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Particle rejection by *Calanus pacificus*: discrimination between similarly sized particles

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Abstract

Particle rejection behavior was studied for late-stage copepodites and adult females of *Calanus pacificus* collected from La Jolla Bay, California (32°N; 117°W) in spring and summer of both 1979 and 1982. Several particles of approximately the same size were offered, both singly and in mixtures, to *C. pacificus*. In the first set of experiments the copepods were presented with choices between a diatom, *Thalassiosira weissflogii* (13.1 μm equivalent spherical diam), and polystyrene beads of two sizes (11.1 and 16.5 μm diam). In the second set of experiments they were presented with choices between one of two dinoflagellates, *Gyrodinium dorsum* (27 μm diam) or *Peridinium trochoideum* (22 μm diam), and polystyrene beads of three sizes (15, 20 and 25 μm diam). When offered mixtures of cells and beads, the filtration and ingestion rates were significantly greater on the cells than on the beads, irrespective of bead size. We also find that *C. pacificus* demonstrates a clear preference for *G. dorsum* relative to *P. trochoideum*. Our results cannot be explained by a model of passive, size-selective feeding.

Introduction

Herbivorous calanoid copepods have, until quite recently, been regarded as strict filter-feeders (Jørgensen, 1966). From the feeding currents generated by their mouthparts (Cannon, 1928), suspended particulate matter was thought to be trapped by the sieve-like mesh of setae and setules which comprise the second maxillae. It was further suggested that the spaces between setules determine the efficiency with which different particle sizes are caught (Nival and Nival, 1976), and this provided an explanation (Boyd, 1976) for the many observations of apparent selection for larger particles (e.g. Mullin, 1963; Frost, 1977; Runge, 1980).

An implicit corollary to this model of copepods as mechanical filter-feeders is that their ingestion rates can be fully characterized by the size and abundance of the particles they encounter, and many predictive models of copepod feeding make this assumption (e.g. Lam and Frost, 1976; Wroblewski and O'Brien, 1976; Conover and Huntley, 1980).

Two relatively recent findings have challenged these interpretations of copepod feeding behavior. First, cinematographic evidence has shown that the feeding appendages are surrounded by a highly viscous layer of water, which causes them to behave as "paddles" rather than as "sieves" (Koehl and Strickler, 1981). The mouthparts do not passively strain particles from the surrounding medium, but instead capture parcels of water containing particles and then remove the water by squeezing it out (Paffenhöfer *et al.*, 1982; Strickler, 1982). Second, a few studies have shown that copepods are able to select particulate food according to its quality. Poulet and Marsot (1978) showed that two species of neritic copepods selected artificial microcapsules enriched with phytoplankton homogenate over unenriched microcapsules of the same size; and Donaghay and Small (1979) demonstrated that *Acartia clausi* could discriminate between phytoplankton cells and plastic beads of approximately the same size.

The emerging model of copepods as perceptive, discriminating particle feeders is, however, far from complete. The filmed observations suggest that copepods are able to detect the chemical microenvironment of an individual particle (Strickler, 1982), and that they respond accordingly by either ingesting or rejecting the particle. Critics could contend that existing cinematographic data show no more than that some particles are successfully captured and others are not. Similarly, experimental studies which purport to demonstrate particle selection on the basis of quality have also been criticized (see Gifford *et al.*, 1981). Finally, it remains true that "the important question of whether particle selection takes place among different particles of similar size, e.g. algal cells, of different nutritional value, still goes largely unanswered" (Hughes, 1980).

We present the results of experiments designed to determine whether or not the copepod *Calanus pacificus* is capable of selecting particulate food according to its quality. Bearing in mind, and attempting to avoid, the potential criticisms which have been leveled at similar studies, we presented *C. pacificus* with a choice of different types of particles in a very narrow size range. We find that the copepod responds not only to a particular type of particle, but also to the combination of available particles.

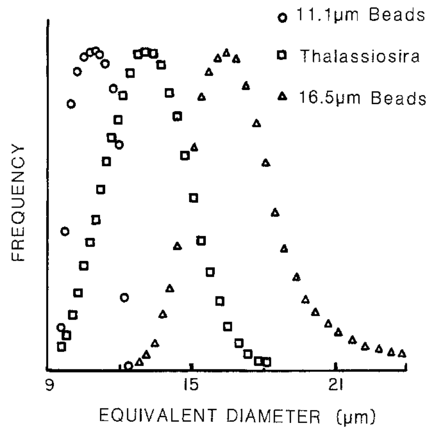

Fig. 1. Relative size–frequency distributions of the three particle types used in diatom grazing experiments (*Thalassiosira weissflogii* of 13.1 μm and different-sized polystyrene beads), as measured with an electronic particle counter (Electrozone Celloscope)

Materials and methods

Diatom experiments

Calanus pacificus were collected 2 km from the coast off La Jolla, California (32°N; 117°W) during the summer of 1979. Stage IV copepodites and adult females of *C. pacificus* were sorted live, and 5 to 10 individuals were placed in experimental glass bottles (950 ml) within 3 h of collection. The bottles were rotated at 1 rpm on a motor-driven wheel at 16 °C under dim light ($<2\times10^{13}$ quanta cm^{-2} s^{-1}, or 0.02% of noon sunlight) for 24 h. At the end of the experiment the copepods were preserved in formalin for later examination.

Particles were suspended in freshly collected seawater which had been filtered through Whatman GF/C filters. We used the diatom *Thalassiosira weissflogii* (= *T. fluviatilis*, Food Chain Research Group Culture Collection, Scripps Institution of Oceanography, La Jolla) and polystyrene beads of two sizes (Duke Scientific Corp., Palo Alto, California). Size distributions of these three particles are shown in Fig. 1; the modal equivalent spherical diameters were 13.1 μm for the diatom, and 11.1 and 16.5 μm for the polystyrene beads.

The experiments were run in quadruplicate. On each of three occasions treatments included: (1) diatom cells alone, (2) large polystyrene beads alone, and (3) a mixture of cells and the larger beads. On the third occasion we used an additional treatment: cells with both smaller and larger beads. We attempted to keep the total volume of particles in each experiment constant at 6×10^{6} μm^{3} ml^{-1} (for *Thalassiosira weissflogii,* this equals ~ 300 μg C l^{-1}). In one experiment the final particle suspensions were preserved in basic Lugol's solution; 100 ml aliquots of the preserved fluids were settled and the faecal pellets were counted and examined microscopically.

Two sets of experiments were conducted with CIV copepodites, and one set with adult females. In the first set of experiments we used two additional treatments: (1) large beads in IMR medium (Eppley *et al.*, 1967), and (2) large beads in "spent" medium (IMR medium in which log-phase *Thalassiosira weissflogii* cells had been growing, which was then filtered through Whatman GF/C filters). The experiments with fresh and "spent" medium were designed to determine whether or not extracellular compounds released by the phytoplankton might stimulate feeding activity by the copepods.

The particle suspensions were counted on an Electrozone Celloscope electronic particle counting system (Particle Data Systems, Inc.) with a 76 μm orifice. For our experiments we analyzed 41 channels of data over a size range of to 9 to 24 μm. Filtration and ingestion rates were calculated on the basis of particle disappearance from each channel using the methods of Frost (1972), where the filtration rate, F, is given by:

$$F=V g/N \text{ (ml copepod}^{-1}\text{ h}^{-1}), \tag{1}$$

where V is the volume of the experimental grazing chamber (ml), g is the grazing constant, and N is the number of copepods in the grazing chamber. The grazing constant, g, is calculated from:

$$g=k-[(\ln C_2^{*}-\ln C_1^{*})/(t_2-t_1)], \tag{2}$$

where k, the algal growth constant is given by:

$$k=[(\ln C_2-\ln C_1)/(t_2-t_1)], \tag{3}$$

and where C_1 and C_2 are the initial and final concentrations (cells ml^{-1}), respectively, in the control chamber which contains no copepods; C_1* and C_2* are the initial and final cell concentrations in the experimental chamber; and (t_2-t_1) is the time elapsed during the experiment.

The ingestion rate, I, is given by:

$$I=F\,C \text{ (cells copepod}^{-1}\text{ h}^{-1}), \tag{4}$$

where F is the filtration rate and C is the mean cell concentration, calculated from:

$$C=\frac{C_1^{*}\,[e^{(k-g)(t_2-t_1)}-1]}{(t_2-t_1)(k-g)} \tag{5}$$

In our experiments, there was no significant change between the initial control (C_1) and final control (C_2) cell concentrations; since the initial concentrations were identical in both the control and experimental treatments (i.e., $C_1=C_1^{*}$), the calculation of filtration and ingestion rates was somewhat simplified.

Dinoflagellate experiments

Some of the experimental results purported to demonstrate the dependence of selection on particle quality have been criticized recently, either for the use of electronic particle counters (Harbison and McAlister, 1980) or for the use of *Thalassiosira weissflogii* as a reference particle (Gifford *et al.,* 1981). Having demonstrated that the presence or absence of chitan spines on *T. weissflogii* could affect the filtration rate of *Calanus finmarchicus,* Gifford *et al.* concluded that "the postulated rejection of plastic spheres in favor of *Thalassiosira* cells may be an artifact of experimental design".

Bearing these criticisms in mind, we fed copepods on spineless dinoflagellate cells, and analyzed samples microscopically rather than with an electronic particle counter. Copepods were collected in the spring of 1982, in the same manner as for the diatom experiments. Stage V copepodites and adult females were sorted live, and 3 to 5 individuals were placed in experimental bottles shortly thereafter. The bottles were rotated on a grazing wheel at 15 °C in the dark for 24 h. Samples of initial and final control (no copepods) and experimental suspensions were preserved in basic Lugol's solution for later microscopic examination.

Particle suspensions were prepared with fresh, GF/C-filtered seawater. We used two species of dinoflagellates, *Gyrodinium dorsum* (27 μm) and *Peridinium trochoideum* (22 μm), both of which are approximately spherical. We also used polystyrene beads of three sizes, with modal diameters of 15, 20 and 25 μm.

Experiments were run in quadruplicate. In two experiments we fed copepods on *Gyrodinium dorsum,* and in one experiment we fed copepods on *Peridinium trochoideum.* On each occasion the treatments included: (1) dinoflagellate cells only, (2) a mixture of small beads and large beads, and mixtures of (3) cells and small beads, (4) cells and medium beads, (5) cells and large beads, and (6) cells, small beads and large beads. We kept the total volume of particles in each experiment constant at approximately $5 \times 10^5\ \mu m^3\ ml^{-1}$ (for the two dinoflagellate cells used, this equals $\sim 50\ \mu g\ C\ l^{-1}$), while also attempting to keep the total volume of each particle type approximately equal in the case of mixtures.

We used CV copepodites in all experiments except the second one with *Gyrodinium dorsum,* in which we used adult female copepods. Preserved particle suspensions were counted on a Zeiss inverted microscope using the Utermöhl method (Lund *et al.,* 1958), and from these data we calculated filtration and ingestion rates following Frost (1972). We also counted faecal pellets at the end of each experiment, and calculated their rates of production.

Results

Diatom experiments

Ingestion rates and filtration rates of *Calanus pacificus* Copepodite CIV are presented in Figs. 2–5. Ingestion rates of adult female *C. pacificus* were approximately 15% higher. When *Thalassiosira weissflogii* cells were the only particles offered, the maximum filtration rate on a size class was 2.5 ml copepod^{-1} h^{-1}, and the maximum ingestion rate reached 600 cells copepod^{-1} h^{-1} (Fig. 2). Filtration and ingestion rates were greatest on the size class where the cell concentration was greatest; there was no apparent production of particles smaller than the cells, as has sometimes been observed (Frost, 1977). In a second treatment, copepods were presented with a suspension containing only 16.5 μm beads (Fig. 3). The filtration rate was <0.3 ml copepod^{-1} h^{-1}, and the ingestion rate (<30 beads copepod^{-1} h^{-1}) was lower by a factor of 20 than when cells were offered alone. In all experiments (including those run in fresh or spent IMR medium) beads offered alone markedly depressed the ingestion rates.

In a third treatment, equal volumes of the *Thalassiosira weissflogii* and the 16.5 μm bead suspension were mixed and offered to the copepods (Fig. 4). Filtration and ingestion rates were maximal on the size classes containing the *T. weissflogii* cells (12 to 15 μm), but approached zero in the region of the bead peak (15 to 18 μm). Diatom cells were ingested from the mixture, but the ingestion rate was about one-third that when cells were offered alone. Filtration rates were <1.1 ml copepod^{-1} h^{-1}, also much lower than in the suspension of cells alone.

In the final treatment from this series, both sizes of polystyrene beads were offered in combination with *Thalassiosira weissflogii* (Fig. 5). Again, ingestion rates on the phytoplankton cells were much greater than on either of the two bead sizes. The maximum ingestion rate on the cells reached only 100 cells copepod^{-1} h^{-1}. These rates are markedly lower than when copepods were offered either cells alone or cells in combination with the large beads.

In treatments where cells were present, the filtration effort was centered on the size classes containing the cells. Bead consumption was low in all of the experiments, even when beads were the only available particles.

Our results further demonstrate that the relative proportions of diatom cells and non-nutritious beads influenced the feeding behavior of the copepods. There is a significant relationship between the filtration rate on the cell size classes and the proportion of cells available in a given particle mixture (Fig. 6; $r^2=0.85$; $n=11$). As the proportion of inert particles in the mixture was increased, the filtration rate on *Thalassiosira weissflogii* cells declined proportionately. However, the filtration rate on beads remained constant, regardless of the proportion at which they were available.

Photomicrographs of the faecal material complement our interpretations of the results from the particle counts. Pellets produced by copepods feeding on mixtures of cells and beads (Fig. 7 a) were approximately the same size as pellets produced by copepods feeding on cells alone (Fig. 7 b); however, pellets produced by copepods feeding on beads alone were substantially smaller (Fig. 7 c). Note that faecal pellets from copepods grazing in the mixtures of cells and beads did in fact contain beads. The rates of

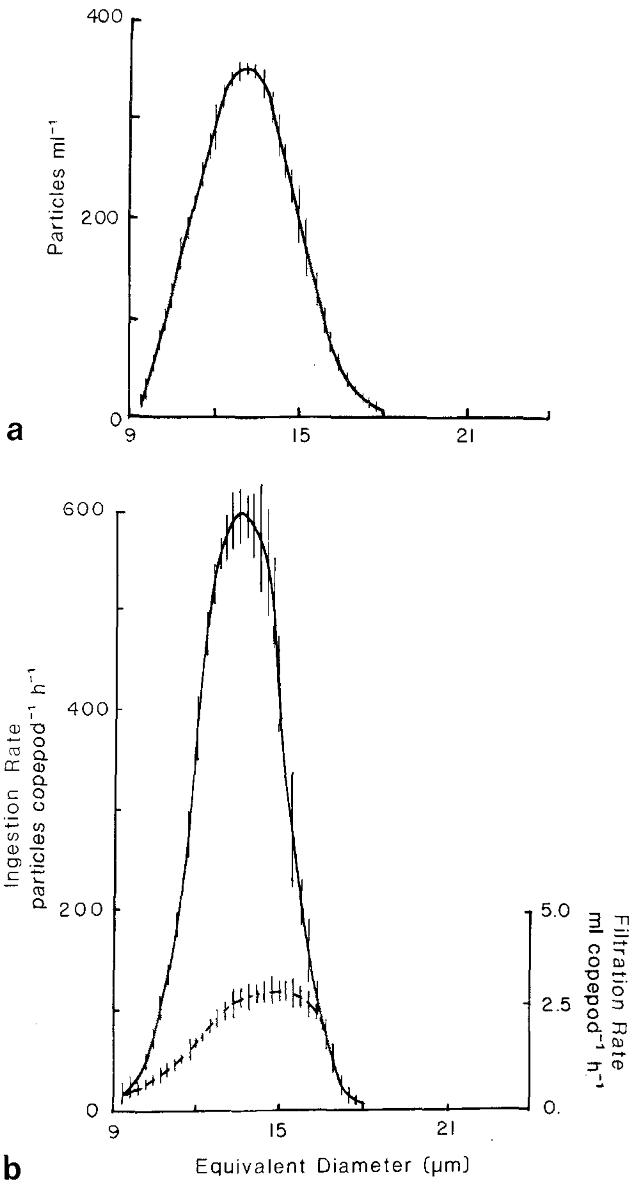

Fig. 2. *Calanus pacificus.* CIV copepodites feeding on *Thalassiosira weissflogii* cells only. (a) Initial size-frequency distribution of particles; (b) ingestion rate (continuous line) and filtration rate (dashed line) as function of particle size. Plotted data are mean of 4 replicates. Error bars are standard deviations

faecal pellet production in the cell suspension, the mixture, and the bead suspension were 3.1, 2.9 and 1.5 pellets copepod^{-1} h^{-1}, respectively. Faecal pellets were intact in all treatments, and the rate of faecal pellet production in the mixtures was not significantly different from that in the cell suspensions; these data suggest that the ingestion and subsequent resuspension of beads was not important.

Dinoflagellate experiments

Results of our feeding experiments with dinoflagellates were comparable to results of the diatom experiments: i.e.,

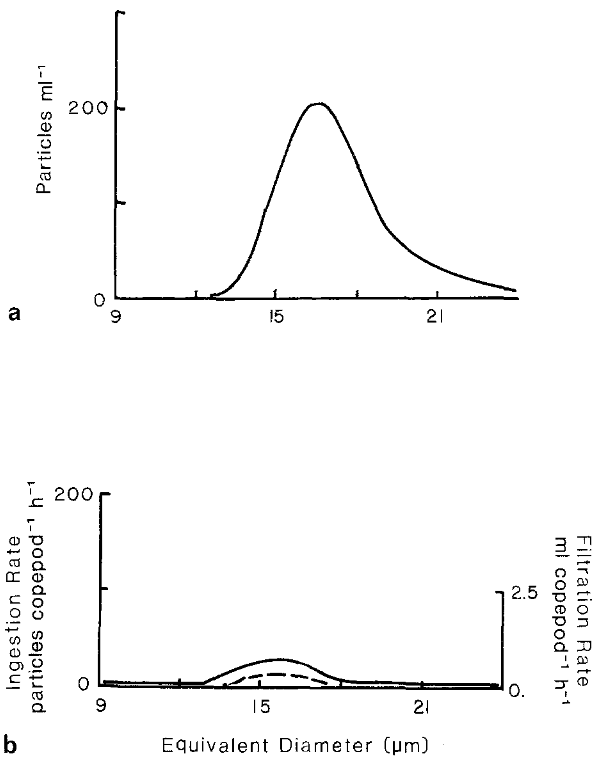

Fig. 3. *Calanus pacificus.* CIV copepodites feeding on 16.5 µm polystyrene beads only. Further details as in Fig. 2

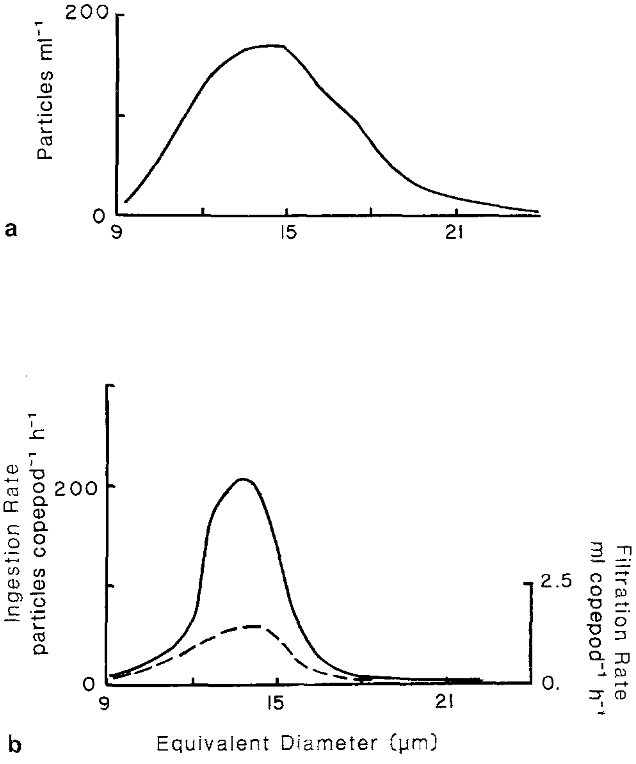

Fig. 4. *Calanus pacificus.* CIV copepodites feeding on *Thalassiosira weissflogii* cells plus 16.5 µm beads. Further details as in Fig. 2

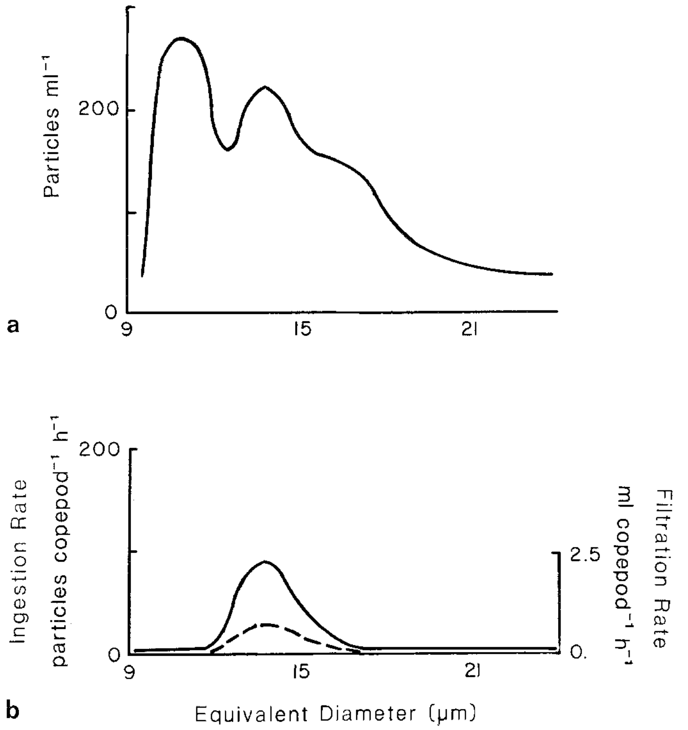

Fig. 5. *Calanus pacificus.* CIV copepodites feeding on *Thalassiosira weissflogii* cells plus 11.1 µm and 16.5 µm polystyrene beads. Further details as in Fig. 2

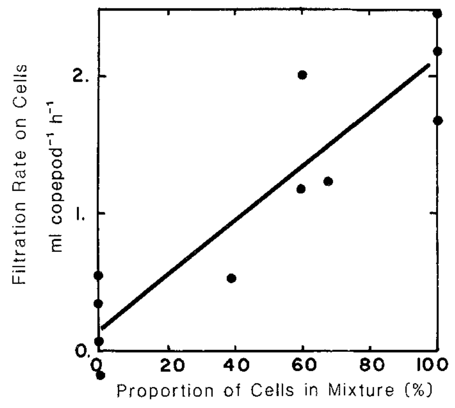

Fig. 6. *Calanus pacificus.* Filtration rate of copepods feeding on diatoms. Rates are averaged over particle size classes in which cells may be found versus the proportion of the total number of particles in a given experiment which were cells. Data from all treatments were pooled for this analysis

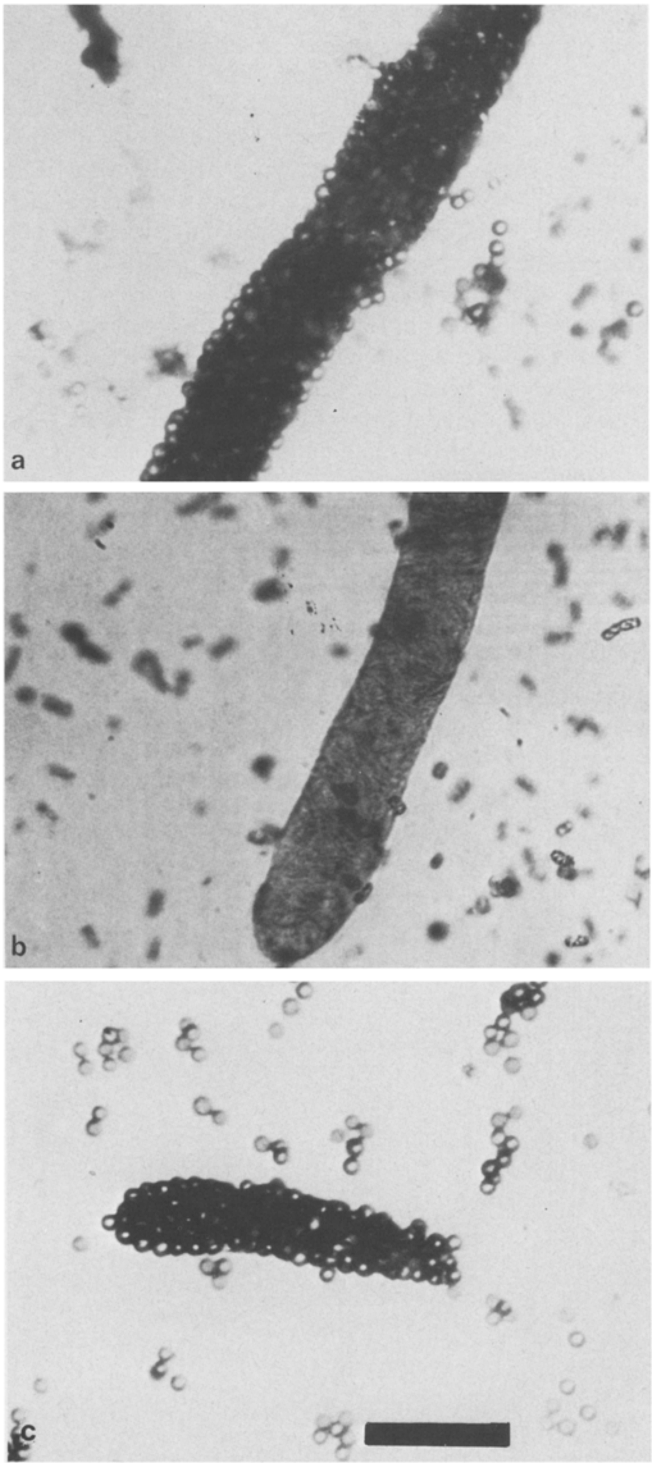

Fig. 7. *Calanus pacificus.* Photomicrographs of faecal pellets produced by CIV copepodites feeding on (a) mixture of *Thalassiosira weissflogii* cells and large beads, (b) cells alone, and (c) large beads alone. Scale bar = 125 µm

Calanus pacificus selects phytoplankton cells in preference to inert plastic beads of similar size and abundance. However, we also found distinct differences in the feeding rates on *Gyrodinium dorsum* and *Peridinium trochoideum*, despite similarities between these two species in size and shape. We did not offer *C. pacificus* a mixture of these two cells because their morphologies are so similar that they cannot be accurately distinguished in routine counting of mixtures.

For CV copepodites feeding on *Gyrodinium dorsum*, the filtration rate on cells ranged from 3.4 to 6.6 ml copepod^{-1} h^{-1}, which was significantly greater than the filtration rate on beads and also greater than the filtration rate on *Peridinium trochoideum.* In the third experiment, where adult females were presented with various suspensions of *G. dorsum* cells and/or beads, filtering rates were again much greater on cells (6.6 to 9.8 ml copepod^{-1} h^{-1}) than on beads (< 2.4 ml copepod^{-1} h^{-1}). In the dinoflagellate experiments, as in those with diatoms, there was some ingestion of beads when they were the only particle offered.

For CV copepodites feeding on *Peridinium trochoideum,* the filtering rate on cells ranged from 0.6 to 2.3 ml copepod^{-1} h^{-1} (Fig. 8). In mixtures containing cells and only one bead size, the filtering rate on cells was consistently greater than on beads; however, in the treatment containing both small and large beads in addition to cells, the filtering rate was lowest on the cells.

In experiments with *Peridinium trochoideum,* the ingestion rates of copepods were not much greater on cells than on beads present at the same concentrations (Fig. 9). By contrast, in experiments with *Gyrodinium dorsum,* the copepods ingested cells at higher rates than beads, regardless of whether these particles were offered singly or in combination (Fig. 10).

Particle preferences of *Calanus pacificus* were further evaluated by Ivlev electivity indices (Ivlev, 1961) for the

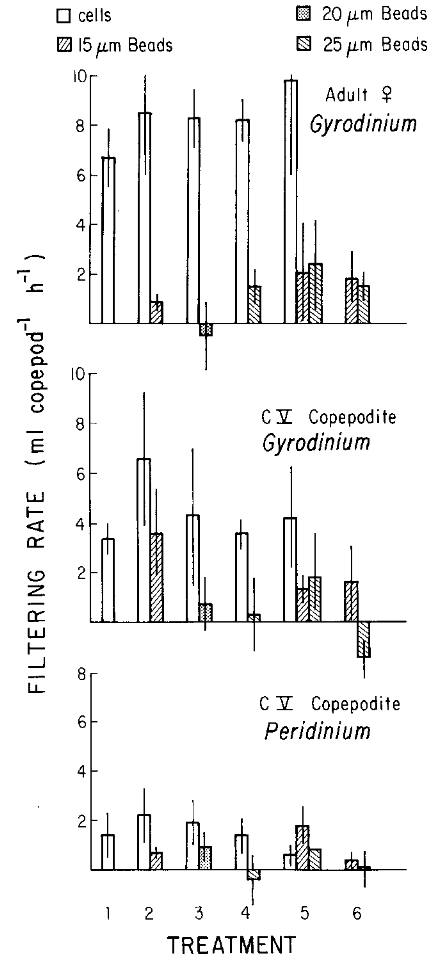

Fig. 8. *Calanus pacificus.* Filtration rates of copepods feeding on dinoflagellates and/or beads (two experiments with *Gyrodinium dorsum* and one experiment with *Peridinium trochoideum*). Means of 4 replicates ± 1 SD are shown. Treatments were: (1) cells only; (2) cells + 15 μm beads; (3) cells + 20 μm beads; (4) cells + 25 μm beads; (5) cells + 15 μm beads + 25 μm beads; (6) 15 μm beads + 25 μm beads. In all but one experiment, *C. pacificus* filtered at greater rates upon dinoflagellate cells than upon polystyrene beads. Furthermore, the copepods filtered *G. dorsum* at greater rates than they did *P. trochoideum*

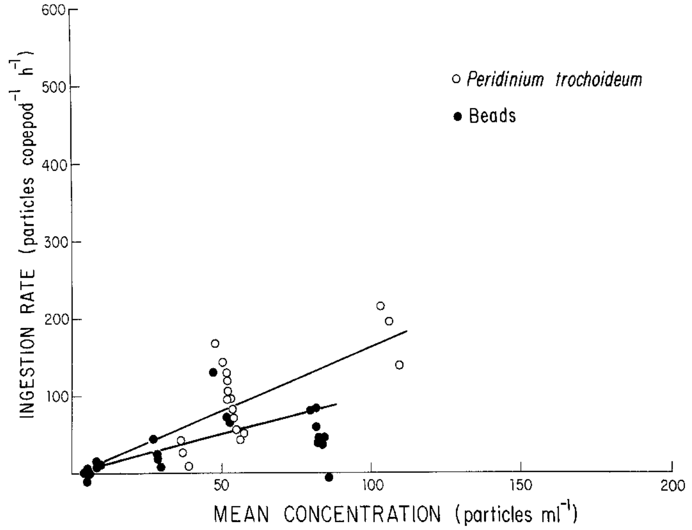

Fig. 9. *Calanus pacificus.* Comparison of ingestion rates of *Peridinium trochoideum* and polystyrene beads, offered either singly or in combination to CV copepodites. Abundances of particles were approximately equal on a volume basis. Lines fitted by eye. Copepodites show no significant selection for *P. trochoideum* in preference to beads offered at comparable concentrations

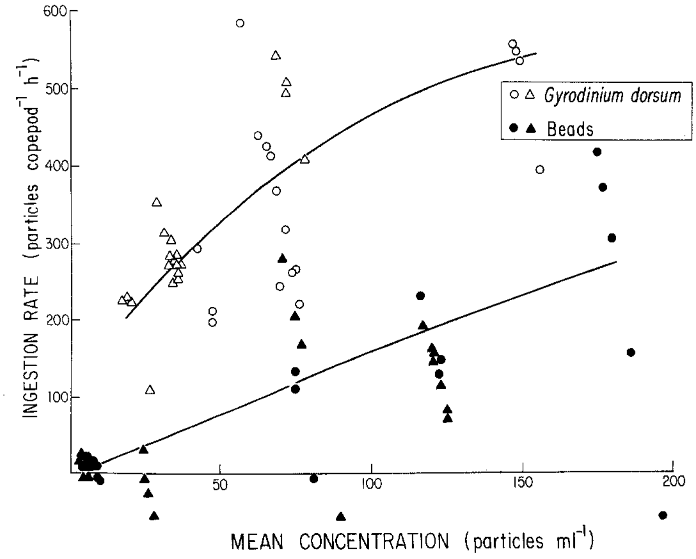

Fig. 10. *Calanus pacificus.* Comparison of ingestion rates of *Gyrodinium dorsum* and polystyrene beads, offered either singly or in combination. Abundances of particles were approximately equal on a volume basis. Triangles: CV copepodites; circles: adult females. Lines fitted by eye. Copepods show strong selection for *G. dorsum* cells in preference to beads offered at comparable concentrations

different particle types used in our experiments (Table 1). The Ivlev electivity index is given by:

$$E=\frac{r_i-P_i}{r_i+P_i} \qquad (6)$$

where r_i is the relative proportion (i.e., percentage) of a prey particle in the ration, and P_i is the relative proportion of the same prey in the water. Positive values (0 to +1) indicate a preference, whereas negative values (0 to −1) indicate an avoidance of a class of particles. Selection for *Gyrodinium dorsum* was positive ($\bar{x}=0.27$), whereas selection for *Peridinium trochoideum* was neither strongly positive nor negative ($\bar{x}=0.02$). Selection for all bead sizes was negative, with means of −0.20, −0.61 and −0.52 for 15 μm, 20 μm and 25 μm beads, respectively.

Our analysis of faecal material reinforces the interpretation of results based on particle counts. First, the pellets produced in the various treatments were similar in size and shape to those produced in comparable treatments from the diatom experiments (Fig. 7). Second, the volume of pellets produced by CV copepodites in the experiment using *Gyrodinium dorsum* was, on average, 1.74 times as large as those produced in the experiment using *Peridinium trochoideum* (Table 2). Furthermore, pellets pro-

Table 1. *Calanus pacificus.* Ivlev electivity indices for copepods feeding on *Gyrodinium dorsum*, *Peridinium trochoideum* and three sizes of polystyrene beads, offered either singly or in combination. (Treatments are described in text and numbered in Table 2) Copepods showed positive selection for *G. dorsum*, no significant selection for *P. trochoideum*, and negative selection for polystyrene beads. SD = standard deviation

Copepodite stage	Treatment No.	Cells		Beads:		
		G. dorsum (27 μm)	*P. trochoideum* (22 μm)	15 μm	20 μm	25 μm
CV	2			0.04[a]		−0.08[a]
	3	0.54		−0.48		
	4	0.28			−1.00	
	5	0.08				−0.066
	6	0.46		−0.29		−0.19
CV	2			0.02[a]		−0.58[a]
	3		0.26	0.28		
	4		0.10		−0.27	
	5		0.05			−1.00
	6		−0.33	0.17		−0.24
CVI♀	2			0.02[a]		−1.00[a]
	3	0.19		−0.10		
	4	0.28			−0.56	
	5	0.05				−0.88
	6	0.32		−0.23		−0.08
Mean:		0.27	0.02	−0.20	−0.61	−0.52
SD:		±0.17	±0.25	±0.22	±0.37	±0.39

[a] Indices from experiment with beads only not included in calculation of mean

Table 2. *Calanus pacificus.* CV copepodites: comparison of faecal pellet volumes of individual pellets produced in experiments using polystyrene beads, offered either singly or in combination with *Gyrodinium dorsum* or *Peridinium trochoideum*. $\bar{x}$ = mean volume; SD = standard deviation; n = number of pellets measured. Pellets produced in the *P. trochoideum* experiment were, on average, smaller than those produced in the *G. dorsum* experiment

Treatment	*G. dorsum*			*P. trochoideum*		
	$\bar{x}$ ($10^3\,\mu m^3$)	SD ($10^3\,\mu m^3$)	n	$\bar{x}$ ($10^3\,\mu m^3$)	SD ($10^3\,\mu m^3$)	n
(1) Cells only	1 701	995	19	1 273	314	3
(2) 15 μm + 25 μm beads	1 616	1 326	18	832	353	39
(3) Cells + 15 μm beads	1 565	962	14	728	413	16
(4) Cells + 20 μm beads	514	417	12	394	312	9
(5) Cells + 25 μm beads	1 084	580	62	541	308	20
(6) Cells + 15, 25 μm beads	1 934	980	116	1 077	482	28
Overall mean =		1 581			803.8	

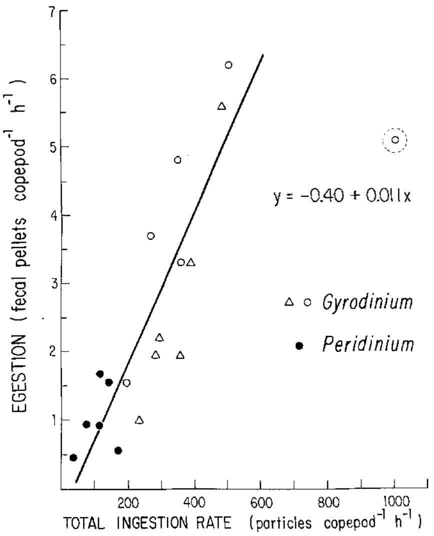

Fig. 11. *Calanus pacificus.* Faecal pellet production rates as a function of total ingestion rate in dinoflagellate experiments. Triangles: CV copepodites feeding on *Gyrodinium dorsum* and beads; filled circles: CV copepodites feeding on *Peridinium trochoideum* and beads; open circles: adult females feeding on *G. dorsum* and beads. Each point represents mean of a single treatment. Faecal pellet volumes calculated from microscopic measurements. One outlying point (circled: from females feeding on a mixture of *G. dorsum* and 15 μm beads) was omitted from regression analysis

duced in suspensions of beads alone tended to be smaller, by about 50%, than pellets produced in any other treatment.

Finally, analysis of the faecal pellet production rates (Fig. 11) indicates that they were significantly correlated with the total ingestion rate ($r^2 = 0.76$; $n = 17$). Thus, low rates of faecal pellet production in the *Peridinium trochoideum* experiment reflect the low ingestion rates calculated from particle counts.

Discussion

The ability of calanoid copepods to select particulate matter according to its size has been well documented (e.g. Mullin, 1963; Frost, 1977; Runge, 1980). Such results have been interpreted as due to passive size-selection based on differential filtration efficiencies of differently sized particles (Boyd, 1976; Steele and Frost, 1977).

Is it possible that our results could be explained by a model of passive size-selection? Gifford *et al.* (1981) suggested that the presence of long chitan spines on *Thalassiosira weissflogii* cells (McLachlan *et al.*, 1965) caused their "effective size to be shifted to the right in the particle size spectrum," with the result that *Calanus finmarchicus* fed on them as if they were larger than indicated by an electronic particle counter. Gifford *et al.* (1981) showed that the filtration rates on spined cells were 1.60 to 1.89 times as great as on cells whose spines had been experimentally removed (their Table 2). The "spine effect" cannot satisfactorily explain our results. In our experiments the filtration rate on cells (2.5 ml copepod^{-1} h^{-1}) was greater by a factor of 8.1 than the rate on similarly sized 16.5 μm beads (< 0.3 ml copepod^{-1} h^{-1}).

Frost (1977) performed a similar series of experiments (using *Calanus pacificus* adult females, *Thalassiosira weissflogii*, and beads of 10.3, 20 and 32 μm diam) and concluded that the copepods exhibited no behavioral component of size-selection. He considered his results to be compatible with a model of passive size-selection. We believe that the methods of analysis may be responsible for the difference between our results and those of Frost (1977). The particle concentrations in his experiments were so high that *C. pacificus* removed < 10% of the total suspended particles. Therefore, being prevented from estimating particle removal based on the initial and final concentrations (as we have done), Frost analyzed the gut contents of the copepods to estimate the relative ingestion of beads. He based his estimates on the size-frequency distribution of different beads in the guts relative to that in the suspension, and concluded that selection was passive. He did not measure the ingestion rates of either diatoms or beads. We found, as did Frost, that *C. pacificus* removed measurable amounts of beads from suspension, but we have demonstrated that ingestion rates on cells were far greater than on beads.

Donaghay and Small (1979) found that *Acartia tonsa* discriminated between *Thalassiosira fluviatilis* (= *T. weissflogii*) and 20 μm latex spheres. They concluded that their results were in part due to rejection of the undesirable particles. Our work on *Calanus pacificus* complements their results, but we also observe that the copepods are not completely efficient at rejecting the undesirable particles. Donaghay and Small's examination of the faecal pellets produced by *A. tonsa* showed that there were very few spheres (1 to 3 spheres in < 5% of the pellets) in faecal matter. By contrast, *C. pacificus* pellets from copepods fed in a mixture of cells and beads contained large numbers of beads (see Fig. 7a). In the results of Donaghay and Small (1979) the distributions of cells and beads were almost completely disjunct, whereas in our diatom experiments the distributions greatly overlapped (Fig. 1). However, Donaghay and Small also conducted experiments with *A. tonsa* in which distributions of cells and beads greatly overlapped (P. L. Donaghay, personal communication), and again found very few spheres in the faecal pellets. Thus, we suggest a species-specific difference: *A. tonsa* may be capable of more efficient discrimination than *C. pacificus.*

We considered the possibility that dissolved compounds excreted by phytoplankton could stimulate *Calanus pacificus* to feed on plastic beads. Lower invertebrates respond to the presence of dissolved compounds: e.g. *Hydra* spp. exhibit typical feeding behavior at low concentrations of dissolved glutathione (Loomis, 1955). Furthermore, copepods have chemosensory abilities which are used in the activities of both feeding (Poulet and Marsot, 1978) and sex (Katona, 1973), but response to dissolved compounds is known certainly only in the latter case. The work of Poulet and Marsot (1978) showed that *Acartia clausii* and *Eurytemora herdmanni* could detect chemical differences between artificial microcapsules containing phytoplankton homogenate and those containing seawater. However, the microcapsules were permeable to low molecular weight compounds and thus it is not certain whether the copepods responded to chemical differences in the microenvironment surrounding the microcapsules or to differences in their content or surface chemistry.

We presented copepods with beads suspended in both fresh IMR medium and in "spent" IMR medium (growth medium from which all *Thalassiosira weissflogii* cells had been filtered), and found no difference between the filtration or ingestion rates in these treatments. If *T. weissflogii* loses long-lived dissolved material to the medium, these compounds did not deceive the copepods into treating beads as phytoplankton cells. Of course, the bead will not have a concentration gradient of some "feeding substance" originating from it. This suggests that chemosensory abilities are probably used in the immediate microenvironment around a food particle rather than in sensing a general characteristic of the environment. Alcaraz *et al.* (1980) and Koehl and Strickler (1981) have reached the same conclusion, based upon cinematographic observations of copepod feeding behavior.

Our evidence for post-capture rejection is indirect. In our diatom experiments, as the proportion of inert particles in the mixture was increased, *Calanus pacificus* reduced its filtration rate on cells proportionately, and consequently the ingestion rate was reduced. This indicates that proportionately more time is required to handle and process a "nutritious" particle as the relative abundance of "non-nutritious" particles is increased. We were not able to detect a similar effect in our dinoflagellate experiments, and we cannot suggest a satisfactory explanation for this discrepancy. One possibility is that *C. pacificus* encounters more difficulty in handling and sorting spinose *Thalassiosira weissflogii* cells from beads than it does in handling and sorting dinoflagellate cells from beads. The results of the diatom experiments are suggestive, but more work is obviously required to ascertain the effect of non-nutritious particles on the ability of copepods to ingest nutritious particles.

The results of our experiments with dinoflagellates demonstrate beyond doubt that *Calanus pacificus* is capable of discriminating between particles of different types, though they may be equal in size and abundance. Not only did the copepods display a preference for *Gyrodinium dorsum* relative to plastic beads, they also apparently preferred *G. dorsum* to *Peridinium trochoideum*. Neither of these results can be explained by a model of passive size-selection. The two dinoflagellate species are virtually indistinguishable in their morphology; *G. dorsum* differs from *P. trochoideum* only in that (a) its modal diameter is slightly larger (27 vs 22 μm) and (b) the girdle is displaced in *G. dorsum* but not in *P. trochoideum*. Thus, we suggest that differences in the chemical characteristics of the two species may have caused *C. pacificus* to feed differently upon them.

The rejection of dinoflagellate cells is known from a limited number of other studies. For example, White (1979) found that *Acartia tonsa* ingested cells from highly luminescent cultures of *Gonyaulax excavata* at much lower rates than it ingested those from low-luminescent cultures of the same species. Huntley (1982) demonstrated that *Calanus pacificus* avoided the ingestion of a naturally occurring bloom of *Gymnodinium flavum*. Fiedler (1982) found that copepods and other particle-feeding zooplankton avoided the biomass maxima of *Gymnodinium splendens* in coastal waters off Southern California and fed poorly in waters from the maximum layers. These observations support the hypothesis that grazing avoidance by zooplankton may be a key factor in the initiation and maintenance of dinoflagellate blooms (Wyatt and Horwood, 1973).

Almost nothing is known of the ecological importance of particle rejection behavior by copepods, but studies such as ours demonstrate that copepods are capable of rejecting phytoplankton in the field. The potential significance of particle rejection cannot be overemphasized. Any primary production rejected by copepod grazers would be available to other parts of the marine ecosystem such as (a) the microheterotrophic food web, already estimated to consume 60% of primary production (Williams, 1981); (b) other, less discriminating, pelagic grazers such as salps or appendicularians (Harbison and McAlister, 1979; Alldredge, 1981); or (c) mesopelagic or benthic particle feeders, after the material had had time to sink out of the photic zone.

Extrapolations from our simple laboratory system are hazardous since we do not know how the discriminatory mechanism operates in natural particle systems. However, our results with *Calanus pacificus* demonstrate that these copepods are capable of making complicated choices. It seems reasonable that this behavior would not have evolved unless it were required in nature, but its trophodynamic significance remains virtually unexplored.

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Literature cited

Alcaraz, M., G.-A. Paffenhöfer and J. R. Strickler: Catching the algae: a first account of visual observations on filter-feeding calanoids. *In:* Evolution and ecology of zooplankton communities, pp 241–248. Ed. by C. Kerfoot. Hanover, New Hampshire: New England University Press 1980

Alldredge, A. L.: The impact of appendicularian grazing on natural food concentration *in situ.* Limnol. Oceanogr. *26,* 247–257 (1981)

Boyd, C. M.: Selection of particle sizes by filtering copepods: a plea for reason. Limnol. Oceanogr. *21,* 175–180 (1976)

Cannon, H. G.: On the feeding mechanism of the copepods *Calanus finmarchicus* and *Diaptomus gracilis.* J. exp. Biol. *6,* 131–144 (1928)

Conover, R. J. and M. E. Huntley: General rules of grazing in pelagic ecosystems. *In:* Primary productivity in the sea, pp 461–485. Ed. by P. Falkowski. New York: Plenum Press 1980

Cullen, J. J., S. G. Horrigan, M. E. Huntley and F. M. H. Reid: Yellow water in La Jolla Bay, California, July 1980. I. A bloom of the dinoflagellate, *Gymnodinium flavum* Kofoid & Swezy. J. exp. mar. Biol. Ecol. *63,* 67–80 (1982)

Donaghay, P. L. and L. F. Small: Food selection capabilities of the estuarine copepod *Acartia clausi.* Mar. Biol. *52,* 137–146 (1979)

Eppley, R. W., R. W. Holmes and J. D. H. Strickland: Sinking rates of marine phytoplankton measured with a fluorometer. J. exp. mar. Biol. Ecol. *1,* 191–208 (1967)

Fiedler, P.: Zooplankton avoidance and reduced grazing responses to *Gymnodinium splendens* (Dinophyceae). Limnol. Oceanogr. *27,* 961–965 (1982)

Frost, B. W.: Effects of the size and concentration of food particles on the feeding behavior of the marine planktonic copepod *Calanus pacificus.* Limnol. Oceanogr. *17,* 805–815 (1972)

Frost, B. W.: Feeding behavior of *Calanus pacificus* in mixtures of food particles. Limnol. Oceanogr. *22,* 472–491 (1977)

Gifford, D. N., R. N. Bohrer and C. M. Boyd: Spines on diatoms: do copepods care? Limnol. Oceanogr. *26,* 971–981 (1981)

Harbison, G. R. and V. L. McAlister: The filter-feeding rates and particle retention efficiencies of three species of *Cyclosalpa* (Tunicata, Thaliacea). Limnol. Oceanogr. *24,* 875–892 (1979)

Harbison, G. R. and V. L. McAlister: Fact and artifact in copepod feeding experiments. Limnol. Oceanogr. *25,* 971–981 (1980)

Hughes, R. N.: Optimal foraging in the marine context. Oceanogr. mar. Biol. A. Rev. *18,* 428–449 (1980)

Huntley, M. E.: Yellow water in La Jolla Bay, California, July, 1980. II. Suppression of zooplankton grazing. J. exp. mar. Biol. Ecol. *63,* 81–91 (1982)

Ivlev, V. S.: Experimental ecology of the feeding of fishes, 302 pp. Translated from Russian by D. Scott. New Haven: Yale University Press 1961

Jørgensen, C. B.: Biology of suspension feeding, 357 pp. Oxford: Pergamon Press 1966

Katona, S.: Evidence for sex pheromones in planktonic copepods. Limnol. Oceanogr. *18,* 574–583 (1973)

Koehl, M. A. R. and J. R. Strickler: Copepod feeding currents: food capture at low Reynolds number. Limnol. Oceanogr. *26,* 1062–1073 (1981)

Lam, R. K. and B. Frost: Model of copepod filtering response to changes in size and concentration of food. Limnol. Oceanogr. *21,* 490–500 (1976)

Loomis, W. F.: Glutathione control of the specific feeding reactions of *Hydra.* Ann. N.Y. Acad. Sci. *62,* 209–228 (1955)

Lund, J. W. G., C. Kipling and E. D. LeCren: The inverted microscope method of estimating algal numbers and the statistical basis of estimations by counting. Hydrobiologia *11,* 143–170 (1958)

McLachlan, J., A. J. McInnes and M. Falk: Studies on chitan (chitin: poly-*n*-acetyl glucosamine) fibers of the diatom *Thalassiosira fluviatilis* Hustedt. Can. J. Bot. *43,* 707–713 (1965)

Mullin, M. M.: Some factors affecting the feeding of marine copepods of the genus *Calanus.* Limnol. Oceanogr. *8,* 239–250 (1963)

Nival, P. and S. Nival: Particle retention efficiencies of an herbivorous copepod, *Acartia clausi* (adult and copepodite stages): effects of grazing. Limnol. Oceanogr. *21,* 24–38 (1976)

Paffenhöfer, G.-A., J. R. Strickler and M. Alcaraz: Suspension-feeding by herbivorous calanoid copepods: a cinematographic study. Mar. Biol. *67,* 193–199 (1982)

Poulet, S. A. and P. Marsot: Chemosensory grazing by marine calanoid copepods (Arthropoda: Crustacea). Science, N.Y. *200,* 1403–1405 (1978)

Runge, J. A. Effects of hunger and season on the feeding behavior of *Calanus pacificus.* Limnol. Oceanogr. *26,* 134–145 (1980)

Steele, J. H. and B. W. Frost: The structure of plankton communities. Proc. R. Soc. (Ser. B) *280,* 485–535 (1977)

Strickler, J. R.: Calanoid copepods, feeding currents, and the role of gravity. Science, N.Y. *218,* 158–160 (1982)

White, H. H.: Effects of dinoflagellate bioluminescence on the ingestion rates of herbivorous zooplankton. J. exp. mar. Biol. Ecol. *36,* 217–224 (1979)

Williams, P. J. LeB.: Incorporation of microheterotrophic processes into the classical paradigm of the planktonic foodweb. Kieler Meeresforsch. (Sonderh.) *5,* 1–28 (1981)

Wroblewski, J. S. and J. J. O'Brien: A spatial model of phytoplankton patchiness. Mar. Biol. *35,* 161–175 (1976)

Wyatt, T. and J. Horwood: Model which generates red tides. Nature, Lond. *244,* 238–240 (1973)

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