Feeding dynamics of the invasive calanoid copepod *Pseudodiaptomus inopinus* in two northeast Pacific estuaries

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**ABSTRACT:** The Asian calanoid copepod *Pseudodiaptomus inopinus*, first observed in the Columbia River Estuary in the early 1990s, has since become the dominant copepod species in many estuaries along the US Pacific coast, but its feeding behavior has not been previously studied. In October 2019 and 2020, when *P. inopinus* was at peak seasonal abundance, we conducted incubation experiments with this species feeding on natural microplankton prey assemblages sampled from 2 invaded estuaries: the Chehalis River estuary, Washington, and the Yaquina River estuary, Oregon. In both estuaries, diatoms were the most numerically abundant prey group, with 11–15 μm *Chaetoceros* sp. and 21–25 μm *Cyclotella* sp. dominating the Chehalis and Yaquina estuaries, respectively. Diatom and ciliate biomass were highest in both estuaries, with all prey cells in the Yaquina estuary typically larger than those in the Chehalis estuary. *P. inopinus* fed omnivorously on microplankton prey, with a preference for prey >20 μm and occasional avoidance of cyanobacteria and cells <10 μm. Ingestion rates were highest on ciliates and diatoms. The omnivory of *P. inopinus* may contribute to its success as an invader in northeast Pacific estuaries.

**KEY WORDS:** Copepod · Feeding ecology · Trophic ecology · Estuaries · Invasive species

1. **INTRODUCTION**

Aquatic invasive species can have large-scale ecological impacts on plankton community composition and food web dynamics in aquatic ecosystems (Bollens et al. 2002, Strayer 2010, Havel et al. 2015, Dexter & Bollens 2020, Dexter et al. 2020a). For example, the spread of zebra and quagga mussels throughout North America has resulted in major community shifts and structural alterations of freshwater environments (Vanderploeg et al. 2002, Cuhel & Aguilar 2013). Ecological impacts such as these are likely to worsen, as some models predict a substantial increase in the distributional range and/or abundance of aquatic invasive invertebrates in response to climate change (Bellard et al. 2013, Dexter et al. 2020b).

Estuaries are often the locations where non-indigenous aquatic taxa are initially introduced, due largely to the global transport of plankton via ballast water (Carlton & Geller 1993, Ruiz et al. 2000) and because in these areas they can reach high enough abundances or otherwise disrupt native communities so as to become invasive. This is especially true for estuaries of the northeast Pacific Ocean, which have experienced numerous aquatic invasions in recent decades (Bollens et al. 2002, Cordell et al. 2008, Dexter et al. 2015). Notably, these estuaries have been invaded by at least 9 species of planktonic copepods native to Asia, most of which first appeared in the San Francisco Estuary (SFE) in California, USA (Bollens et al. 2002, Cordell et al. 2010), as well as the bosminid cladoceran *Bosmina coregoni* (Smits et al. 2013).
2. MATERIALS AND METHODS

2.1. Study sites

Experiments were conducted using copepods and the ambient prey assemblage from 2 estuaries — the Chehalis River estuary and the Yaquina River estuary — each located within 200 km north and south, respectively, along the Pacific coast from the CRE,
where *Pseudodiaptomus inopinus* was first introduced (Fig. 1). These estuaries were selected because both had high abundances of *P. inopinus* and each was within a 2 h drive from our laboratory, which minimized the time between collection of live specimens and the start of the feeding incubation experiments. The Chehalis River estuary is located 100 km north of the mouth of the Columbia River in southwest Washington, USA (Fig. 1). The river runs 200 km from its headwaters in the Willapa Hills to its outlet in Greys Harbor, with discharge being lowest in August and highest in January (Gendaszek 2011). Our sample site on the Chehalis River was near Cosmopolis (46° 57' 27.8'' N, 123° 46' 16.9'' W), approximately 15 river km from the outlet to Greys Harbor, where the tidal range is 5 m and salinity ranges tidally from 0 to 12.

The Yaquina River estuary is located 170 km south of the mouth of the Columbia River along the central coast of Oregon, USA (Fig. 1). The river runs 95 km from its headwaters in the Oregon Coast Range to the Pacific Ocean, with discharge annually averaging 6.9 m$^3$ s$^{-1}$ (Sigleo & Frick 2007). Our sampling site in the Yaquina River was near Toledo (44° 36' 13.8'' N, 123° 54’ 8.8’’ W), approximately 30 river km upstream from the Pacific Ocean, where the tidal range is 1.9 m (Brown & Ozretich 2009) and the salinity ranges tidally from 0 to 15.

2.2. Field sampling

Water and copepods for incubation experiments were collected at one site from each estuary during October 2019 and again in October 2020. At each site in each estuary, sampling occurred within 1 h of high slack tide at a location near the center of the channel, where both the surface and bottom salinities were between 2–5 as measured with a YSI portable salinity–temperature meter. Water depth, surface and bottom temperature, and surface and bottom salinity were recorded immediately before and after plankton sampling. Oblique net tows from near bottom to the surface were conducted from a small (4 m) boat using a 0.5 m diameter, 75 μm mesh plankton net; net contents were immediately rinsed from the net and transferred to a clean bucket. Oblique tows were used to ensure that *P. inopinus* were collected regardless of their vertical position in the water column, which can fluctuate on a diel basis (Bollens et al. 2002). Surface water for the laboratory incubation experiments was then gently filtered into carboys through a 300 μm mesh sieve to remove large grazers. In addition, three 200 ml samples of unfiltered surface water were collected and preserved in 5% Lugol’s solution for later microscopical analysis. All samples were transported to the lab within 6 h of collection and stored in conditions consistent with those recorded during sampling (12 h light:12 h dark cycles and temperature of 16°C).

2.3. Incubation experiments

Following the methods of Rollwagen-Bollens et al. (2013) and Bowen et al. (2015), 40 non-ovigerous adult female *P. inopinus* for each experiment were hand-picked from the plankton net tow samples and transferred into each of four 500 ml incubation bottles containing filtered surface water from the collection site. Additionally, 4 replicate 500 ml bottles were filled with filtered surface water to serve as initial controls and another 4 replicate bottles were filled with filtered surface water as final controls. All replicate bottles were covered with parafilm and sealed to prevent bubbles. All treatment and final control bottles were then incubated for 12 h, overnight, on a ro-

![Fig. 1. Study sites (Chehalis and Yaquina estuaries) where *Pseudodiaptomus inopinus*, microplanktonic prey, and environmental conditions were sampled in October 2019 and 2020, as well as the Columbia River Estuary, the site of several related historical studies](image)
tating (0.5–1 rpm) plankton wheel in the dark and kept at ambient temperature to mimic field conditions.

After incubation, copepods were removed from the treatment bottles by filtering the incubation water over a 300 μm mesh sieve and then preserved in 5% buffered formalin solution in 20 ml glass vials. Initial control bottles were sub-sampled immediately prior to the start of the incubations, and final control and treatment bottles were sub-sampled at the end of the incubations as follows: 200 ml sub-samples were taken from each bottle then preserved in 5% Lugol’s solution for microscopical analysis.

2.4. Microplankton sample processing and analysis

Taxonomic composition, abundance, biomass, and cell size of microplankton prey in each incubation bottle were assessed by settling aliquots of 24.5 ml from each sample bottle overnight in Utermöhl chambers and then observing the contents using an inverted microscope (Leica DMI4000B) at 400× magnification. For each sample (aliquot), cells up to 200 μm in their longest dimension were counted and sized in microscope fields along transects of the settling chamber until at least 300 cells >10 μm were observed (Kirchman 1993). Every cell counted was further identified to the lowest practical taxonomic level using Patterson & Hedley (1992) and Wehr et al. (2015). The cells were then grouped into 1 of 6 major prey taxonomic categories: diatoms, dinoflagellates, flagellates, ciliates, chlorophytes, and cyanobacteria, as well as 4 major size categories based on their longest dimension: 1–10, 11–20, 21–30, and >30 μm. Carbon biomass was calculated for each individual using biovolume calculated from geometric shape (Hillebrand et al. 1999) and estimated from the algorithms of Menden-Deuer & Lessard (2000). Any one of our experiments was considered to have resulted in a feeding effect if there was a significant (p < 0.05) reduction in abundance and/or biomass of any individual prey group between final control and treatment bottles as measured using Student’s t-tests (Rollwagen-Bollens et al. 2013, Bowen et al. 2015).

Copepod clearance rates (ml copepod−1 h−1) and ingestion rates (μg C copepod−1 h−1) were calculated from the changes in abundance (cells ml−1) or biomass (μg C ml−1) of each prey taxonomic category and each size category over the course of the incubation, following the approach of Marin et al. (1986). Feeding selectivity and preference were then estimated using 2 approaches. First, significant differences in clearance rates among prey taxa and size categories were assessed within each experiment using Kruskal-Wallis ANOVA by ranks, since these data did not meet the assumptions of a parametric ANOVA. Second, an electivity index (E*; Vanderploeg & Scavia 1979) was calculated for each prey taxonomic and size category in each experiment, and each mean E* value was tested for being significantly different from zero using 1-sample t-tests. E* values range from +1 to −1; significantly positive values of E* were interpreted to indicate a preference for that particular prey category whereas significantly negative E* values were indicative of avoidance of that particular prey category.

Body mass of each *P. inopinus* specimen was also determined from both incubation experiments from each estuary using length–weight–carbon biomass conversions measured by Ara (2001). Daily weight-specific ingestion rates (μg C prey [μg C copepod]−1 d−1) and daily ration (% body carbon consumed d−1) were then calculated for each experiment to more accurately draw comparisons to feeding rates of other copepods taxa reported in the literature.

3. RESULTS

3.1. Microplankton prey assemblages

In October 2019 and 2020, microplankton abundance in the Chehalis estuary was primarily composed of diatoms (59 and 63%, respectively) and flagellates (31 and 29%, respectively) (Fig. 2A). However, while ciliates only comprised 8% of the microplankton abundance in 2019, these cells contributed 60% of microplankton biomass, with diatoms sub-dominant in terms of biomass (20%). In 2020, this pattern was reversed: diatoms were dominant with a relative biomass of 46% and ciliates sub-dominant, contributing 30% of relative biomass (Fig. 2B). In both years, chlorophytes, cyanobacteria, and dinoflagellates were particularly scarce, each comprising less than 5% of both total abundance and total biomass of microplankton. The dominant prey size category in the Chehalis estuary in 2019 and 2020 was 11–20 μm in the longest dimension, comprising 70.0 and 61.2% of available prey, respectively. This size category primarily contained diatoms, dinoflagellates, flagellates, and ciliates. Sub-dominant to this size category were cells 1–10 μm in their longest dimension (21.3% in 2019 and 20.3% in 2020), primarily comprising flagellates, chlorophytes, and cyanobacteria (Table 1).
In the Yaquina estuary, diatoms were the most abundant microplankton prey taxon in 2019 (77%) followed by flagellates (15%); in 2020, diatoms and flagellates were roughly equal in relative abundance (53 and 45%, respectively) (Fig. 2A). With respect to microplankton biomass, diatoms were consistently dominant in 2019 and 2020 (64 and 58%, respectively), although in 2019, ciliates, which only accounted for 6% of the total prey abundance, contributed 30% of the total prey biomass (Fig. 2B). In both years, chlorophytes, cyanobacteria, and dinoflagellates each comprised less than 5% of both total abundance and total biomass of microplankton. The dominant prey size category in the Yaquina estuary in 2019 and 2020 was that of cells 21–30 μm in their longest dimension, comprising 42.9 and 62.5% of available prey, respectively. This size category primarily contained diatoms, flagellates, and ciliates. Sub-dominant to this size category were cells 11–20 μm in their longest dimension (30.2% in 2019 and 31.0% in 2020), primarily containing dinoflagellates, flagellates, and ciliates (Table 1).

### 3.2. *P. inopinus* prey preferences

In the Chehalis estuary, *P. inopinus* clearance rates on prey taxa cate-

| Group          | Size (μm) | Genus sp.            | Chehalis | Yaquina |
|----------------|-----------|----------------------|----------|---------|
| Diatoms        | 1−10      | Unknown              | ·        | ·       |
|                | 11−20     | Melosira             | ·        | ·       |
|                |           | Skeletonema          | ·        | ·       |
|                |           | Tabellaria           | ·        | ·       |
|                |           | Chaetoceros          | ·        | ·       |
|                |           | Navicula             | ·        | ·       |
|                | 21−30     | Nitzschia            | ·        | ·       |
|                |           | Amphora              | ·        | ·       |
|                |           | Cocconeis            | ·        | ·       |
|                |           | Cyclotella           | ·        | ·       |
|                | 30+       | Asterionella         | ·        | ·       |
|                |           | Gomphonema           | ·        | ·       |
|                |           | Gyrosigma            | ·        | ·       |
|                |           | Synebra              | ·        | ·       |
|                |           | Unknown               | ·        | ·       |
| Dinoflagellates| 11−20     | Gymnodinium          | ·        | ·       |
|                |           | Peridinium           | ·        | ·       |
|                |           | Unknown thecate      | ·        | ·       |
|                | 21−30     | Gynodinium           | ·        | ·       |
|                |           | Peridinium           | ·        | ·       |
|                |           | Unknown thecate      | ·        | ·       |
| Flagellates    | 1−30      | Cryptomonad          | ·        | ·       |
|                |           | Unknown               | ·        | ·       |
| Ciliates       | 11−20     | Mesodinium rubrum   | ·        | ·       |
|                |           | Unknown loricate     | ·        | ·       |
|                | 21−30     | Mesodinium rubrum   | ·        | ·       |
|                |           | Unknown loricate     | ·        | ·       |
| Chlorophytes   | 1−10      | Coelastrum          | ·        | ·       |
|                |           | Crucigenia          | ·        | ·       |
|                |           | Tetrastrum          | ·        | ·       |
|                |           | Scenedesmus         | ·        | ·       |
|                |           | Spirulina           | ·        | ·       |
|                | 11−20     | Ankistrodesmus      | ·        | ·       |
|                |           | Staurastrum         | ·        | ·       |
| Cyanobacteria  | 1−10      | Anabaena            | ·        | ·       |
|                |           | Microcystis         | ·        | ·       |
|                |           | Unknown               | ·        | ·       |

Table 1. Microplankton taxa observed in the Chehalis and Yaquina estuaries during October 2019 and 2020. Asterisks indicate the presence of the listed taxon in the estuary.
Categories ranged from −0.25 to 1.23 ml copepod$^{-1}$ h$^{-1}$ in 2019 and −2.67 to 1.24 ml copepod$^{-1}$ h$^{-1}$ in 2020 (Fig. 3). Clearance rates on prey size categories ranged from −0.04 to 0.34 ml copepod$^{-1}$ h$^{-1}$ in 2019 and −0.17 to 1.96 ml copepod$^{-1}$ h$^{-1}$ in 2020 (Table 2). Statistical analyses indicated that there were no significant differences ($\chi^2_5 = 9.12$, $p > 0.05$) in $P. \text{inopinus}$ clearance rates among prey taxonomic categories in 2019. However, there was a significant difference ($\chi^2_5 = 11.014$, $p < 0.05$) in $P. \text{inopinus}$ clearance rates for prey taxa categories that share letters are not significantly different from each other ($p > 0.05$). $E^*$ values significantly different from zero ($p < 0.05$) are marked with an asterisk.

| Size (μm) | Clearance rate Chehalis 2019 | clearance rate Yaquina 2019 | $E^*$ Chehalis 2020 | $E^*$ Yaquina 2020 |
|----------|-----------------------------|-----------------------------|---------------------|---------------------|
| 1–10     | $-0.037 (±0.058)$            | $-0.064 (±0.086)$           | $1.094 (±0.629)$    | $-0.032 (±0.331)$   |
| 11–20    | $0.382 (±0.069)$             | $0.136 (±0.126)$            | $0.097 (±0.108)$    | $-0.501 (±0.148)$   |
| 21–30    | $0.340 (±0.174)$             | $0.064 (±0.107)$            | $0.027 (±0.118)$    | $-0.412 (±0.248)$   |
| 30+      | $0.192 (±0.616)$             | $-0.688 (±0.312)$           | $0.708 (±0.303)$    | $0.120 (±0.115)$    |
| 1–10     | $-0.169 (±0.302)$            | $-0.636 (±0.211)$           | $0.017 (±0.126)$    | $-1.0 (±0)$         |
| 11–20    | $0.704 (±0.351)$             | $-0.267 (±0.237)$           | $0.280 (±0.096)$    | $-0.515 (±0.182)$   |
| 21–30    | $1.959 (±0.362)$             | $0.306 (±0.403)$            | $1.403 (±0.109)$    | $0.248 (±0.018)$    |
| 30+      | $0.806 (±0.157)$             | $-0.147 (±0.293)$           | $4.168 (±1.952)$    | $0.321 (±0.028)$    |

Table 2. Mean (±SE) clearance rates and electivity ($E^*$) values of $P. \text{inopinus}$ on prey size categories in the Chehalis and Yaquina estuaries in October 2019 and 2020. $E^*$ values significantly different from zero shown in bold.
clearance rates among prey categories in 2020; a post hoc Dunn’s test of the 2020 data revealed that \textit{P. inopinus} cleared chlorophytes at a higher rate than cyanobacteria. With respect to prey size, there were no significant differences in clearance rates among prey size categories in 2019, but in 2020, \textit{P. inopinus} cleared prey in the 21–30 μm size category at a significantly higher rate than prey cells 1–10 μm in size (Table 3).

\( E^* \) values for prey taxa categories in the Chehalis estuary ranged from −0.37 to 0.25 in 2019 and from −0.77 to 0.14 in 2020 (Fig. 3). Among prey size categories in the Chehalis estuary experiments, \( E^* \) values ranged from −0.69 to 0.14 in 2019 and from −0.64 to 0.31 in 2020 (Table 2). \( E^* \) values calculated from the 2019 experiment were significantly positive (\( p < 0.05 \)) for cyanobacteria (\( t_6 = -10.28, p = 0.0005 \)) and ciliates (\( t_6 = -4.06, p = 0.007 \)) and negative for diatoms (\( t_6 = 5.25, p = 0.002 \)) (Fig. 3), and there were no \( E^* \) values significantly different from zero among prey size categories (Table 2). Conversely, in the 2020 Chehalis estuary experiment, \( E^* \) values were significantly negative for both cyanobacteria (\( t_6 = 3.35, p = 0.015 \)) and diatoms (\( t_6 = 3.26, p = 0.017 \)) (Fig. 3) and were significantly positive (\( t_6 = -7.12, p = 0.006 \)) for prey cells 21–30 μm in size (Table 2).

Among prey taxa categories in experiments from the Yaquina estuary, clearance rates ranged from −0.12 to 1.31 ml copepod\(^{-1} \) h\(^{-1} \) in 2019 and from 0.94 to 2.21 ml copepod\(^{-1} \) h\(^{-1} \) in 2020, and \( E^* \) values ranged from −0.66 to 0.34 in 2019 and from −0.47 to 0.20 in 2020 (Fig. 4). Among prey size categories, clearance rates ranged from 0.03 to 1.09 ml copepod\(^{-1} \) h\(^{-1} \) in 2019 and from 0.02 to 4.17 ml copepod\(^{-1} \) h\(^{-1} \) in 2020, and \( E^* \) values ranged from −0.57 to 0.20 in 2019 and from −1 to 0.32 in 2020 (Table 2). In the Yaquina estuary, no significant differences in \( P. inopinus \) clearance rates were observed among prey taxonomic or size categories in the 2019 experiment (Table 4); however, \( P. inopinus \) exhibited significantly positive \( E^* \) for chlorophytes (\( t_6 = -5.93, p = 0.001 \)) and significantly negative \( E^* \) for both flagellates (\( t_6 = 2.79, p = 0.032 \)) and prey cells 11–20 μm in size (\( t_6 = 3.84, p = 0.031 \)) in 2019. In 2020 in the Yaquina estuary, \( P. inopinus \) similarly did not exhibit significantly different clearance rates for any prey taxon but did clear prey >30 μm at a significantly higher rate than prey cells in the 1–10 and 11–20 μm size categories (Table 4). \( P. inopinus \) did not show significant \( E^* \) for any prey taxa but did show significantly positive \( E^* \) for prey cells 21–30 μm (\( t_6 = -14.02, p = 0.001 \)) and 30+ μm in size (\( t_6 = -11.42, p = 0.001 \)) in the 2020 Yaquina experiment (Table 2).

Table 3. Statistical comparison of \textit{Pseudodiaptomus inopinus} clearance rates on microplankton prey taxa and size categories in experiments conducted from the Chehalis River estuary in 2019 and 2020. Significant (\( p < 0.05 \)) values shown in bold.

| Chehalis 2019, taxa; Kruskal-Wallis | Chehalis 2020, taxa; Kruskal-Wallis |
|-----------------------------------|-----------------------------------|
| df  | \( \chi^2 \)  | \( p \)  | df  | \( \chi^2 \)  | \( p \)  |
| Prey 5 | 9.12 | 0.10 | Prey 5 | 11.01 | **0.05** |
| Cil | 0.02 | 0.66 | 1 | 1 | 1 |
| Cyano | 0.77 | 1 | 1 | 1 | 1 |
| Diat | 0.56 | 1 | 1 | 1 | 1 |
| Dino | 0.06 | 1 | 1 | 1 | 1 |
| Flag | 0.06 | 1 | 1 | 1 | 1 |

| Chehalis 2019, size; Kruskal-Wallis | Chehalis 2020, size; ANOVA |
|-----------------------------------|--------------------------|
| df  | \( \chi^2 \)  | \( p \)  | df  | SS  | MS  | \( F \)  | \( p \)  |
| Size 3 | 5.38 | 0.15 | Size 3 | 9.15 | 3.06 | 4.03 | **0.03** |
| 1–10 | 12 | 9.08 | 0.76 |
| 11–20 | 0.51 | **0.01** | 0.42 |
| 21–30 | 0.23 | 0.10 |
| 30+ | 0.29 |

### 3.3. \textit{P. inopinus} ingestion rates

In the Chehalis estuary, average total ingestion rates by \textit{P. inopinus} were 0.021 ± 0.007 and 0.015 ± 0.0008 μg C copepod\(^{-1} \) h\(^{-1} \) in 2019 and 2020, respectively (Fig. 5). There were significant differences in \textit{P. inopinus} ingestion rates among prey categories in both 2019 and 2020 (Fig. 5). In October 2019, ciliate biomass was ingested at the highest rate (0.031 μg C copepod\(^{-1} \) h\(^{-1} \)), and a post hoc Dunn’s test revealed that the biomass of this group was ingested at a significantly higher rate than the biomass of chlorophytes (\( z_3 = 3.03, p = 0.035 \)) and cyanobacteria (\( z_3 = -3.18, p = 0.022 \)) (Table 5). In October 2020 in the Chehalis estuary, ciliate, diatom, and flagellate
biomass was ingested at rates between 0.0031 and 0.0062 μg C copepod⁻¹ h⁻¹, and a post hoc Dunn’s test revealed that both ciliate \( z_3 = -3.40, p = 0.010 \) and diatom \( z_3 = 3.32, p = 0.013 \) biomass were ingested at significantly higher rates than that of cyanobacteria (Table 5).

In the Yaquina estuary, average total ingestion rates by *P. inopinus* were 0.007 ± 0.011 and 0.053 ± 0.001 μg C copepod⁻¹ h⁻¹ in 2019 and 2020, respectively (Fig. 6). Similar to the Chehalis estuary, significant differences in *P. inopinus* ingestion rates on different prey taxa were observed in both years (Fig. 6). In October 2019, diatom biomass was ingested at the highest rate (0.014 μg C copepod⁻¹ h⁻¹) and a post hoc Dunn’s test revealed that this taxon was ingested at a significantly higher rate than cyanobacteria biomass \( z_3 = 3.71, p = 0.003 \) (Table 6). Similarly, in October 2020 in the Yaquina estuary, diatom biomass was again ingested at the highest rate (0.038 μg C copepod⁻¹ h⁻¹), and a post hoc Dunn’s test showed that diatoms were ingested at a significantly higher rate than the biomass of chlorophytes \( z_3 = 3.29, p = 0.014 \); both diatom \( z_3 = 3.75, p = 0.003 \) and dinoflagellate \( z_3 = 2.94, p = 0.043 \) biomass was ingested at a significantly higher rate than cyanobacteria (Table 6).

The average individual biomass of copepods in the Chehalis estuary was higher than in the Yaquina estuary (3.36 and 2.86 μg C copepod⁻¹, respectively). Weight-specific ingestion rates of *P. inopinus* on the total microplankton community in the Chehalis estu-

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**Fig. 4.** Same as Fig. 3, but for the Yaquina estuary.
ary in 2019 and 2020 were 0.147 ± 0.619 and 0.104 ± 0.250 (μg C prey) (μg C copepod)−1 d−1 (respectively), representing a daily biomass ration of 14.8 ± 4.9% in 2019 and 10.3 ± 0.6% in 2020. In the Yaquina estuary, P. inopinus weight-specific ingestion rates in 2019 and 2020 were 0.055 ± 0.495 and 0.441 ± 1.160 (μg C prey) (μg C copepod)−1 d−1, respectively, which resulted in daily rations of 10.3 ± 6.2% in 2019 and 44.0 ± 0.9% in 2020.

4. DISCUSSION

*Pseudodiaptomus inopinus* has invaded and is now highly abundant in at least 11 northeast Pacific estuaries (Dexter et al. 2020c), including the Chehalis and Yaquina estuaries in Washington and Oregon states, respectively, yet this is the first investigation of *P. inopinus* feeding behavior in either its native or invasive range. We found that *P. inopinus* consumed prey omnivorously with a preference for ciliates, diatoms, and prey 21−30 μm. Its ingestion rate was highest on prey that comprised greater relative biomass rather than greater relative abundance. We also provide the first report of abundance and composition of microplankton in the Chehalis estuary, as well as a more comprehensive report of the microplankton assemblage in the Yaquina estuary.

4.1. Microplankton assemblage structure

During October 2019 and 2020, the microplankton prey assemblages in the Chehalis and Yaquina estuaries were dominated by diatoms and flagellates, specifically *Chaetoceros* sp. in the 11–15 μm (Chehalis) and *Cyclotella* sp. in the 21–25 μm (Yaquina) size ranges. In both estuaries, ciliates ranged in size from 10 to 25 μm and flagellates ranged from 6 to 25 μm. Our findings from the Yaquina estuary are consistent with previous studies of this estuary which reported that diatoms dominated in the spring–autumn and dinoflagellates and cyanobacteria increased in abundance in the summer (Karentz & McIntire 1977, Gilbert et al. 2010). Blooms of the non-toxic, red-tide-forming ciliate *Myrionecta rubra* also recur in the Yaquina estuary during spring in the vicinity of our sampling location (Brown & Nelson 2010). To our knowledge, our study is the first to report abundance and composition of the microplankton assemblage in the Chehalis estuary.

Moreover, the microplankton assemblage patterns observed in our study are consistent with many other temperate estuaries. *Cyclotella* and *Chaetoceros* dominated the upper eutrophic and middle transitional sections, respectively, of the Urdaibai estuary of northern Spain (Trigueros & Orive 2001, Revilla et al. 2002), and the autumn assemblage of the Guadiana estuary on the Iberian Peninsula was marked by high diatom abundance and biomass (Domingues & Galvão 2007). In the northeast Pacific, autumn microplankton assemblages in the SFE are dominated by *Chaetoceros* sp. and pennate diatoms (Rollwagen-Bollens & Penry 2003, Rollwagen-Bollens et al. 2006, Bouley & Kimmerer 2006); however, the CRE (located between the Chehalis and Yaquina estuaries along the US Pacific coast) is low in diatom and flagellate abundance in autumn but has occasional blooms of ciliates and consistently high abundance of cyanobacteria at this time of year (Bowen et al. 2015, Breckenridge et al. 2015, Rollwagen-Bollens et al. 2020).

4.2. *P. inopinus* prey selection

Despite the dominance of diatoms, flagellates, and ciliates in the Chehalis and Yaquina River estuaries, *P. inopinus* cleared most prey types at similar rates in both estuaries and both years while showing a slight preference for chlorophytes, ciliates, and cells >20 μm. These results generally align with the

| Prey | df | χ² | p   | Prey | df | χ² | p   |
|------|----|----|-----|------|----|----|-----|
| Yaquina 2019, taxa; Kruskal-Wallis | 5 | 7.36 | 0.20 | Yaquina 2020, taxa; Kruskal-Wallis | 5 | 3.20 | 0.67 |
| Yaquina 2019, size; Kruskal-Wallis | 3 | 4.02 | 0.26 | Yaquina 2020, size; Kruskal-Wallis | 3 | 13.26 | 0.004 |
| Size 1−10 | 0.48 | 0.13 | 0.005 | Size 11−20 | 0.48 | 0.047 |
| Size 21−30 | 0.48 | 0.005 |
| Size 30+ | 0.48 | 0.005 |

Table 4. Statistical comparison of *Pseudodiaptomus inopinus* clearance rates on microplankton prey taxa and size categories in experiments conducted from the Yaquina River estuary in 2019 and 2020. Significant (p < 0.05) values shown in bold
patterns observed in the native range of *Pseudodiaptomus* spp., such as was found by Chen et al. (2018) using a stable isotope approach in Guangyang Bay, Korea, where *P. marinus* and *P. koreanus* as well as other brackish copepod species were observed to feed omnivorously and across a broad size spectrum.

The pattern of prey preference for ciliates is not surprising, as ciliates commonly comprise large portions of calanoid copepod diets, particularly when phytoplankton biomass is low (Rollwagen-Bollens & Penry 2003, Calbet & Saiz 2005, Gifford et al. 2007), and ciliates may be more carbon-rich than diatoms of similar volumes (Menden-Deuer & Lessard 2000). Size selection by calanoid copepods was also observed in a tropical lagoon (Cote d’Ivoire), where Pagano et al. (2003) observed *P. hessei* to generally prefer particles up to 39 μm. In the Bay of Biscay, bordering Spain and France, *Temora longicornis* selected for prey >40 μm (Vincent & Hartmann 2001). In the SFE, *Acartia* spp. preferred prey >15 μm and often targeted prey >25 μm (Rollwagen-Bollens & Penry 2003). Kayfetz & Kim-

Table 5. Statistical comparison of *Pseudodiaptomus inopinus* ingestion rates on multiple categories of microplankton prey taxa in experiments conducted from the Chehalis River estuary. Significant (p < 0.05) values shown in **bold.** Abbreviations as in Table 3

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| Prey          | Chehalis 2019; Kruskal-Wallis df | χ² | p          | Chehalis 2020; Kruskal-Wallis df | χ² | **p**          |
|---------------|---------------------------------|----|------------|--------------------------------|----|----------------|
| Cil           | Chloro                           | 0.035 | 1 | 1 | 1 | Chloro          | 0.45 | 1 | 0.45 | 1 | 1 | **p = 0.003** |
| Cyan          | 0.022                            | 0.30 | 0.46       | Cil                            | **0.010** | 1 | 0.45 | 1 |               |
| Diat          | 1                                | 1    | 1          | Cyan                           | **0.013** | 1 | 0.45 |
| Dino          | 1                                | 1    | 1          | Diat                           | 0.45 | 1 |               |
| Flag          |                                  |      |            | Dino                           | 1    |               |
```
merer (2017) found that \textit{P. forbesi} showed low clearance rates on prey 7−15 μm, and \textit{Limnoithona tetraspina} showed preference for flagellates >15 μm in the SFE. Additionally, the brackish environment of estuaries may impact prey preference of \textit{P. inopinus}, as Galloway & Winder (2015) found that the long-chain essential fatty acid content of chlorophytes correlated positively with salinity, was variable but highest at intermediate salinities for diatoms, and was relatively low for cyanobacteria regardless of salinity. Thus, under brackish conditions, chlorophytes and diatoms could be more nutritious than cyanobacteria (Galloway & Winder 2015).

Although cyanobacteria were scarce in both of our estuaries in October of both years and are relatively low in nutritional value, \textit{P. inopinus} demonstrated a slight preference for this taxon in the Chehalis estuary in October 2019. Conversely, \textit{P. inopinus} showed a distinct avoidance of cyanobacteria in the Chehalis estuary in October 2020. This is of note, as previous studies have shown estuarine calanoid copepods are able to feed on cyanobacteria and overcome the nutritional deficiencies of this prey.

![Graph A](image1.png)

![Graph B](image2.png)

**Fig. 6.** Same as Fig. 5, but for the Yaquina estuary

| Yaquina 2019, Kruskal-Wallis | Yaquina 2020, Kruskal-Wallis |
|-----------------------------|-----------------------------|
| df | χ²  | p | df | χ²  | p |
| Prey | 5 | 12.51 | 0.028 | Prey | 5 | 20.695 | 0.0009 |
| Cil | Cyano | Diat | Dino | Flag | Cil | Cyano | Diat | Dino | Flag |
| Chloro | 1 | 1 | 0.18 | 1 | 1 | Chloro | 0.76 | 1 | 0.014 | 1 | 0.14 |
| Cil | 0.83 | 0.48 | 1 | 1 | Cil | 0.33 | 0.76 | 1 | 1 |
| Cyano | 0.003 | 0.67 | 0.57 | Cyano | 0.003 | 1 | 0.043 |
| Diat | 0.66 | 0.75 | Diat | 0.08 | 1 |
| Dino | 1 | Dino | 0.55 |

**Table 6.** Statistical comparison of \textit{Pseudodiaptomus inopinus} ingestion rates on multiple categories of microplankton prey taxa conducted from the Yaquina River estuary. Significant (p < 0.05) values shown in **bold**. Abbreviations as in Table 3
taxon to sustain growth. For instance, in the Baltic Sea, all field-collected zooplankton taxa (copepods, cladocerans, and rotifers) as well as laboratory-fed specimens of the copepod Acartia tonsa showed the presence of picocyanobacteria DNA in their gut contents, even when alternative food was plentiful, suggesting direct consumption of cyanobacteria (Motwani & Gorokhova 2013), P. hessei (Kâ et al. 2012) and P. forbesi (Bowen et al. 2015, Owens et al. 2019) have been shown to ingest cyanobacteria found in natural prey assemblages, and nauplii of P. marinus were reported to feed on the cyanobacterium Synechococcus sp. under cultured laboratory conditions (Vogt et al. 2013).

4.3. Patterns of P. inopinus prey consumption

In both estuaries, P. inopinus most often exhibited the highest ingestion rates on prey categories that were high in biomass rather than abundance. This may be due to the high abundance of large Cyclorella diatoms in the Yaquina estuary, which could have provided adequate carbon to support the copepods’ diet and lessened the need to ingest heterotrophic prey such as ciliates—which tend to have a slightly lower carbon:nitrogen ratio than autotrophic prey (Broglio et al. 2003), even though they can be a more efficient source of proteins and amino acids (Kierboe et al. 1985).

The omnivorous diet of P. inopinus is not surprising, as copepod omnivory is common, particularly in systems where phytoplankton biomass is low and nanophytoplankton abundance is high (Gifford & Dagg 1988, Rollwagen-Bollens & Penry 2003, Bouley & Kimmerer 2006). The ingestion of heterotrophic taxa (e.g. ciliates and some dinoflagellates) by copepods has been observed in other temperate estuaries and may have cascading impacts on phytoplankton. For instance, in the SFE, the invasive copepods L. tetraspina, P. forbesi, and Acartiella sinensis are well established and broadly distributed (Bollens et al. 2011, 2014), and in the lower salinity zone exhibited grazing impacts on ciliates, which at times released phytoplankton growth from grazing pressure by the microzooplankton (Kratina et al. 2014, York et al. 2014). P. inopinus may have a similar impact in its invaded range, particularly in the Chehalis estuary, where we observed this species to selectively consume ciliates. Seasonal monitoring of both P. inopinus and the microplankton assemblage of this estuary would provide further insight into the potential role of P. inopinus to exert top-down control of the microplankton assemblage. We also note that the significant consumption of ciliate biomass by P. inopinus in our incubations could have resulted in higher growth of small phytoplankton in those experiments through cascading effects (e.g. Nejstgaard et al. 2001, York et al. 2014) and might have artificially reduced the estimation of P. inopinus ingestion rates upon these smaller prey taxa. However, the ambient abundance of such small-sized phytoplankton groups was already quite low in both the Chehalis and Yaquina estuaries; therefore, we consider such cascading effects, if present, to be quite limited in our incubations.

More broadly, the potential trophic implications of omnivory are 2-fold: first, there is the possibility that P. inopinus could exert direct top-down control on both the phytoplankton and microzooplankton assemblages; and second, there are potential bottom-up impacts on higher level consumers, depending on whether P. inopinus is acting as a primary consumer (e.g. ingesting diatoms) or a secondary consumer (e.g. ingesting ciliates). In a laboratory setting with linear 3- and 4-trophic-level food chains, Malzahn et al. (2010) found that when autotrophic quality was low, copepods reared as secondary consumers grew faster than those reared as primary consumers. The authors concluded that if intermediary consumers are in high enough abundance they may, counterintuitively, increase higher trophic level production.

The weight-specific ingestion rates of P. inopinus measured in our study (6-44% body carbon consumed d−1) fell well within the ranges reported for other Pseudodiaptomus species and were especially comparable with rates measured for P. forbesi in its invasive range. Bowen et al. (2015) found total weight-specific ingestion rate of P. forbesi in the CRE to be 0.163 (μg C prey) (μg C copepod)−1 d−1, or 16.3% body carbon d−1, and in the SFE, Kayfetz & Kimmerer (2017) measured the average daily ration of this copepod as ranging from 5 to 22%, primarily on centric diatoms. Similarly, several studies have reported weight-specific ingestion rates for P. hessei that align with those for P. inopinus and P. forbesi, although with a much wider range of daily ration. Specifically, P. hessei in a northern African tropical lagoon (Cote d’Ivoire) exhibited daily rations between 13 and 100% based on incubations with the natural assemblage of microplankton (Pagano et al. 2003), and from 5 to 150% based on the gut fluorescence technique (Kouassi et al. 2001); and in a southern African estuary, Froneman (2004) measured daily biomass rations of P. hessei ranging from 4 to 64%.
Our measurements of *P. inopinus* ingestion rates in the Chehalis and Yaquina estuaries also align generally with rates reported for marine and estuarine copepods more broadly, although the weight-specific ingestion rates for the genus *Pseudodiaptomus* appear to be on the low end of the range based on reviews by Saiz & Calbet (2007, 2011) as well as 24 additional studies of calanoid and small cyclopoid copepod feeding rates published between 1992 and 2017 using a range of experimental approaches. For instance, daily biomass rations of mixed copepod assemblages estimated using the gut fluorescence technique (sensu Mackas & Bohrer 1976) have been reported to range from as low as 4 to >1000% (and in some cases, biologically impossible rates of >7000%) (e.g. Hansen & van Boekel 1991, Debes et al. 2008, Calliari & Tiselius 2009). When provided with cultured algae as prey in the laboratory, calanoid and small cyclopoid copepods have been observed to consume prey biomass from 0.5 to 250% of body carbon d−1 (e.g. Durbin & Durbin 1992, Koski et al. 1998, Dam & Lopes 2003, Garrido et al. 2013, Zamora-Terol & Saiz 2013, Saiz et al. 2014, van Someren Gréve et al. 2017). In feeding experiments using wild-caught copepods incubated with natural prey assemblages (such as in our study), daily rations have ranged from 1 to 150% (Pagano et al. 2003, Zamora-Terol et al. 2014, Bowen et al. 2015, Kayfetz & Kimmerer 2017). There are numerous biological explanations for such large differences in weight-specific ingestion rates by copepods, including past feeding history, availability of preferred prey taxa, temperature, season, latitude, etc. However, the variation in these rates also highlights the differences between experimental approaches used to assess feeding dynamics and the assumptions upon which they are based.

### 4.4. Co-occurrence and potential competition with congeners

*P. inopinus* is now the dominant zooplankton taxon in many northeast Pacific estuaries (Dexter et al. 2020c), yet this species has not been observed in the CRE (where it first arrived in the northeast Pacific) since 2002 (Cordell et al. 2008, Dexter et al. 2020c). Its congener species, *P. forbesi*, however, is now the second-most dominant zooplankton taxon along the northeast Pacific coast (Dexter et al. 2020c) and since 2002 has been present in very high abundance during August and September in the CRE (Cordell et al. 2008, Dexter et al. 2020c). In the CRE, *P. forbesi* is omnivorous, ingesting ciliates, algae, and cyanobacteria, and it consumes these prey taxa non-selectively (Bowen et al. 2015). Similarly, in the upper SFE, *P. forbesi* has been reported to ingest diatoms, ciliates, and flagellates at the highest rates (Kayfetz & Kimmerer 2017). Although *P. forbesi* is found to co-occur with other estuarine calanoid copepods in the CRE, such as *Eurytemora affinis* (Bollens et al. 2012, Bowen et al. 2015), it has not been observed to co-occur with *P. inopinus* (Bouley & Kimmerer 2006, Dexter et al. 2020c). *P. inopinus*, however, has been found to co-occur with another congener, *P. poplesia*, in the Mankyung River estuary, South Korea, where it is native (Park et al. 2013).

Both *P. inopinus* and *P. forbesi* are at peak abundance during the autumn wherever they are found. In the lower CRE, cyanobacteria become the most highly abundant taxon during this time, while other prey taxa decline in abundance (Bowen et al. 2015, Rollwagen-Bollens et al. 2020). Our current results from the Chehalis and Yaquina estuaries show that *P. inopinus* occasionally avoids cyanobacteria and shows preference for ciliates and diatoms—prey taxa that are not highly abundant in the lower CRE during autumn. Thus, the lower CRE may not be conducive to supporting a *P. inopinus* population due to prey limitation, a hypothesis that merits testing. It is also possible, given the overlap in prey preferences between *P. inopinus* and *P. forbesi* as well as the greater tolerance of broader environmental conditions by *P. forbesi* (Cordell et al. 2010, Dexter et al. 2015), that the subsequent introduction of *P. forbesi* to the CRE may have contributed to the disappearance of *P. inopinus* from this estuary, as suggested by Dexter et al. (2015). Indeed, the issue of potential congeneric displacement in zooplankton is not novel to *P. inopinus* and *P. forbesi*. In the Gulf of Finland, Baltic Sea, the native *E. affinis* and its invasive congener *E. carolleeae* currently co-exist, but multiple biotic factors—in particular, similarities in population dynamics and differences in body size—point to the potential for *E. carolleeae* to displace native *E. affinis* (Sukhikh et al. 2019). Further studies to address the possibility of resource competition or competitive exclusion between *P. inopinus* and *P. forbesi* would provide insight into the current and future invasions of this type.

### 5. CONCLUSIONS

This study is the first, to our knowledge, to determine the prey preferences and feeding rates of the Asian calanoid copepod *Pseudodiaptomus inopinus*.
on natural prey assemblages, in either its native or invasive range. We found that *P. inopinus* is omnivorous in these estuaries, primarily consuming ciliates and diatoms and selecting for prey in the 21–30 μm size range. Its ingestion rates were highest on prey taxa that comprised the largest proportion of the available biomass in these estuaries, and when compared to other congeneric species, *P. inopinus* had a weight-specific ingestion rate similar to that of *P. forbesi*. This study expands our understanding of the trophic role of *P. inopinus* in 2 invaded northeast Pacific estuaries where it has become dominant by furthering our knowledge of the feeding rates, prey selection, and potential impacts of *P. inopinus* in these invaded systems.

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**LITERATURE CITED**

Ara K (2001) Length−weight relationships and chemical content of the planktontic copepods in the Cananeia Lagoon estuarine system, Sao Paulo, Brazil. Plankton Biol Ecol 48:121−127

Bellard C, Thuiller W, Leroy B, Genovesi P, Bakkenes M, Courchamp F (2015) Will climate change promote future invasions? Glob Change Biol 19:3740−3748

Bollens SM, Cordell JR, Avent S, Hooff R (2002) Zooplankton invasions: a brief review, plus two case studies from the northeast Pacific Ocean. Hydrobiologia 480:87−110

Bollens SM, Breckenridge JK, Vanden Hooff RC, Cordell JR (2011) Mesozooplankton of the lower San Francisco Estuary: spatio-temporal patterns, ENSO effects, and the prevalence of non-indigenous species. J Plankton Res 33:1358−1377

Bollens SM, Breckenridge JK, Cordell JR, Rollwagen-Bollens G, Kalata O (2012) Invasive copepods in the lower Columbia River Estuary: seasonal abundance, co-occurrence and potential competition with native copepods. Aquat Invasions 7:101−109

Bollens SM, Breckenridge JK, Cordell JR, Simenstad CA, Kalata O (2014) Zooplankton of tidal marsh channels in relation to environmental variables in the upper San Francisco Estuary. Aquat Biol 21:205−217

Bouley P, Kimmerer WJ (2007) *Ecology of a highly abundant, introduced cyclopoid copepod in a temperate estuary. Mar Ecol Prog Ser* 324:219−228

Bowen A, Rollwagen-Bollens G, Bollens SM, Zimmerman J (2015) Feeding of the invasive copepod *Pseudodiaptomus forbesi* on natural microplankton assemblages within the lower Columbia River. J Plankton Res 37:1089−1094

Breckenridge JK, Bollens SM, Rollwagen-Bollens G, Roeger GC (2015) Plankton assemblage variability in a river-dominated temperate estuary during late spring (high-flow) and late summer (low-flow) periods Estuar Coasts 38:93−103

Broglio E, Jónasdóttir SH, Calbet A, Jakobsen HH, Saiz E (2003) Effect of heterotrophic versus autotrophic food on feeding and reproduction of the calanoid copepod *Acartia tonsa*: relationship with prey fatty acid composition. Aquat Microb Ecol 31:267−278

Brown CA, Nelson WG (2010) Chapter 5: Case studies. 7: Yaquina Estuary. In: Gilbert PM, Madden CJ, Boynton W, Flemer D, Heil C, Sharp J (eds) Nutrients in estuaries: a summary report of the National Estuarine Expert Workgroup, 2005−2007. US Environmental Protection Agency, Washington, DC, p 127−135

Brown CA, Ozretich RJ (2009) Coupling between the coastal ocean and Yaquina Bay, Oregon: importance of oceanic inputs relative to other nitrogen sources. Estuar Coasts 32:219−237

Calbet A, Saiz E (2005) The ciliate−copepod link in marine ecosystems. Aquat Microb Ecol 38:157−167

Calliari D, Tiselius P (2009) Organic carbon fluxes through the mesozooplankton and their variability at different timescales in the Gullmarsfjord, Sweden. Estuar Coast Shelf Sci 85:107−117

Carlton JT, Geller JB (1993) Ecological roulette: the global transport of nonindigenous marine organisms. Science 261:78−82

Chen M, Kim D, Liu H, Kang CK (2018) Variability in copepod trophic levels and feeding selectivity based on stable isotope analysis in Gwangyang Bay of the southern coast of the Korean Peninsula. Biogeosciences 15:2055−2073

Cordell JR, Morrison SM (1996) The invasive Asian copepod *Pseudodiaptomus inopinus* in Oregon, Washington, and British Columbia estuaries. Estuaries 19:629−638

Cordell JR, Morgan CA, Simenstad CA (1992) Occurrence of the Asian calanoid copepod *Pseudodiaptomus inopinus* in the zooplankton of the Columbia River Estuary. J Crustac Biol 12:260−269

Cordell JR, Rasmussen M, Bollens SM (2007) Biology of the introduced copepod *Pseudodiaptomus inopinus* in a northeast Pacific estuary. Mar Ecol Prog Ser 332:211−227

Cordell JR, Bollens SM, Draheim R, Sytsma M (2008) Asian copepods on the move: recent invasions in the Columbia−Snake River system, USA. ICES J Mar Sci 65:753−758

Cordell JR, Tear LM, Bollens SM (2010) Modelling physico-chemical factors affecting occurrences of a non-indigenous planktonic copepod in northeast Pacific estuaries. Biol Invasions 12:1427−1445

Counts CL (1986) The zoogeography and history of the invasion of the United States by *Corbicula fluminea* (*Bivalvia: corbiculidae*). Am Malacol Bull (Spec Ed)2:7−39

Cuhel RL, Aguilar C (2013) Ecosystem transformations of the Laurentian Great Lake Michigan by nonindigenous biological invaders. Annu Rev Mar Sci 5:289−320

Dam HG, Lopes RM (2003) Omnivory in the calanoid copepod *Temora longicornis*: feeding, egg production and egg hatching rates. J Exp Mar Biol Ecol 292:119−137

Dehes H, Elasen K, Gaard E (2008) Seasonal variability in copepod ingestion and egg production on the Faroe shelf. Hydrobiologia 600:247−265

Dexter E, Bollens SM (2020) Zooplankton invasions in the early 21st century: a global survey of recent studies and recommendations for future research. Hydrobiologia 847:309−319
Dexter E, Bollens SM, Rollwagen-Bollens G, Emerson J, Zimmerman J (2015) Persistent vs. ephemeral invasions: 8.5 years of zooplankton community dynamics in the Columbia River. Limnol Oceanogr 60:527–539

Dexter E, Bollens SM, Cordell J, Soh HY and others (2018) A genetic reconstruction of the invasion of the calanoid copepod Pseudodiaptomus inopinus across the North American Pacific Coast. Biol Invasions 20:1577–1595

Dexter E, Katz S, Bollens SM, Rollwagen-Bollens G, Hampton S (2020a) Modeling the trophic impacts of invasive zooplankton in a highly invaded river. PLOS ONE 15: e0243002

Dexter E, Bollens SM, Rollwagen-Bollens G (2020b) Native and invasive zooplankton show differing responses to decadal-scale increases in maximum temperatures in a large temperate river. Limnol Oceanogr Lett 5:403–409

Dexter E, Bollens SM, Cordell J, Rollwagen-Bollens G (2020c) Zooplankton invasion on a grand scale: insights from a 20-year time-series across 38 Northeast Pacific estuaries. Ecosphere 11:03040

Domingues RB, Galvão H (2007) Phytoplankton and environmental variability in a dam regulated temperate estuary. Hydrobiologia 586:117–134

Durbin EG, Durbin AG (1992) Effects of temperature and food abundance on grazing and short-term weight change in the marine copepod Acartia hudsonica. Limnol Oceanogr 37:361–378

Eyun SL, Lee YH, Suh HL, Kim S, Soh HY (2007) Genetic identification and molecular phylogeny of Pseudodiaptomus species (Calanoida, Pseudodiaptomidae) in Korean waters. Zoolog Sci 24:265–271

Froneman PW (2004) In situ feeding rates of the copepods, Pseudodiaptomus hessei and Acartia longipellata, in a temperate, temporarily open/closed eastern Cape estuary. S Afr J Sci 100:577–583

Galloway AWE, Winder M (2015) Partitioning the relative importance of phylogeography and environmental conditions on phytoplankton fatty acids. PLOS ONE 10:e0130053

Garrido S, Curz J, Santos AMP, Ré P, Saiz E (2013) Effects of temperature, food type and food concentration on the grazing of the calanoid copepod Centroflagipes chierchiae. J Plankton Res 43:458–468

Gendaszek AS (2011) Hydrogeologic framework and groundwater/surface-water interactions of the Chehalis River Basin, southwestern Washington. Scientific Investigations Report No. 2011–5160. US Geological Survey, Reston, VA

Gifford DJ, Dagg MJ (1988) Feeding of the estuarine copepod Acartia tonsa Dana: carnivory vs. herbivory in natural microplankton assemblages. Bull Mar Sci 43:458–468

Gifford SM, Rollwagen-Bollens G, Bollens SM (2007) Mesozooplankton omnivory in the upper San Francisco Estuary. Mar Ecol Prog Ser 348:33–46

Gilbert PM, Madden CJ, Dettmann ED, Boynton W and others (2010) Chapter 4: a framework for developing nutrient criteria. In: Gilbert PM, Madden CJ, Boynton W, Flemer D, Heil C, Sharp J (eds) Nutrients in estuaries: a summary report of the National Estuarine Expert Working Group, 2005–2007. US Environmental Protection Agency, Washington, DC, p 43–71

Hanssen FC, van Boekel WHM (1991) Grazing pressure of the calanoid copepod Temora longicornis on a Phaeocystis dominated spring bloom in a Dutch tidal inlet. Mar Ecol Prog Ser 78:123–129

Hassett W, Bollens SM, Counihan TD, Rollwagen-Bollens G, Zimmerman J, Emerson J (2017) Veligers of the invasive Asian clam Corbicula fluminea in the Columbia River Basin: broadscale distribution, abundance and ecological associations. Lake Reserv Manage 33:234–248

Hillebrand H, Dürselen C-D, Kirschdel T, Pollingher U, Zohary T (1999) Biovolumecalulation for pelagic and benthic microalgae. J Phycol 35:404–424

Kâ S, Mendoza-Vera JM, Bouvy M, Champalbert G, N’Gom-Kâ R, Pagano M (2012) Can tropical freshwater zooplankton graze efficiently on cyanobacteria? Hydrobiologia 679:119–138

Karentz D, McIntire CD (1977) Distribution of diatoms in the plankton of Yaquina Estuary, Oregon. J Phycol 13:379–388

Kayleitz K, Kimmerer W (2017) Abiotic and biotic controls on the copepod Pseudodiaptomus forbesi in the upper San Francisco Estuary. Mar Ecol Prog Ser 581:85–101

Kiarboe T, Mehlberg F, Hamburger K (1985) Bioenergetics of the planktonic copepod Acartia tonsa: relation between feeding, egg production and respiration, and composition of specific dynamic action. Mar Ecol Prog Ser 26: 85–97

Kirchman DL (1993) Statistical analysis of direct counts of microbial abundance. In: Kemp PF (ed) Handbook of methods in aquatic microbial ecology. CRC Press, Washington, DC, p 117–119

Koski M, Klein Breterel W, Schogt N (1998) Effect of food quality on rate of growth and development of the pelagic copepod Pseudocalanus elongatus (Copepoda, Calanoida). Mar Ecol Prog Ser 170:169–187

Kouassi E, Pagano M, Saint-Jean L, Arfi R, Bouvy M (2001) Vertical migrations and feeding rhythms of Acartia clausi and Pseudodiaptomus hessei (Copepoda: Calanoida) in a tropical lagoon (Ebrîê, Côte d’Ivoire). Estuar Coast Shelf Sci 52:715–728

Kratinia P, Nally RM, Kimmerer WJ, Thomson JR, Winder M (2014) Human-induced biotic invasions and changes in plankton interaction networks. J Appl Ecol 51:1066–1074

Mackas D, Bohrer R (1976) Fluorescence analysis of zooplankton gut contents and an investigation of diet feeding patterns. J Exp Mar Biol Ecol 25:77–85

Malzahn AM, Hantzsche F, Schoo KL, Boersma M, Aberle N (2010) Differential effects of nutrient-limited primary production on primary, secondary or tertiary consumers. Oecologia 162:33–48

Marin V, Huntley ME, Frost B (1986) Measuring feeding rates of pelagic herbivores: analysis of experimental design and methods. Mar Biol 93:49–58

Matsui H, Sasaki T, Kobari T, Waqalevu V, Kikuchi K, Ishikawa M, Kotani T (2021) DHA accumulation in the polar lipids of the euryhaline copepod Pseudodiaptomus inopinus and its transfer to red sea bream Pagrus major larvae. Front Mar Sci 8:e632876

Menden-Deuer S, Lessard EJ (2000) Carbon to volume relationships for dinoflagellates, diatoms, and other protist plankton. Limnol Oceanogr 45:569–579

Motwani NH, Gorokhova E (2013) Mesozoooplankton grazing on picocyanobacteria in the Baltic Sea as inferred from molecular diet analysis. PLOS ONE 8:e79230

Nejstgaard JC, Naustvoll LJ, Szahin A (2001) Correcting for underestimation of microzooplankton grazing in bottle incubation experiments with mesozoooplankton. Mar Ecol Prog Ser 221:59–75
