Freshwater Biology

Editor-in-Chief: Belinda Robson

Associate Editors: Stefan Bertilsson, Lisette de Senerpont Domis, Andy J. Green, Angus McIntosh, Mariana Meerhoff, Alan D. Steinman

ORIGINAL ARTICLE

Freshwater Biology   WILEY

Non-native Chinook salmon add nutrient subsidies and functional novelty to Patagonian streams

Nicolas J. Muñoz[1] 🆔 | Brian Reid[2] 🆔 | Cristian Correa[3,4] 🆔 | Bryan D. Neff[5] |
John D. Reynolds[1]

[1]Earth to Ocean Research Group, Simon Fraser University, Burnaby, BC, Canada

[2]Centro de Investigación en Ecosistemas de la Patagonia, Coyhaique, Chile

[3]Instituto de Conservación Biodiversidad y Territorio, Universidad Austral de Chile, Valdivia, Chile

[4]Centro de Humedales Río Cruces, Universidad Austral de Chile, Valdivia, Chile

[5]Department of Biology, Western University, London, ON, Canada

Correspondence
Nicolas J. Muñoz, Earth to Ocean Research Group, Simon Fraser University, Burnaby, BC, Canada.
Email: nico_munoz@sfu.ca

Abstract

1. The impacts of non-native species are hypothesised to be proportional to the functional distinctiveness of invaders in their invaded ecosystems. Throughout the Patagonia region of southern South America, Chinook salmon (*Oncorhynchus tshawytscha*) have recently established non-native populations, and their anadromous, semelparous life cycle could be functionally unique such that marine-derived nutrients are delivered to streams which have historically lacked such a resource linkage with the ocean.

2. We tested the hypothesis that salmon subsidise biofilm-associated algae in streams throughout the Aysén province of southern Chile. Using spatial and temporal variation in the presence of salmon among multiple streams and across two spawning seasons, we found strong evidence of salmon-subsidised algae in three out of four streams examined that have spawning salmon populations.

3. The biofilm of subsidised streams had enriched stable isotopic ratios of nitrogen and carbon, indicating that marine-derived nutrients were incorporated by biofilms. This nutrient uptake translated into increased algal biomass and percent of total biofilm biomass composed of algae, indicating that the incorporation of marine-derived nutrients stimulated autotrophic production of biomass.

4. In one stream, the incorporation of marine-derived nutrients by biofilm occurred in only one of the two studied spawning seasons. Incorporation occurred in a year with low flows of water throughout salmon spawning (4.59 m³/s) and did not occur in a year with much higher flows (41.6 m³/s), suggesting that inter-annual variation in discharge can mediate the subsidising effect of salmon.

5. These results indicate that Chinook salmon have bridged the historical gap between productive marine ecosystems and nutrient-poor stream ecosystems in Patagonia. Anadromous salmon can be a significant source of nutrients in nutrient-limited catchments, and their ongoing expansion in southern South America is likely to entail ecological impacts in stream and riparian food webs.

KEYWORDS
eutrophication, invasive species, *Oncorhynchus*, periphyton, stable isotopes

Bryan D. Neff and John D. Reynolds should be considered joint senior authors

1 | INTRODUCTION

The movement of energy and nutrients among ecosystems is fundamental to the productivity and composition of biological communities (Polis et al., 1997). These resources are often transported into ecosystems through the movement of animals and they can increase the biomass within recipient food webs beyond that which can be supported by in situ productivity alone (e.g. Atlas et al., 2013; Graham et al., 2018). Such subsidies are recognised as having a vital ecological function. For example, seabirds and anadromous fish bring marine-derived nitrogen and phosphorus into terrestrial and freshwater ecosystems (e.g. Graham et al., 2018; Hood et al., 2019; Walsh et al., 2020), and the widespread collapse of seabird and anadromous fish populations has resulted in the global transfer of phosphorus from sea to land declining over 96% from historic levels (Doughty et al., 2016). Whereas the loss of historic connections among ecosystems disrupts nutrient cycles and recipient food web productivity (Gresh et al., 2000), the emergence of novel resource linkages among ecosystems that occur due to the establishment of non-native species could be equally disruptive.

One of the best described systems of resource subsidisation concerns Pacific salmon (*Oncorhynchus* spp.) in freshwater and terrestrial ecosystems in Pacific North America. Pacific salmon are anadromous, gaining 99% of their biomass in the ocean before returning to their natal streams to spawn en masse and subsequently die of rapid senescence (Quinn, 2005). Aquatic and terrestrial ecosystems are typically nutrient limited in that the biomass of primary producers is limited by the availability of nitrogen and phosphorus (Elser et al., 2007). As such, the annual pulse of marine-derived nitrogen and phosphorus from salmon carcasses can strongly subsidise autotrophic and heterotrophic production in both streams and the surrounding riparian habitat (Gende et al., 2002; Janetski et al., 2009; Walsh et al., 2020). Salmon-derived nutrients enter these food webs through a wide variety of pathways. Direct consumption of salmon carcasses increases the biomass of collector and shredder invertebrates (Chaloner & Wipfli, 2002; Lessard & Merritt, 2006), and consumption of salmon eggs increases the biomass of a variety of stream fishes (Armstrong et al., 2010; Bailey & Moore, 2020; Bentley et al., 2012; Hermann et al., 2020; Scheuerell et al., 2007; Swain et al., 2014). Carcasses that are deposited in the riparian zone are heavily consumed by terrestrial insects (Hocking & Reimchen, 2002) and cause soils to accumulate nitrogen, affecting the composition of riparian plant assemblages (Hocking & Reynolds, 2011; Morris & Stanford, 2011) and enhancing the growth of spruce trees (Quinn et al., 2018). Some carcasses are flushed downstream into estuaries, where they are scavenged by gulls and subtidal invertebrates (Field & Reynolds, 2013; Reimchen, 2017). Alternatively, salmon-derived nutrients can enter stream food webs through the uptake of dissolved nutrients in the water column by the microbial communities comprising benthic biofilms.

Biofilms are a complex aggregation of algae, bacteria, protozoa, fungi, and archaea found on submerged rocks. They can be subsidised by salmon at multiple time scales, both during spawning (i.e. weeks; Chaloner et al., 2007; Schuldt & Hershey, 1995) as well as months after spawning due to retention of salmon-derived nutrients within the ecosystem (Harding et al., 2014; O'Keefe & Edwards, 2002). Conversely, salmon can have a negative effect on biofilm biomass during spawning due to the physical disturbance of the streambed caused by females digging nests (i.e. redds) (e.g. Holtgrieve & Schindler, 2011; Moore & Schindler, 2008), although streams that experience such disturbance during spawning can still retain salmon nutrients (Rex & Petticrew, 2008) and can be subsidised by salmon months after spawning (Harding et al., 2014). Subsidised biofilm can propel salmon-derived nutrients up the food web. For example, biofilm biomass and macroinvertebrate density are 15 and 25 times higher, respectively, in a salmon subsidised stream in Alaska relative to a non-subsidised stream (Wipfli et al., 1998). In British Columbia, salmon spawner density among streams is positively associated with the abundance of biofilm-grazing mayflies (Verspoor et al., 2011) and insectivorous forest birds (Wagner & Reynolds, 2019). Indeed, as the resource base for higher trophic levels, biofilms can have strong, bottom-up effects, as key sites of primary production and the transfer of carbon from autotrophic to heterotrophic organisms (Risse-Buhl et al., 2012; Rosemond et al., 2000).

One of the few instances of a Pacific salmon establishing self-sustaining populations outside of their native range has recently occurred in Patagonia, southern South America. Introduction attempts for commercial purposes in the 1970s and 1980s seeded the establishment of Chinook salmon (*Oncorhynchus tshawytscha*) populations in a few streams in southern Chile (Correa & Gross, 2008). Since these efforts, Chinook have colonised seemingly all inhabitable catchments in southern Chile, from 39 to 55°S, and have even crossed the Strait of Magellan and colonised Atlantic-draining catchments in Argentine Patagonia (Di Prinzio & Pascual, 2008). Most of the research to date on Patagonian Chinook has focused on genetic and life history diversity among populations (e.g. Araya et al., 2014; Correa & Moran, 2017; Gomez-Uchida et al., 2018; Narum et al., 2017). Conversely, the food web and ecosystem impacts of these non-native populations have received little study (Arismendi & Soto, 2012; Ciancio et al., 2008; Soto et al., 2007).

Chinook are the least abundant species of Pacific salmon in North America and require the largest streams and substrate sizes for spawning habitat (Gottesfeld et al., 2008; Kondolf &Wolman, 1993). Larger substrates are harder to dislodge from the streambed and are therefore less prone to disturbance during salmon spawning (Holtgrieve et al., 2010; Janetski et al., 2009, 2014). The larger substrates and lower densities in which Chinook spawn (relative to other salmon species) could mean that, in general, their disturbance (i.e. negative) effects are smaller than their subsidy (i.e. positive) effects on stream biofilms. For example, low densities of spawning Chinook salmon (<0.001 spawners/m^2) caused a 46% increase in the gross primary production of three streams in Washington, U.S.A. (Benjamin et al., 2016), whereas moderate densities of spawning sockeye (*Oncorhynchus nerka*) salmon (>0.6 spawners/m^2) caused a 72% reduction in

gross primary production in three streams in Alaska (Holtgrieve & Schindler, 2011). Although Chinook can have dramatic disturbance effects through the creation of spawning *dunes* in the streambed (Gottesfeld et al., 2008), this behaviour is observed only in the largest rivers in which they spawn and has not been reported in Patagonia, suggesting that they could be more likely to have a subsidy effect in their South American range.

If Chinook salmon subsidise stream biofilms in Patagonia, these subsidies would represent a novel resource linkage between marine and freshwater ecosystems. The delivery of marine-derived nutrients to these ecosystems has historically not occurred due to the apparent absence of any native anadromous fish that spawn in the upper reaches of Andean catchments (Alò et al., 2019). As such, Chinook are functionally unique in Patagonia. The ecological impacts of invasive species are hypothesised to be proportional to the functional distinctiveness of the invader in the recipient community (Ricciardi & Atkinson, 2004; Ricciardi et al., 2013; Schittko et al., 2020; Vitousek, 1990), meaning the novel function performed by Chinook would be predicted to have large impacts. Moreover, southern Chile has one of the lowest rates of atmospheric nitrogen deposition in the world (Dentener et al., 2006). An annual pulse of marine-derived nutrients in these otherwise nutrient-poor systems (Diaz et al., 2007; Perakis & Hedin, 2002) could therefore have a disproportionately strong ecological effect (Flecker et al., 2010).

The aim of this study was to test the hypothesis that nutrients from non-native Chinook salmon subsidise biofilm-associated algae in Patagonian streams. We sampled biofilm in the presence and absence of senescent salmon across 2 years and multiple streams. We used stable isotope analysis to assess the incorporation of marine-derived carbon and nitrogen by biofilms. Stable isotope analysis is a powerful, time-integrated technique for assessing sources of nutrients in food webs (Peterson & Fry, 1987). Salmon tissue is highly enriched in ^{13}C and ^{15}N due to their trophic interactions in the marine food web, allowing the use of carbon and nitrogen isotopic ratios to test for the incorporation of marine-derived nutrients in consumer tissue (Gende et al., 2002; Naiman et al., 2002). To infer the presence or absence of a subsidy effect (i.e. an increase in biomass due to incorporation of an externally produced, donor-controlled resource), we also tested whether marine-derived nutrients increase the biomass of algae by measuring biofilm chlorophyll *a* (a measure of algal biomass) and ash-free dry mass (a measure of total organic matter). We tested our hypothesis using two experimental designs that utilised spatial and temporal differences in the presence of senescent salmon.

2 | METHODS

2.1 | Study sites

Field surveys were conducted in streams throughout the Aysén province of southern Chile (Figure 1). This area is in the Valdivian temperate rainforest ecoregion and is characterised by relatively low levels of human-modified land cover, forests dominated by southern beech trees (*Nothofagus* spp.), a seasonal climate, and strong longitudinal gradients in rainfall. Chinook salmon were first reported in this province in the early 2000s (Correa & Gross, 2008) and now have annual spawning runs in many tributaries of its Andean catchments. The main point sources for the Chinook invasion of Chile were stocking efforts between 1976 and 1987 in a few streams to the north (42°S) and south (51°S) of the Aysén province, suggesting that catchments here were colonised by straying salmon from established populations (Correa & Gross, 2008; Correa & Moran, 2017).

Five streams were used in this study: four in the Río Aysén catchment (El Toqui, Ñirehuao, Emperador Guillermo, and Huemules) and one in the Río Baker catchment (Jaramillo; Figure 1). These streams are dominated by non-native rainbow trout (*Oncorhynchus mykiss*) and brown trout (*Salmo trutta*), with native fishes being rare (N. J. Muñoz, unpublished data) probably because of predation and competition imposed by trout (Correa & Hendry, 2012). The streams range from third to fifth order and from 6 to 65 km in total length, the largest being Ñirehuao, which drains nearly 2,000 km^2 (Table 1). Flows in these streams generally follow a rainfall–snowmelt transitional hydrologic regime and are typically high during the salmon spawning season due to heavy autumn rains. In Jaramillo, flows are regulated by a small, nearby lake. These streams were selected for the study due to the presence of a barrier to salmon migration in each one, which was key to the study design (described below). Additional criteria included accessibility, evidence of salmon runs, and similar land use upstream versus downstream of the barrier.

2.2 | Control–Impact study design

In February–May 2016, we conducted a *Control–Impact* (CI) study in four streams (El Toqui, Ñirehuao, Emperador Guillermo, and Jaramillo, with Huemules omitted due to logistical constraints). *Control* and *Impact* refer to paired sites within a stream that differ in the presence of salmon. In El Toqui, Ñirehuao, Huemules, and Jaramillo, adult Chinook salmon arrive throughout February and March and, over the course of several weeks, aggregate at distinct spawning sites and eventually die of senescence. Upstream of these spawning sites in each stream is habitat that is not occupied by salmon due to a natural barrier to salmon migration in the form of a waterfall (except for El Toqui, see below). Thus, upstream sites were paired with downstream sites such that they differ in the presence of salmon. This upstream–downstream approach has been used by several other studies (e.g. Chaloner et al., 2007; Christie & Reimchen, 2008; Harding et al., 2014; Hocking & Reimchen, 2002; Lessard & Merritt, 2006; Mitchell & Lamberti, 2005) and is powerful in that it controls for the physical and chemical attributes of the streams (see below for relevance of variables that affect biofilm).

In El Toqui, a small, run-of-the-river hydroelectric facility diverts water at a small dam located directly upstream of a steep gradient in the stream. It releases the water back into the stream at the bottom
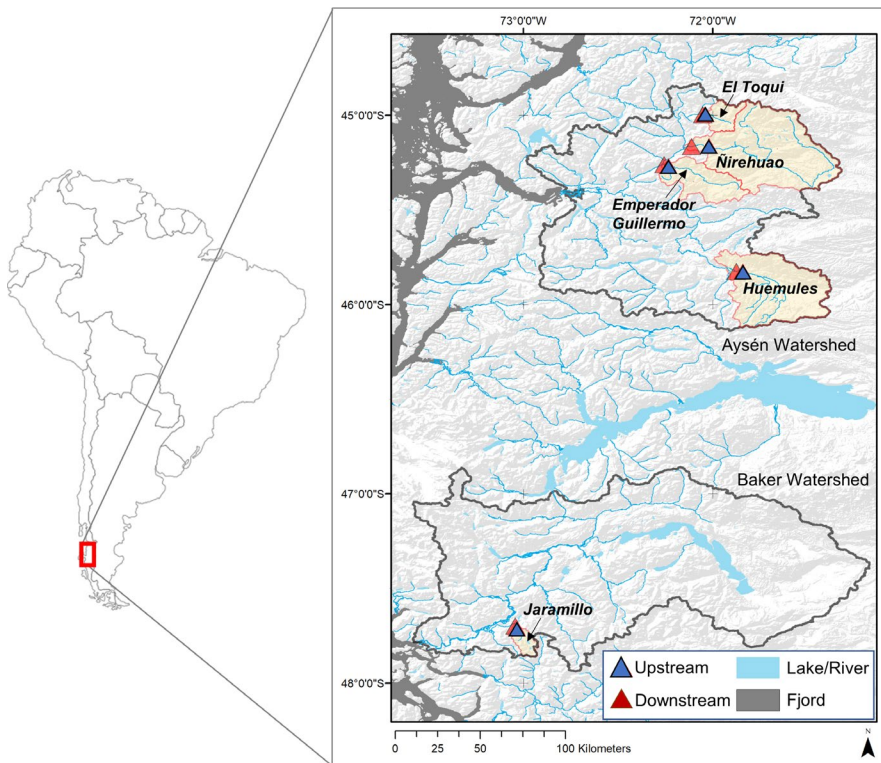
FIGURE 1 Map of study streams located throughout the Aysén province of southern Chile. Shown are the upstream and downstream sites that were sampled in this study, as well as the drainage areas of each study stream (outlined in red) and of the two catchments in which study streams were located (outlined in grey) [Colour figure can be viewed at wileyonlinelibrary.com]

of the gradient, upstream of the observed spawning habitat. The habitat between the release site and the dam consists of pool-riffle habitat caused by the steep elevation gain. Such habitat is unsuitable for Chinook spawning, and visual surveys found no evidence of spawning throughout this section. This allowed us to place the Control site below the gradient and shortly downstream of the release site, thereby maintaining the key feature of the CI design of having only the presence of salmon being different between the sites.

Emperador Guillermo (Emperador hereafter) was used as a reference stream in the CI study because it has a waterfall shortly upstream (c.2 km) of its confluence with the main stem and seemingly no salmon that spawn in the available habitat below this waterfall. Discussions with several local people yielded mixed responses as to whether adult salmon enter the reach below the waterfall. In a snorkel survey in 2018, we identified juvenile Chinook in the reach below the waterfall (N. J. Muñoz, unpublished data). However, multiple foot surveys were conducted at each stream during the CI study and Emperador was the only one in which there were no live or dead adult salmon observed at any point. Although it is possible that salmon spawn in the downstream portion of Emperador, we concluded that it would have to be limited to far fewer individuals than at the other streams, thereby allowing us to use it as a reference system for the upstream versus downstream comparisons.

We deployed unglazed ceramic tiles (30 × 20 cm) to measure biofilm biomass and stable isotope composition at each Control and Impact site. Ceramic tiles reduce the sampling variability inherent in sampling biofilm from natural rock surfaces while accurately representing the algal, bacterial, and macroinvertebrate densities and species compositions of rocks after only a few weeks of incubation

(Lamberti & Resh, 1985). The timing of tile deployment was based on the salmon spawn timing observed in each stream. Spawn timing was assessed using repeated visual surveys on foot, beginning in early February. The number of live and dead salmon observed during each survey was recorded (Table 1); while not exhaustive, these surveys were sufficient to reveal temporal patterns of spawner presence and behaviour. In general, fish occupied pool habitats in February and early March, then occupied riffle-run transition zones in mid-March to April and displayed mating behaviours such as redd construction and maintenance. Impact sites were chosen at locations downstream of the majority of observed spawning (0.13–0.2 km downstream of closest spawning location across streams). Tile deployment occurred part way through the spawning season, shortly after the first observation of mating behaviours (mid-March to mid-April across all four streams). Tiles were deployed in the corresponding Control sites within 24 hr of Impact site deployment. At each site, tiles were arrayed across the width of the stream in multiple rows, directly upstream of riffle habitat. In more narrow streams, tiles were deployed as such in front of two adjacent riffle habitats. Each tile was glued to a heavy brick, anchoring them to the streambed. Nine to 12 tiles were deployed at each site.

We selected the location of each Impact and Control site with the intention of minimising any longitudinal (i.e. upstream to downstream) variation in physical characteristics among sites. An important longitudinal effect in rivers is that water generally increases in temperature as it moves from headwaters to the terminal mouth (Vannote et al., 1980). To minimise any temperature differences, paired sites were placed as closely together as possible, with no significant tributaries entering streams between sites (as confirmed through stream surveys and satellite imagery). Sites were

TABLE 1 Stream and survey characteristics for study systems in Patagonia, southern Chile

Stream name	Latitude (S)	Longitude (W)[a]	Drainage area (km²)	Order	Mean discharge (m³/s)[b]	Total length (km)	Length of surveys for salmon (km)	Maximum no. salmon observed[c]	Mean NO_3^{2-} (μg/L)[d]	Mean PO_4^- (μg/L)[d]
El Toqui	44°59'50"	72°03'15"	207	4	4.6	31	1.5	25	0.081 (0.077)	0.010 (0.013)
Ñirehuao	45°09'57"	72°06'46"	1,971	5	31.4	65	2.3	26	0.048 (0.055)	0.011 (0.010)
Emperador	45°15'41"	72°15'27"	566	5	13.4	29	2.1	0	0.048 (0.049)	0.011 (0.010)
Huemules	45°49'33"	71°52'34"	1,423	5	15.8	34	1.8	34	N/A	N/A
Jaramillo	47°41'51"	73°02'43"	117	3	2.8	6	1.0	63	0.032 (0.017)	0.004 (0.002)

Latitudes and longitudes indicate the downstream (i.e. Impact) sites used in each stream. See Figure 1 for location of upstream sites.

[b]Mean discharge represents the annual mean discharge and was calculated using long-term mean precipitation and drainage area.

[c]Maximum no. salmon observed is the maximum number of live and dead salmon observed on an individual survey date.

[d]Mean (and standard deviation) nitrate (NO_3^{2-}) and phosphate (PO_4^{2-}) concentrations in each stream were obtained from the Government of Chile's water quality monitoring programme. These values were calculated using measurements taken across seasons and in multiple years (n = 4 to 35 measurements per stream per nutrient). No nutrient data were available for Huemules.

1.5–2.2 km apart in El Toqui, Emperador, and Jaramillo (Table S1; Figure 1). Long, inaccessible reaches throughout Ñirehuao necessitated that sites there were 9.2 km apart. To evaluate if temperatures differed among sites, we recorded hourly temperatures at each site in Ñirehuao and Jaramillo using HOBO temperature loggers (Onset, Bourne, MA, U.S.A.) from March to May. Problems with logger and data retrieval prevented temperatures in the other two streams from being recorded. Also, to account for variation in light availability, we measured percent open canopy at each tile using a spherical densiometer as well as the depth of each tile in the water column on the day of sampling (Table S1).

Biofilm samples were collected 10 weeks after deployment and more than 4 weeks after all spawning had occurred (i.e. no live salmon in the streams, only carcasses). Control sites were sampled either on the same day or within 24 hr of their corresponding Impact site. Any invertebrates present on the tiles at the time of collection were removed. A portion of tiles were lost in all but two sites, often due to water levels decreasing over the study period such that tiles placed in shallower locations (e.g. near banks) were out of water at some point (tiles sampled at each site: n = 5–12, Table S1).

Samples for stable isotope analysis were taken from approximately 576 cm² of tile surface area. Stream water and a brush were used to transfer the biofilm in this area into a plastic container. Containers were kept in the dark and on ice for 2–8 hr, during which they were transported to laboratory facilities at the Centro de Investigación en Ecosistemas de la Patagonia (CIEP) in Coyhaique. Upon arrival at CIEP, samples were dried at 70°C, coarsely ground, and stored until further processing. Later, samples were ground into a fine powder and a subsample (2–3 mg dried weight) was analysed for nitrogen and carbon isotope abundance at the UC Davis Stable Isotope laboratory (http://stableisotopefacility.ucdavis.edu/). Analysis employed a PDZ Europa ANCA-GSL elemental analyser interfaced to a PDZ Europa 20-20 isotope ratio mass spectrometer (Sercon Ltd.). Stable isotopes are expressed in delta notation (δ) as ratios of isotopes in the sample relative to that in a standard (atmospheric N_2 for nitrogen, Vienna Pee Dee Belemnite for carbon). This ratio is expressed in parts per thousand following:

$$\delta^{15}N \ \text{ or } \ \delta^{13}C = \left(\frac{R_{sample}}{R_{standard}} - 1 \right) \times 1,000$$

where R is the ratio of the heavy isotope (^{15}N or ^{13}C) to light isotope (^{14}N or ^{12}C). One value of $\delta^{15}N$ and $\delta^{13}C$ was obtained for each sampled tile. Tiles at the Jaramillo Control site had low levels of biofilm biomass and only four tiles were able to be sampled for isotope analysis. Of these four samples, three contained insufficient nitrogen content for $\delta^{15}N$ determination (but enough carbon for $\delta^{13}C$ determination).

Chlorophyll a (chl a) is the most abundant photosynthetic pigment in plants and provides an estimate of algal (i.e. autotrophic) biomass (Steinman et al., 2017). Ash-free dry mass (AFDM) is a measurement of the loss of mass upon oxidation and estimates the biomass of all organic matter in biofilm including algae, bacteria, and detritus. Samples for chl a and AFDM were taken from

a circular area (23.8 cm^2) in the centre of each tile. Samples were collected and transported to CIEP as described above. At CIEP, samples were filtered onto glass fibre filters (Whatman, 47 mm, 0.7 μm) and stored in the dark at −20°C. Pigments were extracted into 10 ml of 90% acetone for 24 hr and measured using the spectrophotometry method described in Steinman et al. (2017) whereby absorbance measurements are taken before and after acidification to account for chlorophyll degradation products. Using the remaining material (i.e. after pigment extraction), filters were dried, weighed, oxidised at 500°C for 1 hr, cooled to room temperature, and then weighed again to estimate AFDM (following Steinman et al., 2017). One value of chl *a* (μg/cm^2) and AFDM (mg/cm^2) was obtained for each sampled tile. Because the samples that we used to estimate AFDM were washed with acetone beforehand, our measure of AFDM estimates the total organic material in the sample apart from non-polar organic matter such as chlorophylls and accessory pigments (e.g. Chaloner et al., 2007; Mitchell & Lamberti, 2005). To assess the relative amount of chl *a* to total (non-polar) organic matter, the percent of total biofilm biomass composed of chl *a* (chl *a* %) was calculated by converting AFDM to μg/cm^2 and using the following formula:

$$chla\% = \frac{chla}{chla + AFDM} \times 100$$

Higher chl *a* % values indicate that algae (and cyanobacteria, which are photosynthetic bacteria containing chl *a*) make up a greater proportion of total biofilm biomass relative to heterotrophic or detrital components of biofilm. Because we hypothesised that salmon-derived nutrients would increase autotrophic biomass, we predicted that chl *a* % would be higher in Impact sites relative to Control sites.

2.3 | Before–After–Control–Impact study design

In addition to the CI study, we conducted a Before–After–Control–Impact (BACI) study in January–May 2018 to further assess the effect of salmon on stream biofilms. Before–After–Control–Impact designs are broadly used in environmental monitoring programmes because they provide rigorous tests of the impacts of specific activities (Downes et al., 2002). Here, this design included the CI comparison of the CI study as well as a Before–After comparison at each site to account for any confounding differences between upstream and downstream sites (e.g. temperature differences).

The BACI study was conducted in Ríos Ñirehuao and Huemules. El Toqui was originally included in this design, however all tiles were lost in this stream after a storm during the study. Indeed, the summer and autumn of 2018 had much more precipitation than in 2016, resulting in substantially higher levels of discharge during the BACI study (e.g. monthly mean discharge in April 2016 and April 2018 was 4.59 and 41.6 m^3/s, respectively, in Ñirehuao). In the Before period, 15 tiles were deployed at each site in early January and sampled after 5 weeks, before salmon had returned to spawn. Tiles were

returned to the stream after sampling. Visual surveys downstream of the migration barriers were again used to monitor the progression of spawning in February through April. Initiation of the After period occurred in the second week of April, when carcasses had begun accumulating in both streams (in a ratio of dead to live fish of about 1:2). At this time, all tiles were thoroughly cleaned using a large brush, removing the biofilm that had accumulated since the end of the Before period. Control and Impact sites were cleaned within 24 hr of each other. Tiles were then incubated in the streams for 5 weeks, after which they were sampled, processed, and analysed for biomass and isotopic composition as described above. Site and sampling characteristics of the BACI study are described in Table S2. Hydrographs of Ñirehuao and Huemules during the study period are shown in Figure S1, with reference to the timing of tile deployment.

2.4 | Data analysis

Within each stream in the CI study, we assessed whether tile depth or percent open canopy were to be included as covariates in the full models by testing for differences in these variables between upstream and downstream sites and by regressing the five response variables (δ^{13}C, δ^{15}N, chl *a*, AFDM, and chl *a* %) onto depth and canopy cover. Tile depth and percent open canopy were not significantly different between upstream and downstream sites (Table S1) and were not significantly associated with response variables ($0.001 > r^2 > 0.22$) except for a negative relationship between percent open canopy and AFDM in Emperador ($r^2 = 0.31, p = 0.024$). Because this relationship was found in only one stream and was in the opposite direction to that expected and thus probably spurious, we did not include canopy cover (or depth) as a covariate in the full models.

For the CI study, we used linear models with *stream* (four levels) and *site* (two levels) as fixed effects to analyse the variation in the response variables. We also included the *site* × *stream* interaction because we predicted that the reference stream (Emperador) would not differ between sites. Tukey's post hoc tests were used to evaluate stream-specific differences between Control and Impact sites. Because only one δ^{15}N value was obtained from the Jaramillo Control site, Jaramillo was not included in the δ^{15}N analysis. The assumptions of normality were inspected, and Box Cox transformations were used for δ^{13}C, chl *a*, AFDM, and chl *a* % to improve normality (λ = 0.75, 0.26, 0.06, and 0.63, respectively). The relationship between chl *a* and δ^{15}N within streams was assessed using linear regression. These relationships were used as an additional line of evidence as to whether differences in algal biomass between Control and Impact sites were driven by salmon. Specifically, if the relationship between chl *a* and δ^{15}N within streams was positive, and if this relationship was stronger in salmon-bearing streams compared to the reference stream, this would suggest that the incorporation of salmon-derived nitrogen increased algal biomass.

In the BACI study, there were no significant relationships (within streams and periods) between any of the response variables and percent open canopy or tile depth ($0.001 > r^2 > 0.12$). In Ñirehuao during the After period, tile depth was significantly lower in the downstream site than in the upstream site (Table S2); however, because this difference was not associated with variation in any of the response variables, we did not include depth (or canopy cover) as a covariate in the full models. We used linear models with *site*, *period*, and *stream* as fixed factors (all with two levels) to analyse the variation in the five response variables. Given the stream-specific effects observed in the CI study (described below), we included all interaction terms. In this BACI design, an impact of salmon would be indicated by a significant *site × period* effect (Downes et al., 2002). If the two streams were differentially impacted by salmon (e.g. an impact in only one of the streams), this would be indicated by a significant *site × period × stream* effect. The other interaction effects (i.e. *site × stream* and *period × stream*) were not of direct interest but were included in the models because stream-specific effects were expected. The assumptions of models were inspected, and Box Cox transformations were used for chl *a*, AFDM, and chl *a* % to improve normality ($\lambda = 0.14$, 0.42, and 0.63, respectively). Tukey's post hoc tests were used to evaluate differences between Control and Impact sites within streams and periods. The relationship between chl *a* and δ^{15}N within streams and periods was assessed using linear regression. All linear models were analysed using the *lm()* function in R (R Core Team, 2020).

3 | RESULTS

3.1 | Control–impact study

The results of the CI study support a nutrient subsidy effect of salmon in El Toqui and Jaramillo but not in Ñirehuao. In general, biofilm isotopic composition and biomass differed between upstream and downstream sites, and this difference depended on the stream (Table 2). As predicted, biofilm δ^{13}C and δ^{15}N were significantly enriched in the downstream (Impact) sites of each salmon-bearing stream relative to upstream (Control) sites, and they were not different between sites in the salmon-free reference stream (i.e. Emperador; Figure 2a, b). The salmon-impacted sites of Jaramillo and El Toqui had significantly higher chl *a* than upstream sites, whereas there was no difference in chl *a* between sites at both Ñirehuao and the reference stream (Figure 2c). Ash-free dry mass was significantly higher at the salmon-impacted site of Jaramillo relative to the upstream site (Figure 2d). Chlorophyll *a* and AFDM were strongly and positively associated in El Toqui ($r^2 = 0.83$, $p < 0.001$), Ñirehuao ($r^2 = 0.89$, $p < 0.001$), Jaramillo ($r^2 = 0.90$; $p < 0.001$), and, to a lesser extent, the reference stream ($r^2 = 0.23$, $p = 0.058$; Figure S2). Chlorophyll *a* % was significantly higher in the salmon-impacted sites of El Toqui and Jaramillo relative to upstream sites, increasing by an average of 25 and 74%, respectively (Figure 2e).

Chlorophyll *a* and δ^{15}N were strongly and positively associated in El Toqui ($r^2 = 0.78$, $p < 0.001$), whereas they were not associated in Ñirehuao ($r^2 = 0.02$) or the reference stream ($r^2 = 0.01$; Figure 3a).

TABLE 2 Effects of site (upstream or downstream) and stream on biofilm biomass and isotopic traits in the Control–Impact study of Chinook salmon in Patagonia, southern Chile

Trait	df	SS	F	P
δ^{13}C				
site	1	34.5	59.3	**<0.001**
stream	3	61.2	35.0	**<0.001**
site × stream	3	7.88	4.51	**0.008**
residual	41	23.9		
δ^{15}N				
site	1	14.4	87.4	**<0.001**
stream	2	7.23	22.0	**<0.001**
site × stream	2	15.7	47.8	**<0.001**
residual	33	5.43		
chl *a*				
site	1	0.18	21.6	**<0.001**
stream	3	4.34	179	**<0.001**
site × stream	3	0.11	4.55	**0.006**
residual	54	0.44		
AFDM				
site	1	0.001	5.64	**0.021**
stream	3	0.17	261	**<0.001**
site × stream	3	0.001	2.21	0.097
residual	54	0.01		
chl *a* %				
site	1	0.07	36.6	**<0.001**
stream	3	0.76	126	**<0.001**
site × stream	3	0.06	10.6	**<0.001**
residual	54	0.11		

Note: The results of linear models are summarised for effects on biofilm isotopic ratios of carbon (δ^{13}C) and nitrogen (δ^{15}N) as well as algal biomass (chl *a*), total biofilm biomass (AFDM), and percent of total biomass composed of chl *a* (chl *a* %). Significant effects are bolded ($p < 0.05$).

While being limited by having only one δ^{15}N value from the Control site, this relationship was also positive in Jaramillo ($r^2 = 0.22$).

The mean ($\pm$ *SD*) temperature at the downstream site in Ñirehuao was 0.25 ± 0.30°C warmer than at the upstream site, whereas temperatures at the downstream site in Jaramillo were very similar to those at the upstream site (0.03 ± 0.22°C warmer; Table S1). Because they are similar distances apart, the sites in Emperador and El Toqui are likely to have negligible temperature differences, as with those in Jaramillo.

3.2 | Before–After–Control–Impact study

The results of the BACI study support a nutrient subsidy effect of salmon in Huemules but not in Ñirehuao. The effect of
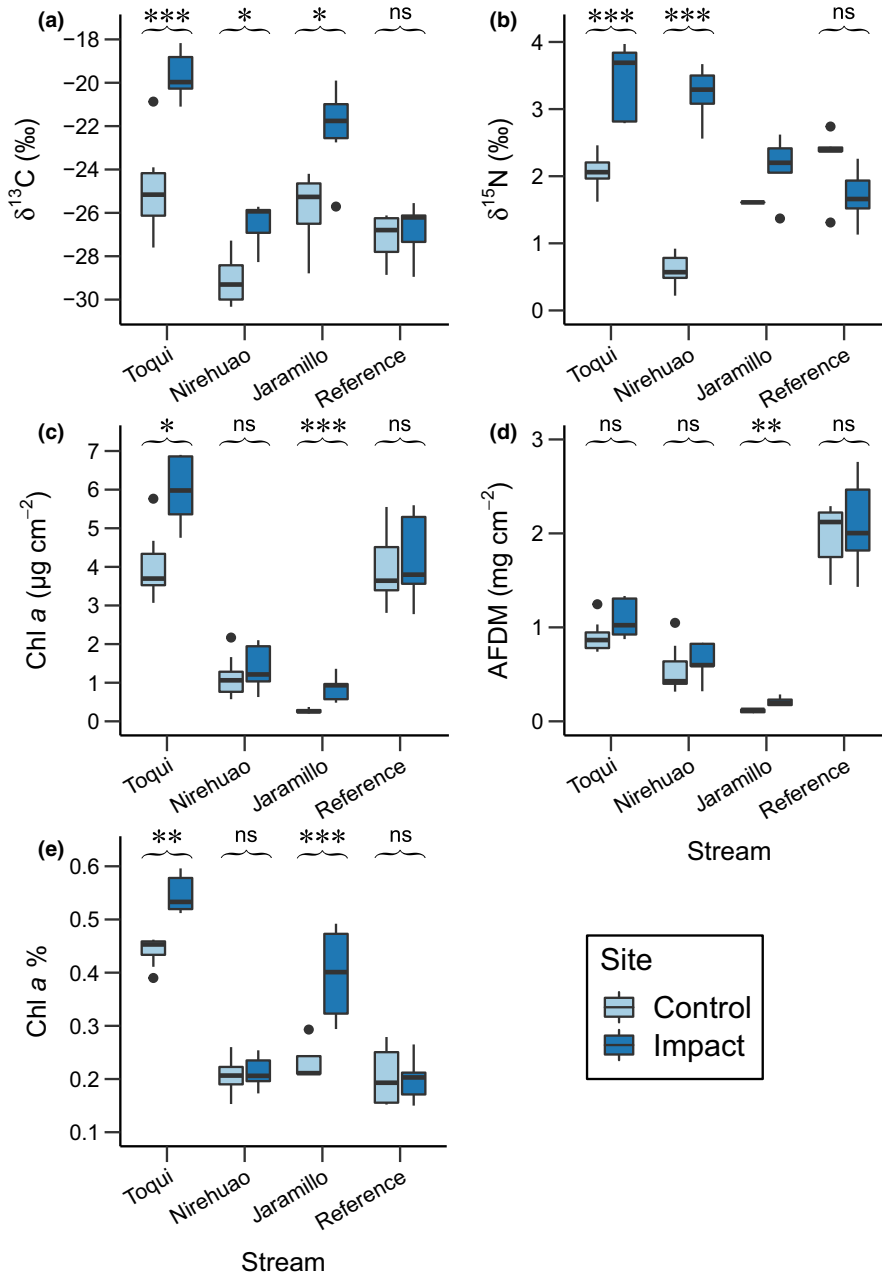
FIGURE 2 Effect of Chinook salmon on (a) biofilm isotopic ratio of carbon (δ^{13}C), (b) biofilm isotopic ratio of nitrogen (δ^{15}N), (c) algal biomass (chl *a*), (d) total biofilm biomass (AFDM), and (e) percent of biofilm biomass composed of chlorophyll *a* (chl *a* %) among streams in the Control–Impact study. Biofilm was sampled in Control (upstream, salmon-free) and Impact (downstream, salmon-impacted) sites of four streams, one of which (the reference stream) had no salmon in the downstream site. Only one δ^{15}N value was collected from the Jaramillo Control site, preventing comparison of δ^{15}N for this stream. The results of Tukey's post hoc comparisons of Control and Impact sites are displayed (***$p < 0.001$; **$p < 0.01$; *$p < 0.05$; ns $p > 0.05$) [Colour figure can be viewed at wileyonlinelibrary.com]

salmon on biofilm stable isotopic composition differed between streams, with significant *site × period × stream* effects on biofilm δ^{13}C ($F_{1,83} = 55.6$, $p < 0.001$) and δ^{15}N ($F_{1,83} = 22.1$, $p < 0.001$; Table S3). In Huemules, after the senescence of salmon, biofilm δ^{13}C was more enriched in the downstream (Impact) site than in the upstream (Control) site, whereas the opposite was true before the arrival of salmon (Figure 4a). δ^{15}N did not differ between sites at Huemules before the arrival of salmon, but, after salmon senescence, the downstream site was significantly enriched in δ^{15}N relative to the upstream site (Figure 4b). These data indicate that salmon-derived carbon and nitrogen were incorporated into biofilm in Huemules. Conversely, δ^{13}C and δ^{15}N were not different between sites at Ñirehuao after salmon senescence (Figure 4a, b), indicating that salmon-derived carbon and nitrogen were not incorporated into biofilm in this stream. Instead, there was a

depletion in δ^{13}C and an enrichment in δ^{15}N from the Before to the After period, irrespective of site.

There was an effect of salmon on algal and biofilm biomass that was similar in the two streams, with significant *site × period* effects on biofilm chl *a* ($F_{1,87} = 10.5$, $p = 0.002$), AFDM ($F_{1,87} = 9.69$, $p = 0.003$), and chl *a* % ($F_{1,87} = 8.42$, $p = 0.005$) and non-significant *site × period × stream* effects on these traits (Table S3). Indeed, the median value of each of these three traits was consistently higher in the downstream sites than in the upstream sites following salmon senescence (Figure 4c-e). However, post hoc tests found that, after salmon senescence, chl *a* and chl *a* % were significantly higher in the downstream site of Huemules only, whereas AFDM was significantly higher in Ñirehuao only (Figure 4c-e). Before the arrival of salmon, there were no differences in chl *a*, AFDM, or chl *a* % between sites in both streams. There was also a strong temporal (i.e. *period*) effect on these three traits, with each one being significantly
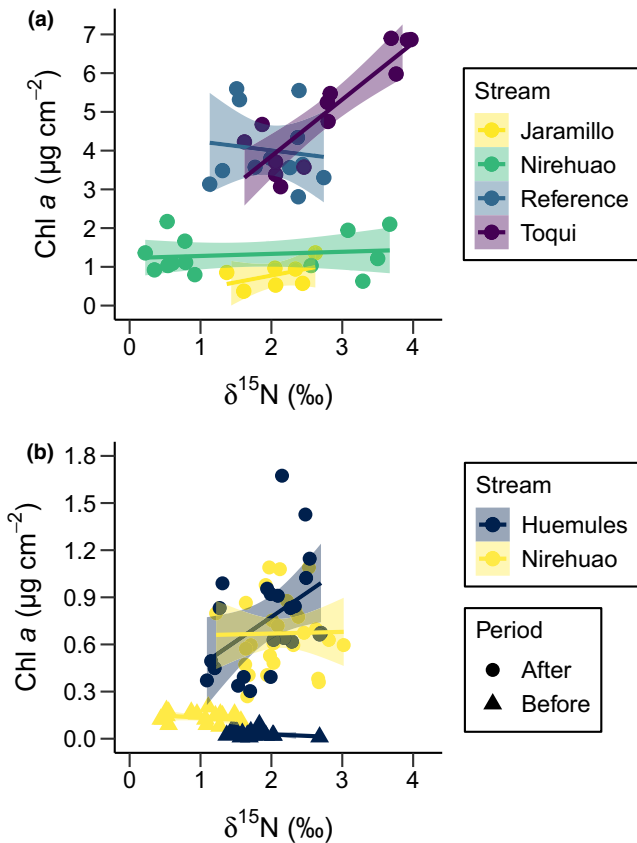
FIGURE 3 Relationship between biofilm isotopic ratio of nitrogen (δ¹⁵N) and algal biomass (chl *a*): (a) among streams in the Control–Impact study; and (b) among streams and periods in the Before–After–Control–Impact study. Shading represents 95% confidence intervals for predictions from a linear model [Colour figure can be viewed at wileyonlinelibrary.com]

higher in the autumn (after salmon senescence) relative to the summer (before salmon senescence) (Table S3, Figure 4c-e). Chlorophyll *a* and AFDM were strongly and positively associated within both time periods in both streams (Figure S3).

Delta ¹⁵N and chl *a* were positively and significantly associated in Huemules after salmon senescence ($r^2 = 0.20$, $p = 0.032$) but were not associated beforehand in Huemules ($r^2 = 0.08$) or in Ñirehuao during either period ($r^2 ≤ 0.06$; Figure 3b).

4 | DISCUSSION

The results of the two studies presented here provide evidence of salmon-mediated nutrient subsidies in three out of the four streams examined that have spawning Chinook salmon populations. These subsidies represent a novel resource linkage between marine and freshwater ecosystems in Patagonia, with non-native salmon contributing marine-derived nutrients to systems in which nutrients have historically only flowed downstream, from stream to sea. By performing a novel function in these invaded systems, salmon in Patagonia are likely to have ecological impacts (Ricciardi & Atkinson, 2004; Ricciardi et al., 2013; Vitousek, 1990).

The effects of salmon-mediated nutrient additions can be limited by the environmental context in which salmon spawning occurs, such as the temperature, light, discharge, and nutrient limitation status of streams (Armstrong et al., 2010; Bentley et al., 2012; Chaloner et al., 2007). If this context is poorly suited for additional growth of algae (e.g. nutrient-rich streams), salmon nutrients can have little effect on algal biomass (e.g. Janetski et al., 2014; Rand et al., 1992) and, subsequently, fail to support higher trophic levels (e.g. Lessard & Merritt, 2006). In three streams (El Toqui, Jaramillo, and Huemules), we found evidence that marine-derived nutrients were incorporated by biofilms and increased the biomass of algae (chl *a*; including cyanobacteria) as well as the proportion of biofilm biomass that is composed of algae (chl *a* %). We also found that total biofilm biomass (AFDM) and chl *a* were strongly and positively associated in all salmon-bearing streams and, in the BACI study, streams had higher chl *a*, AFDM, and chl *a* % in the autumn (after spawning) relative to the summer. These results suggest that: (1) algae are major components of biofilm in these streams; (2) marine-derived nutrients enter these streams at a time of year when algae are relatively abundant and comprise a greater proportion of total biofilm biomass, indicating that stream conditions are well-suited for algal growth; and (3) marine-derived nutrients significantly increase algal biomass after spawning in the autumn (except for in Ñirehuao, discussed below). The positive response of algae to salmon in El Toqui, Jaramillo, and Huemules is probably underlain by the strong nutrient limitation that is found in southern Chile (Perakis & Hedin, 2002) and represents the first described ecological impact of Patagonian salmon.

One of the environmental variables that modulates the effects of nutrient additions in streams is discharge. Temporary bouts of high stream flows (i.e. spates) or sustained high flows can scour the streambed and flush carcasses and dissolved nutrients downstream, thereby preventing salmon nutrients from being incorporated by biofilms (Chaloner et al., 2007; Richey et al., 1975). Inter-annual variation in discharge could explain the differences in biofilm incorporation of marine-derived nutrients observed in Ñirehuao during the CI and BACI studies here. During the BACI study in 2018, flows were high in Ñirehuao throughout the autumn spawning season (41.6 m³/s for all of April, compared to 4.89 m³/s in Huemules) and marine-derived nutrients were not incorporated by biofilm in this stream. Conversely, flows were exceptionally low during the CI study in 2016 (April discharge of 4.59 m³/s in Ñirehuao) and, unlike in 2018, the salmon-impacted site at Ñirehuao was enriched in biofilm δ¹⁵N and δ¹³C relative to the upstream site. These results suggest that the low flows of 2016 could have facilitated the incorporation of marine-derived carbon and nitrogen by biofilm. However, the incorporation of marine-derived nutrients in 2016 did not cause an increase in algal biomass in Ñirehuao, indicating that algal biomass in this stream is limited by some environmental variable other than nutrients. The lack of a subsidy effect in Ñirehuao and the difference in salmon nutrient incorporation among years are in line with other studies demonstrating that the effect of spawning salmon differs
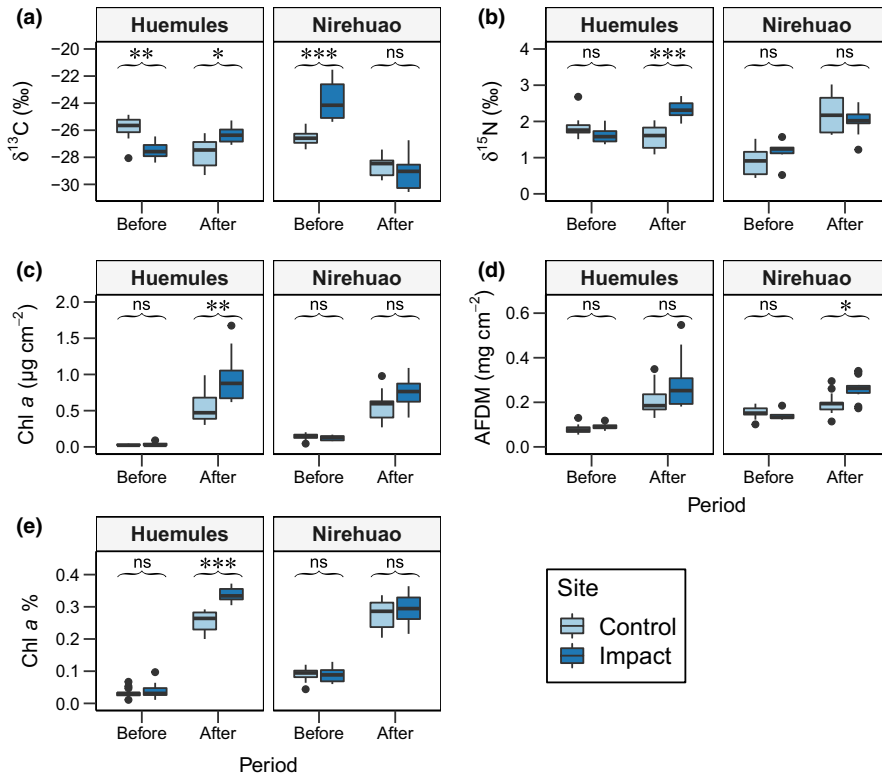
FIGURE 4 Effect of Chinook salmon on: (a) biofilm isotopic ratio of carbon (δ^{13}C); (b) biofilm isotopic ratio of nitrogen (δ^{15}N); (c) algal biomass (chl a); (d) total biofilm biomass (AFDM); and (e) percent of biofilm biomass composed of chlorophyll a (chl a %) among streams in the Before–After–Control–Impact study. Biofilm was sampled in Control (upstream, salmon-free) and Impact (downstream, salmon-impacted) sites of two streams (Huemules and Ñirehuao), before and after salmon spawning. The results of Tukey's post hoc comparisons of Control and Impact sites within periods are displayed (***$p < 0.001$; **$p < 0.01$; *$p < 0.05$; ns $p > 0.05$) [Colour figure can be viewed at wileyonlinelibrary.com]

among streams and among years in individual streams (e.g. Chaloner et al., 2007; Mitchell & Lamberti, 2005). As such, the vulnerability of individual streams in Patagonia to the subsidising effect of salmon is probably set by an inter-annual interplay between discharge and other environmental variables such as baseline nutrient regime, geomorphology, and temperature.

The abiotic and biotic characteristics of lotic ecosystems change predictably from headwaters to terminal mouths (Vannote et al., 1980) and can potentially confound upstream–downstream comparisons. During the CI study here, the stream with the largest distance between study sites (Ñirehuao, 9.2 km apart) was $0.25 \pm 0.30°$C warmer at the downstream site relative to the upstream site. This small difference in temperature could slightly confound our between-site comparisons of biofilm biomass and isotopic signatures in this stream. Nevertheless, there were no significant differences in chl a or AFDM between sites in Ñirehuao during the CI study, and the magnitude of difference in δ^{15}N between sites was large, so this small difference in temperature does not change our interpretation of these results. Another factor that could confound these comparisons is differences in grazing intensity. Grazers directly remove an average of 55% of total algal biomass (Hillebrand, 2009) but also indirectly increase algal biomass by increasing light penetration and nutrient availability to underlying algae (Andre et al., 2003; Power, 1990). Grazing intensity was not quantified at our study sites and could confound the comparisons of biofilm biomass in the CI study. However, because all paired study sites were of the same stream size (i.e. order), and because longitudinal changes in grazer abundance occur at the level of stream order (Vannote

et al., 1980), we assume that differences in grazing intensity were negligible.

Many studies have reported that the principal effect of spawning salmon on biofilm is a positive, nutrient subsidy effect (e.g. Chaloner et al., 2007; Mitchell & Lamberti, 2005; Wipfli et al., 1998), whereas many others have reported a negative, disturbance effect (e.g. Holtgrieve & Schindler, 2011; Moore & Schindler, 2008; Rüegg et al., 2020). Whether a given salmon population has a net positive or negative effect on stream biofilm biomass is probably mediated by substrate size, spawner density, and background nutrient levels (Holtgrieve et al., 2010; Janetski et al., 2009, 2014). Such influence of environmental conditions on responses to spawning salmon has been observed in the catchments of the Laurentian Great Lakes, where Chinook salmon have similarly established non-native populations. There, some streams are nutrient-limited and have increased algal biomass in reaches downstream of Chinook spawning relative to upstream (Schuldt & Hershey, 1995), whereas other streams have negligible subsidy effects and strong disturbance effects in salmon-impacted reaches, probably due to high background nutrient levels and small substrate sizes (Janetski et al., 2014). Although we found a nutrient subsidy effect, our methods probably would not have captured a disturbance effect; we generally did not observe Chinook spawning in the immediate vicinity of the tiles that we used to measure biofilm, and if a tile had been turned over by a spawning salmon, it would not have been sampled for biofilm. As such, our study does not rule out disturbance effects of salmon in these streams. Disturbance of the streambed is likely to happen to some degree in these streams, and at the whole-stream scale, this would reduce the net benefit of salmon to algal biomass (e.g. Holtgrieve et al., 2010). However, because Chinook build redds in

specific microhabitats that comprise a small proportion of total stream habitat, and because their spawning densities seem generally low in Patagonia, we suspect that subsidy effects are stronger than disturbance effects in these streams.

In resource-limited ecosystems that receive a pulsed subsidy, organisms optimise resource use when resources become available (Sears et al., 2004). Because the streams in southern Chile are highly nutrient-limited, salmon subsidies that increase algal biomass are likely to have bottom-up effects such that some heterotrophic species also increase in biomass (Risse-Buhl et al., 2012; Rosemond et al., 2000; Wipfli et al., 1998). Increased algal biomass could benefit consumers such as biofilm-grazing invertebrates (e.g. Verspoor et al., 2011), stream fishes (e.g. Kohler et al., 2012), and insectivorous forest birds (e.g. Wagner & Reynolds, 2019). Benefits to heterotrophic species can extend beyond the autumn spawning season due to the variety of mechanisms that retain salmon-derived nutrients in stream and riparian food webs year-round (Gende et al., 2002; O'Keefe & Edwards, 2002). Indeed, 6 months after salmon spawning in British Columbia, algal biomass is higher in salmon-impacted stream reaches (Harding et al., 2014). The subsidisation of algae can represent a positive feedback link between adult salmon and their progeny such that nutrients from adult salmon carcasses increase in situ primary productivity, causing increased productivity among the invertebrate prey of juvenile salmon in the spring and, ultimately, resulting in enhanced recruitment of juvenile salmon (Benjamin et al., 2020; Kaylor et al., 2020). As such, the subsidy effect described here could help sustain Chinook salmon populations in Patagonia.

The functional novelty described here should increase the degree of ecological change in Patagonia (Schittko et al., 2020). This change will probably involve some native species benefitting from the novel availability of marine-derived nutrients. It will probably also involve benefits to non-native species. Rainbow trout (*O. mykiss*) and American mink (*Neovison vison*) are invasive in Patagonia and co-evolved with and strongly benefit from Pacific salmon subsidies in their native ranges (Ben-David et al., 1997; Scheuerell et al., 2007). Both of these invaders have negative effects on the abundance and distribution of their native prey in Patagonian streams and forests (e.g. Correa & Hendry, 2012; Habit et al., 2010; Schüttler et al., 2009; Valenzuela et al., 2013), and salmon subsidies could strengthen these negative effects through apparent competition (Sears et al., 2004). The native aquatic fauna in Patagonia is characterised by low species richness but high levels of endemism (Cussac et al., 2016; Valdovinos, 2006), making the increasing abundance of non-native species a threat to local biodiversity (Habit & Cussac, 2016). At the same time, recreational fisheries in Patagonia are a key contributor to the regional tourism industry, and Chinook have added value to these fisheries. Nevertheless, it is clear that non-native salmonids are contributing to ecological change in Patagonia. Conservation planning that uses hydrologic isolation and barriers to salmonid migration to identify priority water bodies for protection could be an effective way of ensuring the persistence of salmonid-free ecosystems and the many unique species that inhabit them.

CONFLICTS OF INTEREST

The authors have no conflicts of interest to declare.

ORCID

Nicolas J. Muñoz https://orcid.org/0000-0002-8360-180X
Brian Reid https://orcid.org/0000-0002-4274-3350
Cristian Correa https://orcid.org/0000-0002-8608-6858

REFERENCES

Alò, D., Correa, C., Samaniego, H., Krabbenhoft, C. A., & Turner, T. F. (2019). Otolith microchemistry and diadromy in Patagonian river fishes. *PeerJ, 7*, e6149. https://doi.org/10.7717/peerj.6149

Andre, E. R., Hecky, R. E., & Duthie, H. C. (2003). Nitrogen and phosphorus regeneration by cichlids in the littoral zone of Lake Malawi, Africa. *Journal of Great Lakes Research, 1330*(03), 190–201. https://doi.org/10.1016/S0380-1330(03)70548-6

Araya, M., Niklitschek, E. J., Secor, D. H., & Piccoli, P. M. (2014). Partial migration in introduced wild Chinook salmon (*Oncorhynchus tshawytscha*) of southern Chile. *Estuarine, Coastal and Shelf Science, 149*, 87–95. https://doi.org/10.1016/j.ecss.2014.07.011

Arismendi, I., & Soto, D. (2012). Are salmon-derived nutrients being incorporated in food webs of invaded streams? Evidence from southern Chile. *Knowledge and Management of Aquatic Ecosystems, 405*, 01. https://doi.org/10.1051/kmae/2012009

Armstrong, J. B., Schindler, D. E., Omori, K. L., Ruff, C. P., & Quinn, T. P. (2010). Thermal heterogeneity mediates the effects of pulsed subsidies across a landscape. *Ecology, 91*(5), 1445–1454. https://doi.org/10.1890/09-0790.1

Atlas, W. I., Palen, W. J., Courcelles, D. M., Munshaw, R. G., & Monteith, Z. L. (2013). Dependence of stream predators on terrestrial prey fluxes: Food web responses to subsidized predation. *Ecosphere, 4*(6), 1–16. https://doi.org/10.1890/ES12-00366.1

Bailey, C. J., & Moore, J. W. (2020). Resource pulses increase the diversity of successful competitors in a multi-species stream fish assemblage. *Ecosphere, 11*(9), e03211. https://doi.org/10.1002/ecs2.3211

Ben-David, M., Hanley, T. A., Klein, D. R., & Schell, D. M. (1997). Seasonal changes in diets of coastal and riverine mink: The role of spawning Pacific salmon. *Canadian Journal of Zoology, 75*(5), 803–811. https://doi.org/10.1139/z97-102

Benjamin, J. R., Bellmore, J. R., & Watson, G. A. (2016). Response of ecosystem metabolism to low densities of spawning Chinook salmon. *Freshwater Science, 35*(3), 810–825. https://doi.org/10.1086/686686

Benjamin, J. R., Bellmore, J. R., Whitney, E., & Dunham, J. B. (2020). Can nutrient additions facilitate recovery of Pacific salmon? *Canadian*

Journal of Fisheries and Aquatic Sciences, 77(10), 1601–1611. https://doi.org/10.1139/cjfas-2019-0438

Bentley, K. T., Schindler, D. E., Armstrong, J. B., Zhang, R., Ruff, C. P., & Lisi, P. J. (2012). Foraging and growth responses of stream-dwelling fishes to inter-annual variation in a pulsed resource subsidy. *Ecosphere*, 3(12), art113. https://doi.org/10.1890/es12-00231.1

Chaloner, D. T., Lamberti, G. A., Cak, A. D., Blair, N. L., & Edwards, R. T. (2007). Inter-annual variation in responses of water chemistry and epilithon to Pacific salmon spawners in an Alaskan stream. *Freshwater Biology*, 52(3), 478–490. https://doi.org/10.1111/j.1365-2427.2006.01715.x

Chaloner, D. T., & Wipfli, M. S. (2002). Influence of decomposing Pacific salmon carcasses on macroinvertebrate growth and standing stock in southeastern Alaska streams. *Journal of the North American Benthological Society*, 21(3), 430–442. https://doi.org/10.2307/1468480

Christie, K. S., & Reimchen, T. E. (2008). Presence of salmon increases passerine density on Pacific Northwest streams. *The Auk*, 125(1), 51–59. https://doi.org/10.1525/auk.2008.125.1.51

Ciancio, J. E., Pascual, M. A., Botto, F., Frere, E., & Iribarne, O. (2008). Trophic relationships of exotic anadromous salmonids in the southern Patagonian Shelf as inferred from stable isotopes. *Limnology and Oceanography*, 53(2), 788–798. https://doi.org/10.4319/lo.2008.53.2.0788

Correa, C., & Gross, M. R. (2008). Chinook salmon invade southern South America. *Biological Invasions*, 10(5), 615–639. https://doi.org/10.1007/s10530-007-9157-2

Correa, C., & Hendry, A. P. (2012). Invasive salmonids and lake order interact in the decline of puye grande *Galaxias platei* in western Patagonia lakes. *Ecological Applications*, 22(3), 828–842. https://doi.org/10.1890/11-1174.1

Correa, C., & Moran, P. (2017). Polyphyletic ancestry of expanding Patagonian Chinook salmon populations. *Scientific Reports*, 7(1), 14338. https://doi.org/10.1038/s41598-017-14465-y

Cussac, V. E., Habit, E., Ciancio, J., Battini, M. A., Riva Rossi, C., Barriga, J. P., … Crichigno, S. (2016). Freshwater fishes of Patagonia: Conservation and fisheries. *Journal of Fish Biology*, 89(1), 1068–1097. https://doi.org/10.1111/jfb.13008

Dentener, F., Drevet, J., Lamarque, J. F., Bey, I., Eickhout, B., Fiore, A. M., … Wild, O. (2006). Nitrogen and sulfur deposition on regional and global scales: A multimodel evaluation. *Global Biogeochemical Cycles*, 20(4), GB4003. https://doi.org/10.1029/2005GB002672

Di Prinzio, C. Y., & Pascual, M. A. (2008). The establishment of exotic Chinook salmon (*Oncorhynchus tshawytscha*) in Pacific rivers of Chubut, Patagonia, Argentina. *Annales De Limnologie - International Journal of Limnology*, 44(1), 25–32. https://doi.org/10.1051/limn:2008020

Diaz, M., Pedrozo, F., Reynolds, C., & Temporetti, P. (2007). Chemical composition and the nitrogen-regulated trophic state of Patagonian lakes. *Limnologica*, 37(1), 17–27. https://doi.org/10.1016/J.LIMNO.2006.08.006

Doughty, C. E., Roman, J., Faurby, S., Wolf, A., Haque, A., Bakker, E. S., … Svenning, J.-C. (2016). Global nutrient transport in a world of giants. *Proceedings of the National Academy of Sciences of the United States of America*, 113(4), 868–873. https://doi.org/10.1073/pnas.1502549112

Downes, B. J., Barmuta, L. A., Fairweather, P. G., Faith, D. P., Keough, J., Lake, P. S., & Quinn, G. P. (2002). *Monitoring ecological impacts: Concepts and practice in flowing waters*. Cambridge University Press.

Elser, J. J., Bracken, M. E. S., Cleland, E. E., Gruner, D. S., Harpole, W. S., Hillebrand, H., … Smith, J. E. (2007). Global analysis of nitrogen and phosphorus limitation of primary producers in freshwater, marine and terrestrial ecosystems. *Ecology Letters*, 10(12), 1135–1142. https://doi.org/10.1111/j.1461-0248.2007.01113.x

Field, R. D., & Reynolds, J. D. (2013). Ecological links between salmon, large carnivore predation, and scavenging birds. *Journal of Avian Biology*, 44(1), 009–016. https://doi.org/10.1111/j.1600-048X.2012.05601.x

Flecker, A. S., McIntyre, P. B., Moore, J. W., Anderson, J. T., Taylor, B. W., & Hall, R. O. (2010). Migratory fishes as material and process subsidies in riverine ecosystems. *American Fisheries Society Symposium*, 73, 559–592.

Gende, S. M., Edwards, R. T., Willson, M. F., & Wipfli, M. S. (2002). Pacific salmon in aquatic and terrestrial ecosystems. *BioScience*, 52(10), 917–928.

Gomez-Uchida, D., Cañas-Rojas, D., Riva-Rossi, C. M., Ciancio, J. E., Pascual, M. A., Ernst, B., & Seeb, L. W. (2018). Genetic signals of artificial and natural dispersal linked to colonization of South America by non-native Chinook salmon (*Oncorhynchus tshawytscha*). *Ecology and Evolution*, 8, 6192–6209. https://doi.org/10.1002/ece3.4036

Gottesfeld, A. S., Hassan, M. A., & Tunnicliffe, J. F. (2008). Salmon bioturbation and stream process. *American Fisheries Society Symposium*, 65, 175–193.

Graham, N. A. J., Wilson, S. K., Carr, P., Hoey, A. S., Jennings, S., & MacNeil, M. A. (2018). Seabirds enhance coral reef productivity and functioning in the absence of invasive rats. *Nature*, 559, 250–253. https://doi.org/10.1038/s41586-018-0202-3

Gresh, T., Lichatowich, J., & Schoonmaker, P. (2000). An estimation of historic and current levels of salmon production in the Northeast Pacific ecosystem: Evidence of a nutrient deficit in the freshwater systems of the Pacific Northwest. *Fisheries*, 25(1), 15–21. https://doi.org/10.1577/1548-8446(2000)025<0015:AEOHAC>2.0.CO;2

Habit, E. & Cussac, V. (2016). Conservation of the freshwater fauna of Patagonia: An alert to the urgent need for integrative management and sustainable development. *Journal of Fish Biology*, 89(1), 369–370. https://doi.org/10.1111/jfb.12882

Habit, E., Piedra, P., Ruzzante, D. E., Walde, S. J., Belk, M. C., Cussac, V. E., … Colin, N. (2010). Changes in the distribution of native fishes in response to introduced species and other anthropogenic effects. *Global Ecology and Biogeography*, 19, 697–710. https://doi.org/10.1111/j.1466-8238.2010.00541.x

Harding, J. N., Harding, J. M. S., & Reynolds, J. D. (2014). Movers and shakers: Nutrient subsidies and benthic disturbance predict biofilm biomass and stable isotope signatures in coastal streams. *Freshwater Biology*, 59(7), 1361–1377. https://doi.org/10.1111/fwb.12351

Hermann, N. T., Chaloner, D. T., Gerig, B. S., & Lamberti, G. A. (2020). Ecological consequences of Great Lakes salmon subsidies for stream-resident brook and brown trout. *Canadian Journal of Fisheries and Aquatic Sciences*, 77(11), 1758–1771. https://doi.org/10.1139/cjfas-2020-0086

Hillebrand, H. (2009). Meta-analysis of grazer control of periphyton biomass across aquatic ecosystems. *Journal of Phycology*, 45(4), 798–806. https://doi.org/10.1111/j.1529-8817.2009.00702.x

Hocking, M. D., & Reimchen, T. E. (2002). Salmon-derived nitrogen in terrestrial invertebrates from coniferous forests of the Pacific Northwest. *BMC Ecology*, 2, 4. https://doi.org/10.1186/1472-6785-2-4

Hocking, M. D., & Reynolds, J. D. (2011). Impacts of salmon on riparian plant diversity. *Science*, 331(6024), 1609–1612. https://doi.org/10.1126/science.1201079

Holtgrieve, G. W., & Schindler, D. E. (2011). Marine-derived nutrients, bioturbation, and ecosystem metabolism: Reconsidering the role of salmon in streams. *Ecology*, 92(2), 373–385. https://doi.org/10.1890/09-1694.1

Holtgrieve, G. W., Schindler, D. E., Gowell, C. P., Ruff, C. P., & Lisi, P. J. (2010). Stream geomorphology regulates the effects on periphyton of ecosystem engineering and nutrient enrichment by Pacific salmon. *Freshwater Biology*, 55(12), 2598–2611. https://doi.org/10.1111/j.1365-2427.2010.02489.x

Hood, E., Fellman, J. B., Edwards, R. T., D'Amore, D. V., & Scott, D. (2019). Salmon-derived nutrient and organic matter fluxes from a coastal catchment in southeast Alaska. *Freshwater Biology*, 64(6), 1157–1168. https://doi.org/10.1111/fwb.13292

Janetski, D. J., Chaloner, D. T., Moerke, A. H., Levi, P. S., & Lamberti, G. A. (2014). Novel environmental conditions alter subsidy and engineering

effects by introduced Pacific salmon. *Canadian Journal of Fisheries and Aquatic Sciences*, 71(4), 502–513. https://doi.org/10.1139/cjfas-2013-0292

Janetski, D. J., Chaloner, D. T., Tiegs, S. D., & Lamberti, G. A. (2009). Pacific salmon effects on stream ecosystems: A quantitative synthesis. *Oecologia*, 159(3), 583–595. https://doi.org/10.1007/s00442-008-1249-x

Kaylor, M. J., White, S. M., Sedell, E. R., & Warren, D. R. (2020). Carcass additions increase juvenile salmonid growth, condition, and size in an interior Columbia River Basin tributary. *Canadian Journal of Fisheries and Aquatic Sciences*, 77(4), 703–715. https://doi.org/10.1139/cjfas-2019-0215

Kohler, A. E., Pearsons, T. N., Zendt, J. S., Mesa, M. G., Johnson, C. L., & Connolly, P. J. (2012). Nutrient enrichment with salmon carcass analogs in the Columbia river basin, USA: A stream food web analysis. *Transactions of the American Fisheries Society*, 141(3), 802–824. https://doi.org/10.1080/00028487.2012.676380

Kondolf, G. M., & Wolman, M. G. (1993). The sizes of salmonid spawning gravels. *Water Resources Research*, 29(7), 2275–2285. https://doi.org/10.1029/93WR00402

Lamberti, G. A., & Resh, V. (1985). Comparability of introduced tiles and natural substrates for sampling lotic bacteria, algae and macro invertebrates. *Freshwater Biology*, 15(1), 21–30. https://doi.org/10.1111/j.1365-2427.1985.tb00693.x

Lessard, J. A. L., & Merritt, R. W. (2006). Influence of marine-derived nutrients from spawning salmon on aquatic insect communities in southeast Alaskan streams. *Oikos*, 113(2), 334–343. https://doi.org/10.1111/j.2006.0030-1299.14512.x

Mitchell, N. L., & Lamberti, G. A. (2005). Responses in dissolved nutrients and epilithon abundance to spawning salmon in southeast Alaska streams. *Limnology and Oceanography*, 50(1), 217–227. https://doi.org/10.4319/lo.2005.50.1.0217

Moore, J. W., & Schindler, D. E. (2008). Biotic disturbance and benthic community dynamics in salmon-bearing streams. *Journal of Animal Ecology*, 77(2), 275–284. https://doi.org/10.1111/j.1365-2656.2007.01336.x

Morris, M. R., & Stanford, J. A. (2011). Floodplain succession and soil nitrogen accumulation on a salmon river in southwestern Kamchatka. *Ecological Monographs*, 81(1), 43–61. https://doi.org/10.1890/08-2296.1

Naiman, R. J., Bilby, R. E., Schindler, D. E., & Helfield, J. M. (2002). Pacific salmon, nutrients, and the dynamics of freshwater and riparian ecosystems. *Ecosystems*, 5(4), 399–417. https://doi.org/10.1007/s10021-001-0083-3

Narum, S. R., Gallardo, P., Correa, C., Matala, A., Hasselman, D., Sutherland, B. J. G., & Bernatchez, L. (2017). Genomic patterns of diversity and divergence of two introduced salmonid species in Patagonia, South America. *Evolutionary Applications*, 10(4), 402–416. https://doi.org/10.1111/eva.12464

O'Keefe, T. C., & Edwards, R. T. (2002). Evidence for hyporheic transfer and removal of marine-derived nutrients in a sockeye stream in southwest Alaska. *American Fisheries Society Symposium*, 33, 99–107.

Perakis, S. S., & Hedin, L. O. (2002). Nitrogen loss from unpolluted South American forests mainly via dissolved organic compounds. *Nature*, 415(6870), 416–419. https://doi.org/10.1038/415416a

Peterson, B. J., & Fry, B. (1987). Stable isotopes in ecosystem studies. *Annual Review of Ecology and Systematics*, 18(1), 293–320. https://doi.org/10.1146/annurev.es.18.110187.001453

Polis, G. A., Anderson, W. B., & Holt, R. D. (1997). Toward an integration of landscape and food web ecology: The dynamics of spatially subsidized food webs. *Annual Review of Ecology and Systematics*, 28(1), 289–316. https://doi.org/10.1146/annurev.ecolsys.28.1.289

Power, M. E. (1990). Resource enhancement by indirect effects of grazers: Armored catfish, algae, and sediment. *Ecology*, 71(3), 897–904. https://doi.org/10.2307/1937361

Quinn, T. P. (2005). *The behavior and ecology of Pacific salmon and trout*. American Fisheries Society.

Quinn, T. P., Helfield, J. M., Austin, C. S., Hovel, R. A., & Bunn, A. G. (2018). A multidecade experiment shows that fertilization by salmon carcasses enhanced tree growth in the riparian zone. *Ecology*, 99(11), 2433–2441. https://doi.org/10.1002/ecy.2453

R Core Team (2020). *R: A language and environment for statistical computing*. R Foundation for Statistical Computing.

Rand, P. S., Hall, C. A. S., McDowell, W. H., Ringler, N. H., & Kennen, J. G. (1992). Factors limiting primary productivity in Lake Ontario tributaries receiving salmon migrations. *Canadian Journal of Fisheries and Aquatic Sciences*, 49(11), 2377–2385. https://doi.org/10.1139/f92-262

Reimchen, T. E. (2017). Diverse ecological pathways of salmon nutrients through an intact marine–terrestrial interface. *Canadian Field-Naturalist*, 131(4), 350. https://doi.org/10.22621/cfn.v131i4.1965

Rex, J. F., & Petticrew, E. L. (2008). Delivery of marine-derived nutrients to streambeds by Pacific salmon. *Nature Geoscience*, 1(12), 840–843. https://doi.org/10.1038/ngeo364

Ricciardi, A., & Atkinson, S. K. (2004). Distinctiveness magnifies the impact of biological invaders in aquatic ecosystems. *Ecology Letters*, 7(9), 781–784. https://doi.org/10.1111/j.1461-0248.2004.00642.x

Ricciardi, A., Hoopes, M. F., Marchetti, M. P., & Lockwood, J. L. (2013). Progress toward understanding the ecological impacts of nonnative species. *Ecological Monographs*, 83(3), 263–282. https://doi.org/10.1890/13-0183.1

Richey, J. E., Perkins, M. A., & Goldman, C. R. (1975). Effects of kokanee salmon (*Oncorhynchus nerka*) decomposition on the ecology of a subalpine stream. *Journal of the Fisheries Research Board of Canada*, 32(6), 817–820. https://doi.org/10.1139/f75-109

Risse-Buhl, U., Trefzger, N., Seifert, A. G., Schönborn, W., Gleixner, G., & Küsel, K. (2012). Tracking the autochthonous carbon transfer in stream biofilm food webs. *FEMS Microbiology Ecology*, 79(1), 118–131. https://doi.org/10.1111/j.1574-6941.2011.01202.x

Rosemond, A. D., Mulholland, P. J., & Brawley, S. H. (2000). Seasonally shifting limitation of stream periphyton: Response of algal populations and assemblage biomass and productivity to variation in light, nutrients, and herbivores. *Canadian Journal of Fisheries and Aquatic Sciences*, 57(1), 66–75. https://doi.org/10.1139/f99-181

Rüegg, J., Chaloner, D. T., Ballantyne, F., Levi, P. S., Song, C., Tank, J. L., … Lamberti, G. A. (2020). Understanding the relative roles of salmon spawner enrichment and disturbance: A high-frequency, multi-habitat field and modeling approach. *Frontiers in Ecology and Evolution*, 8, 19. https://doi.org/10.3389/fevo.2020.00019

Scheuerell, M. D., Moore, J. W., Schindler, D. E., & Harvey, C. J. (2007). Varying effects of anadromous sockeye salmon on the trophic ecology of two species of resident salmonids in southwest Alaska. *Freshwater Biology*, 52(10), 1944–1956. https://doi.org/10.1111/j.1365-2427.2007.01823.x

Schittko, C., Bernard-Verdier, M., Heger, T., Buchholz, S., Kowarik, I., Lippe, M., … Jeschke, J. M. (2020). A multidimensional framework for measuring biotic novelty: How novel is a community? *Global Change Biology*, 26(8), 4401–4417. https://doi.org/10.1111/gcb.15140

Schuldt, J. A., & Hershey, A. E. (1995). Effect of salmon carcass decomposition on Lake Superior tributary streams. *Journal of the North American Benthological Society*, 14(2), 259–268. https://doi.org/10.2307/1467778

Schüttler, E., Klenke, R., McGehee, S., Rozzi, R., & Jax, K. (2009). Vulnerability of ground-nesting waterbirds to predation by invasive American mink in the Cape Horn Biosphere Reserve, Chile. *Biological Conservation*, 142(7), 1450–1460. https://doi.org/10.1016/j.biocon.2009.02.013

Sears, A. L., Holt, R. D., & Polis, G. A. (2004). Feast and famine in food webs: the effects of pulsed productivity. Chapter 23. In: G. A. Polis,

M. E. Power, & G. R. Huxel (Eds.), *Food webs at the landscape level* (pp. 359–386). University of Chicago Press.

Soto, D., Arismendi, I., Di Prinzio, C., & Jara, F. (2007). Establishment of Chinook salmon (*Oncorhynchus tshawytscha*) in Pacific basins of southern South America and its potential ecosystem implications. *Revista Chilena De Historia Natural*, *80*(1), 81–98. https://doi.org/10.4067/S0716-078X2007000100007

Steinman, A. D., Leavitt, P. R., & Uzarski, D. G. (2017). Biomass and pigments of benthic algae. Chapter 12. In: F. R. Hauer, & G. A. Lamberti (Eds.), *Methods in stream ecology* (3rd ed., pp. 223–241). Academic Press.

Swain, N. R., Hocking, M. D., Harding, J. N., & Reynolds, J. D. (2014). Effects of salmon on the diet and condition of stream-resident sculpins. *Canadian Journal of Fisheries and Aquatic Sciences*, *71*(4), 521–532. https://doi.org/10.1139/cjfas-2013-0159

Valdovinos, C. (2006). Invertebrados dulceacuícolas. In CONAMA (ed.), *Biodiversidad de Chile* (pp. 202–223). Ocho Libros.

Valenzuela, A. E. J., Raya Rey, A., Fasola, L., Sáenz Samaniego, R. A., & Schiavini, A. (2013). Trophic ecology of a top predator colonizing the southern extreme of South America: Feeding habits of invasive American mink (*Neovison vison*) in Tierra del Fuego. *Mammalian Biology*, *78*(2), 104–110. https://doi.org/10.1016/j.mambio.2012.11.007

Vannote, R. L., Minshall, G. W., Cummins, K. W., Sedell, J. R., & Cushing, C. E. (1980). The river continuum concept. *Canadian Journal of Fisheries and Aquatic Sciences*, *37*(1), 130–137. https://doi.org/10.1139/f80-017

Verspoor, J. J., Braun, D. C., Stubbs, M. M., & Reynolds, J. D. (2011). Persistent ecological effects of a salmon-derived nutrient pulse on stream invertebrate communities. *Ecosphere*, *2*(2), art18. https://doi.org/10.1890/ES10-00011.1

Vitousek, P. M. (1990). Biological invasions and ecosystem processes: Towards an integration of population biology and ecosystem studies. *Oikos*, *57*(1), 7. https://doi.org/10.2307/3565731

Wagner, M. A., & Reynolds, J. D. (2019). Salmon increase forest bird abundance and diversity. *PLoS One*, *14*(2), e0210031. https://doi.org/10.1371/journal.pone.0210031

Walsh, J. C., Pendray, J. E., Godwin, S. C., Artelle, K. A., Kindsvater, H. K., Field, R. D., ... Reynolds, J. D. (2020). Relationships between Pacific salmon and aquatic and terrestrial ecosystems: Implications for ecosystem-based management. *Ecology*, *101*(9), e03060. https://doi.org/10.1002/ecy.3060

Wipfli, M. S., Hudson, J., & Caouette, J. (1998). Influence of salmon carcasses on stream productivity: Response of biofilm and benthic macroinvertebrates in southeastern Alaska, U.S.A. *Canadian Journal of Fisheries and Aquatic Sciences*, *55*(6), 1503–1511. https://doi.org/10.1139/f98-031

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.