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Histological Observation on the Hypothalamo-Hypophyseal System and the Thyroid Gland of the Sailfish, *Istiophorus platypterus*

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Abstract The hypothalamo-hypophyseal system and the thyroid gland of the adult and immature sailfish caught off Sado Island in the Sea of Japan were studied histologically. Seven types of glandular cells were distinguishable in the adenohypophysis. The thyrotroph of immature fish was distinct and the thyroid gland showed a hyperactive state. Due to postspawning a large number of gonadotrophs in the adult fish were found in the process of vacuolization and degranulation. A considerable amount of aldehyde fuchsin stainable neurosecretory material was laden in the cells of the nucleus preopticus, axonal tract and pars nervosa. The nucleus lateralis tuberis consisted mainly of two portions, the pars lateralis and pars ventromedialis. The cells of the pars ventromedialis in the adult fish were characterized by the presence of a large vesicular structure filled with acidophil coarse granules.

The sailfish, *Istiophorus platypterus* (Shaw et Nodder), is one of the major epipelagic fish of a comparatively large size occurring widely in the warm waters of the Indo-Pacific Ocean. Among the billfishes, this species is the only one that exhibits feeble euryhalinity and frequently appears in the coastal waters where salinity is slightly lower. However, little is known of the endocrine glands and physiology of the sailfish that migrates to the Sea of Japan. There are so far only two reports dealing with the thyroid (Honma, 1956) and endocrine pancreas (Hirano and Honma, 1971). In order to gain more knowledge of this field, this study was designed to investigate the hypothalamo-hypophyseal system and the thyroid gland of this species.

Material and methods

Three fresh specimens of the sailfish, *Istiophorus platypterus*, one adult (1.8 m in total length) and two immature (87 cm), were obtained by the senior author on August 4, 1978 and on September 11, 1979, respectively. They were caught in a commercial set net that was installed off the Sado Marine Biological Station of Niigata University, Sado Island in the Sea of Japan. The sexes of the specimens were not determined, because the visceral organs including the gonads had already been removed by the fishermen to keep the fish fresh.

The brain with its hypophysis and the thyroid gland were fixed in Bouin-Hollande-sublimate solution for a week. Then the organs were dehydrated, embedded in wax and cut into 7 to 10-μm thick sections in both sagittal and transverse directions. The sections were stained with azan trichrome, Gomori's aldehyde fuchsin (AF)-fast green-orange G, periodic acid Schiff (PAS)-alcian blue (AB)-orange G, AB-acid fuchsin, and lead hematoxylin (PbH).

Results

Hypophysis. The general structural pattern of the hypothalamo-hypophyseal region of the sailfish is schematically shown in Fig. 1. The hypophysis, ovoid in shape, is connected to the diencephalic floor by a thin wall of the infundibulum. Three lobes, i.e., the rostral pars distalis (RPD), the proximal pars distalis (PPD), and the pars intermedia (PI), are visible in the glandular portion. The PI is the most voluminous part of the gland. From the antero-dorsal portion of the gland the pars nervosa (PN) invades deeply into the

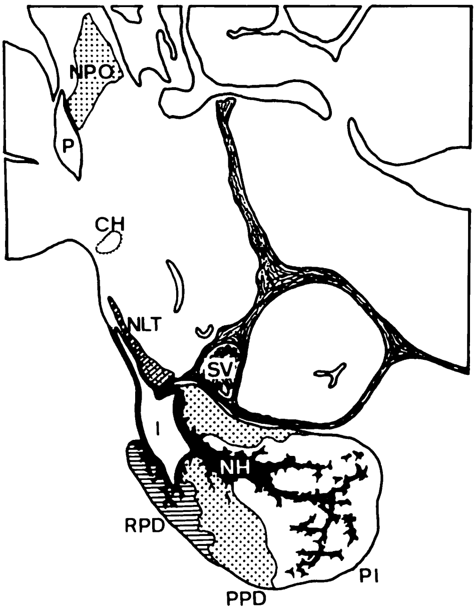

Fig. 1. Schematic illustration of the hypothalamo-hypophyseal region of the sailfish, *Istiophorus platypterus*. CH, commisure holizontalis; I, infundibular recess; NH, neurohypophysis; NLT, nucleus lateralis tuberis; NPO, nucleus preopticus; P, preoptic recess; PI, pars intermedia; PPD, proximal pars distalis; RPD, rostral pars distalis; SV, saccus vasculosus.

adenohypophysis, and sends its ramifications extensively toward the PI. A considerable amount of AF stainable granules is laden in the PN. The so-called Herring bodies are scattered here and there in the PN of the adult fish.

The RPD, the smallest part in the glandular portion, occupies an antero-ventral part of the gland. This lobe consists of two types of acidophil cells that are considered adrenocorticotrophs and the prolactin cells. The adrenocorticotrophs form a fine palisade layer facing the neurohypophyseal branches (Fig. 2). The cell is columnar in shape, and contains a round or oval nucleus with a somewhat indistinct nucleolus. The finely granulated cytoplasm, selectively stained with PbH, is considerably rich in amount (Fig. 3). The cell is also tinged with yellowish orange by azan trichrome stain or pale green by PAS-fast green-orange G stain. The prolactin cells are considerably numerous and closely packed in the ventral region of the corticotrophs (Fig. 2). These cells, polyhedral or round in shape, have a round nucleus bearing a rather round nucleolus. The size, cytoplasmic granulation and morphological aspect of the nucleus vary from cell to cell. A great number of the cells have their rich cytoplasm containing a great deal of carminophil granules. Cells with poor granulation are also encountered in the periphery of RPD. In addition, there exist some small cells, which are characteristic in having a flat nucleus and scanty cytoplasm lacking granules, interspersed with the prolactin cells (Fig. 3). These cells may be equivalent to the so-called stellate cells.

The PPD is highly vascularized. The volume of PPD increases remarkably with age. Three main types of cells are identified in this lobe: two types of basophil (=mucoid) cells and acidophil cells. The first type of basophil cell, diagnosed as thyrotroph, is located in the antero-dorsal portion of PPD adjacent to the neural tissue and also in RPD. As a rule, cells of this type gather in small clusters or cords (Fig. 4). Some groups of the cells lie just beneath the ependyma on the floor of the infundibulum, while others intersperse with the corticotrophs. Each cells appears pear- or club-shaped and its basal elongation reaches the basement membrane. The cytoplasm is finely granular and stained intensely with AF, AB, PAS and aniline blue. The nucleus, round in shape, is situated in the apical swelling of the cell. Unlike the thyrotroph, the second type of basophil cell, considered to be a gonadotroph, is widely distributed in the PPD and intermingles with the remaining acidophil cells (Fig. 5). This cell, usually polygonal in shape, has a round nucleus and a various amount of coarse granules of glycoproteinaceous nature. Occasionally, the cell projects a slender process toward the basement membrane. The cells of the adult fish surpass those of the immature fish in size, number and the amount of cytoplasmic granules (Figs. 4, 5). In the adult fish, there are several types of mucoid cells probably derived from the gonadotroph: 1) remarkably hypertrophied cells with paucity of stainable granules (Fig. 6), 2) signet ring

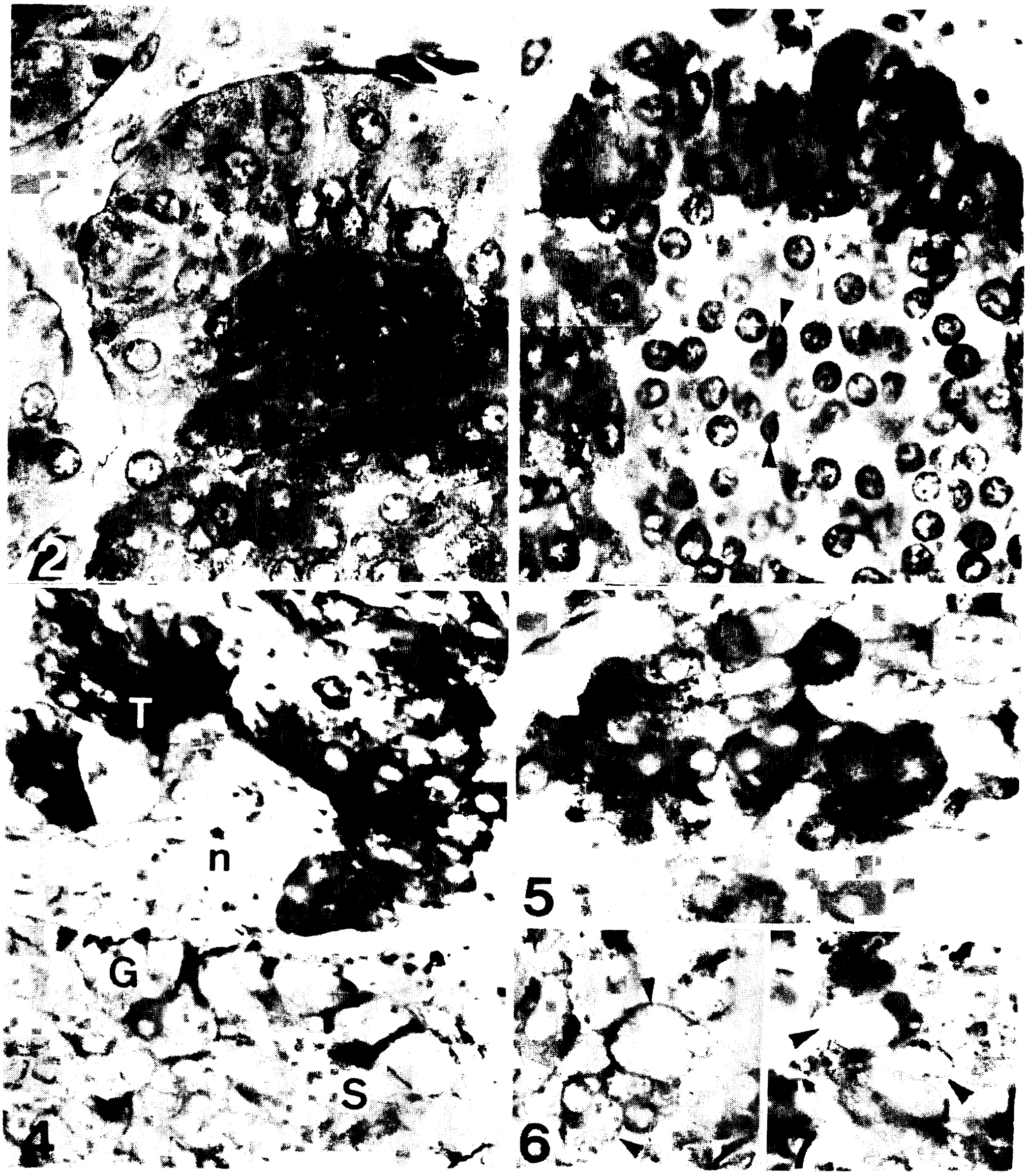

Fig. 2. A part of RPD from adult sailfish showing two types of cells, the corticotroph (light) and prolactin cell (dark). Azan stain. ×1,000.

Fig. 3. A part of RPD from adult sailfish showing PbH-positive corticotroph (dark) and PbH-negative prolactin cell (light) and agranular stellate cell (arrow head). PbH stain. ×1,000.

Fig. 4. The PPD of immature sailfish showing three types of cells, the thyrotroph (T), gonadotroph (G) and somatotroph (S). n, neural tissue. AF stain. ×1,000.

Figs. 5∼7. A part of PPD from adult sailfish. AF-fast green-orange G stain in 5 and 6, and AB-acid fuchsin in 7. ×1,000. Fig. 5. Somatotroph (light) and heavily granulated gonadotroph (dark). Fig. 6. Degranular and enlarged gonadotrophs (arrow head). Fig. 7. Signet ring gonadotrophs under intense vacuolization (arrow head).

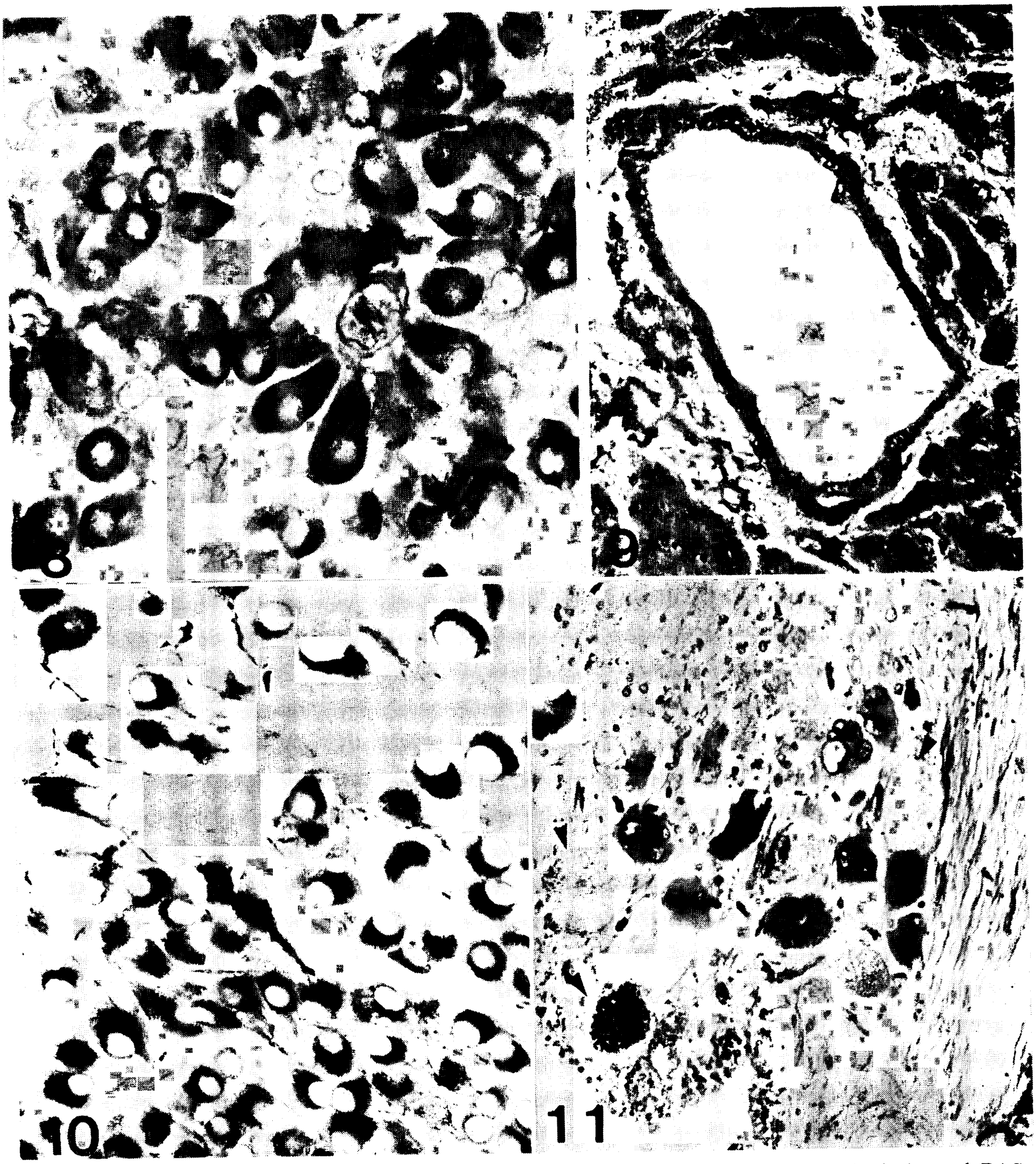

Fig. 8. The PI of adult sailfish showing two types of cells, the PAS-positive cell (dark) and PAS-negative cell (light). PAS-fast green-orange G stain. ×1,000.

Fig. 9. A part of PI from adult sailfish showing a large lacuna filled with amorphous material. Azan stain. ×1,000.

Fig. 10. The NPO pars magnocellularis of immature sailfish showing AF-positive neurosecretory cells. AF stain. ×400.

Fig. 11. Cross section of the NLT pars ventromedialis showing acidophil perikarya. Note the large vesicular structure filled with coarse granules (arrow head). Azan stain. ×200.

cells characterized by a huge vacuole occasionally accompanied by acidophil globules (Fig. 7), and 3) shrunken cells with a pycnotic nucleus. The remaining acidophil cells may correspond to the somatotroph. This type of cell, cuboid or ovoid in shape, has a round nucleus. The cytoplasm is rich in amount and finely granulated, and shows an intense affinity for orange G, acid fuchsin and azocarmine. The cells are disposed in clusters or branching cords. A considerable number of lacunae of various sizes were noticed in PPD of the adult fish. In these lacunae, a fibrous material and small cells were seen together with the disintegrating glandular cells.

The PI comprises two main types of cells: PAS positive cells of ovoid, elliptic or round form, and PAS negative cells of polygonal or clavate form (Fig. 8). By means of azan trichrome stain, these two cell types were differentiated into orangenophil and cyanophil cells, respectively. The PAS negative cells show no affinity for PbH in the present examination. The shape and size of the nucleus in these cells vary from cell to cell. Strikingly, a single large space (400 μm in long axis) was noticed in PI of the adult fish (Fig. 9). This space is lined with a thin layer of membrane-like structures and single or partly two-tier glandular cells. Moreover, this space was occupied with amorphous (fluffy) cyanophilic material and a few free cells.

Hypothalamic neurosecretory center. Two neurosecretory centers were defined: the nucleus preopticus (NPO) located on both sides of the preoptic recess and the nucleus lateralis tuberis (NLT) located in the ventro-caudal region of the hypothalamus, extending partly into the infundibular recess. The NPO is composed of two contiguous portions, a rostroventrally shifted pars parvocellularis (12.5 μm in mean cell diameter) and a dorso-caudally shifted pars magnocellularis (27.9 μm). The NPO cell, polygonal in shape, contains a round or oval nucleus with a prominent nucleolus, although a pycnotic or multilobate nucleus is seldom encountered. The perikaryon was stained intensely or moderately with AF (Fig. 10). The axones in beaded fibers are traceable toward the neurohypophysis. The NLT extends over two parts: pars lateralis and pars ventromedialis. The pars lateralis is well-vascularized and composed of large cells, 46.9 μm in mean diameter. It lies more laterally on the tuber cinereum in close proximity to the meningeal connective tissue. The cell is polygonal in shape with rich cytoplasm stained with acidic dyes, but its axon is poorly defined. A considerable amount of AF and PAS positive coarse granules is demonstrated in some of the cells in the adult fish. The nucleus of the cell is round in the immature fish, while polymorphic or multilobate in the adult fish. The pars ventromedialis is situated between the ependyma facing the third ventricle and the ventral portion of the pars lateralis. There is no clear demarcation between these two parts, however. The cell of the pars ventromedialis is closely similar to that of the pars lateralis in appearance, but the former is larger in size (61.1 μm in mean diameter) and its nucleus is more chromophilic and more irregularly outlined than that of the latter. Cells of the adult ventromedialis are characterized by having a large vesicular structure (54.8 μm in mean diameter) in the perikaryon (Fig. 11). Usually, it is filled inside with acidophil coarse granules. Occasionally, the entire cell is replaced by this structure.

Thyroid gland. The macroscopic and microscopic structures of the thyroid gland of the immature fish are nearly identical with those of the adult fish which was described by Honma (1956). The gland with rich vascularization consists of thyroid follicles, ranging from 28 to 152 (mean 60.9) μm in diameter. The follicle is oval or elliptic in shape, but frequently exhibits an irregular outline. The colloid in the lumen is homogeneous or finely granular and strongly positive to PAS reaction. Occasionally, small vacuoles were seen in the margin of the colloid. The follicular epithelium is composed of columnar or cuboid cells (10.4 μm in mean height) with oval or elliptic nuclei containing prominent nucleoli (Fig. 12). The mitotic figures and PAS positive colloid droplets in the apical portion of the cells were seldom observed. These pictures were considered as an indication of active condition.

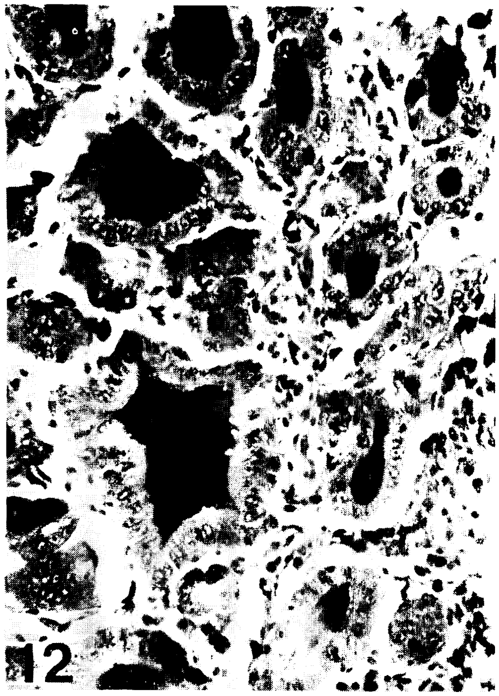

Fig. 12. Part of an active thyroid gland from immature sailfish showing the follicle lined with columnar epithelial cells. Azan stain. ×400.

Discussion

The hypophysis of sailfish has never been studied, although several species of related scombrid fishes were described by Honma (1960) and Kawamoto (1967). The cytology of teleostean hypophysis has been extensively documented and reviewed by several authors (Ball and Baker, 1969; Sage and Bern, 1971; Schreibman et al., 1973; Holmes and Ball, 1974). So far, seven or eight types of cells are identifiable with either light or electron microscopy. The present study of the sailfish has also demonstrated seven types of granular cells in the adenohypophysis. Among them, the PbH positive cell in the dorsal region of RPD corresponds well with the adrenocorticotroph of other teleostean fish that was demonstrated by administration of corticosteroidogenesis blocking agent (metopirone) (Ball and Olivereau, 1966; Mattheij, 1968; Yoshie and Honma, 1978) or by means of immunocytochemistry (McKeown and Overbeeke, 1969; Follenius and Dubois, 1976). It is well established that the acidophil cell in the RPD is the source of prolactin which exerts on osmoregulation in freshwater environments (Ball, 1969; Olivereau, 1969; Bern, 1975; Honma and Yoshie, 1974; Honma et al., 1976). In the sailfish caught off the coast of Sado Island, the prolactin cell is rather rich in carminophil granules. However, the exact role of the prolactin cell is still obscure in the marine teleosts.

Two types of basophil cells were clearly differentiated in the PPD of the sailfish hypophysis. The polygonal cell occurring widely in this lobe is considered to be the gonadotroph, since this cell is more numerous and more heavily granulated in the adult fish than in the immature fish. Moreover, some of these cells of adult fish were markedly hypertrophic with simultaneous occurrence of degranulation or vacuolization. These features possibly represent the over-activated or exhausted phase of the gonadotrophs as demonstrated in the spent fish by Honma and Tamura (1965). Therefore, it is likely that the present adult specimen was a postspawning fish.

The intense affinity of another basophil for basic dyes indicates the presence of much secretory granules in the cell diagnosed as thyrotroph. This appearance also implies an active condition of the thyroid gland. The present description of the active gland of an immature sailfish closely resembles that of the adult fish that was reported previously by Honma (1956).

No noteworthy feature was obtained in the present histological design to study the NPO of the sailfish, when compared with those of other teleostean species (Charlton, 1932). Two portions were discernible in the NLT of the sailfish, although the microscopic structure and subdivision of this nucleus differ considerably from species to species (Terlou and Ekengren, 1979). Contrary to the immature fish, the cell of the adult pars ventromedialis was characterized by having the large vesicular structure filled with coarse granules. Nearly identical structure has already been reported in the NLT pars medialis and pars ventrolateralis of the tench, *Tinca vulgaris*, by Brehm (1958). Being associated with the reproductive activity, "die Blase" of the tench increased in number toward summer and decreased in autumn. The granules stored in the vesicular structure was suggested to be a secretory product originally elaborated in the nucleus of the cell (Brehm, 1958). If this is the case, the presence of the vesicular structure in the NLT may imply that the presently studied adult specimen is a post-spawning fish. Further studies on various organs are now in planning to learn the physiological condition of migrating sailfish.

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バショウカジキの視床下部—下垂体系および甲状腺の組織学的観察

千葉　晃・本間義治

佐渡島沖から得られたバショウカジキの成魚と未成魚を用い，視床下部—下垂体系および甲状腺を光顕レベルで検索した．下垂体には7種類の腺細胞が検出されたが，未成魚の甲状腺刺激細胞は塩基性色素で強染され，甲状腺は機能亢進状態を呈していた．成魚の生殖腺刺激細胞には脱顆粒や空胞化した肥大細胞が認められ，経産個体と推定された．神経葉と視束前核およびその軸路には，AF 陽性の神経分泌物が相当量検出された．隆起部外側核は側部と腹内側部の二部からなり，成魚の腹内側部の細胞には酸好性顆粒を充満した小嚢構造がみられたことが特異的であった．

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