

Germ Cell Differentiation, Sexual Maturation And
Changes In The Epithelial Cells Of The Seminal
Vesicle Of The Male *Neptunea Cumingii*
(Gastropoda: Buccinidae)

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INTRODUCTION

This species is especially found in silty sand of the subtidal zone of the west coast of Korea. Recently, as the standing stock of this species gradually decreased due to extensive reclamation projects and reckless over-harvesting, it has been designated as one of the important organisms in need of natural resources management.

On *Neptunea* spp. in foreign countries and Korea, previously, there have been some studies on aspects of reproduction including the reproductive cycle (Fujinaga, 1985) and spawning. Little information is available on ultrastructural study on germ cell differentiations during spermatogenesis, the reproductive cycle of *N. cumingii*. Understanding the reproductive cycle and the spawning period of this species will provide necessary information for natural spat collections or the recruitment period and age determination of this population. In addition, data on first sexual maturity and reproductive strategy of this population are very useful information for natural resource management. Therefore, the main aim of the present study is to understand germ cell differentiations, the reproductive cycle, first sexual maturity, changes in epithelial cells of the seminal vesicle of this species.

MATERIALS AND METHODS

1) Sampling

Specimens of the male *Neptunea cumingii* were collected monthly at the subtidal zone of Maldo, Chollabuk-do, Korea, from January 2002 to September 2003 (Fig. 1) The snails ranging from 41.0 to 106.8 mm in shell height were

used for the present study. After the snails were transported alive to the laboratory, shell heights and total body weights were immediately measured.

2) Gonadosomatic index (GSI)

A total of 486 individuals were used for calculation of the GSI. Monthly changes in the mean gonadosomatic index (GSI) were calculated by the following equation (Chung et al., 1993, 2002):

$$\text{GSI} = \frac{\text{The thickness of the gonad} \times 100}{\text{Diameter of posterior appendage including the gonad and digestive gland (Fig. 2C)}}$$

Diameter of posterior appendage including the gonad and digestive gland (Fig. 2C).

1) Germ Cell Differentiation by the electron microscop observations

For electron microscopical observations, 1) Prefixation: 2.5 % paraformaldehyde-glutar- aldehyde in 0.1 M phosphate buffer (pH 7.4) for 2 hours at 4°C. Postfixation: 1 % osmium tetroxide dissolved in 0.2 M phosphate buffer solution (pH 7.4) for 1 hour at 4°C. 3) Ultrathin section of Epon-embedded specimens: Sorvall MT-2 microtome. 4) Staining: doubly stained with uranyl acetate followed by lead citrate. Examination: JEM 100 CX-2 (80 kv) electron microscope.

2) Histological Observations

For light microscopic examination of histological preparations, a total of 456 individuals were used for Histological analysis of the gonads and seminal vesicle tissues from January to December, 2002. 1) Fixation: Bouin's fixative for 24 h. 2) Embedded in paraffin molds. 3) Staining: Hansen's hematoxylin-0.5 % eosin, Mallory's triple stain and PAS stain. Examination: using a light microscope.

RESULTS AND DISCUSSION

The spermatozoon is approximately 20 μm in length. The axoneme of the tail flagellum consists of nine pairs of microtubules at the periphery and one pair at the center. A considerable amount of spermatozoa make their appearance in testicular lobules and some of them were transported from the testis towards the seminal vesicles until May.

Monthly changes in the GSI began to increase in September and reached the maximum in January (2.83) in males. Thereafter, their values rapidly decreased February in males. The reproductive cycle with gonadal

development of this species can be classified into four stages in males: developing stage (August to September), ripe stage (October to May), copulation stage (April to June), and recovery stage (July to August).

Copulation in males occurred between April and June when the seawater temperature rose to approximately 16-23 °C.

The process of the cyclical changes of the epithelial cells of the seminal vesicles could be classified into three phases: (1) S-I phase (resting), (2) S-II phase (accumulating), and (3) S-III phases (spent).

REFERENCES

- FUJINAGA, K., 1985. The reproductive ecology of the neptune whelk (*Neptunea arthritica* Bernardi) population, with special reference to the reproductive cycles, depositions of egg masses and hatchings of juveniles. *Bulletin of Faculty of Fisheries Hokkaido University*, 36: 87-98. (In Japanese with English summary).