

Gonadal Maturation in Pen Shells *Pinna rudis* and *Pinna nobilis* Stimulated by Reproductive Neuropeptides

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Abstract : The pen shell *Pinna nobilis* population has declined dramatically since 2016 due to die-off events observed in the whole extent of the Mediterranean Sea associated with the protozoan *Haplosporidium pinnae*. As of 2019, it is considered a critically endangered species. Due to its ecological importance and its endangered status, several initiatives have been developed for its salvation and recovery. This research is an effort to understand and control its reproduction under captivity. As a limited number of *Pinna nobilis* individuals could be used for experimentation, the possibility of using the *Pinna rudis* as a model animal was explored. The molecular mechanism that regulates the reproduction of both species is unknown; consequently, transcriptomic analysis was performed to identify neuropeptides that are expressed in the key regulatory tissues of the visceral ganglia and gonads of both species. Neuropeptides form an important group of signaling peptides that regulate reproductive, behavioral and physiological functions in molluscs. In total, 17 neuropeptide precursors were identified in *P. nobilis* and 14 in *P. rudis* transcriptomes; 14 of them were identical in both species. This affinity verified the genetic similarity of these species at the reproduction level. APGWamide, buccalin, ELH and GnRH were tested in *P. rudis* and demonstrated their capacity to advance gonadal maturation and trigger spawning while spawning was recorded in *P. nobilis* after the usage of APGWamide and buccalin. The neuropeptides were administered using intramuscular injection and cholesterol implants following relative literature as well as a new method was developed for external administration without the use of anesthesia using a mathematical model. The know-how of this research will not only lead to the survival of the species but also will narrow the horizons of broodstock conditioning of other similar species.

Keywords : neuropeptides, *Pinna nobilis*, reproduction, transcriptomics

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