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HATCHING OF EGGS OF *PONTELLA MEDITERRANEA* CLAUS (COPEPODA : CALANOIDA) (1)

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COPEPODE
PONTELLIDÉS
NEUSTON
OEUF AU REPOS

RÉSUMÉ. - Il est bien connu que *Pontella mediterranea* produit deux types d'œufs qui diffèrent par leurs morphologie et caractéristiques d'éclosion. Il a été démontré que les œufs lisses produits par les femelles à la fin de l'été éclosent simultanément à 21°C deux jours après la ponte, tandis que ceux de forme épineuse pondus en automne sont des œufs de diapause qui n'éclosent pas avant qu'un certain temps favorable se soit écoulé. Ces œufs au repos permettent le repeuplement des régions où l'espèce n'est pas pérenne. L'éclosion des œufs épineux n'est pas affectée par une incubation de plusieurs semaines à des températures de laboratoire variant de 4° à 16°C, mais est asynchrone après trois mois d'incubation à 18°C. Des œufs en diapause incubés dans les eaux de Woods Hole Harbor durant quatre mois ont éclos simultanément quand ils furent réchauffés à 18°C.

COPEPOD
PONTELLID
NEUSTON
RESTING EGGS

ABSTRACT. - *Pontella mediterranea* are known to produce two types of eggs which differ in morphology and hatching characteristics. It was demonstrated that the smooth eggs produced by females in late summer are subitaneous, because they hatch at 21°C within two days of being laid; while the spiny form laid in fall are diapause eggs which do not hatch until a suitable period of time has elapsed. These resting eggs permit the repopulation of areas where planktonic stages of the species cannot exist throughout the year. Spiny egg hatching was not affected by incubation for several weeks at reduced laboratory temperatures of from 4° to 16° C, but these eggs hatched non-synchronously after three months of incubation at 18° C. Diapause eggs incubated in Woods Hole Harbor waters for four months hatched synchronously when warmed to 18° C.

INTRODUCTION

The presence of dormant eggs in the life cycle of the calanoid copepod *Pontella mediterranea* in the Black Sea was suggested by Sazhina (1968). During summer months Sazhina observed that females of this species produced smooth eggs which hatched within two days, but towards the end of August and early September spiny eggs were produced that did not hatch over observational periods of up to two months. These spiny eggs were also observed in autumn and winter plankton samples from depths of 150-200 m. Planktonic stages of the species were absent in winter; nauplii first appeared in late May. Since the spiny eggs were not induced to hatch despite prolonged experiments and observations,

there was only circumstantial evidence for Sazhina's hypothesis that they represented an over-wintering mechanism for *P. mediterranea*, especially as diapause eggs had never been demonstrated for any marine copepod. Since that study, however, resting egg production has been shown to be a common phenomenon among marine calanoids, including species of Pontellidae (see Marcus, 1979 for a brief review of the literature).

We became interested in Sazhina's report of smooth and spiny eggs laid by *P. mediterranea* after studies with *P. meadi* (Grice and Gibson, 1977) and *Labidocera aestiva* (Grice and Gibson, 1975; Grice and Lawson, 1976) revealed that although both species in the western North Atlantic produced subitaneous and diapause eggs the two types were morphologically similar. *P. mediter-*

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ranea is common in the Mediterranean as well as the Black Sea (Zaitsev and Zelezinskaia, 1976), and from September to November of 1979 we investigated aspects of the reproductive biology, especially egg production and hatching, of this species from the waters off Cap Ferrat, France. We are indebted to Dr. Paul Bougis, Director of the Station Zoologique, Villefranche-sur-Mer for permitting us to use the facilities and to Dr. Robert Fenaux for arranging for laboratory space and collecting. We also thank Dr. Nancy Marcus for criticizing the manuscript.

METHODS

Neuston tows with a 333 μm net were made on 10 occasions from 13 September to 6 November 1979. During this time surface water temperatures varied between about 20 °C and 23 °C. Three other neuston tows were made on 3 and 4 January 1980 (water temperature 13.2 °C), but yielded no adult *P. mediterranea*. Large numbers of this species were obtained in all but two of the fall collections (17, 19 October) which were made following periods of intense rain and runoff. In the laboratory individual adult females were removed from the collections to 125-ml dishes containing sea water of comparable salinity to that where the adults were collected, fed *Artemia* nauplii, and placed at room temperature (18 to 23 °C) overnight. The following day the female was removed and eggs examined, counted and distributed at approximately 4° (dark), 10° (dark), 16 °C (12 hr. photoperiod) and room temperature for periods of 10 to 45 days in 15-cc and 75-cc jars (7 to 65 eggs per jar). The jars contained filtered (100 μm mesh) sea water and each had a screw type lid to prevent evaporation. The sea water was not changed during the incubation period. Although flagellates and, presumably, bacteria occurred in some jars, contamination appeared not to be significant. In a few cases eggs were used from females held for three days. To reach the lower incubation temperatures the eggs were gradually chilled over a two-day interval and at the end of the incubation period were slowly raised to 20° or to 22 °C. The jars were examined periodically and the number of hatched nauplii enumerated. Following these experiments at Villefranche, 43 sets of eggs from 25 females were returned to Woods Hole for further incubation and observation. In Woods Hole eggs were incubated at approximately 11°, 15°, 18 °C and room temperature (18°-21 °C). In addition, four sets of eggs from three females were placed beneath the Institution pier in Woods Hole Harbor as described previously (Grice and Gibson, 1975).

RESULTS

A single female generally laid either spiny or non-spiny eggs, although a few clutches from the same

individual contained both types. The eggs agreed with the description of Sazhina (1968) in size (spiny eggs, approximately 0.14 mm; non-spiny, 0.13-0.14 mm) and morphology. The smooth eggs or, rarely, eggs with small spines, hatched within two days at approximately 21 °C. These subitaneous eggs were produced by four of the females removed from collections obtained on 13 and 17 September. Other spawning females from these two collections, and over 90% (40 females) of the females isolated from 24 September through 6 November samples, produced spiny eggs which did not hatch in two days at about 21 °C. Of the approximately 2900 such eggs observed in Villefranche only four hatched in two days. Incubations at 4°, 10°, 16 °C for 10 to 45 days with slow increase of temperature to 20-22 °C resulted in no hatching of spiny eggs. Incubation at room temperatures (17.5-22 °C) for up to 45 days likewise resulted in no hatching.

Hatching of nauplii was observed from eggs (all spiny) in 16 of the 43 sets of eggs brought to Woods Hole. Eggs hatched in jars that were maintained in darkness as well as in light. Nauplii that hatched in 13 of these sets were incubated at 18 °C. Nauplii hatched in two sets incubated at 15 °C and one set that was incubated in Woods Hole Harbor. Considering the 15 sets of eggs incubated at laboratory temperatures, the shortest incubation from egg laying to hatching of the first nauplius was 96 days; other times were 131, 139, 140, 142, 143 (two sets), 149, 152, 163, 164, 177, 179 and 196 days. Twelve of the 15 sets which successfully hatched had experienced incubation temperatures in Villefranche no lower than 10 °C. Three sets of eggs hatched after exposure to 4 °C. No synchronous hatching of these laboratory incubated eggs was observed. Within a given set of eggs, nauplii typically hatched at the rate of one or two a day, with intervening days of no hatching. The maximum number hatched in a given set was about one-third of the total.

The Woods Hole Harbor set which hatched had been suspended from the pier from December through March (120 days). During this time the eggs experienced temperatures as low as 2°-3 °C in February and March. When transferred to the laboratory and placed at 18 °C, hatching was initiated within four days. Synchronous hatching occurred; 37 of 57 eggs hatched within two days. Eggs from two other females did not hatch when similarly treated and were presumably nonviable.

DISCUSSION

These observations and experiments on viability and longevity of eggs of *P. mediterranea* confirm the presence of subitaneous and diapause eggs in the life history of this species in the Northern Mediterranean Sea. As reported by Sazhina (1968) subitaneous eggs produced in summer readily hatch, while diapause eggs laid in fall do not hatch in the same length of time. Although

she did not observe hatching in fall eggs, our laboratory work confirms that these fall eggs are viable and hatch after three months and, therefore, are true diapause eggs. Hatching of diapause eggs was not influenced by exposure to reduced temperatures for periods of several weeks (followed by warming) in the laboratory, as was the case with *P. meadi* (Grice and Gibson, 1977), nor did hatching occur synchronously. Marcus (1979) considered synchronous hatching as characteristic of diapause eggs. Prolonged incubation temperatures of 15° and 18 °C could have led to hatching of the eggs similar to the observed sporadic hatching of diapause eggs of *Labidocera aestiva* when incubated at 19 °C (Marcus, 1979). The synchronous hatching of the eggs incubated for four months in Woods Hole Harbor indicates that the refractory period of these diapause eggs was completed and that after exposure to favorable temperatures (18 °C) they were able to readily hatch.

The presence of winter diapause eggs and their hatching after several months of dormancy permits the species to repopulate areas where, because of unfavorable conditions, it cannot survive throughout the year in the plankton.

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