Ecology and Taxonomy of an Epizooic Diatom[1]

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ABSTRACT: An epizooic diatom, *Sameioneis carinae* gen. et sp. nov., has been found in the inland marine waters of northwestern Washington. It was found attached only to the copepod *Corycaeus affinis*. Attachment of the diatom occurs mainly on the second antennae and thoracic segments of male animals, and on the carapace and thoracic segments of females, which are the areas of contact during copulatory and noncopulatory clasping. It is probable that the diatoms are transferred to other animals during these activities.

COPEPODS, often the commonest of planktonic animals, are the hosts for a variety of parasitic and epizooic animals and plants. In the inland marine waters of northwestern Washington we have studied a marine copepod, *Corycaeus affinis*, and the diatoms that attach to it. One diatom in particular is abundant on most specimens of this animal, and we have investigated this diatom in detail, for it is not known to occur on any other substrate.

Although the genus *Corycaeus* Dana occurs in all the seas, it was first reported with epizooic diatoms by Giesbrecht (1892) from specimens collected in the Adriatic Sea. Giesbrecht's monograph of copepods included an illustration of a female *Corycaeus elongatus* which had stalked diatoms attached to the first four thoracic segments and the caudal ramus. There was no comment in Giesbrecht's text on this association.

Steuer (1910) found stalked diatoms on *Corycaeus brehmi, C. rostratus,* and *C. obtrusus* from collections in the Adriatic Sea. Schröder (1911) reported these diatoms as *Licmophora lyngbyei* (Kutz.) Grun., but in a later paper Schröder (1914) described epizooic diatoms of another collection as *Cymbella pusilla.*

Klevenhusen (1933) reported the only record of epizooic diatoms on *Corycaeus* from outside the Adriatic Sea when he found diatoms in the South Atlantic Ocean similar to those described by Giesbrecht. Klevenhusen tentatively identified these diatoms as *Amphora* sp. They were found on *Corycaeus speciosus, C. gracilis, C. lautus,* and *C. flaccus.*

The most detailed examination of stalked diatoms on *Corycaeus* was reported by Jurilj (1957) who studied specimens from the Adriatic Sea. Among other diatoms on these animals Jurilj reported two new species belonging to the new genus *Hormophora.* It is likely that these two species are the same as those that were earlier placed in the genera *Licmophora, Cymbella,* and *Amphora.*

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MATERIALS AND METHODS

Sampling was carried on at irregular periods from April 9, 1968 to December 4, 1968. Most of the specimens were obtained at a station in the northern part of Puget Sound two miles northwest of Edmonds, Washington. In addition, one sample was obtained from Admiralty Inlet near the entrance to Hood Canal, and two from Speiden Channel in the San Juan Archipelago, north of San Juan Island.

Specimens were obtained by vertical net tows to approximately 12 fathoms. The net samples were diluted in seawater in gallon-size jars,

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packed in ice, and returned to the laboratory where they were transferred to 200 mm stacking dishes and stored in culture chambers maintained on a 12-hour photoperiod at 10° C.

It was necessary to count and record most of the information on the living specimens within 12 hours after their collection because deterioration of the animals and diatoms often began at this time.

Attempts to culture the diatom in a seawater enrichment medium (Provasoli 1968) were unsuccessful. No other media were tried.

Preparations of cleared diatom frustules were made using the technique of Werff (1955). Fine detail of the diatom frustules was intensified by staining them with 0.1 percent alcohol solution of neutral red. Nuclei of the diatoms were stained with acetic orcein.

RESULTS AND DISCUSSION

Taxonomy

The stalked diatom that we find on *Corycaeus affinis* in the Puget Sound region has a form and structure that without doubt allies it with the genus *Hormophora* Jurilj (1957). Our diatom is distinct from the two Adriatic species in size and seems to represent an undescribed species in this genus. The name *Hormophora* Jurilj, however, is invalid because it is a later homonym of *Hormophora* J. G. Agardh (1892), a genus of red algae from Australia currently assigned to the Kallymeniaceae. We propose the new name, *Sameioneis,* for this diatom genus.

Sameioneis nom. nov.

Gr. σημειον, banners; νεώς, ship)

Basionyms

Homophora Jurilj, 1957, Acta Bot. Cro., vol. 16, p. 96, fig. 2

Sameioneis rogallii (Jurilj) comb. nov.

Basionym

Hormophora zavodnikia Jurilj 1957, Acta Bot. Cro., vol. 16, p. 98, fig. 3

Type Species

Sameioneis rogallii (Jurilj) comb. nov.

Sameioneis carinaes sp. nov.

(Latin *carina,* a keel; *aes,* copper)

Fig. 1*a*

Cellulae aspectu sicut lunares. Valvae 28–70 μ long., 9–15 μ lat. Utraque valva duas raphes breves, longitudine inaequas, una utraque in extremitate, e 5–9 poris parvis (0.5 μ) distinctis in ordine sitis constantes habens. Raphes prope utramque extremitatem ad marginem valvae ventralem oriuntur, et versus apicem dorsaliter curvant. Utraque extremitas cellulae raphem et longam et brevem habet, omnibus quattuor, cellula a ventre visa, visibilibus. Noduli nulli. Valvae punctis parvis vix visibilibus (ca. 2 in omni micro) quae in ordinibus ad pseudoraphem, in linea cellulae media sitam, perpendiculariter ordinatis disponuntur tectae. Puncta in ordinibus longitudinalibus non disposita, in ordinatione, autem, velut fractiflexam interruptam efficientia. Pseudoraphe poro dorsali raphis ad unam valvae extremitatem connexa, ad alteram extremitatem ad marginem dorsalem accedit, deinde abrupte curvat et ad porum ventralem raphis desinit. Pseudoraphe in altera valva eadem est, inversa, autem. Axis apicalis in plana valvari curvatus, heteropolaris, una extremitate paululo angustiore quam altera. Axis pervalvaris in plana transapicali curvatus, heteropolaris, raphibus pseudoraphibusque non aequis symmetricisve. Axis cellulae isopolaris nullus. Chromatophora flavo-virentia ad brunnea ellipsoidea, in cellula laxe distributa, omni unam pyrenoidem refractivam habente. Cellulae substrato cauliculis flexibilibus ramosis liquidis affixae. Cellulae in copopodo pelagico marino *Corycaeo affini,* in loco Puget Sound, Washington dicto, epibioticae.

Valves are crescent-shaped (Fig. 1*a*), 28–70 μ long by 9–15 μ wide. Two short raphes of unequal lengths are at each end of each valve (Figs. 1*d*, 2*b*). Raphes consist of 5–9 small pores (0.5 μ diameter) in a row and extend from the ventral edge of the valve in a dorsal curve toward the valve apex (Figs. 1*a*, 2*a*). The valves at each end of the cell have a long and a short raphe and all four raphes are visible from the ventral side of the cell. Nodules are not present. The valves are covered with small, almost invisible, puncta (approximately 2 per micron) that are arranged in rows perpendicular to a pseudoraphe that lies along the midline of the valve (Figs. 1*a*, 2*a*). The puncta are not in distinct longitudinal rows but give the appear-

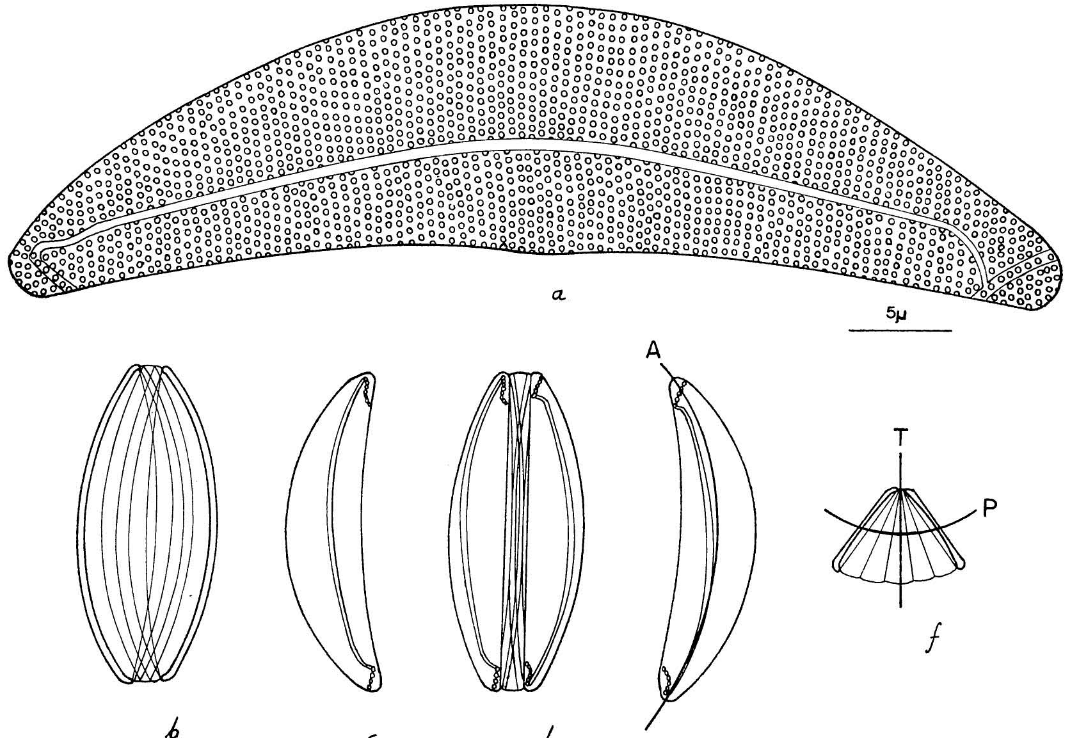


FIG. 1. *Sameioneis carinaes* gen. et sp. nov. *a,* Valve view showing puncta; *b,* dorsal girdle view; *c,* valve view; *d,* ventral girdle view; *e,* opposing valve view; A, apical axis; *f,* apical view; T, transapical axis; P, pervalvar axis.

ance of an interrupted zigzag pattern. The pseudoraphe connects to the dorsal pore of the raphe at one end of the valve, traverses the valve and curves abruptly, ending at the ventral pore of the other raphe. The pseudoraphe on the other valve of the cell is identical in form, but reversed in position (Fig. 1*c, d, e*).

The apical axis is curved in the valvar plane, and is heteropolar, one end usually being narrower than the other (Fig. 1*c, e*). The transapical axis is straight (Fig. 1*f*) but heteropolar because the dorsal girdle contains bands that overlap at angles near the apices and become narrower on the ventral girdle making the cell wedge-shaped (Fig. 1*b, d, f*). The pervalvar axis is curved in the transapical plane (Fig. 1*f*) and is heteropolar because the raphes and pseudoraphes of the valves are not equal or symmetric. Cells are attached to the substratum by clear, branching flexible stalks that usually have septa (Figs. 2*c*, 3*b*).

Many ellipsoidal, loosely packed, light yellow-green to brown chromatophores line the valves (Figs. 2*c;* 3*a, b*). Each chromatophore has a refractive pyrenoid on the interior surface.

Sameioneis carinaes is found epizooic only on the marine pelagic copepod, *Corycaeus affinis.* It has never been found attached to any other object.

Type Locality

Type locality is Puget Sound, near Edmonds, Washington.

Ecology

Seasonal occurrence: *Corycaeus affinis* was not present in the plankton samples taken between April and August. On August 21 a sample made in Speiden Channel contained *Corycaeus affinis,* but there were no diatoms attached to the animals. All subsequent samples, taken during the autumn season, were from Puget Sound

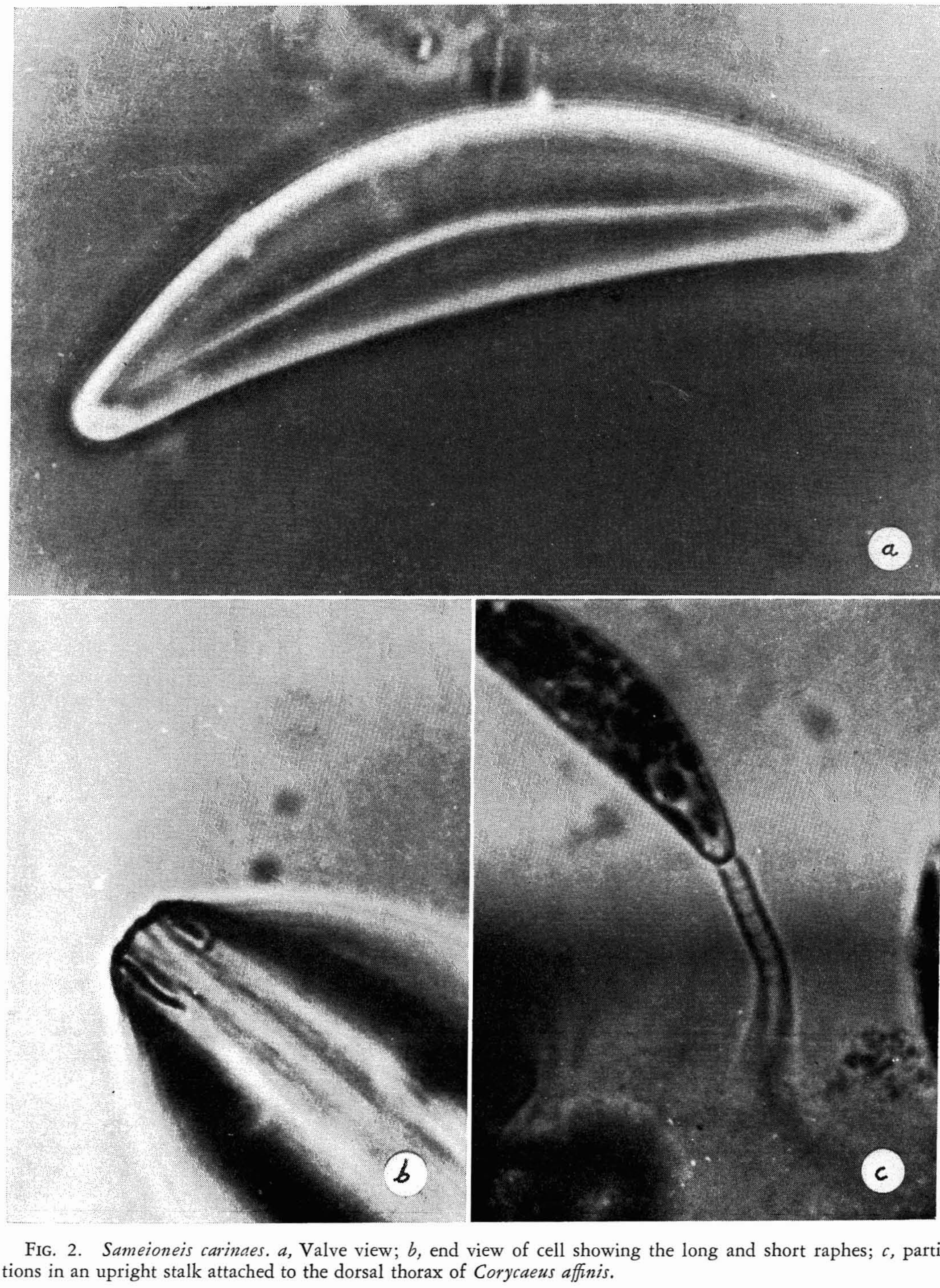


FIG. 2. *Sameioneis carinaes*. *a*, Valve view; *b*, end view of cell showing the long and short raphes; *c*, partitions in an upright stalk attached to the dorsal thorax of *Corycaeus affinis*.

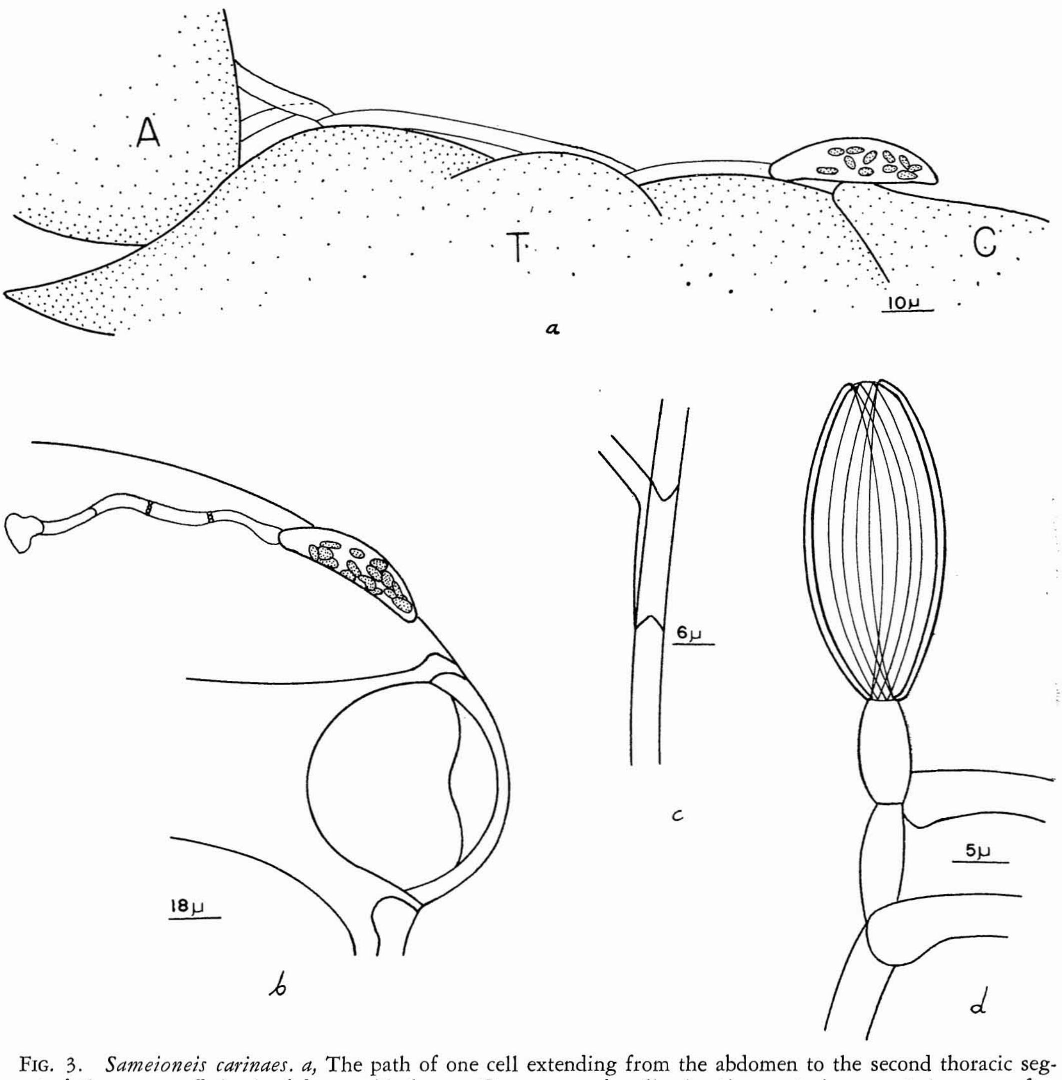


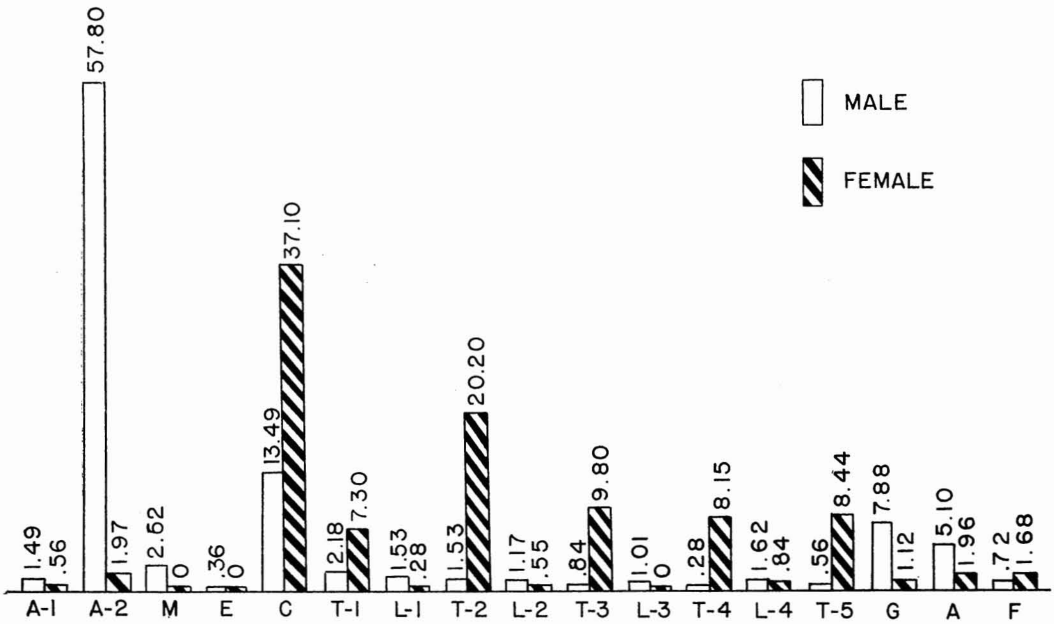
FIG. 3. *Sameioneis carinaes. a,* The path of one cell extending from the abdomen to the second thoracic segment of *Corycaeus affinis;* A, abdomen; T, thorax; C, carapace; *b,* cell migrating posterior to anterior soon after its attachment to the carapace of *Corycaeus affinis;* c, partitions found in the stalk after cell division; *d,* partitions found in the stalk after two cell divisions.

off Edmonds, and all contained *Corycaeus affinis* with *Sameioneis* attached. The percentage of copepods infected with *Sameioneis* increased from 55 percent in September to as high as 82 percent in October. Our records of the seasonal occurrence of *Corycaeus affinis* in the Puget Sound region corresponds with the data of Hebard (1956).

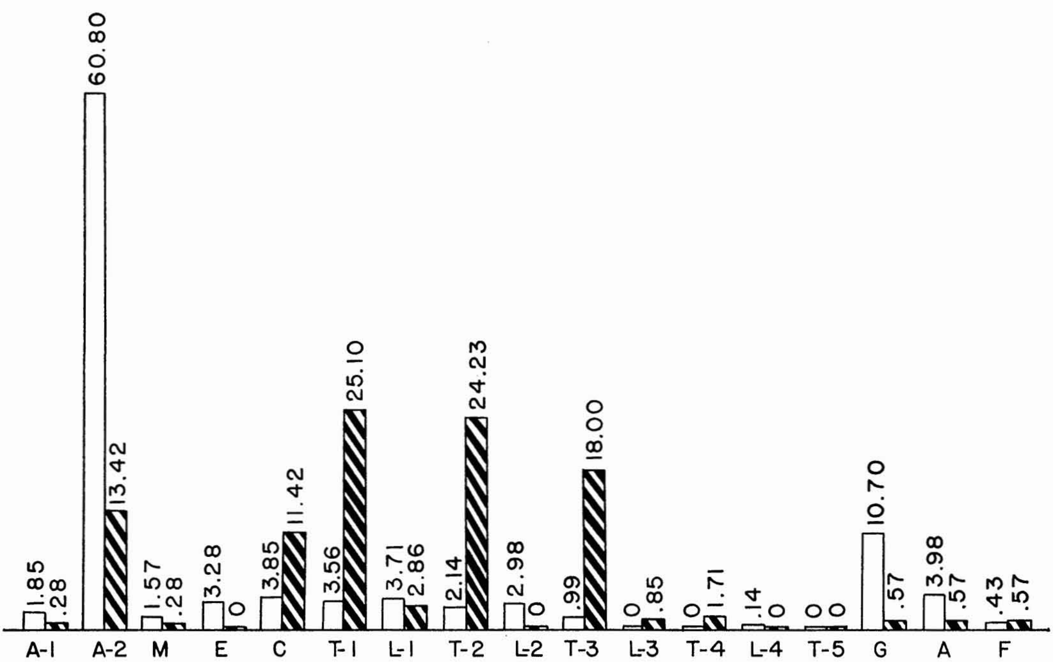
Distribution on the Host

Table 1 summarizes the information in the literature on the distribution of stalked diatoms on species of *Corycaeus.*

Male animals were consistently more abundant than females throughout the sampling period (85 percent male, 15 percent female). Contrary to Fruchtl's report (1924) showing male infections much higher than female infections with *Sameioneis* in the Adriatic Sea, the male copepods in Puget Sound carried only about 1.4 diatoms more per animal than the females (Table 2).


MALE
FEMALE
A-1 1.49 .56
A-2 57.80 1.97
M 2.62 0
E .36 0
C 13.49 37.10
T-1 2.18 7.30
L-1 1.53 .28
T-2 1.53 20.20
L-2 1.17 .55
T-3 .84 9.80
L-3 1.01 0
T-4 .28 8.15
L-4 1.62 .84
T-5 .56 8.44
G 7.88 1.12
A 5.10 1.96
F .72 1.68

%
4A


A-1 1.85 .28
A-2 60.80 13.42
M 1.57 .28
E 3.28 0
C 3.85 11.42
T-1 3.56 25.10
L-1 3.71 2.86
T-2 2.14 24.23
L-2 2.98 0
T-3 .99 18.00
L-3 0 .85
T-4 0 1.71
L-4 .14 0
T-5 0 0
G 10.70 .57
A 3.98 .57
F .43 .57

%
4B

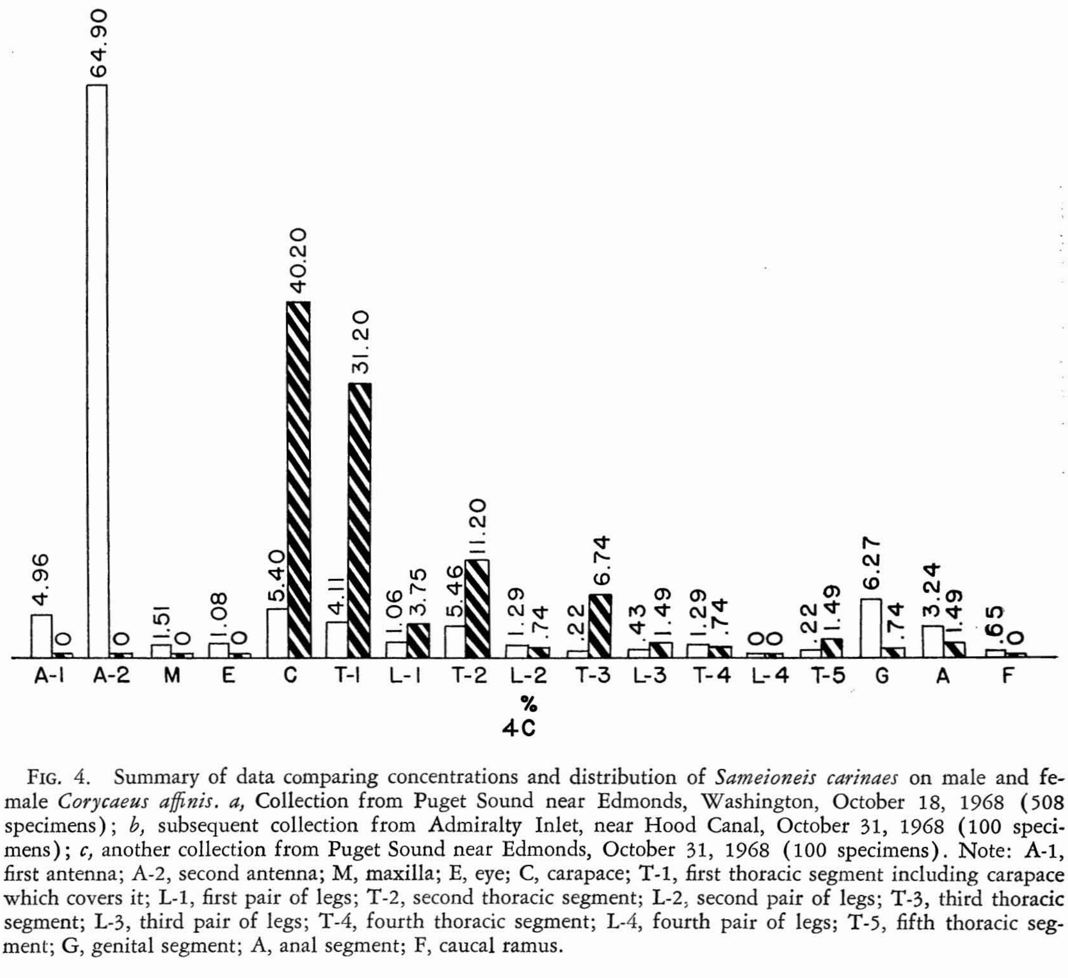


FIG. 4. Summary of data comparing concentrations and distribution of *Sameioneis carinaes* on male and female *Corycaeus affinis*. *a,* Collection from Puget Sound near Edmonds, Washington, October 18, 1968 (508 specimens); *b,* subsequent collection from Admiralty Inlet, near Hood Canal, October 31, 1968 (100 specimens); *c,* another collection from Puget Sound near Edmonds, October 31, 1968 (100 specimens). Note: A-1, first antenna; A-2, second antenna; M, maxilla; E, eye; C, carapace; T-1, first thoracic segment including carapace which covers it; L-1, first pair of legs; T-2, second thoracic segment; L-2, second pair of legs; T-3, third thoracic segment; L-3, third pair of legs; T-4, fourth thoracic segment; L-4, fourth pair of legs; T-5, fifth thoracic segment; G, genital segment; A, anal segment; F, caucal ramus.

It was easily recognizable from our collection of October 18 that male animals have a different distribution of *Sameioneis* than do the female animals. A percentage comparison between male and female infections showed a much higher concentration of diatoms on the second antennae of the male than on the second antennae of the female. The females have much higher concentrations of diatoms on the carapace and thoracic segments than do the males. The data obtained from this and two subsequent collections were similar. This information is summarized in Figure 4. The distribution of diatoms on *C. affinis* copepodites was similar to that of the females.

Corycaeus affinis was the host for several other epizooic organisms besides *Sameioneis carinaes,* although none of the others were as abundant as this diatom. *Synedra investiens* W. Smith and an undetermined species of *Synedra, Cymbella pusilla* Grun, *Cymbella* sp., *Cocconeis scutellum* Ehr., and *C. scutellum* var. *staureiformis* W. Smith, as well as a brown algal filament and isolated brown algal cells, were observed on *Corycaeus affinis.* The most common protozoans on *C. affinis* were *Vorticella,* clumps of monads, and cysts of an apostome ciliate.

Two factors are most important in an attempt to analyze the reasons for the particular distribution of diatoms on the animals: (1) the diatom's means of attachments by a mucilaginous stalk, and (2) habits of the animals that may

TABLE 1

DISTRIBUTION OF STALKED DIATOMS ON *Corycaeus* SPP., AS OBSERVED BY EARLIER AUTHORS

AUTHOR	*Corycaeus* SPP.	DIATOM DIST.	LOCATION
Jurilj (1957)	*Corycaeus* sp.	ventral	Adriatic Sea
Früchtl (1924)	*C. gibbulus*	2nd antenna tail	Adriatic Sea
Klevenhusen (1933)	*C. speciosus*	furca, tail	S. Atlantic
Klevenhusen (1933)	*C. gracilis*	furca, tail	S. Atlantic
Klevenhusen (1933)	*C. latus*	furca, tail	S. Atlantic
Klevenhusen (1933)	*C. lautus*	furca, tail	S. Atlantic
Klevenhusen (1933)	*C. flaccus*	furca, tail	S. Atlantic
Giesbrecht (1892)	*C. elongatus*	thorax, tail	Adriatic Sea

TABLE 2

NUMBER OF *Sameioneis* CELLS ON MALE AND FEMALE *Corycaeus affinis* COLLECTED FROM PUGET SOUND NEAR EDMONDS, WASHINGTON, OCTOBER 18, 1968

SEX	TOTAL *C.*	TOTAL *C.* WITH *S.*	PERCENT C. WITH S.	TOTAL *S.*	*S.* / EACH *C.*
Male	431	286	66.5	2479	8.65
Female	76	49	64.5	356	7.25

be responsible for transferring diatoms from one animal to another.

Description of Stalk

Sameioneis cells are attached to the copepods by clear, flexible stalks similar to those of *Gomphonema* sp., and may consist of polysaccharides (Fee and Drum, 1965). The stalks may be upright or prostrate and vary in length from a rudiment to almost the full length of *Corycaeus affinis* (1 mm). The stalk diameters average 3.5 μ and correspond to the distance between the raphes on each valve (3.6 μ) at the ends of the cells. The mucilage is probably secreted through these raphes.

Periodic changes in the structure and density of the stalk material produced by *Sameioneis* form partitions in the stalk (Figs. 2*c*, 3*b*). An adhesive mucilage occurs at the partitions. Although partitions may be formed at almost any time, they seem to be regularly associated with division of cells. Before division a partition is formed, and during the division process one of the mother cell valves begins stalk formation on the outer surface of the original stalk. Upon completion of cell division, another partition is made on the original stalk and the new stalk with its daughter cell branches from it (Figs. 3*c, d*).

Although actual movement of *Sameioneis* cells was not observed, a change in their position on the animal was indicated by the paths of prostrate stalk material. Such a path of one diatom extended from the abdomen of the animal to its second thoracic segment (Fig. 3*a*). At this point the diatom appeared to be bridging the gap between the thorax and the carapace. This was a feat considerably easier than the one it had already accomplished when, as indicated by its stalk, it crossed a larger gap between the thorax and the abdomen—an area of considerable animal movement. Other diatoms showed short prostrate migrations before assuming upright positions. Most diatoms migrated from the anterior part of the animal toward the posterior, but a few went in the opposite direction against the current caused by the swimming copepod. In migrating diatoms the chromatophores are located at the forward end of the cell.

The diatom secretes a mucilage which seems to emerge as a watery sticky material and soon becomes the firm mucilage of the stalk. As a result, it is possible for the cells to become separated from the stalk at this place, the free

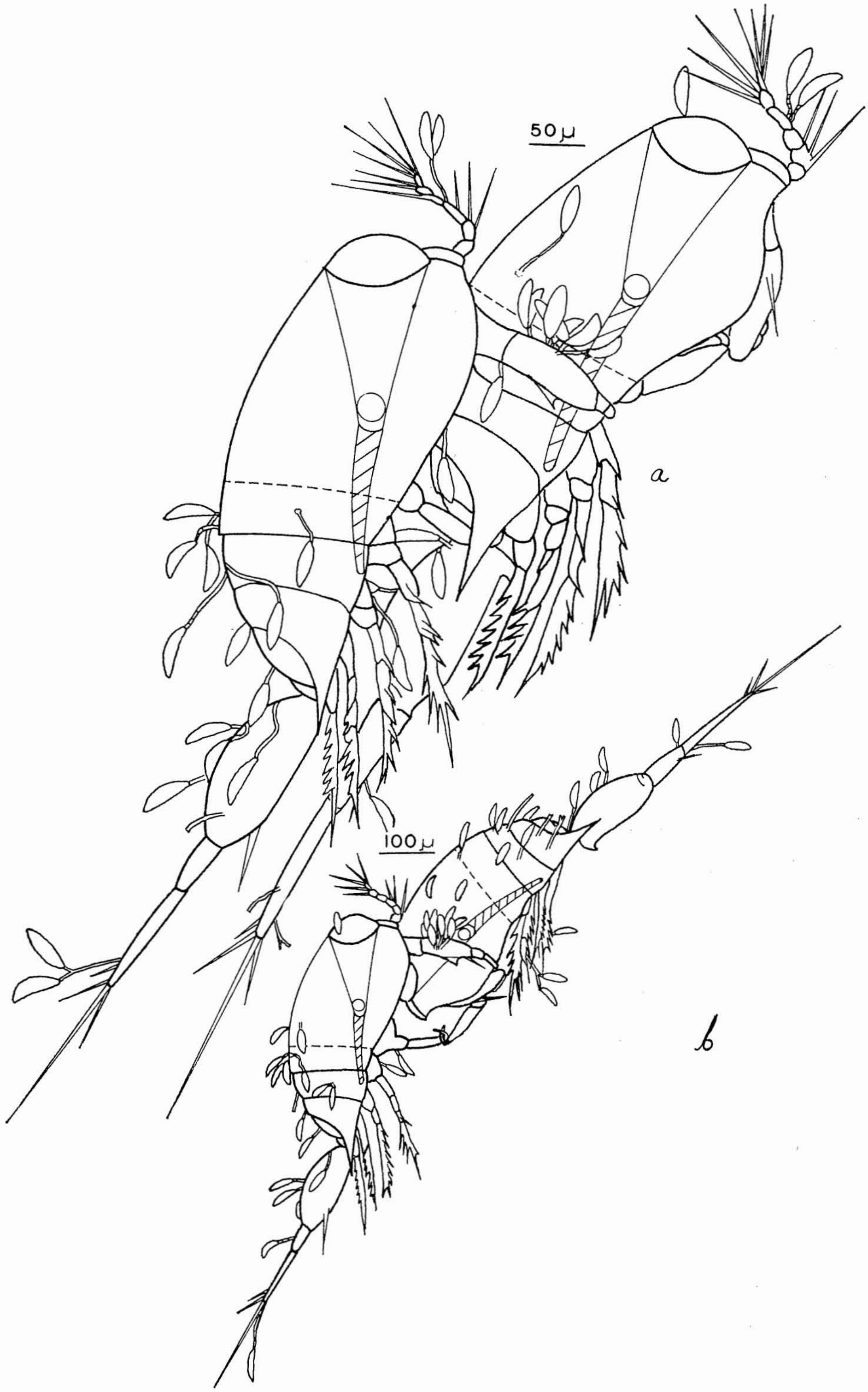


FIG. 5. *Corycaeus affinis*. *a*, Male clasping female in copulation; *b*, noncopulatory clasping, male clasping female.

cell carrying with it the sticky mucilage that undoubtedly is important in reattaching the cell. Some *Sameioneis* cells secrete an abundance of this watery mucilage which fans out into the water from their unattached ends. It seems likely that this mucilage increases the chances of a cell being transferred to another animal.

Habits of Corycaeus affinis

In copulation, the male animal clasps the female around the first and second thoracic segments with the movable setae of his second antennae. At the same time he clasps the female between the abdomen and thorax with the setae of his maxillipeds (Fig. 5*a*). Male copepods sometimes clasp other males in the same manner.

Male copepods were frequently observed swimming rapidly with another copepod of the same species, either male or female, in a noncopulatory clasping position (Fig. 5*b*). In this position the male hooks his second antennae around his captive's carapace directly behind the eyes, and the setae of his maxillipeds hook into the corresponding setae of the trapped copepod. The females do not appear to clasp other members of their species.

Transfer of *Sameioneis* from one copepod to another probably occurs during contact of two animals. At this time the cell may be dislodged and the sticky mucilage that is usually present at one or both ends of the diatom cell may attach it to the other animal. After it is attached it may form an upright stalk or it may migrate on the animal by means of a prostrate stalk.

The main points of contact of the copepods during copulatory and noncopulatory clasping all show high concentrations of diatoms (Figs. 4, 5*a*, 5*b*). In the males the main claspers are the second antennae. In the females the main points of contact with males are the first and second thoracic segments and the carapace (the latter area is also in contact during noncopulatory clasping). Male copepods may also have numerous diatoms attached to their first and second thoracic segments, probably because of homosexual mating attempts. Males also show concentrations of diatoms on their genital and anal segments, whereas, on the females, the spermatophores and egg masses in these areas probably interfere with diatom attachment. Diatom migration and inadvertent contacts between copepods probably account for the limited distribution of diatoms on other parts of the copepods.

Clasping of copepodites by mature *Corycaeus affinis* was not observed, but this behavior probably does occur because *Sameioneis* was found on the copepodites.

As Klevenhusen (1933) hypothesized, it is possible that the added surface area *Sameioneis* provides may aid in suspending the copepods in the water. On the other hand, the diatoms may also hinder action of the swimming legs of *Corycaeus* and be a distinct disadvantage. There is no indication that the diatoms are a food for this predaceous animal.

Means of maintenance of the diatom population throughout the year on this seasonally appearing copepod remains unknown. It is possible that a few infected animals remain in the plankton the year around but were too few to be found in our sampling. Also, it seems possible that the infected animals may migrate with water currents along the Pacific Coast of North America, appearing in our region only during the autumn season.

Although our evidence from cultures and field observations suggests that *Sameioneis carinaes* does not grow except on *Corycaeus affinis,* it remains possible that the diatom may live in a benthic habitat during at least part of the year. The fact that *Corycaeus affinis* often has other diatoms attached to it that are also known in benthic habitats lends support to this hypothesis.

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