

REPRODUCTIVE CYCLE AND GAMETOGENESIS OF THE CAENOGASTROPOD *Melongena melongena* IN THE GOLFETE DE UARE, VENEZUELA.

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(Nicida Noriega from Venezuela won the 2003 Constance Boone Grant to Malacology. The research we helped fund was her Master's Thesis. She is a Venezuelan citizen and a student at the Simon Bolivar University in Caracas. Her studies have an ecological approach and she is (and has been) very interested in Venezuelan marine environments, their value, man's impact and conservation.)

The Caenogastropod *Melongena melongena* is distributed along the Venezuelan coast and established in muddy places of salty lagoons and in muddy areas of open waters, where it lives buried under the sediment. *M. melongena* is a species with economic importance within the Golfete de Cuare (zone belonging to the Cuare Wildlife Reserve) where it is exposed to an intense fishing pressure. The *M. melongena* is a dioic species with internal fertilization whose sex is not distinguishable by the shell's external characteristics. In this species imposex does not exist, since the presence of sexual cells (spermatozoids and oocytes) coincides with the presence of a penis in the male and a gonoduct in the female with the absence of a deferent vessel or penis. The minimal sexual maturity size for the females was 5.1 cm shell-length and 5.8 cm shell-length for the males.

In both sexes the gonad is not clearly differentiated from the hepatopancreas, since the gonad is made up of tubular follicles which penetrate the hepatopancreas. The gonad presents the same color in both sexes as gametogenesis progresses. During the first stages of development, the gonad presented a yellowish color and was characterized by the presence of spermatogonia and spermatocytes on the follicular wall in the males and of oogonia in the case of the females. In the mature stage, the gonad presented a yellowish-orange color which coincided with a great abundance of spermatozoids in the follicular lumen in the males and of oocytes of great length in the females. During spawning, or gamet liberation, the gonad presented a white color and microscopic analysis revealed a diminution in the amount of spermatozoids and oocytes in the follicles.

Gametogenesis and reproductive cycle

Oogenesis was classified in the following stages: early active, late active, ripe and spawning/reabsorption. The gonads in the early active stage were characterized by the presence of previtelogenic oogonia and oocytes with lengths which varied between 8-9 μ m and 10-30 μ m respectively, both attached to the follicular wall. The previtelogenic oocytes presented a close relation with the follicle's epithelial tissue and accompanying cells, and had a nucleus which measured 5 μ m and a nucleolus which measured 1 μ m. During the late stage there were found vitelogenic oocytes whose length varied between 60-80 μ m; these oocytes presented an elongated shape and were attached to the follicular wall by means of a peduncle. Once the oocytes reach maturity, they separate themselves from the follicular wall and migrate to the follicle's lumen; these oocytes are characterized by presenting a length >80 μ m and having an abundant vitellus content in their interior. Finally the gonads in the spawning stage were characterized by few mature oocytes in the lumen's center; the oocytes which were not liberated were reabsorbed, and during the process of reabsorption there was observed a vacuolae formation within the oocytes as well as the presence of yellow bodies inside the follicles, which bodies are in charge of disintegrating those oocytes which were not liberated (Fig. 1)

Spermatogenesis was classified into three stages: early active, ripe and spawning. During the growth phase (early stage) the gonad characterized itself by the presence of spermatogonia located on the follicle's wall; then the gonads enter a maturity stage and are characterized by the presence of follicles filled with spermatozoids, which in turn form dense masses with their tails always directed toward the lumen's center and their heads oriented toward the follicular wall. Likewise there was observed a presence of spermatogonia, spermatocytes and spermatids, indicating that gametogenesis went on near the follicular wall. The gonads in the spawning stage showed few spermatozoids in the follicular lumen and the spermatocytes and spermatids formed a very thin layer (Fig.2).

During the reproductive cycle of *Melongena melongena* it was observed that the largest percentage of females with gonads in the early stage occurred in the months of May 2002 and January 2003, with 60% and 70% respectively, while the males showed 100% of individuals in such stage in the months of May, July and October 2002. The late active stage was observed to occur only in the females, with the largest percentages in the months of September and October 2002, with 50% and 70% respectively. There were observed males with mature gonads in various months along the year, but the largest percentages showed themselves in August and March 2002 with 80% and 100% of individuals respectively. As for the females, those examined in the months of June and November 2002 and in February 2003 showed 100% of individuals to be in such stage. On the other hand, the females with gonads in the spawning stage were found in July 2002 and March 2003; in fact, the totality of the examined females presented gonads in this stage, while for the males the largest percentages of individuals with gonads at the spawning stage were found in the months of April and September 2002 and January 2003.

Therefore, during *M. melongena*'s reproductive cycle, it was found that the spawning times of the females occurred in the months of July and December 2002 and March 2003, while in the males the spawning was observed to take place in the months of April and September 2002 and January 2003. After each reproduction event the gonads recuperate immediately and enter the gamet growth stage up to gamet maturity. No synchronization was observed for *M. melongena* as regards reproductive timing and gametogenic activity between males and females. However, it is important to point out that prior to the females' spawning events, there were found males which liberated gametes. Neither was there found any relation between the events mentioned above and environmental factors such as temperature, salinity and sunlight hours.

Protein content embryonary level and fluid capsule

During the embryonary development of *M. melomgena* there was a slight increase in the protein content from the egg stage (11.23 µg/egg) to the trocophore (12.37 µg/larvae), veliger (12.47 µg/larvae) and pediveliger (13.03 µg/larvae) stages, and fell significantly at the hatching stage (5.95 µg/juvenile). Significant differences were found (one-way ANOVA, $p < 0.01$) in the protein content per embryo for the *M. melongena* embryonary stages. An a posteriori test (Tukey's Multiple Comparison, $p < 0.05$) showed differences in the intracapsular protein content for the stages egg, veliger and pediveliger with respect to the hatching stage ($p < 0.05$), while no significant differences were found among the stages egg, veliger and pediveliger ($p < 0.05$).

A slight decrease was observed in the protein concentration of the intracapsular fluid as the embryo developed. In the egg stage, the protein concentration in the intracapsular fluid was 0.18 µg/µl, while in the pediveliger stage the concentration was 0.13 µg/µl. However, no significant differences were found ($p < 0.05$) in the intracapsular fluid's protein concentration as the organisms developed.