Short Paper

Diurnal Changes in Plasma 17α , 20β -Dihydroxy-4-pregnen-3-one Levels during Spawning Season in the Red Sea Bream *Pagrus major*

Hirohiko Kagawa,*¹ Hideki Tanaka,*
Koichi Okuzawa,* Michiya Matsuyama,*²
and Keiji Hirose*¹

(Received September 14, 1990)

Many studies on final oocyte maturation have been carried out mainly on salmonids and cyprinids, suggesting that 17α , 20β -dihydroxy-4-pregnen-3-one (17α , 20β -diOHprog) acts as an important mediator of oocyte maturation in these species.¹⁾ Plasma levels of 17α , 20β -diOHprog of the fish increase at the time when final oocyte maturation occurs. However, there is little information on oocyte maturation of marine teleosts so far, especially on changes in plasma levels of maturation-inducing steroid. The present study was undertaken to clarify diurnal changes in plasma 17α , 20β -diOHprog levels during final oocyte maturation of red sea bream.

Three-year-old female red sea bream were kept in 1 or 10 t tanks with running sea water (5 females per 1 t with the same number of males). Under these conditions, most of the female fish spawned every day between 16:00 and 19:00. After ascertaining the daily spawning during one week, 5–10 fish were sampled at 4 hours intervals from 8:00. After they were anesthetized with 2-phenoxyethanol (300 ppm), blood samples were taken from the caudal vessel. The plasma were stored at -20°C until 17α , 20β -diOHprog was measured with radioimmunoassay described previously.²⁾ Developmental stages of the oocytes at each sampling time were identified according to the previous classification³⁾ with ordinary light microscopy.

The most advanced oocytes in the ovary after ovulation and spawning were in tertiary yolk globule stage at 16:00 and 20:00. The migration of the germinal vesicle was observed between 20:00 and 24:00, and the germinal vesicle breakdown was completed until 8:00. Ovulation was observed in all of the fish sampled at 12:00. Plasma 17α , 20β -diOHprog levels (Fig. 1) gradually increased from 20:00 when the migration of the germinal vesicle was observed and reached a peak (389 ± 98 pg/ml) at 4:00 ($p < 0.01$) when the pre-mature and mature stage oocyte occurred. Thereafter, their levels decreased to minimum levels (39 ± 5 pg/ml) at 16:00 ($p < 0.01$).

The levels of plasma 17α , 20β -diOHprog detected in the present study were the same order as reported in Japanese whiting,⁴⁾ a marine daily-spawning teleost, but one or two order magnitude lower than those observed in flatfish induced maturation with human chorionic gonadotropin.⁵⁾ In the present study, however, 17α , 20β -diOHprog levels at 4:00 increased to about 10 times as high as the minimum levels at 16:00 and this increase of the steroid correlated well with final oocyte maturation. 17α , 20β -diOHprog was one of the most potent inducers of *in vitro* final oocyte maturation in the red sea bream,⁶⁾ inducing over 90%

Fig. 1. Diurnal changes in plasma 17α , 20β -dihydroxy-4-pregnen-3-one (17α , 20β -DiOHprog) levels in the red sea bream.

Data were expressed as mean $\pm$ SEM. Statistic differences ($p < 0.01$) were observed between 4:00 and others, except 8:00, using Duncan's multiple range test.

even at a concentration of 100 pg/ml. Moreover, thin-layer chromatography showed that this steroid was found as one of the metabolites when ovarian fragments were incubated with 17α -hydroxyprogesterone as an precursor (Asahina, unpublished data). These data suggest that 17α , 20β -diOHprog is the major steroid which plays an important role in inducing final oocyte maturation in red sea bream. A recent study indicates that 17α , 20β , 21-trihydroxy-4-pregnen-3-one is a maturation-inducing steroid in the Atlantic croaker *Micropogonias undulatus*.⁷⁾ Because this steroid was also potent inducer of *in vitro* oocyte maturation in red sea bream (Hirose, unpublished data), more detailed studies are necessary to clarify whether this steroid is implicated in oocyte maturation in the red sea bream.

This study was supported in part by a grant-in-Aid (Bio Media Program) from the Ministry of Agriculture, Forestry and Fisheries. (BMP 90-II-2-6)

References

- 1) Y. Nagahama: *Develop. Growth and Differ.*, **29**, 1–12 (1987).
- 2) G. Young, L. W. Crim, H. Kagawa, A. Kambe-gawa, and Y. Nagahama: *Gen. Comp. Endocrinol.*, **51**, 96–105 (1983).
- 3) M. Matsuyama, S. Adachi, Y. Nagahama, and S. Matsuura: *Aquaculture*, **73**, 357–372 (1988).
- 4) M. Matsuyama, S. Adachi, Y. Nagahama, K. Maruyama, and S. Matsuura: *Fish Physiol. Biochem.*, **8**, 329–338 (1990).
- 5) A. V. M. Canario and A. P. Scott: *Gen. Comp. Endocrinol.*, **77**, 177–191 (1990).
- 6) S. Adachi, K. Ouchi, K. Hirose, and Y. Nagahama: *Nippon Suisan Gakkaishi*, **54**, 1665 (1988).
- 7) J. M. Trant, P. Thomas, and C. H. L. Shackleton: *Steroids*, **47**, 89–99 (1986).

*¹ National Research Institute of Aquaculture, Nansei, Mie 516–01, Japan (香川浩彦, 田中秀樹, 奥澤公一, 広瀬慶二: 養殖研究所).

*² Faculty of Bioresources, Mie University, Tsu 514, Japan (松山倫也: 三重大学生物資源学部).