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REPRODUCTION OF *Mytilus galloprovincialis* (Lmk) IN THE ROMANIAN WATERS OF THE BLACK SEA

Corina CIOCAN
National Institute for Marine Research and Development
"Grigore Antipa" Constanta

ABSTRACT

The reproductive cycle of the common mussel *Mytilus galloprovincialis* was carried out during one year, from May 1995 to May 1996, using histological preparations from the mantle. Two spawning periods were identified, in April and late summer (July-August), strongly related to seawater temperature. Individuals in spawning conditions were found throughout the year. The sex ratio was 1:1. Only seven specimen out of 1200 mussels studied were hermaphrodites.

KEY WORDS : Black Sea, *Mytilus galloprovincialis*, reproductive cycle, hermaphrodites

INTRODUCTION

Marine bivalve molluscs have attracted considerable attention in the last two decades. Much of the studies has concentrated on studies of reproduction: *Mytilus galloprovincialis* (DIMOFTACHE & TELEMBICI , 1972), *M. edulis* (SEED, 1975; SUNILA, 1981; LOWE *et al.*, 1982; KING *et al.*, 1982), *Macoma balthica* (PEKKARINEN, 1986), *Mytilus californianus* (NORMAN KELLEY *et al.*, 1982), unionid mussels (PEKKARINEN, 1993).

There have been recognized four major stages in the reproductive cycle of *Mytilus edulis*: development, ripe, spawning and rest. Development and spawning stages can be also underclassified to ten total stages:

Stage 0 - Immature or already spawned, resting gonads. No trace of sexuality observed in the mantle. Spawned gonads can be thin and transparent, or thick and opaque, depending on local available food. Glycogen and lipid are accumulated in the adipogranular cells and vesicular connective tissue cells.
Developmental stage 1 - The beginning of the gametogenesis, germinal tissue spots appear in the mantle tissue.
Developmental stage 2 - Some ripe sperm and ova appear in the middle of the follicles, but early stages of gametogenesis are present.
Developmental stage 3 - Fast gametogenesis stage, the follicles are about half-filled with ripe gametes.
Developmental stage 4 - Maximum proliferation of the gonad. There are almost only ripe gametes in the follicles.
Developmental stage 5 - The highest stage of sexual maturity is achieved. Compacted ova have an angular configuration, the morphologically ripe sperm dilates male follicles.
Spawning stage 4 - The release of gametes into seawater has begun and there is a consequently gamete density decrease in the follicles.
Spawning stage 3 - Follicles are half full of gametes, ripe ova is rather spherical than angular and, in the mantle, the germinal area is suffered severe decrease.
Spawning stage 2 - Follicles contain less than half ripe gametes. Connective tissue is expanding, genital ducts are still shrinking.
Spawning stage 1 - Some residual sperm and ova are left, cytolyse occurs in the ducts and follicles.

The mussel (*Mytilus galloprovincialis*) is one of the most important and characteristic benthic invertebrate in the Black Sea. It is a major food source for fish and a potential source for human food. Mussel gonad is located in mantle tissue. Sex determination is possible (but not certain) only by macroscopic observation of the gonad: a white-yellowish gonad is characteristic for male, a red-yellowish gonad for female.

Microscopic examination of the histological slides or squash preparations is indispensable to establish developmental or spawning stages and to identify the hermaphrodites. The aim of this study was to establish the mussel spawning periods in the Black Sea Romanian waters, using histological techniques.

MATERIAL AND METHODS

Mussels have been sampled from the Constanta station, in front of NIMRD (former Romanian Marine Research Institute) at a depth of about 2 m. In the laboratory, mussels sampled monthly from May 1995 to May 1996, have been selected by size (only 50-60 cm length) and by number (100 mussels / sample). Small pieces of gonad (5x5 mm) were quickly fixed in Backer-formaldehyde (pH = 7), by boiling for few minutes, before they were cut at 5 µm on a freezing microtome, and stained by Toluidine blue 1% solution.

There are some criteria which allowed us to identify the developmental / spawning stages under microscopic examination, but sometimes a partially spawning may be followed by a new development of the gonad, and here we deal with a subjective estimation. For each sample, a gonad index calculated according to SUNILA (1981), expresses the reproductive status of the population: the number of mussels at each stage was multiplied by the numerical score of the stage, products were added and the result divided by the number of individuals in the sample. A resting gonad is defined with value 0 and the gonad index varies from zero, when no sexual activity was noted, to five, when all the individuals are mature.

RESULTS AND DISCUSSION

Mussels sampled from NIMRD station showed a reproductive cycle illustrated in figure 1.

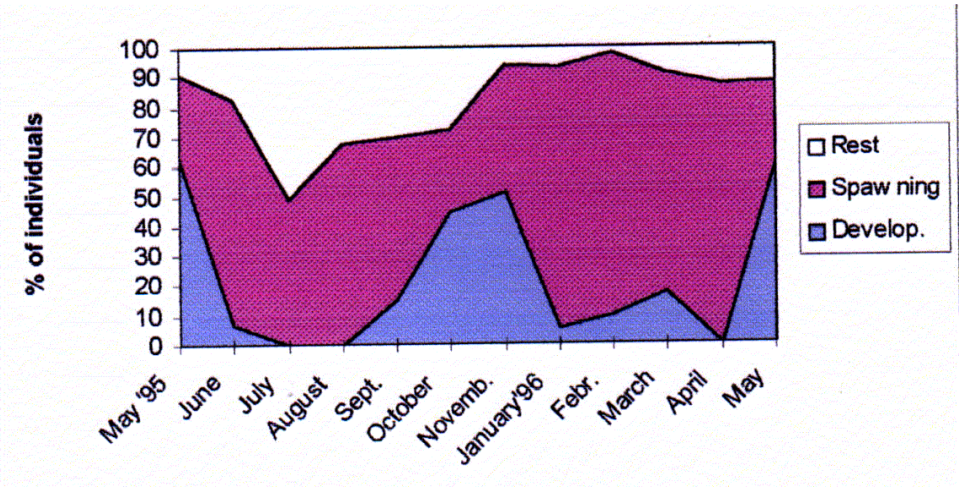

Fig.1 - Reproductive cycle of the common mussel, NIMRD station, 1995-1996

There is a gametogenesis period from September to February, a spawning period in March-April, followed by a recovery and a new development of the follicles, culminating in a major spawn in summer (July-August).

The highest percentages of resting gonads were found in late summer (end of July - 51%, August - 32%). Gametogenesis begind in autumn when the number of individuals with immature gonads is low (6-7 %). A new gamete release occurs in March-April together with worming seawater process, especially when the water temperature rises over 15^0C. No mussels with developmental gonads were found in April sample. At the beginning of May a new gametogenesis starts (Fig.2).

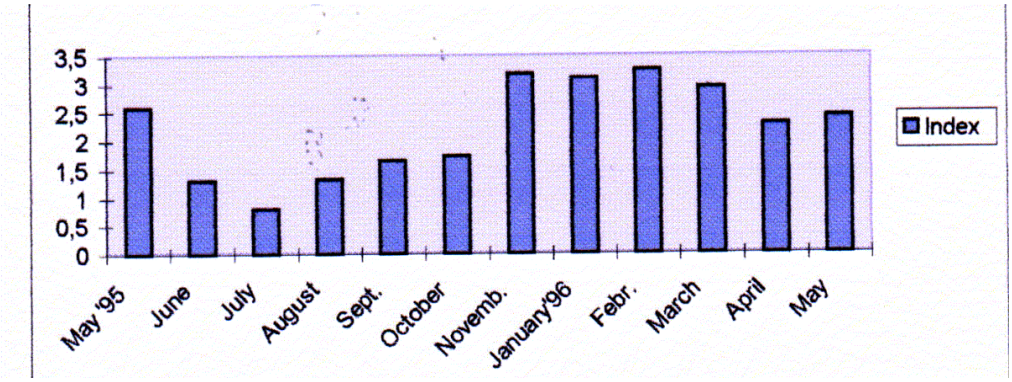

Fig.2 - Variation of the NIMRD mussel population gonad index during May 1995 - May 1996

Only seven of the 1200 investigated individuals were hermaphrodites (0.58 %). The sex ratio was 1:1, when calculated from monthly samples, except August 1995 when the sex ratio was 2:1 (male:female).

The frequency and seasonality of the reproductive cycle in *Mytilus edulis* varies according to geographical distribution (LOWE *et al.,* 1982). In the north part of England, mussel populations spawn only once a year (late spring) whereas those living in the south-west, an area of milder winters and warmer summers, may spawn twice a year, in spring and late summer (SEED, 1976). On the west coast of Ireland, mussels (*Mytilus edulis*) spawn in July-August. Gametogenesis starts in September and a second spawn occurs in March-April (KING *et al.*,1989). A single spawning period (in the middle of the summer) is characteristic for mussel populations from the Finnish coast of Baltic Sea (SUNILA, 1981).

According to DIMOFTACHE & TELEMBICI (1972), the sea water temperature is an important factor which leads the gamete release. The best interval for spawning, for mussel populations on the rocky shore of the Black Sea is between $+15^0$ - $+18^0$C.

In our study, the first spawned mussel occur at 15.3^0C water temperature (in springtime) and in summer, the major spawn takes place at 22^0 - 25^0C water temperatures.

Spontaneous gamete release can be induced by some unexpected phenomenon like changes in temperature or salinity, shaking (storms or strong streams), chemical stimulation (SUNILA, 1981).

CONCLUSIONS

Mussel populations from the Romanian Black Sea coast spawn twice a year, the reproductive cycle being strongly related to seawater temperature.

Generally, sex ratio was 1:1, the exception from August sample (2:1, male:female) being attributed to high number of individuals already spawned (inactive gonads make indistinguishably sex). Thus males and females seem to reach the resting stage at different times.

Hermaphroditism is not wide spread among the mussel populations - we count only seven individuals from 1200 analysed mussels.

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