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STUDIES ON THE REPRODUCTIVE BIOLOGY OF THE MUD CRAB, *RHITHROPANOPEUS HARRISII* (GOULD): INDUCTION OF SPAWNING DURING THE NON-BREEDING SEASON (DECAPODA, BRACHYURA)

BY

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INTRODUCTION

Most temperate brachyuran crabs breed only during the warmer part of the year, thus preventing year-round experimentation on their reproductive and development biology. Although there have been reports of chance matings by brachyurans in the laboratory (Hartnoll, 1969), most of these matings occurred during the normal breeding season. There is little information available on the induction of mating and spawning of decapod crustaceans in the laboratory during the non-breeding season. Little (1968) induced winter breeding in the grass shrimp, *Palaemonetes pugio* Holthuis, and Sulkin et al. (1976) were able to induce winter spawning in blue crab, *Callinectes sapidus* Rathbun females which had previously mated. The only brachyuran crab that has successfully been induced to mate and spawn during the non-breeding season has been the stone crab, *Menippe mercenaria* (Say) (McConaugha et al., 1980).

The normal breeding season for *Rhithropanopeus harrisii* (Gould, 1841) in North Carolina is from May to September (Costlow et al., 1966), but over the last five or six years we have found it to sometimes extend from mid-April to mid-October in both the Newport and Neuse Rivers. The mud crab, *Rhithropanopeus harrisii*, is widely distributed in estuarine waters with an original range from New Brunswick, Canada to Veracruz, Mexico and northeastern Brazil and with introductions into Coos Bay, Oregon, San Francisco Bay, California, northwest Europe and the Black Sea (Williams, 1974). The adults of *R. harrisii* are small-sized crabs that are easy to maintain, readily obtainable, hardy and fecund. Their larvae show a high and relatively consistent

survival rate through all developmental stages compared to most species of crabs. Therefore, the development of *R. harrisii* has been one of the most extensively studied of all brachyuran species in relation to environmental factors (e.g., Costlow et al., 1966; Christiansen & Costlow, 1975; Forward & Cronin, 1980) and to toxic substances (e.g., Rosenberg & Costlow, 1976; Laughlin & Guard, 1981; Bookhout et al., 1981). The present study was conducted to determine if *R. harrisii* could be induced to breed non-seasonally in the laboratory producing a continuous stock of larvae to be used for experimentation throughout the year.

METHODS AND MATERIALS

Adult mud crabs were collected from the Neuse River Estuary, North Carolina, on 1 October 1979. These were stocked at a density of 30 females to 15 males in an artificial "habitat" assembled from a Sears off-white copolymer tub 58.5 cm wide by 63.5 cm deep, which was equipped with an undergravel filter constructed from plastic egg-crate ceiling material covered with plastic window screening and with two PVC standpipes 30 cm long and 3 cm in diameter. The "habitat" was provided with a 4 cm deep substrate of crushed oyster shell, with larger shells and parts of shells to provide cover for the crabs. It was aerated by airstones placed inside the PVC standpipes and connected to an air pump. A 12:12 day:night photoperiod was provided by four fluorescent bulbs placed one meter above the bottom of the "habitat". A Hagen 200W submersible aquarium heater maintained the water temperature at $24 \pm 1°C$, and the salinity ranged from 8 to 20‰ with a mean of 12‰. The "habitat" was cleaned and the water changed biweekly, while the crabs were fed weekly on Ralston Purina Marine Ration # 25 and San Francisco (lot # 3288) *Artemia*, with minced fish and clam added occasionally to supplement their diet.

The "habitat" was checked weekly for ovigerous crabs, which were placed individually in an 8 cm diameter culture dish with 12‰ seawater to hatch their eggs. All ovigerous females were removed and any mortalities were replaced with individuals of the appropriate sex in order to maintain the initial sex ratio of 30 females to 15 males.

Small hatches from the ovigerous crabs were counted, while larger hatches were estimated. Fifty zoeae from each hatch were reared 10 per 3.5 cm diameter bowl at 25°C and 20‰ under a 12:12 day:night photoperiod in a temperature-control cabinet. Larvae were fed daily with an excess of newly hatched San Francisco *Artemia* nauplii (lot # 3288) after their water was changed. A daily record was kept on larval molting and survival until the first crab stage was reached.

RESULTS

In 1979, the breeding season of *R. harrisii* from the Neuse River Estuary extended from mid-April to the end of September. On the first of October the

TABLE I

Summary of reproductive data for 1979 "habitat" *Rhithropanopeus harrisii*

Egg production:	
Mean no. ovigerous females/week	4.3
Hatching efficiency:	
Mean % females that hatched eggs	51.9
Hatch size:	
Mean hatch size	800
Larval variability:	
Mean % survival, hatch to megalopa	88.0
Mean % survival, megalopa to crab	97.5
Mean % survival, hatch to crab	69.3
Mean duration (days), hatch to megalopa	11.5
Mean duration (days), megalopa to crab	5.8
Mean duration (days), hatch to crab	15.0

"habitat" was stocked with field collected animals and the first ovigerous females appeared five weeks later. The female crabs continued to produce sponges until 17 December 1979 when the study was discontinued. Over this seven-week period a total of 27 egg masses were produced with a mean sponge production of 4.3 per week. Of these 27 ovigerous females though, 13 dropped their eggs before the larvae hatched. Data concerning egg production, hatching efficiency, hatch size and larval viability are summarized in table I.

DISCUSSION

The mud crab, *Rhithropanopeus harrisii*, was induced to breed and spawn during the non-breeding season by maintaining a 12:12 day:night photoperiod and a water temperature that would be encountered by this organism in the field during the normal breeding season. The only other brachyuran crab that has been induced to breed by using elevated temperature and a 12:12 day:night photoperiod has been *Menippe mercenaria* (cf. McConaugha et al., 1980). However, a diurnal light cycle or a long daylength is not necessary to initiate reproduction in *R. harrisii*, which only relies on an elevated temperature to induce breeding (in preparation).

The mean larval duration and survival of *R. harrisii* presented here were within the limits reported by Costlow et al. (1966) for larvae reared during the normal breeding season. This is especially relevant, since only the best hatches were chosen for their experiments, while during the present study all hatches were reared.

If egg production can be increased by increasing the stocking density and/or the sex ratio, a greater supply of larvae could be obtained with a minimal amount of effort. The hatching efficiency and hatch size might also be in-

creased by placing some type of substrate in the culture dishes with the gravid females. Ovigerous females collected from the "habitat" were usually buried in the substrate, which confirms the recorded observation that immediately before egg laying, *Rhithropanopeus harrisii* females bury themselves in sand (Turoboyski, 1973). Hatching of eggs from females in culture dishes without a suitable substrate may weaken the binding of the eggs to the adult's pleopod resulting in egg loss before the larvae can hatch. Laboratory conditions might also be stressful to the female causing her to pick off eggs with her chelipeds.

Maintaining a reproductive population of *R. harrisii* in the laboratory greatly reduces the number of time-consuming and occasionally unsuccessful trips to the field to collect ovigerous crabs during the breeding season. Furthermore, the population can be kept reproductively active throughout the non-breeding season to provide a continuous supply of larvae for experimentation. We have overwintered populations of *R. harrisii* in the laboratory both in 1980 and 1981, and obtained similar or better results to our first trial in 1979. The present study is one of a series of papers on the reproductive biology of the xanthid mud crab, *Rhithropanopeus harrisii*. The major part of our data for 1980 and 1981 will be presented in forthcoming publications in such areas as: influence of photoperiod and temperature on the induction of spawning; effects of sex ratio and stocking density on egg production; and the retention of sperm plugs with resulting effects on clutch size and larval viability.

RÉSUMÉ

Des *Rhithropanopeus harrisii* recueillis à la fin de la période normale de reproduction ont été maintenu dans un système d'eau de mer fermé à 24 ± 1°C, avec un cycle jour-nuit de 12:12 et une salinité moyenne de 12‰. Les premières femelles ovigères de la saison de non-reproduction ont apparu 5 semaines après le début de l'étude. Les larves, écloses après une période additionnelle de 7 semaines, ont montré un taux de survivance dans les limites normales pour les élevages en laboratoire de cette espèce. Cette étude montre qu'il est possible de maintenir toute l'année en laboratoire une population reproductive de *R. harrisii*, et d'obtenir une production continue de larves à des fins expérimentales.

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