Phytoplankton, not allochthonous carbon, sustains herbivorous zooplankton production

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Terrestrial organic matter inputs have long been thought to play an important role in aquatic food web dynamics. Results from recent whole lake ^{13}C addition experiments suggest terrestrial particulate organic carbon (t-POC) inputs account for a disproportionate portion of zooplankton production. For example, several studies concluded that although t-POC only represented ≈20% of the flux of particulate carbon available to herbivorous zooplankton, this food source accounted for ≈50% of the C incorporated by zooplankton. We tested the direct dietary impact of t-POC (from the leaves of riparian vegetation) and various phytoplankton on *Daphnia magna* somatic growth, reproduction, growth efficiency, and lipid composition. By itself, t-POC was a very poor quality resource compared to cryptophytes, diatoms, and chlorophytes, but t-POC had similar food quality compared to cyanobacteria. Small additions of high quality *Cryptomonas ozolinii* to t-POC-dominated diets greatly increased *Daphnia* growth and reproduction. When offered alone, t-POC resulted in a *Daphnia* growth efficiency of 5 ± 1%, whereas 100% *Cryptomonas* and *Scenedesmus obliquus* diets resulted in growth efficiencies of 46 ± 8% (± SD) and 36 ± 3%, respectively. When offered in a 50:50 mixed diet with *Cryptomonas* or *Scenedesmus*, the t-POC fraction resulted in a partial growth efficiency of 22 ± 9% and 15 ± 6%, respectively. *Daphnia* that obtained 80% of their available food from t-POC assimilated 84% of their fatty acids from the phytoplankton component of their diet. Overall, our results suggest *Daphnia* selectively allocate phytoplankton-derived POC and lipids to enhance somatic growth and reproduction, while t-POC makes a minor contribution to zooplankton production.

Daphnia | fatty acids | nutritional ecology | planktonic food web

It has long been recognized that terrestrial carbon inputs dominate the carbon flux of many lakes, particularly small nutrient poor lakes with heavily vegetated watersheds (1, 2). The classic perspective (3) is that the flux of carbon from terrestrial sources to lakes can be quite substantial, but that this flux is predominantly as terrestrial dissolved organic carbon (t-DOC), which dominates the overall DOC pool within many lakes. However, because t-DOC is the residual carbon that was not metabolized by bacteria within watershed soils, this DOC source is mostly recalcitrant and is therefore used much less efficiently by lake bacterioplankton than is DOC produced by phytoplankton (3). Within lakes, t-DOC is converted to the particulate phase, and thereby made available to zooplankton, by bacteria uptake and production. This large but slowly metabolized pool of t-DOC may dampen the ecosystem fluctuations induced by the highly variable primary production dynamics in most lakes (3). More recently, Pace, Carpenter, Cole, and colleagues (4–6), presented a fundamentally different model of allochthonous carbon contributions to aquatic ecosystem production. Based on mass balance calculations for several whole lake $NaH^{13}CO_3$ addition experiments, these authors inferred t-POC was the predominant terrestrial contributor to herbivorous zooplankton production and of the terrestrial carbon incorporated by zooplankton <4% originated from t-DOC (6). Furthermore, although t-POC only represented ≈20 ± 10% of the flux of particulate carbon available to herbivorous zooplankton this food source accounted for 48 ± 22% of the C incorporated by *Daphnia* (6). In another striking example, these authors calculated t-POC loading to a different lake was equivalent to 2% of net primary production by phytoplankton, whereas 31% of cladoceran zooplankton production was supported by allochthony (7, 8). These conclusions represent a dramatic reassessment of the relative contributions and pathways by which terrestrial carbon is used in the pelagic food webs of lakes. This and related research (9–11) has contributed to the increasingly widespread view that terrestrial carbon inputs are the main contributor to overall upper trophic level production (i.e., zooplankton and fish) in many lakes (12).

While considerable research seems to suggest zooplankton obtain a large proportion of their carbon from terrestrial sources, for physiological reasons, the terrestrial carbon of higher plant origin is an incongruous source for sustaining zooplankton and ultimately fish production. A great deal has been learned about the biochemical basis for zooplankton and especially fish production during the last two decades, and it is now well-established that the nutritional physiology of both is strongly dependent on the dietary highly unsaturated fatty acids eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (13–17). The ω-3 series of polyunsaturated fatty acids (PUFA) include EPA and DHA, as well as the shorter carbon chain molecules α-linolenic acid (α-LA) and stearidonic acid (SDA) (17). Diatom and cryptophyte phytoplankton readily synthesize EPA and DHA (17), and cryptophytes also synthesize large amounts of α-LA and SDA (17). Green algae and cyanobacteria synthesize very little EPA and DHA, but most green algae and some cyanobacteria synthesize appreciable amounts of α-LA and some SDA (17, 18). Higher plants can synthesize α-LA, but lack the enzymes necessary to elongate and desaturate this molecule to EPA and DHA (19). Similar to cyanobacteria, higher plants often have a very high proportion of saturated fatty acids. Bacteria synthesize a wide variety of fatty acids, but usually produce very little ω-3 and ω-6 PUFA (20). Animals cannot synthesize ω-3 and ω-6 fatty acids de novo because they lack the enzymes necessary to synthesize α-LA and linoleic acid (18:2ω6) from the monounsaturated oleic acid (18:1ω9). However, animals can to varying degrees elongate and desaturate α-LA to form EPA and DHA, which are the physiologically active ω-3 molecules in animals (13, 14). Thus, although zooplankton and especially fish production is very EPA and DHA intensive, the terrestrial carbon that is available to support aquatic production is almost entirely devoid of these molecules regardless if this carbon is incorporated directly as t-POC or indirectly as t-DOC

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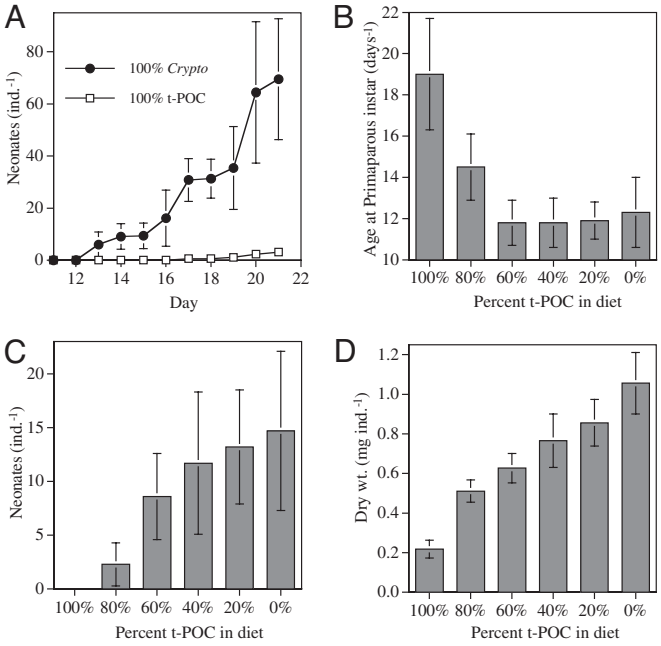

Fig. 1. The reproductive and size responses to the t-POC and *Cryptomonas* diets in the first two life table experiments. (*A*) Cumulative reproduction (mean ± 1 SD) for the first life table experiment. (*B*) Age at first reproduction for the second life table experiment (the age of reproduction for the 100% t-POC treatment was taken from the first experiment). (*C*) Cumulative reproduction during 14 days for the second experiment. (*D*) Mean *Daphnia* dry weight at age 14 days from the second life table.

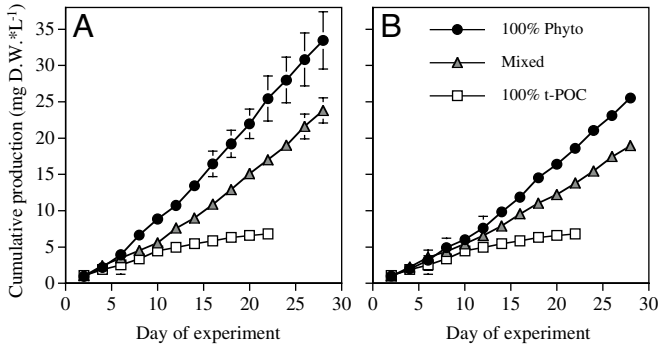

Fig. 2. Cumulative production (mean ± 1 SD) for the batch experiment. *A* represents the results for the *Cryptomonas* treatments and *B* represents the *Scenedesmus* treatments. The 100% t-POC treatment (shown in both panels for comparison) was terminated 1 week early so that there would be sufficient *Daphnia* biomass for fatty acid biomaker determinations. The outcome for the *Microcystis* treatment is not depicted in this Figure, but this treatment had a cumulative production of 4.7 ± 1.3 mg D.W. L^{-1}, which was 31% less than the 100% t-POC treatment.

via bacteria production. However, terrestrial vegetation can contribute precursors for these molecules to aquatic systems.

Because the relative importance of terrestrial carbon to upper trophic level production in pelagic food webs is a classic question in aquatic ecology, limnology and fisheries research (1–12), we conducted a series of experiments to directly test the contributions of allochthonous and autochthonous food sources to the somatic growth, reproduction, energetic efficiency, and lipid composition of herbivorous zooplankton. We conducted four experiments to test the hypotheses that 1) t-POC inputs of higher plant origin have a disproportionately high impact on zooplankton production as suggested by the results of Pace, Carpenter, and Cole (4–6), or alternatively 2) that this t-POC food source is a low food quality resource that only has a minor impact on zooplankton production as suggested by the biochemical reasoning outlined above.

Results

In the first life table experiment, *Daphnia* fed a 100% red alder t-POC diet first reproduced at an age of 19.4 ± 2.5 days and averaged 3.1 ± 2.7 (± SD) neonates $ind.^{-1}$ when this experiment was terminated on day 22. In contrast, *Daphnia* fed the *Cryptomonas* diet first reproduced at 13.7 ± 1.3 days and produced a cumulative total of 69.5 ± 23.2 neonates $ind.^{-1}$ during the entire experiment (Fig. 1). By comparison, *Daphnia* fed *Navicula* reproduced at 14.2 ± 2.8 days and had 30.0 ± 13.3 neonates $ind.^{-1}$, and *Daphnia* fed *Scenedesmus* reproduced at 15.2 ± 3.7 days and had 36.2 ± 34.6 neonates $ind.^{-1}$. Almost all *Daphnia* fed *Microcystis* in this experiment died without reproducing. In the t-POC gradient life table experiment, *Daphnia* fed 100 and 80% t-POC had significantly delayed reproduction, and individuals fed the 100, 80, and 60% t-POC diets had significantly reduced reproduction (Fig. 1). Each 20% *Cryptomonas* increment in the t-POC gradient experiment resulted in clearly larger *Daphnia* compared to the next lower level of *Cryptomonas* in the diet (Fig. 1), with a more than doubling in *Daphnia* size when only 20% *Cryptomonas* was included in the diet. Overall, 87% of the variability (= ANOVA model SS/total SS) in the size outcomes for this experiment could be explained by the six treatments (Fig. 1 and Fig. S1).

In the batch experiment, *Daphnia* fed a 100% t-POC diet had declining productivity as the experiment progressed and ended with an average productivity that was equivalent to a growth efficiency (*Daphnia* productivity/food ration) of 5.4 ± 1.3% (Fig. 2). In fact, the total *Daphnia* biomass harvested from this treatment did not exceed the initial population size, so this growth efficiency value may be an over-estimate. *Daphnia* consuming the 100% *Cryptomonas* diet had rapidly increasing productivity initially and then stable productivity subsequently, which resulted in an average growth efficiency of 46 ± 8%. *Daphnia* consuming the mixed diet (i.e., 50:50 t-POC and *Cryptomonas*) averaged 72 ± 9% of the productivity of *Daphnia* consuming 100% *Cryptomonas* (Fig. 2). *Daphnia* consuming 100% *Scenedesmus* had an average growth efficiency of 33 ± 3%. If we assume *Daphnia* consuming the mixed diet had the same growth efficiency for the phytoplankton portion of their diet as did *Daphnia* consuming pure phytoplankton diets, these results suggest the t-POC fraction of the 50:50 diets resulted in partial growth efficiencies of 22 ± 9% and 15 ± 6%, respectively, for the mixed diets with *Cryptomonas* and *Scenedesmus*. These results suggest t-POC by itself was a very poor diet, but when mixed with high quality phytoplankton this food source contributed to *Daphnia* production.

We also carried out a validation experiment to test whether different allochthonous and autochthonous food sources would yield similar experimental outcomes to those described above. In this experiment, *Daphnia* fed mixed t-POC from leaves of the most common riparian trees in the Pacific northwest of North America (21) and the cyanobacterium *Anabaena*, produced considerably fewer neonates and were much smaller than *Daphnia* that consumed *Rhodomonas*, *Fragilaria*, or *Chlamydomonas* (Fig. 3). In contrast to the first two life table experiments, the mixed t-POC used in this experiment was allowed to accumulate within the feeding vials during the experiment which allowed bacteria more time to colonize and modify this material.

Because the fatty acid composition of daphnids is strongly influenced by diet (22, 23), we used a fatty acid trophic marker approach (24) to determine which lipids were incorporated when consuming mixed diets. The fatty acid composition data for the

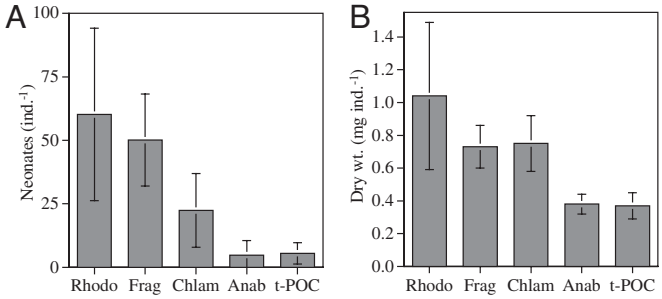

Fig. 3. The reproductive and size responses to t-POC and phytoplankton diets in a validation life table experiment. This experiment was designed to test whether t-POC is generally much lower food quality that phytoplankton, or was just lower food quality than the phytoplankton we used for our first two experiments. For this experiment, we selected representatives of cryptophytes, diatoms, green algae, and cyanobacteria which were from different genera than those used in previous experiments; that is, *Rhodomonas lacustris* (Rhod), *Fragilaria crotonensis* (Frag), *Chlamydomonas reinhardti* (Chlam), and *Anabaena flos-aquae* (Anab). We also used a new t-POC source that was derived from equal dry weights of red alder, black cottonwood, big leaf maple, and willow, which were subsequently milled and sieved together. In this experiment *Daphnia magna* fed *Rhodomonas*, *Fragilaria*, and *Chlamydomonas* were significantly larger and more fecund ($P < 0.05$ for individual *t* tests after Bonferroni correction) than *Daphnia* fed mixed t-POC, however, *Daphnia* fed *Anabaena* had similar outcomes compared to mixed t-POC. The fecundity results accounted for mortality differences, whereas the size differences were only for surviving individuals.

Daphnia from the second life table experiment show the *Cryptomonas* component of the mixed diets had an inordinate impact on *Daphnia* fatty acids even when t-POC strongly dominated the available food (Fig. 4). Simple mixing calculations indicate that when *Daphnia* obtained 80% of their available food from t-POC they assimilated 84% of their fatty acids from phytoplankton. *Daphnia* that consumed any amount of *Cryptomonas* had quite high proportions of SDA and EPA relative to total fatty acids, and much higher ω3:ω6 fatty acid ratios. *Daphnia* that consumed 100% t-POC had high proportions of the saturated fatty acid 16:0, the monounsaturated fatty acids 16:1ω7 and 18:1ω9, as well as the ω6 PUFA 18:2ω6 (Fig. 4). *Daphnia* that consumed the 100% t-POC diet also had a substantially lower proportion of saturated fatty acids and a higher proportion monounsaturated fatty acids than their diet. These *Daphnia* also had 78% and 40% higher proportions ω6 and ω3 PUFA, respectively. These results are consistent with previous reports that *Daphnia* generally have

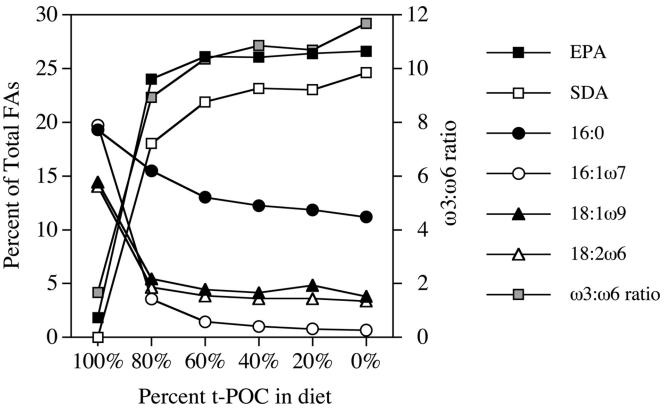

Fig. 4. *Daphnia magna* fatty acid composition for the second life table experiment. The dominant fatty acids (EPA, SDA, 16:0, 16:1ω7, 18:1ω9, and 18:2ω6) are expressed on the left-hand *y* axis and the ω3:ω6 fatty acid ratio is expressed on the right-hand *y* axis.

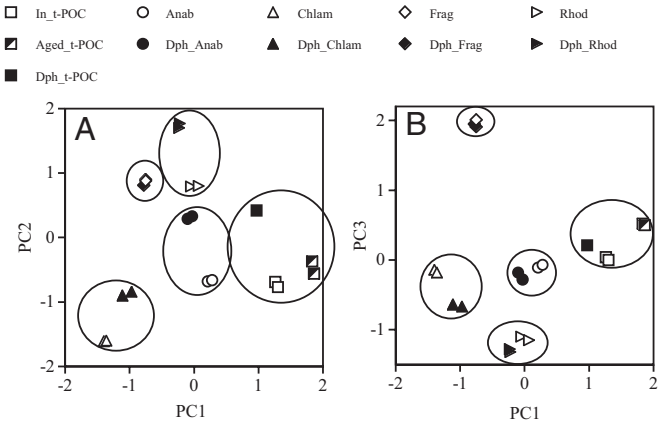

Fig. 5. A principal component analysis (PCA) of fatty acid composition for the diet and *Daphnia* samples from the third life table experiment (see Table S1). The first principal component (PC), see *x* axis of *A* and *B*, explained 30.7% of the overall variability and was positively correlated with the SAFA stearic acid (18:0) as well as the sum of long chain (i.e., C_{20}, C_{22}, and C_{24}) SAFAs ($r = 0.83$ and 0.88, respectively), and negatively correlated with C_{16} PUFAs ($r = -0.93$). The second PC, see *y* axis of *A*, explained 27.6% of the variability and was positively correlated with EPA ($r = 0.93$) and negatively correlated with C_{18} ω6 PUFAs ($r = -0.84$). The third PC, see *y* axis of *B*, explained an additional 20.3% of variability and was positively correlated with C_{16} MUFAs ($r = 0.94$) and negatively correlated with C_{18} ω3 PUFAs ($r = -0.94$). In the legend for this figure, In_t-POC represents the initial mixed t-POC, aged_t-POC represents the mixed t-POC that accumulated within the feeding vials during this experiment, Anab represents *Anabaena*, Chlam represents *Chlamydomonas*, Frag represents *Fragilaria*, Rhod represents *Rhodomonas*, and Dph represents *Daphnia*. For example, Rhod represents the FA composition of *Rhodomonas* and Dph_Rhod represents the FA composition of *Daphnia* fed *Rhodomonas*. All values are based on duplicate samples, except for the *Daphnia* consuming aged mixed t-POC and *Fragilaria* diet samples where one replicate was lost due to contamination.

less saturated, and more mono- and polyunsaturated fatty acids than their diets (22, 23). The fatty acid α-LA, which was prevalent in both t-POC and *Cryptomonas*, comprised ≈ 20% of *Daphnia* fatty acids in all treatments. The saturated fatty acids 18:0, 20:0, 22:0 and 24:0, which are mostly derived from cuticular wax in higher plants (25), were prevalent in t-POC but were not found even in those *Daphnia* that consumed 100% t-POC. A principal component analysis of the diet and *Daphnia* samples from the third life table experiment showed t-POC had a very distinctive FA composition and *Daphnia* FAs were very strongly influenced by their diet (Fig. 5 and Table S1). This analysis also showed the *Daphnia* usually had more EPA and/or less C_{18} ω6 PUFAs than their diets.

Discussion

Our results suggest t-POC of higher plant origin is a very poor quality food resource which is, however, sufficiently nutritionally complete to allow *Daphnia* to produce small numbers of viable offspring. The outcome of our experiments may have been as much due to the recalcitrant (i.e., lignin and cellulose rich) nature of the t-POC as to the high food quality of most of the phytoplankton tested. Even when consuming a diet strongly dominated by t-POC, *Daphnia* acquired and selectively retained the majority of their physiologically important fatty acids from the phytoplankton component of their diets. However, our batch growth experiments show the efficiency with which t-POC is used by herbivorous zooplankton is also influenced by the simultaneous availability of more nutritious phytoplankton. When offered as a component of a mixed diet, t-POC did support *Daphnia* production more than its very low food quality (when offered as a single food source) would suggest. The fact that *Daphnia* fed a mixed diet realized a clear benefit of the t-POC

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component fraction of their diet, but obtained a very small proportion of their fatty acids from t-POC, suggests *Daphnia* may catabolize the low quality part of their diet and use the high quality part for new production when consuming mixed diets. Such an inference would fundamentally change our understanding of the contributions allochthonous and autochthonous food sources make to the upper trophic levels of pelagic food webs. These results also suggest terrestrial carbon incorporation into aquatic food webs may be regulated by the availability and preferential utilization of high food quality phytoplankton such as EFA rich cryptophytes and diatoms. This is particularly relevant to oligotrophic forest lakes where phytoplankton may comprise only a small proportion of the sestonic particulate carbon available to herbivorous zooplankton (26).

Although the aquatic ecosystem allochthonous literature usually only differentiates between terrestrial carbon and "phytoplankton," our results suggest further differentiation is warranted regarding autochthonous food resources. Our results, in fact, show t-POC is similar or perhaps higher food quality than cyanobacteria, but much lower food quality than the representatives of cryptophytes, green algae, and diatoms that we tested in our experiments. While this conclusion is a refinement for the allochthonous C literature, it is consistent with the well-established large food quality differences between the major phytoplankton groups (17, 22).

While our results are quite different from those of Pace and colleagues (4–6) regarding allochthonous contributions to *Daphnia* production, they are similar to recent inferences regarding the relative roles of allochthonous and autochthonous contributions to bacteria metabolism in lakes. Kritzberg et al. (27) concluded bacteria preferentially use phytoplankton derived DOC and convert this carbon to bacteria biomass at a greater efficiency than they do allochthonous DOC. These authors further argued that even in lakes where the flux of DOC is strongly dominated by terrestrial carbon, bacterial production was closely coupled to phytoplankton production due to its higher nutritional value. Karlsson (28) showed allochthonous contributions to bacterial respiration were considerably higher than to zooplankton production, and concluded allochthonous carbon may primarily contribute to catabolic metabolism, whereas autochthonous carbon plays a much more important relative role in the anabolic metabolism of lakes.

In light of our results, we suggest an alternative explanation for the results reported by Pace, Carpenter, and Cole (4–6). It should be noted that these authors' mass balance calculations indicate the *Daphnia* $\delta^{13}C$ values observed were due to a mixed diet of strongly labeled phytoplankton from the epilimnia of their experimental lakes and some other unlabeled food source. Pace and colleagues inferred this unlabeled food source was t-POC, but did not account for the fact that it quite possibly could have been unlabeled phytoplankton from the metalimnia of their lakes. Our inference is supported by the fact that we found t-POC is a very poor quality food source and it therefore should not have supported a high proportion of *Daphnia* productivity as suggested by Pace and colleagues. It is also noteworthy that it has previously been shown that *Daphnia* in several of the lakes sampled by these authors vertically migrate and spend the daylight period of the day in the metalimnion (29) where high food quality phytoplankton are prevalent (30). Furthermore, data from several studies of stratified lake systems show metalimnic phytoplankton have depleted $\delta^{13}C$ values (31, 32). Therefore, studies that use ^{13}C analyses to infer allochthonous contributions to zooplankton production may produce misleading results when zooplankton obtain a substantial fraction of their carbon from metalimnetic primary production particularly if the epilimnion has been labeled with $H^{13}CO_3^-$.

Finally, it is necessary to consider whether the t-POC loading rates assumed for the lakes studied by Pace and colleagues (4–6) are reasonable when compared to t-POC loading actually measured in other lakes. A recent review summarized t-POC loading data for eight lakes (8), and showed that ≈40–45% of total t-POC loading comes as small sized particles (i.e., < 153 μm in diameter). If this is taken into account, total loading of small sized t-POC to these lakes can be estimated to range between 1 and 19, and average 5 mg C m^{-2} d^{-1}. In contrast, Pace and colleagues assumed much higher t-POC loading rates for their calculations, for example, 50–100, 71 ± 30, and 107 ± 72 mg C m^{-2} d^{-1} (4–6). Thus, the t-POC inputs on which Pace et al. (4–6) based their model calculations were considerably higher than the t-POC loading rates that have on average been observed in lakes. If this likely over-estimate of t-POC inputs is taken into account, it suggests very small relative t-POC loading rates (e.g., ≈1–2% of phytoplankton production) support ≈50% of zooplankton production. This result, and the previously mentioned case of 2% relative t-POC loading supporting 31% of cladoceran zooplankton production (7, 8), is energetically implausible. However, t-POC availability in lakes may be somewhat higher than suggested by the exogenous loading rates summarized by Preston et al. (8), if as suggested by von Wachenfeldt and Tranvik (33), t-DOC is readily converted to t-POC in high DOC lakes via flocculation.

While our study was designed to directly test Pace and colleagues (4–7) assertion that small amounts of t-POC loading to lakes play an inordinate role in zooplankton production, our findings also suggest a reassessment of several of the most highly cited papers on this topic (9–11) is warranted. The fundamental limitation of the studies by Jones, Gray, Karlsson, and colleagues (9–11) is that they all used indirect means to estimate the $\delta^{13}C$ values of the phytoplankton fraction of the sestonic particulate matter for their stable isotope mixing model calculations (9–11). For example, Jones et al. (9) estimated the $\delta^{13}C$ values for phytoplankton within the seston of the 12 lakes they studied by separating the phytoplankter *Gonyostomum semem* from the seston of two lakes and assuming this very large phytoplankter had $\delta^{13}C$ values which were representative of the algae zooplankton actually consumed in all of their lakes. Gray et al. (10) estimated the $\delta^{13}C$ values of the phytoplankton consumed by zooplankton in Loch Ness from large diatoms, which they separated from the detritus in their samples by sedimentation. These authors also concluded that in Loch Ness, *Daphnia* production was "almost completely reliant on algal production" despite the fact that "allochthonous carbon is vastly more available than algal carbon at all times of the year." Karlsson et al. (11) estimated the $\delta^{13}C$ values of the phytoplankton in their lakes by assuming photosynthetic fractionation factors which were much larger than a recent literature review on this topic (34) indicates; that is, $\varepsilon_p = -20$ to -30‰ for Karlsson et al. (11) versus 0–15‰ for Bade et al. (34). If lower photosynthetic fractionation factors are assumed for the Karlsson et al. dataset, allochthonous support of zooplankton production is not indicated.

Our experimental approach differs from past studies looking at allochthonous contributions to zooplankton production in that we directly determined the impact of this resource type on zooplankton growth, reproduction, energetic efficiency, and lipid composition in a controlled laboratory setting. In contrast, other studies have indirectly assessed allochthonous contributions to zooplankton production using stable isotope analyses and various debatable modeling inferences (4–12). Stable isotope based field studies of this topic are currently constrained by the fact that is it very difficult determine the actual $\delta^{13}C$ values of the phytoplankton component of the seston. Our laboratory experiments alleviated this particular problem, but they lack the inherent complexity of the "real-world." Ultimately, our laboratory-based inferences regarding the very low quality of t-POC will need to be validated in appropriately designed field studies. Our results indicate t-POC of higher plant origin will likely make

a much smaller relative contribution to zooplankton and fish production than will autochthonous production by phytoplankton rich in essential fatty acids. The results of this study also suggest the lower-quality t-POC component of the diet may be catabolized for metabolic demands for energy, whereas the phytoplankton component is selectively used for production of new somatic material. Furthermore, our results suggest the availability of high food quality phytoplankton regulates the incorporation of low-quality terrestrial carbon into pelagic food web production. Because zooplankton and fish production is essential fatty acid intensive (13–17), upper trophic level production in pelagic food webs will be primarily supported by phytoplankton, benthic algae, or terrestrial animal prey.

Methods

We used the herbivorous zooplankter *Daphnia magna* as the consumer. t-POC of a size suitable for *Daphnia* ingestion (35) was generated by milling and sieving (Fig. S2) fall senesced leaves from red alder (*Alnus rubra*), as well as an equal mixture (i.e., mixed t-POC) of leaves from alder, black cottonwood (*Populus trichocarpa*), big leaf maple (*Acer macrophyllum*), and willow (*Salix* spp.), which are the most prevalent deciduous trees in the riparian zones of lakes and rivers in the Pacific northwest region of North America (21). The cryptophytes *Cryptomonas ozolinii* and *Rhodomonas lacustris*, the diatoms *Navicula pellicosa* and *Fragilaria crotonensis*, the chlorophytes *Scenedesmus obliquus* and *Chlamydomonas reinhardtii*, and non-toxic strains of the cyanobacteria *Microcystis aeruginosa* and *Anabaena flos-aquae* were used as autochthonous food sources. These allochthonous and autochthonous food sources differ greatly in their essential fatty acid composition (Table S1).

In the first experiment, *Daphnia* were fed ad libitum diets comprised of 100% red alder t-POC, *Cryptomonas*, *Navicula*, *Scenedesmus*, or *Microcystis* in a life table design. In the second experiment, *Daphnia* were fed a gradient of red alder t-POC and *Cryptomonas* diets varying by 20% increments; that is, 100% t-POC; 80:20 t-POC; and *Cryptomonas*; etc., using a life table design. In the third experiment, *Daphnia* were maintained in batch culture, fed consistent rations and harvested at a rate of 10% day^{-1} for 28 days (Fig. S3). *Daphnia* in this experiment were fed either 100% red alder t-POC, *Cryptomonas*, *Scenedesmus*, and *Microcystis*, or 50:50 mixtures of red alder t-POC and the three phytoplankters. In a fourth validation experiment, we repeated the first life table using mixed t-POC, *Rhodomonas*, *Fragilaria*, *Chlamydomonas*, and *Anabaena* as food sources. We also used the *Daphnia* collected from the second and last life table experiments to determine how their fatty acid composition was influenced by the availability of t-POC and phytoplankton in their diets.

A detailed description of our methods is presented in *SI Text*.

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