RAPID COMMUNICATIONS / COMMUNICATIONS RAPIDES

First observations of tumor-like abnormalities (exophytic lesions) on Lake Michigan zooplankton

M. Omair, Henry A. Vanderploeg, David J. Jude, and Gary L. Fahnenstiel

Abstract: Tumor-like abnormalities (exophytic lesions) were found on a variety of planktonic calanoid copepods and cladocerans (*Diaptomus* spp., *Epischura lacustris*, *Limnocalanus macrurus*, *Polyphemus pediculus*, *Diaphanosoma* sp., and *Daphnia galeata mendotae*) collected from inshore (3-m depth contour) and offshore (100- to 110-m depth contours) stations of eastern Lake Michigan. The abnormalities, which were quite large relative to animal size and variable in shape, are documented in photographs. Abnormality incidences among species ranged between 0 and 72%. Predatory species of calanoids and cladocerans had higher incidences of tumors than herbivorous species. The abnormalities on some copepods were very similar to cysts described for calanoid copepods in Lago Maggiore, Italy, which like Lake Michigan is undergoing oligotrophication. The recent appearance of the lesions in Europe and North America may indicate an emerging global phenomenon that has a common cause.

Résumé : Des anomalies de type tumoral (lésions exophytiques) ont été observées chez divers copépodes calanoïdes et cladocères planctoniques (*Diaptomus* spp., *Epischura lacustris*, *Limnocalanus macrurus*, *Polyphemus pediculus*, *Diaphanosoma* sp. et *Daphnia galeata mendotae*) capturés à des stations du littoral (isobathe de 3 m) et du large (isobathes de 100 m et 110 m) dans l'est du lac Michigan. Les anomalies, qui étaient plutôt grosses par rapport à la taille des animaux et de taille variable, ont été photographiées. L'incidence de ces anomalies, d'une espèce à l'autre, allait de 0 % à 72 %. Les espèces prédatrices présentaient une plus forte incidence de tumeurs que les espèces herbivores. Les tumeurs présentes chez certains copépodes étaient très semblables aux kystes décrits chez des copépodes calanoïdes du lac Majeur, en Italie, qui comme le lac Michigan est en voie d'oligotrophisation. La récente apparition des lésions en Europe et en Amérique du Nord peut révéler la présence d'un phénomène qui se développe à l'échelle mondiale et dont la cause est la même partout.

[Traduit par la Rédaction]

Introduction

We found tumor-like abnormalities (exophytic lesions) on *Limnocalanus macrurus*, a large predacious copepod that is a dominant of the hypolimnion of offshore Lake Michigan (Vanderploeg et al. 1998), in the first archived sample that we examined from collections made during 1995–1998 in the food web and water-quality monitoring program of the Great Lakes Environmental Research Laboratory (GLERL). Subsequently, we found a high incidence of abnormalities in a variety of nearshore species in zooplankton samples collected by the Center for Great Lakes and Aquatic Sciences (CGLAS), University of Michigan, from the nearshore zone.

Despite extensive collections of zooplankton from all the Great Lakes and intensive collections in Lake Michigan, no such lesions have been reported for North American freshwaters. In fact, very few authors have claimed to have ever seen tumors or tumor-like abnormalities in zooplankton in the oceans or lakes. Crisafi and Crescenti (1975, 1977) found tumor-like anomalies in 40 species of copepods from

Received April 27, 1999. Accepted July 30, 1999.
J15115

M. Omair.[1] Center for Great Lakes and Aquatic Sciences, 2021 C.C. Little Building, University of Michigan, 501 East University Avenue, Ann Arbor, MI 48109-1090, U.S.A.
H.A. Vanderploeg. National Oceanic and Atmospheric Administration, Great Lakes Environmental Research Laboratory, 2205 Commonwealth Boulevard, Ann Arbor, MI 48105, U.S.A.
D.J. Jude. Center for Great Lakes and Aquatic Sciences, 717 Dennison Building, University of Michigan, 501 East University Avenue, Ann Arbor, MI 48109-1090, U.S.A.
G.L. Fahnenstiel. National Oceanic and Atmospheric Administration, Great Lakes Environmental Research Laboratory, Lake Michigan Field Station, 1431 Beach Street, Muskegon, MI 49441-1098, U.S.A.

[1]Author to whom all correspondence should be addressed. e-mail: momair@umich.edu

polluted seas of the world. Silina and Khudolei (1994) reported tumor-like anomalies on zooplankton, but no photographs or descriptions of the anomalies on the zooplankton from a polluted area of the Baltic were ever presented. Here, we document incidences of tumors in a preliminary study of samples from 1995 to 1999 and present photographs of the abnormalities to encourage zooplankton ecologists to examine their samples for such abnormalities to determine geographical distribution of the lesions and stimulate research as to what is causing the lesions. Moreover, we show that some of the lesions bear a striking resemblance to cysts reported by Manca et al. (1996) in their study of epibionts on calanoid copepods in Lago Maggiore, an Italian lake undergoing oligotrophication.

Methods

Zooplankton in GLERL's offshore Lake Michigan monitoring program were collected with a 0.5-m-diameter, 153-mm-mesh net towed vertically from 95 m to the surface. One sample each from 22 June 1995 at a 110-m-deep site 15 km west of Muskegon, Michigan, and from 25 June 1998 at a 100-m-deep site 21 km west of Grand Haven, Michigan, were examined. Zooplankton samples were narcotized with club soda and preserved in a sugar formaldehyde solution (Haney and Hall 1973).

Nearshore zooplankton samples were collected as part of a yellow perch (*Perca flavescens*) recruitment study at 3-m depth contours near Muskegon, Michigan. A push net (0.5-m diameter, 363-mm-mesh net) mounted in front of a small boat and a 0.75-m-diameter, 363-mm-mesh net towed behind the boat from near bottom to the surface were used to collect larval fish and zooplankton. We report on samples preserved in 70% ethyl alcohol that were collected on 7, 8, and 19 June 1998.

To count the preserved samples, we split the samples with a Folsom plankton splitter with the goal of examining about 1000 individual zooplankton. For some of the rare taxa (nearshore cladocerans), we examined the entire sample to observe as many individuals as possible. Results of counts at all dates were combined so that we could report an overall lesion frequency for nearshore samples collected in June 1998.

Results

Appearance of abnormalities

The abnormalities on *Limnocalanus* were very prominent and found on the prosome (Figs. 1*a* and 1*b*) or occasionally on the urosome. Some large abnormalities were highly multilobed (Fig. 1*a*) or more typically large and almost spherical, attached by a stalk-like structure to *Limnocalanus* (Fig. 1*b*). Adult copepods affected were young adults because by June, the old generation had disappeared and the new generation was appearing (Vanderploeg et al. 1998). This is also borne out by the lack of infestation by the suctorian epibiont *Tokophrya quadripartita*, which infests older adults (Vanderploeg et al. 1998). The large lesions on young (recently molted) adults may imply especially rapid growth of the abnormalities if we assume that they appeared after molting. Abnormalities on *Epischura lacustris*, although not as large relative to body size as those on *Limnocalanus*, were quite prominent. Abnormalities on *Diaptomus* (Figs. 1*c* and 1*d*) were variable in size and some were almost as large relative to body size as those seen on *Limnocalanus*. Abnormalities on three species of cladocerans, *Polyphemus pediculus*, *Diaphanosoma* sp., and *Daphnia galeata mendotae*, were quite prominent and found in a variety of locations on the carapace and even on the abdominal claw (Figs. 1*e*, 1*f*, and 1*g*).

Frequency of infestation

We found abnormalities at appreciable frequencies in a variety of copepods (*Epischura lacustris*, *Eurytemora affinis*, and *Diaptomus* spp.) and *Polyphemus* (Table 1) in the nearshore sample collected at Muskegon. Percent occurrence was quite high, especially for *Epischura* (71.4%). Although we found abnormalities on *Limnocalanus* in the June 1995 offshore sample, we found none in the offshore sample collected in June 1998. Interestingly, no abnormalities were found on any species other than *Limnocalanus* at offshore sites. No abnormalities were found on *Cyclops* spp. at any site.

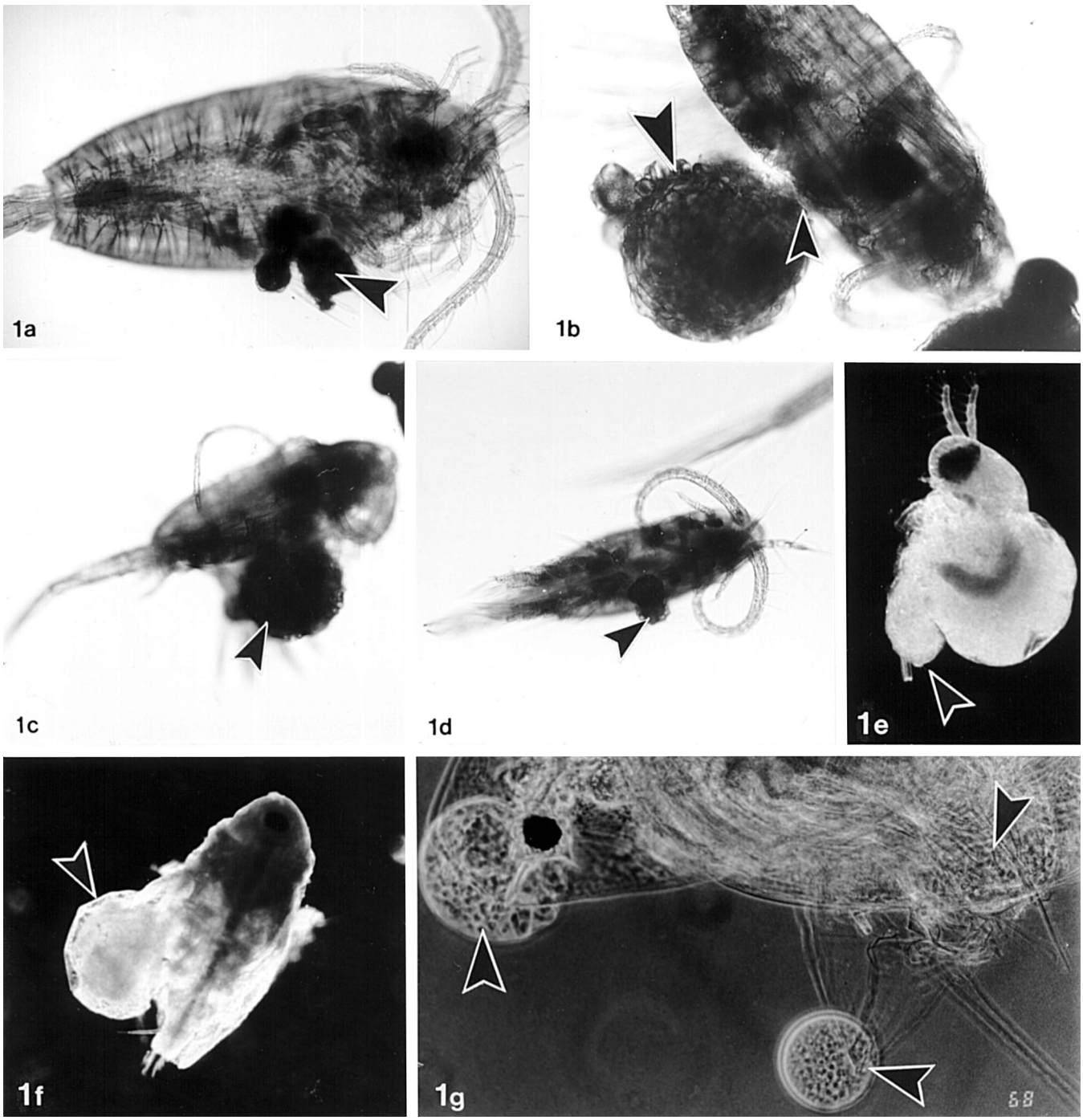
Affected zooplankton species showed a variety of taxonomic diversity (both cladocerans and copepods), habitats, feeding habits, and life histories (Table 2). That *Epischura* and *Polyphemus* had the highest frequency of infestation in the nearshore zone and *Limnocalanus* was the only species affected in the offshore zone suggests that predatory calanoid copepods and cladocerans are more likely to be affected than herbivorous or omnivorous species. A preliminary examination of samples from 1999 showed that abnormalities were present at a variety of sites in the southern basin of Lake Michigan as well as Green Bay, where abnormalities were found on *Diaptomus* spp.

Discussion

A tumor, in general terms, is a swelling or protuberance; however, in medical terms, it implies a neoplasm, which implies growth of cells independent of the normal growth of the organism (Sparks 1972). Verification of a neoplasm can only be done at the cellular level, which we have not done here. Therefore, we have used the conservative description of tumor-like abnormality or exophytic lesion. Tumors and tumor-like abnormalities have been found in a variety of invertebrates: molluscs, arthropods (including insects and shrimp), flatworms, and cnidarians (Sparks 1972; Sparks and Lightner 1973; Overstreet and Van Devender 1978; Harshbarger 1997). However, reports of such abnormalities on crustaceans are rare and their causes and consequences to the affected organisms are obscure (Sparks and Lightner 1973; Overstreet and Van Devender 1978). Sparks and Lightner (1973) reported a tumor-like papilliform growth on the sixth abdominal segment of a brown shrimp that was tentatively diagnosed as a benign neoplasm. Overstreet and Van Devender (1978) reported an overgrowth (hamartoma) of muscle protruding through the sixth abdominal segment of postlarvae of two species of commercial shrimps. This nonneoplastic growth was thought to be an interaction between a pollutant and the normal growth process of the shrimp because the affected shrimp were found at a polluted site.

As far as zooplankton are concerned, Crisafi and Crescenti (1975, 1977) reported finding tumor-like anomalies in 40 species of copepods from polluted seas of the world. No frequencies of infestation or histological descrip-

Fig. 1. Appearance of tumor-like abnormalities (arrowheads) on zooplankton. (*a*) *Limnocalanus* (prosome length = 1.7 mm) with a large multilobed tumor on the ventral surface of the prosome between the swimming feet and mouthparts; (*b*) dorsal view of *Limnocalanus* (prosome length = 1.7 mm) with a large, nearly spherical abnormality (large arrowhead) attached by a stalk-like structure (small arrowhead) to the right side of the prosome; (*c* and *d*) tumor-like abnormalities on the ventral prosome of *Diaptomus* spp. (prosome length = 0.7 mm); (*e*) *Polyphemus pediculus* (body length = 0.7 mm); (*f*) dorsal view of *Diaphanosoma* sp. (body length = 0.9 mm); (*g*) *Daphnia galeata mendotae* (body length = 1.1 mm) with abnormalities on the helmet near the compound eye, abdominal claw, and carapace of the body.



tions were given. Some of the abnormalities that Crisafi and Crescenti (1975) photographed bear at least a superficial resemblance to the lesions that we describe. Silina and Khudolei (1994) found abnormalities on nauplii of cyclopoids and *Limnocalanus* in the Gulf of Finland (Baltic Sea) and nearby lakes. No descriptions of the gross anatomy or histology were reported. Frequencies of abnormalities were as high as 42–80% in a few samples from the southwest part of Neva Inlet, a polluted section of the Gulf of Finland. Further, Silina and Khudolei (1994) suggested that the con-

Table 1. Frequency of occurrence of tumor-like abnormalities among zooplankton species at offshore (100-m Grand Haven site in 1995 and 110-m Muskegon site in 1998) and inshore (3-m Muskegon site) sites in June 1995 and 1998 in eastern Lake Michigan.

	Offshore 1995		Offshore 1998		Inshore 1998	
Taxon	%	*N*	%	*N*	%	*N*
Cyclopoida						
Cyclops spp. copepodids and adults	0	214	0	237	0	49
Calanoida						
Diaptomus spp. copepodids and adults	0	602	0	798	9.3	236
Epischura lacustis copepodids and adults	—	0	0	12	71.4	899
Eurytemora affinis copepodids	—	0	—	0	4.8	41
Limnocalanus macrurus copepodids and adults	10	300	0	55	0	1
Cladocera						
Daphnia galeata mendotae	—	0	0	30	6.2	16
Diaphanosoma sp.	—	0	—	0	16.7	6
Polyphemus pediculus	—	0	—	0	33.3	3

Table 2. Ecology and life history traits of tumor-infested species.

Taxon	Habitat	Feeding habits	Life history
Copepoda			
Cyclops spp.	Inshore and offshore	Predators of microzooplankton and small crustaceans	Three generations per year?
Diaptomus spp.	Inshore and offshore	Omnivores with strong herbivorous tendencies	One or two generations per year; two of four species produce resting eggs
Epischura lacustris	Inshore and epilimnion	Omnivore, primarily predator of microzooplankton	One or two generations per year; resting eggs
Limnocalanus macrurus	Offshore hypolimnion	Omnivore, primarily predator of small crustaceans	One generation per year; no resting eggs
Cladocera			
Daphnia galeata mendotae	Inshore and offshore	Filter-feeder	Parthenogenic during summer; resting eggs
Diaphanosoma spp.	Inshore	Filter-feeder	Parthenogenic during summer; resting eggs
Polyphemus pediculus	Inshore	Predator of microzooplankton and small cladocerans	Two generations per year; parthenogenic during summer; resting eggs

Note: Accounts generalized from Balcer et al. (1984) and Vanderploeg et al. (1998).

centrations of cancer-causing chemicals such as polycyclic aromatic hydrocarbons and *N*-nitroso compounds as well as blue-green algal toxins, suspected carcinogens, should be investigated to see if they were the causal agents.

The spherical lesions on *Diaptomus* and *Limnocalanus* bear a striking resemblance to "cysts" in a paper describing epibionts on copepods in Lago Maggiore, a large Italian subalpine lake (Manca et al. 1996). Especially noteworthy is the similarity of the stalk-like attachment of the lesions in both studies. Up to 15% of the calanoid copepod *Eudiaptomus padanus* had cysts, and as appears to be the case from our limited data collected so far for Lake Michigan, the abnormalities were more common in summer. The cysts ranged between 35 and 140 mm with the average cyst being about 100 mm. The cysts were first observed in 1992 in this regularly monitored lake. Like Lake Michigan, Lago Maggiore is undergoing oligotrophication. Manca et al. (1996) speculated that the larger cysts might have had a negative impact on *Eudiaptomus padanus* because larger cysts were associated with a constriction of the body. Even if there was no direct harm to body tissues caused by the abnormalities, the large size of some of the Lake Michigan tumors would have interfered with swimming and feeding. Until we know more about the nature of the abnormalities and their effects on zooplankton fitness and health, we can only speculate as to what harm they are doing to the zooplankton and food web.

We suspect that occurrence of the abnormalities is a recent phenomenon in the Great Lakes, in general, and in Lake Michigan, in particular, because no abnormalities have been reported despite many monitoring and life history studies. Monthly zooplankton abundance was reported for the years 1971–1982 at nearshore sites out to the 40-m depth contour in a series of studies arising from the Cook Power Plant monitoring program in southeastern Lake Michigan (e.g., Evans 1986). No abnormalities were found on *Limnocalanus* in a lipid and life cycle study that carefully examined individual *Limnocalanus* monthly during 1987–1990 (Vanderploeg et al. 1998). Zooplankton abundance was also reported from the U.S. Environmental Protection Agency's (EPA) Lake Michigan Surveillance Plan for an extensive grid of offshore stations during the years 1983–1992. The U.S. Environmental Protection Agency collections were limited to the upper 20 m during spring and late summer; therefore, this program would have missed the nearshore zooplankton

as well as the deep-dwelling *Limnocalanus* during summer. No lesions were ever noticed (O. Johannsson, Great Lakes Laboratory for Fisheries and Aquatic Sciences, Burlington, Ont., personal communication) in the extensive biomonitoring program on Lake Ontario during 1981 to the present.

Our findings along with those of Manca et al. (1996) and Silina and Khudolei (1994) suggest the intriguing possibilities that the abnormalities are a recent global phenomenon and that there is a common cause. Carcinogenic chemicals, ultraviolet radiation, viral or bacterial infections, or injury can cause tumors or tumor-like anomalies (e.g., Harshbarger 1997).

Histological examination of the tissues and cells of the tumor-like abnormalities now in progress will be helpful for determining whether they are neoplastic and whether a viral or bacterial agent is responsible. Whether or not the abnormalities are neoplastic, are caused by an environmental carcinogen, or are the result of an infectious agent, the consequences to the food web could be significant. We hope that this communication encourages further work. Further information is required to determine the first occurrence of the lesions in Lake Michigan, define their spatial distribution and seasonal timing, and ascertain their etiology.

Acknowledgements

This is GLERL publication No. 1136 and CGLAS publication No. 606. This research was conducted and supported by GLERL through a contract to M. Omair. Michigan Sea Grant (grant R/GLF-45 to D.J. Jude) provided funds for collection of nearshore zooplankton. We thank T. Rizki for useful discussions on tumors in invertebrates and for taking the photographs used in Fig. 1. We thank S. Hensler for helping sort zooplankton for tumors, N. Andresen and M. Edlund for taking photographs and reviewing the manuscript, P. Landrum and two anonymous reviewers for reviewing the manuscript, and J. Reid for a literature search. We thank M. Manca for contacting us to inform us of her work on cysts and B. Belonger and J. Dettmers for sending us zooplankton with lesions from Green Bay.

References

Balcer, M.D., Korda, N.L., and Dodson, S.I. 1984. Zooplankton of the Great Lakes — a guide to the identification and ecology of the common crustacean species. University of Wisconsin Press, Madison, Wis.

Crisafi, P., and Crescenti, M. 1975. Coseguenze delle attivita umane sullo zooplancton del Mare di Taranto. Boll. Pesca Piscic. Idrobiol. **30**: 207–218.

Crisafi, P., and Crescenti, M. 1977. Confirmation of a certain correlation between polluted areas and tumorlike conditions as well as tumours growths in pelagic copepods provening from numerous seas of the world. Rapp. Comm. Int. Mer Medit. **24**(10): 155.

Evans, M.S. 1986. Recent declines in zooplankton populations in the inshore region of Lake Michigan: probable causes and consequences. Can. J. Fish. Aquat. Sci. **43**: 154–159.

Haney, J.F., and Hall, D.J. 1973. Sugar-coated *Daphnia*: a preservation technique for *Daphnia*. Limnol. Oceanogr. **18**: 331–333.

Harshbarger, J.C. 1997. Invertebrate and cold-blooded vertebrate oncology. *In* Spontaneous animal tumors: a survey. *Edited by* L. Rossi, R. Richardson, and J. Harshbarger. Press Point di Abbiategrasso, Milano, Italy. pp. 41–53.

Manca, M., Beltrami, M., and Sonvico, D. 1996. On the appearance of epibionts on the crustacean zooplankton of a large subalpine lake undergoing oligotrophication (L. Maggiore, Italy). Mem. Ist. Ital. Irobiol. **54**: 161–171.

Overstreet, R.M., and Van Devender, T. 1978. Implication of an environmentally induced hamartoma in commercial shrimps. J. Invertebr. Pathol. **31**: 234–238.

Silina, N.I., and Khudolei, V.V. 1994. Tumorlike anomalies in planktonic copepods. Hydrobiol. J. **30**: 52–55.

Sparks, A.K. 1972. Invertebrate pathology — noncommunicable diseases. Chap. VII. Academic Press, New York.

Sparks, A.K., and Lightner, D.V. 1973. A tumor-like papilliform growth in the brown shrimp *Penaeus aztecus*. J. Invertebr. Pathol. **22**: 203–212.

Vanderploeg, H.A., Cavaletto, J.F., Liebig, J.R., and Gardner, W.S. 1998. *Limnocalanus macrurus* (Copepoda: Calanoida) retains a marine arctic lipid and life cycle strategy in Lake Michigan. J. Plankton Res. **20**: 1581–1597.