects on primary production between benthic and pelagic habitats. Ecology 82:1065–1077. https://doi.org/10.1890/0012-9658(2001)082%5b1065:WLFEOD%5d2.0.CO;2

Vander Zanden MJ, Vadeboncoeur Y (2002) Fishes as integrators of benthic and pelagic food webs in lakes. Ecology 83:2152–2161. https://doi.org/10.1890/0012-9658(2002)083%5b2152:FAIOBA%5d2.0.CO;2

Vinyard GL, O'Brien WJ (1976) Effects of light and turbidity on the reactive distance of bluegill (*Lepomis macrochirus*). J Fish Board Can 33:2845–2849. https://doi.org/10.1139/f76-342

Weidel BC, Baglini K, Jones SE, Kelly PT, Solomon CT, Zwart JA (2017) Light climate and dissolved organic carbon concentration influence species-specific changes in fish zooplanktivory. Inland Waters 7:210–217. https://doi.org/10.1080/20442041.2017.1329121

Weishaar JL, Aiken GR, Bergamaschi BA, Fram MS, Fujii R, Mopper K (2003) Evaluation of specific ultraviolet absorbance as an indicator of the chemical composition and reactivity of dissolved organic carbon. Environ Sci Technol 37:4702–4708. https://doi.org/10.1021/es030360x

Wetzel RG (1995) Death, detritus, and energy flow in aquatic ecosystems. Freshw Biol 33:83–89. https://doi.org/10.1111/j.1365-2427.1995.tb00388.x

Wilkinson GM, Carpenter SR, Cole JJ, Pace ML, Yang C (2013) Terrestrial support of pelagic consumers: patterns and variability revealed by a multilake study. Freshw Biol 58:2037–2049. https://doi.org/10.1111/fwb.12189

Williamson CE, Morris DP, Pace ML, Olson OG (1999) Dissolved organic carbon and nutrients as regulators of lake ecosystems: resurrection of a more integrated paradigm. Limnol Oceanogr 44:795–803. https://doi.org/10.4319/lo.1999.44.3_part_2.0795