ระบบนิวโรเอ็นโดครายน์ ในสัตว์กลุ่มครัสเตเชีย

Neuroendocrine System of the Crustacea

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บทคัดย่อ

ครัสเตเชียน เป็นสัตว์ไม่มีกระดูกสันหลังกลุ่มที่มีเปลือกหุ้ม ขาและลำตัวเป็นปล้อง ร่างกายทั้งสองข้างสมมาตร การสืบพันธุ์และการลอกคราบของสัตว์กลุ่มนี้เป็นปรากฏการณ์ที่เกิดขึ้นอย่างเกี่ยวเนื่องและสลับกันไปตามฤดูกาลภายใต้สภาวะแวดล้อมที่เหมาะสมตลอดอายุขัย กระบวนการที่เกิดขึ้นอย่างสอดรับกันนี้อาศัยการทำงานร่วมกันของทั้งระบบประสาทและฮอร์โมนหลายชนิด โดยการสร้างและหลั่งสารเคมีออกสู่ช่องว่างระหว่างเซลล์หรือระบบไหลเวียนโลหิต เรียกว่า นิวโรฮอร์โมน หรือ ฮอร์โมน ไปออกฤทธิ์ยังเซลล์หรืออวัยวะเป้าหมายที่มีตัวรับของ นิวโรฮอร์โมน หรือ ฮอร์โมนนั้นๆ สำหรับระบบ นิวโรเอ็นโดครายน์ ประกอบด้วยเซลล์ประสาทที่สามารถสร้างและหลั่งสารเคมีออกสู่ระหว่างเซลล์หรือระบบไหลเวียนได้เช่นเดียวกับระบบต่อมไร้ท่อที่สร้างและหลั่งฮอร์โมน เรียกเซลล์ประสาทชนิดนี้ว่า นิวโรซีครีทอรี่เซลล์ หรือ นิวโรเอ็นโดครายน์เซลล์ ซึ่งอาจอยู่รวมกันเป็นอวัยวะ หรือกระจายอยู่ตามเนื้อเยื่อต่างๆ และเรียกสารเคมีที่สร้างและหลั่งโดยเซลล์ชนิดนี้ว่า นิวโรฮอร์โมน ภายหลังพบว่า เซลล์ที่มีบทบาทเป็น นิวโรเอ็นโดครายน์เซลล์ไม่ได้เป็นเซลล์ประสาทอย่างเดียว แต่มีเซลล์อื่นๆ ด้วย จึงมีความพยายามศึกษาลักษณะเฉพาะพร้อมทั้งมีข้อเสนอเกณฑ์ต่างๆ ในการพิจารณานิวโรเอ็นโดครายเซลล์อย่างกว้างขวาง ซึ่งได้สรุปไว้ในบทความนี้แล้ว นอกจากนี้ยังแสดงตำแหน่งของเซลล์ที่ทำหน้าที่สร้างนิวโรฮอร์โมนและบทบาทของนิวโรฮอร์โมนที่สำคัญที่นักวิจัยท่านอื่นๆ และผู้เขียนบทความได้ทำการศึกษาวิจัย

คำสำคัญ : นิวโรเอ็นโดครายน์, ครัสเตเชียน

Abstract

Crustaceans are a group of animals that have a hard exoskeleton, jointed legs, and a segmented body that is bilaterally symmetrical. Reproduction and molting phenomena occur entwined during a large part of crustacean life, which need to be in optimal balance with the environment. These synchronous activities involve multihormonal mechanisms, which act synergistically. Both the nervous and endocrine systems play key roles as regulators of these mechanisms. They produce chemical signals called **neurohormones** or **hormones,** which are released into intercellular spaces or into circulation and affect target cells that express hormone receptors. The neurosecretory cells constitute **neuroendocrine system,** which produce neurohormones and aggregate in form of organs, clusters or is a single cell dispersing among other tissues. Criteria of the neuroendocrine system and special techniques used to investigate functions of the neuroendocrine cells are provided in this review. Mapping of neuroendocrine cells and roles of neurohormones and non-neurohormones are discussed.

Keywords : Neuroendocrine, Crustacea

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Introduction

Crustaceans are a group of animals that have a hard exoskeleton, jointed legs, and a segmented body that is bilaterally symmetrical. They have two pairs of sensory antennae, one pair of mandibles for chewing food, and two pairs of maxillae to help the mandibles in positioning the food. Crustaceans are invertebrates and classified as arthropods, in which insects also belong to this group. Most crustaceans live in water, but some live on land. Crustaceans are the most numerous animals in the oceans, but some crustaceans live on freshwater. There are about 30,500 known species of crustaceans around the world.

Some Characteristics of Crustaceans:

- A hard exoskeleton made of calcium - no internal skeleton.
- The head has two compound eyes, two pairs of antennae, and three pairs of mouthparts.
- A pair of green glands excretes wastes near the base of antennae.
- The abdominal segments have swimmerets (swimming legs)
- The sexes are separate.
- The tail is fan-shaped, and ends in uropods and a telson.
- The circulatory system is open.
- The nervous system consists of a primitive ventral nerve cord and ganglia system (similar to those of an earthworm).

Classification: Kingdom Animalia, Phylum Arthropoda, Subphylum Crustacea, Classes:

- Class **Cephalocarida** (primitive, shrimp-like, discovered in 1955) - 9 species
- Class **Branchiopoda** (with flattened gill-carrying appendages) - about 800 species, including brine shrimp, fairy shrimp, water fleas, etc.
- Class **Malacostraca** - 18000 species, including lobsters, shrimp, crabs, woodlice, isopods, amphipods, krill, etc.
- Class M**axillopoda** (ostracods, copepods, barnacles)
- Class **Remipedia** (primitive crustaceans discovered in submerged caves by Jill Yager in 1980)

(Source: http://www.enchantedlearning.com/subjects/invertebrates/crustacean)

Crustaceans have particularly complex physiological processes that may overlap and influence each other. These processes include dramatically different life stages from embryo to adult. Two phenomena that are entwined during a large part of crustacean adult life are reproduction and molting; these two processes need to be optimal balance with the environment. Somatic growth (molting) and gonadal growth (reproduction) are alternate events of cyclical mobilization of metabolic reserves to the body and gonads. These synchronous activities involve multihormonal mechanisms, which act synergistically (Adiyodi and Adiyodi, 1970) (Fig. 1). The cyclical cellular and biochemical transformations, which a crustacean undergoes between one molt and the next are externally characterized by precise morphological features. They aid in the recognition of the physiological stages of the cycle, which are generally referred to:

Postmolt

Early postmolt (A1-A2)

Late postmolt (B1-B2)

Intermolt (C1-C4)

Premolt (D0-D4)

Molt (E)

The intermolt is a period during which excess metabolites are diverted to reserved food, particularly in the hepatopancreas (Adiyodi and Adiyodi, 1970; Kurup, 1972).

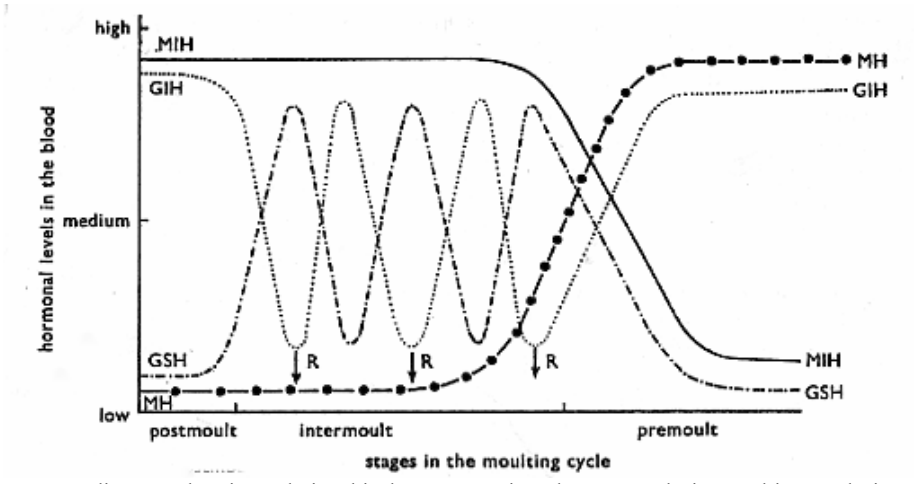

Figure 1. A diagram showing relationship between various hormones during molting cycle in crustaceans. MIH, molt inhibiting hormone; MH, molting hormone or ecdysones; GIH, gonad inhibiting hormone; GSH, gonad stimulating hormone. (from Adiyodi and Adiyodi, 1970)

The molting cycles could be divided into two types; one that does not lead to breeding and the one that does. During intermolt stage of breeding season, the gonads of female crustaceans undergo a sequence of morphological and physiological transformations and entering reproductive cycle. Such changes exhibit a number and classes of oocytes that are undergoing various steps of cellular differentiation. Various stages of oocytes development in female crustaceans have been described for several species. The oocytes development in the giant freshwater prawn *Macrobrachium rosenbergii* have been classified by Meeratana and Sobhon (2007) into four different phases: 1) oogonia, 2) primary oocytes, 3) secondary oocytes, and 4) mature oocytes, while the cycle of ovarian development was classified into five stages based on the number and types of oocytes occupying in each stage: Stage 0, spawn; stage I, spent; stage II, proliferative; stage III, premature; and stage IV, mature.

Neuroendocrine system

Cells in multicellular animals communicate through signaling mechanisms that take place at direct intercellular contacts, or that involve signals released systemically into the extracellular space where they diffuse over large distances and are able to affect targets far from the signaling source. The nervous system and the endocrine system play a key role as regulators of development and almost physiological functions of the processing system in the body. Neurons in the nervous system communicate by specialized cell-cell direct contact to each other via **synapses,** where electrical (nerve impulses) or chemical (neurotransmitters) signals transmitted. The other way of cell-cell communication defined as endocrine mechanism, where chemicals signal called **hormones** released into intercellular spaces or into circulation and affect target cells whose express hormone receptors (Hartenstein, 2006). The endocrine system of animals consists of multiple specialized cell populations, sometimes compact into glands and sometimes disperse in organs. Endocrine glands regulate a large number of homeostatic mechanisms. These include the activity of neurons, muscles, and pigment cells during specific behaviors (food intake, fight and flight, and reproduction), the activity of visceral muscle and exocrine glands (digestion), the control of major metabolic pathways (synthesis, storage, and release of carbohydrates and lipids), the control of the ionic milieu through absorption and excretion, the formation and maturation of gametes, and growth and regeneration of the body. In many instances, endocrine glands form an

integrated neuroendocrine system in which neurohormonal production and releasing is controlled through feed back loops.

In addition to endocrine glands, many neurons of central and peripheral nervous system produce **neurohormones,** which are released locally into extracellular space as well as into circulation. The neurohormones are peptides, glycoproteins and amines (amino acid derivatives), which are also produced by other endocrine cells. Neurons that produce neurohormones are called **neurosecretory cells** or **neuroendocrine cells** or **paraneurons** (Table 1) or amines precursors uptake and decarboxylation **(APUD) cells.** Neurosecretory cells which produce neurohormones, may aggregate in form of organs, clusters or single cell dispersed among other tissues constituted the **neuroendocrine system.** In vertebrates, the neuroendocrine system includes the hypothalamus and pituitary gland (Table 2), as well as peripheral neurons of the autonomic nervous system in the adrenal medulla, the intestinal wall APUD cells and the pancreatic endocrine part (islet of Langerhans) (Hartenstein, 2006) (Table 3). Neuroendocrine cells form a distinct population of neurons, which are recognized by 4 criterions. **First,** they produce a neurotransmitter/ neuromodulator or neuropeptide hormone. **Second,** their products are contained within membrane bound vesicles or granules, i.e. >80 nm dense core secretory granules and 40-80 nm small clear vesicles, from which they are released by a process of exocytosis in response to external (neural) stimuli. **Third,** their mode of transmission is by endocrine or paracrine activities. **Fourth,** neuroendocrine cells share many specific properties and express several markers (amines, amine precursors, amino acid decarboxylase, choline esterase, alpha glycerol-phosphate dehydrogenase, chromogranin, synaptophysin, neural cell adhesion molecule; NCAM etc.) (Langley, 1994).

Table 1 Main members of the paraneuron family (from Langley, 1994)

1. Chromaffin cells of the adrenal medulla.
2. SIF in the autonomic nerve ganglia.
3. Chief cells of the carotid body.
4. Parafollicular cell of the thyroid gland.
5. Parathyroid cell.
6. Anterior pituitary cell.
7. Pancreatic islet cell.
8. Basal granulated cell in the gastroenteric mucosa, pancreatic and bile ducts and urogenital tracts.
9. Gustatory cell.
10. Basal granulated cell in the bronchial epithelium.
11. Hair cells of the inner ear and lateral line organ.
12. Photoreceptor cells of the retina and pinealocyte.
13. Olfactory cell.
14. Liquor contacting neurons.
15. Merkel cell of the skin.
16. Melanocyte.
17. Mast cell.

Table 2 Central neuroendocrine system of vertebrates (from Pearse and Takor, 1979)

Cell type	Peptide products	Amine products
Pineal	Arginine vasotocin, lutropon releasing hormone	Melatonin, 5-hydroxytryptamine
Hypothalamic magnocellular	Arginine vasotocin	Dopamine, norepinephrine, 5-hydroxytryptamine
Hypothalamic parvocellular	Releasing factors, releasing inhibiting factors	-
Pituitaty pars distalis	FSH, LH, TSH, STH, PRL, ACTH, MSH, B-LPH, B-endorphin, gastrin, calcitonin	Dopamine, nor-epinephrine, 5-hydroxytryptamine, histamine (tyramine)
Pituitary pars intermedia	ACTH, MSH, B-LPH, B-endorphin, calcitonin	-

Table 3 Peripheralral neuroendocrine system of vertebrates (from Pearse and Takor, 1979)

Organ	Cell type	Peptide products	Amine products
Pancreas	B, A	Insulin, glucagons	5-hydroxytryptamine
	D, D_2 (F)	Somatostatin, PP	Dopamine
Stomach	G	Gastrin, enkephalin, ACTH	-
	AL	Glucagon	-
	EC_1	Substance P	5-hydroxytryptamine
	ECL	-	Histamine
	D	Somatostatin	-
Intestine	EC_1	Substance P	5-hydroxytryptamine, melatonin
	EC_2(M)	Motillin	-
	L, S	Glicentin(enteroglucagon), secretin	-
	I, D_1(P)	CLK (bombesin)	-
	D, K	Somatostatin, GIP	-
	N	Neurotensin	-
	H	VIP	-
Lung	K(Feyrter)	Bombesin	-
Parathyroid	Chief	Parathyrin	-
Adrenomedullary	E, NE	-	Epinephrine, nor-epinephrine
Sympathetic	Ganglion	VIP	Nor-epinephrine
	SIF	-	Dopamine, nor-epinephrine
Carotid body	Type I	-	Dopamine, norepinephrine
Epidermis	Melanoblast, cyte	-	Premelanin
Thyroid,	C ultimobranchial	Calcitonin, somatostatin	5-hydroxytryptamine
Urogenital tract	EC, V	-	-

PP; pancreatic polypeptides, ECL; enterochromaffin, SIF; small intensely fluorescent,

Furthermore, releases of neurohormones occur through mechanism of fusion of the secretory vesicles with the cell membrane at synapses or anywhere along the soma and axon into intercellular spaces or circulation. In this way, the released neurohormone affects multiple cells containing specific receptors (Fig. 2).

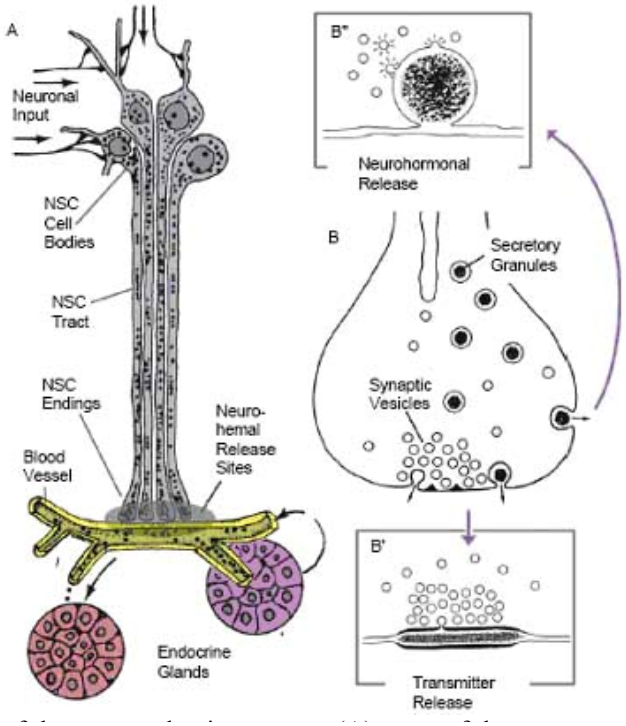

Figure 2 Structure of the neuroendocrine system. (**A**) soma of the neurosecretory cells (NSC) are located in the central nervous system and receive neuronal input from presynaptic neurons. NSC axons project to peripheral neurohemal release sites that are frequently in close contact with endocrine cells targeted by the neurohormones release at the NSC terminals. (**B**) Ultrastructural aspects of neurotramsmitter release (**B'**) and neurohemal release (**B"**). Neurotransmitter release occurs exclusively at presynaptic site from 50 nm vesicles. Neurohormones are store in large vesicles found throughout the NSC and released outside synapses. (from Hartenstein, 2006.)

Investigations on various aspects of neuroendocrine cells could be carried out by a variety of techniques, such as by special histological staining, immunocytochemistry, electron microscopy, *in situ* hybridization and by molecular biology techniques.

It has been proposed that the neuroendocrine system developed from specialized epithelial cells integrated into the epidermis and intestinal lining and react to certain stimuli, e.g. chemicals or physicals, by secreting metabolites that diffuse to target cells via interstitial fluid or blood circulation and evoke adaptive responses (Fig. 3) (Hartenstein, 2006).

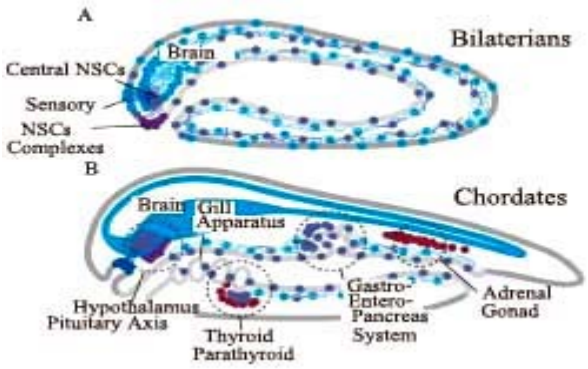

Figure 3 Hypothetical stages in the evolution of the endocrine and neuroendocrine systems. Populations of sensory neurosecretory cells (NSCs) involved in the regulation of fundamental biological processes, such as feeding and reproduction may have formed specialized complexes in the brain, pharynx and gut of early bilaterians (**A**). During later stages of evolution for the chordate lineage (**B**), NSCs and endocrine cells in general show tendency of losing their sensory function, delaminating from the surface epithelium (epidermis, pharynx and intestinal epithelium), and undergoing morphogenetic changes that produced delicate endocrine glands, such as the pituitary, thyroid, parathyroid and pancreatic islets. (from Hartenstein, 2006.)

In invertebrates, the neuroendocrine system is organized and modified in different patterns according to body forms and functions of individual organs (Fig. 4).

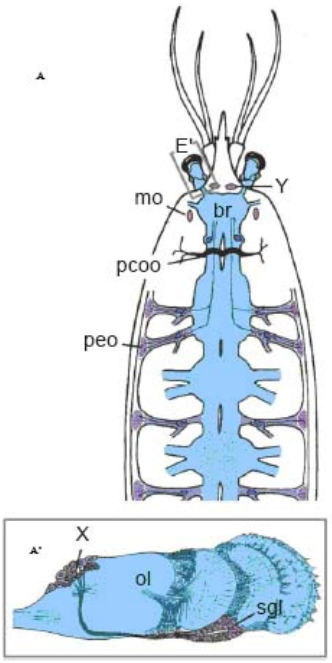

Figure 4 Important elements of the neuroendocrine system in crustaceans. The upper panel shows general view of the organization and the lower panel shows details of the optic lobe (ol). Neurosecretory cells (NSCs) are mostly located in the brain (br) and X-organ (X) in the optic lobe. The X-organ send axons to the sinus gland (sgl) where several types of neurosecretory hormones are released at the axon terminals. Axons from NSCs of the brain, X-organ and other places also terminate and release special neurosecretory hormones at postcommissural organ (pcoo), pericardial organ (peo), mandibular organ (mo) and Y-organ (Y). The pericardial organ is possibly homologous to the insect corpora cardiaca. The Y-organ produces ecdysone and represents a homolog of the insect thoracic gland; likewise, the mandibular organ secretes methyl farnesoate which is chemically and functionally similar to insect juvenile hormone. (from Hartenstein, 2006)

In crustaceans, neurosecretory cells have been identified in the brain, ganglia (Fig. 5), optic lobes and ventral nerve cord. The central neuroendocrine cells of crustaceans are considerably diversified; they send processes to neuropiles (the region of the central nervous system where the synapses and neurons branching and reorganization occur), which have specialized projection to neurohemal organs. A conspicuous group of crustacean neuroendocrine cells is **X-organ** at proximal part of optic lobe in the eyestalk. Axons of X-organ and of neuroendocrine cells in the brain project to the eyestalk neurohemal organ called the **sinus gland,** located on ventral surface of optic lobe. The peripheral neurohemal organs of crustacean called **postcommissural organ** and **pericardial organ,** received projections from neuroendocrine cells in the brain and ventral nerve cord (Figs. 6 and 7).

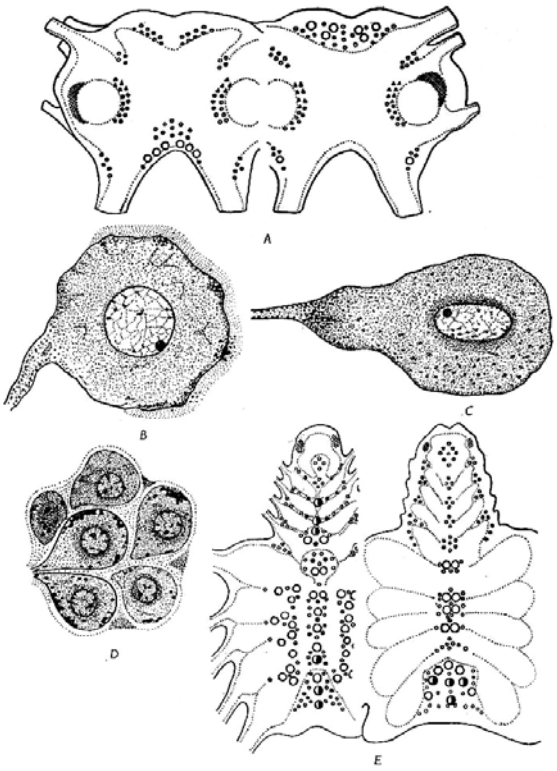

Figure 5 Neurosecretory cells and their distribution in the brain and thoracic ganglion of crabs. **A,** ventral (left) and dorsal (right) views of the brain of Potamon dehanni (White). Larger circles represent the A-cells, smaller circles the B-cells and closed triangles the E-cells. **B,** A-cells of Potamon dehanni; **C,** A'-cells of chionoecetes oplio (Fabricius); **D,** E-cells of Potamon dehanni; **E,** ventral (left) and middle (right) views of the thoracic ganglion of Potamon dehanni. Half closed large and small circles represent the A'- and Bอ-cells respectively. (from Adiyodi and Adiyodi,1970)

Crustaceans use external factors, predominantly temperature and photoperiodicity for timing of their biological activities. Precise timing of biological processes is very important in most crustacean species. For instance, reproduction and molting are two phenomena, which need to be balance with the environment. The modulation of these synergistic biological activities is based on a multi-hormonal system.

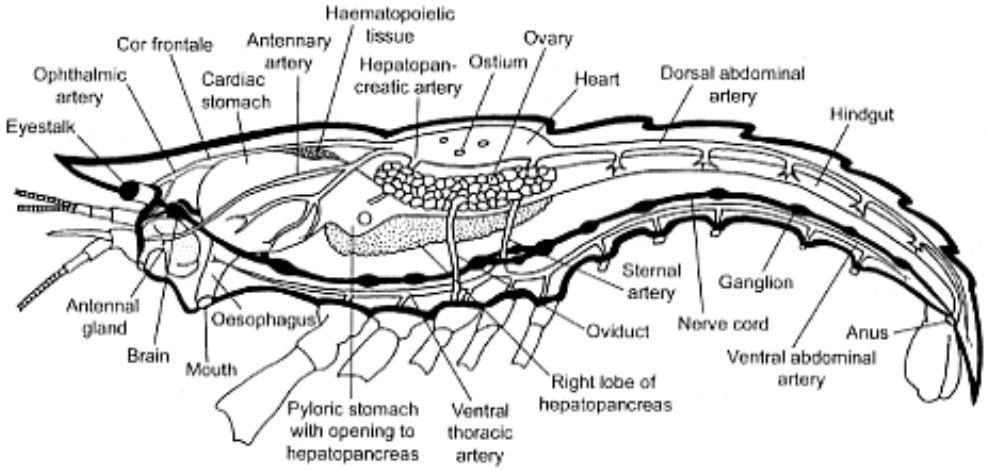

Figure 6 A diagram of a female crayfish showing main organ systems, particularly the nervous and circulatory systems

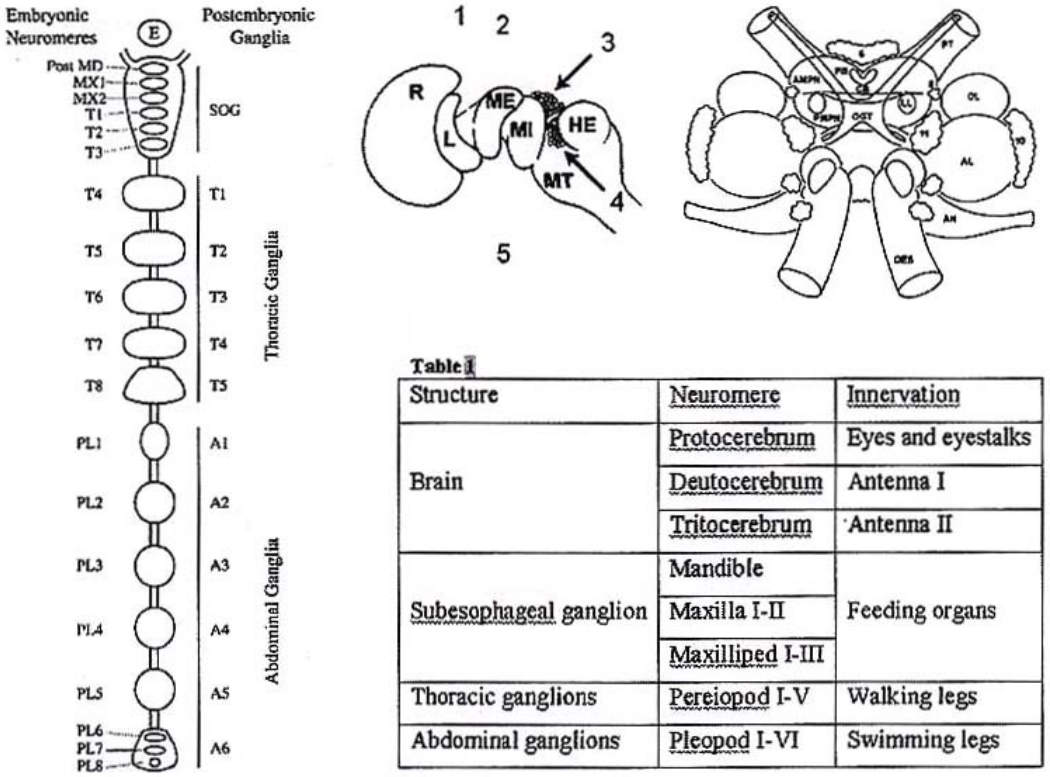

Table 1

Structure	Neuromere	Innervation
Brain	Protocerebrum	Eyes and eyestalks
	Deutocerebrum	Antenna I
	Tritocerebrum	Antenna II
Subesophageal ganglion	Mandible	Feeding organs
	Maxilla I-II	
	Maxilliped I-III	
Thoracic ganglions	Pereiopod I-V	Walking legs
Abdominal ganglions	Pleopod I-VI	Swimming legs

Figure 7 A diagram showing decapod ventral nerve cord (left), eyestalk (middle) and brain (right). The table indicates division and groups of neurons in the nervous system with organs of innervation. A, abdominal ganglion; MD, mandible; MX, maxilla; PL, pleopod; T, thoracic ganglion; SOG, subesophageal ganglion; HE, hemi-ellipsoid body; L, lamina ganglionaris; ME, medulla externa; MI, medulla interna; MT, medulla terminalis; R, retina; 3, X-organ; 4, sinus gland. (from Utting et al., 2000)

The neuroendocrine system in crustacean could be categorized into central and peripheral neuroendocrine organs. The central neuroendocrine organs lie in central nervous system, brain and ventral nerve cord, including X-organ sinus gland complex and diffused neuroendocrine cells. The peripheral neuroendocrine organs are structures composed of cells that are differentiated from the neuroectoderm, or are axon terminals of neuroendocrine cells, of which cell bodies are located in central nervous system. These organs lie external to the central nervous system.

The Central Neuroendocrine Organs

The central neuroendocrine organs are composed of X-organ-sinus gland complex (XOSG) and diffused endocrine cells. The XOSG is the major neuroendocrine structures that store and release a wide variety of neurohormones. Three major groups of neurohormones produced by XOSG are reproductive, molting and metabolic, and color regulating groups.

The reproductive neurohormones are vitellogenesis inhibiting hormone (VIH) and mandibular organ inhibiting hormone (MOIH). The VIH, also known as gonad inhibiting hormone (GIH), is a peptide that inhibits growth and differentiation of oocytes by decreasing vitellogenin (egg yolk protein) synthesis in the ovary and hepatopancreas and inhibiting vitellogenin uptake by the oocytes. Removal of eyestalk, which removes the XOSG and concomitantly VIH, results in sexual maturation (Van Herp and Soyez, 1997) (Fig. 8).

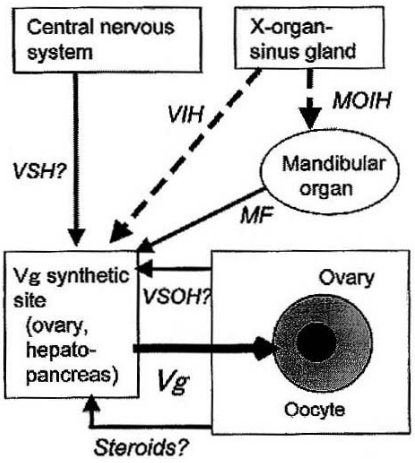

Figure 8 The endocrine contributing to vitellogenesis in shrimp. MF, Methyl farnesoate; MOIH, mandibular organ inhibiting hormone; Vg, vitellogenin; VIH, vitellogenin inhibiting hormone; VSH, vitellogenin stimulating hormone; VSOH, vitellogenin stimulating ovarian hormone. (from Okumura, 2004)

The MOIH is a peptide that inhibits methyl farnesoate secretion in the mandibular organ. Eyestalk removal (ablation) resulted in hypertrophy of the mandibular organ and increase hemolymph level of methyl farnesoate, which stimulate ovarian maturation in some species (Van Herp and Soyez, 1997).

The molting and metabolic neurohormones are composed of molt inhibiting hormone (MIH) and crustacean hyperglycemic hormone (CHH). The MIH is synthesized in the X-organ of medulla terminalis of the optic lobe. Its bulbous axonic terminals ended in the sinus gland (Huberman, 2000). Eyestalk ablation results in significant acceleration of molting cycle, while injection of XOSG extract delays molting of eyestalk-ablated animal (Naya et al., 1989). MIH also inhibits ecdysteroid (ecdysone) synthesis in Y-organ. Decreasing in MIH level permits increased ecdysteroid synthesis, leading to permissive entry of premolt and subsequently molting (Chung and Webster, 2005). During molting, MIH and gonad stimulating hormone (GSH) levels are low, while GIH is high (Wongsawang et al., 2005). Thus, MIH is important not only in controlling molting, but also in regulating reproductive cycle throughout the female life.

The CHH molecule is highly conserved among crustaceans. The CHH family synthesized in the neuroendocrine cells of the medulla terminalis X-organ and transported via axon bundle toward the storage and releasing site in the sinus gland (De Keijn et al., 1998). CHHs are primary involved in regulation of blood sugar level, regulates release of glucose from hepatopancreas and may play a stimulatory role in reproduction. In the lobster, *Homarus americanus*, CHH has two isoforms producing different in functions of osmoregulation and oocytic growth (Van Herp and Soyez, 1997) and several neurotransmitters influence CHH release (Fig. 9).

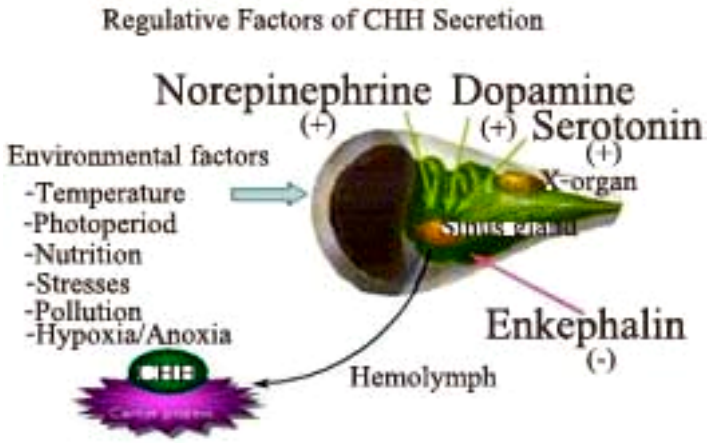

Figure 9 Factors influencing x-organ-sinus gland complex on CHH release. DA, dopamine; 5-HT, 5-hydroxytryptamine, NE, norepinephrine; (+), stimulation; (-), inhibition. (from Ching-Ming Kuo, 2004)

The color regulating neurohormones, also known as chromatophorotropins with primary effects on pigmentation, are composed of red pigment concentrating hormone (RPCH) and distal retinal pigment dispersing hormone (DRPH). Both hormones are members of pigment concentrating hormone (PCH) family, is synthesized by neurosecretory cells in the XOSG. The RPCH is an octapeptide, whereas the DRPH is an octadecapeptide. The RPCH mediates calcium influx, which stimulates aggregation of pigmentary matrix in erythrophores (Alvaraldo-Alvarez et al., 1999). The RPCHergic neurons also have neuromodulatory roles regulating the somatogastric nervous system in crustaceans (Dickinson et al., 2001). RPCH seems to be highly conserved in crustaceans and present only one form in each species (Van Herp and Soyez, 1997). For the DRPH, one species may contain more than one form. Its function is related to light adaptation of the eye and it also acts as dark-adapting pigment hormone; in some species, it may control proximal and reflecting pigments (Van Herp and Soyez, 1997).

Besides the color effect, PDH also inhibits gonadal development in spiny lobster Palinurus argus (Quakenbush and Herrnkind, 1983). Moreover, RPCH could stimulate methyl farnesoate production in mandibular organ (Landau et al., 1989). These actions suggest that chromatophorotropins may play some roles in crustacean reproduction as well.

The XOSG also produces neurotransmitters/ neuromodulators like serotonin, norepinephrin, FMRFamide, APGW-amide, enkephalins and substance P. These substances regulate varieties of crustacean physiological activities including gonadal and somatic growth. Releases of the XOSG neurohormones are induced by nerve impulses to the neuroendocrine cells and environmental stimuli (day length, temperature, osmolarity), stress, limb autonomy as well as an internal oscillator.

In addition to neuroendocrine cells that are well-organized in the central nervous system, there are many neurons that are distributed in various parts of the brain and ventral nerve cord of crustaceans. These diffused cells send their axons to terminate near hemolymphatic vessels or in specialized neurohemal organs. One of the significant neurohormones produced by this kind of neuroendocrine cells in crustacean thoracic ganglia is gonad stimulating hormone (GSH), which is also known as vitellogenesis stimulating hormone (VSH). The hormone may be synthesized by type A neurons in the brain and thoracic ganglion and related to ovarian development since the neurons are apparently active during ovarian developmental period (Adiyodi and Adiyodi, 1970). The contribution of the thoracic ganglia to reproduction has been shown in several studies (Hinsch, 1972, Hasegawa et al., 1993, Meeratana et al., 2006, Rodriguez et al., 2007). For instances, implantation of thoracic ganglia into the freshwater crab *Polumon dehaani* broodstock caused ovarian maturation (Otsu, 1963) and incubation of ovarian tissue with media taken from thoracic ganglion culture resulted in oocytic maturation in the giant freshwater prawn *Macrobrachium rosenbergii* (Meeratana et al., 2006). The GSH appears to have dual activities in promoting oocytes growth and, at the same time, suppressing Y-organ function. The suppression of Y-organ, which is the site of ecdysteroid synthesis, causes an inhibition of the molting process.

Other diffused cells are gonadotropin-releasing hormone (GnRH) cells, which were found to localize as medium-sized neurons of anterior protocerebrum and in nerve fibers extending to other brain areas, thoracic and abdominal ganglia and follicular cells of previtellogenic, vitellogenic and mature ovary of the black tiger shrimp *Penaeus monodon*. The hormone was shown to induce ovarian maturation (Ngernsoungnern, 2007).

Neurons that produce egg laying hormone (ELH), a peptide found in the neural tissue, antennal gland and follicular cells of P. *monodon* (Liu et al., 2006). Levels of ELH increases prior to shrimp spawning and the hormone is co-localized with GnRH in medium-sized neurons of protocerebrum (Ngernsoungnern, 2007).

Serotonin or 5HT, a biogenic amine synthesize from amino acid, tryptophan, is involved in higher-order behavior e.g. feeding, aggression and sexual maturation of vertebrates and invertebrates. 5-HT plays roles as neurotransmitter, neuromodulator and neurohormone in crustacean. 5-HT-like immunoreactivity was found in neurons and neuropils of *M. rosenbergii* brain, subesophageal, thoracic, and abdominal ganglia (Soonthornsumrith, 2006, Meeratana, et al., 2006). It was claimed to induce ovarian maturation indirectly via GSH stimulation in the thoracic ganglia, as well as shortening the embryonic developmental period and early appearance of primordial germ cells in *M. rosenbergii,* probably by triggering on signal transduction and subsequently potentiating differentiation and proliferation processes of the embryonic germ cells (Meeratana, 2001).

The Peripheral Neuroendocrine Organs

These are organs or glands that contain neuroendocrine cells or are axon terminals of neuroendocrine cells, of which cell bodies are located in central nervous system; and they lie external to the central nervous system. The peripheral neuroendocrine organs are composed of the post commissural organ and the pericardial organ.

The post-commisural organ (PCO) is a neurohemal organ, which means it is closed to or surrounded by hemolymphatic pool. It consists mainly axon terminals of neuroendocrine cells, and functions as a storage and release site of neurohormones. Most of the axon terminals emerge from the tritocerebral commissure that lies posterior to the esophagus. Extracts of the PCO and tritocerebral commissure of *Penaeus* spp., *Palaemon* spp. and *Uca pugilator* contained red and white pigment concentrating hormones (Fingerman, 1992).

The pericardial organ (PO) is also a neural plexuses containing terminals of the neuroendocrine axons, of which their cell bodies are in thoracic and subesophageal ganglia (Pulver and Mareder, 2002). These axons branch from segmental nerve cord and terminate on lateral wall of the pericardial cavity. The PO produces neurohormones to regulate the heart. Pericardial organ also contains intrinsic endocrine cells producing crustacean hyperglycemic hormone (CHH) that regulates hemolymph sugar and fatty acid levels, and probably affects heart beat and molting as well (Hartenstein, 2006).

Besides the neuroendocrine cells described, there are several organs that are not neurons. These neuroendocrine organs, which are Y-organ, mandibular organ, androgenic gland, ovary and organ of Bellonci, are closely related to endocrine glands in mammals.

Y-organ has an ectodermal origin and is attached to the epidermis in maxillary segment of the head. This gland has typical compact ovoid shaped lying ventral to the insertion of lateral portion of the external adductor muscle of the mandible, above the attachment point of branchiotegite and cuticle at the anterior end of the gill chamber (Figs. 6 and 10). The organ converts dietary cholesterol into **ecdysone,** which is secreted into the hemolymph. Ecdysone is then transport to the peripheral tissues, where it is converted to the bioactive hemolymphatic form, **20-hydroxyecdysone,** titers of which is increased just prior to molting (Lachaise et al., 1993). The role of ecdysteroids on regulation of female reproduction remains unclear.

The mandibular organ is a multi-lobed organ attached to the ventral epidermis at the posterior surface of each mandible and just posterior to the base of the posterior mandibular abductor muscle (Figs. 6 and 10). The mandibular organs produce **methyl farnesoate (MF)** and **farnesoic acid,** which play an important role on crustacean morphogenesis and reproduction. MF may be one of the driving forces behind the mating behavior of male crustaceans. The roles of MF on ovarian development are not universal in crustaceans (Okumura, 2000). Production activity of MF in mandibular organs is regulated by MOIH from the XOSG (Fig. 10).

The androgenic gland is found in almost all Malacostraca Order. The gland is attached posteriorly to the distal part of the vas deferens (Fig. 6) and produces **androgenic gland hormone,** which regulates spermatogenic activity in the testes and are also responsible for the development and maintenance of male secondary sexual characteristics (Fig. 10) (Adiyodi and Adiyodi, 1970).

The ovary is the source of a hormone that induces formation of female secondary sexual characteristics. The ovarian hormone called **vitellogenin stimulating ovarian hormone (VSOH)** contributes to the stimulation of vitellogenin synthesis in fat bodies (Figs. 8 and 10). The site of hormone synthesis in ovary is the primary follicular cells (Okumura, 2004).

The organ of Bellonci is also known as the **onion bodies,** a compact structure consisting of several packed lobules, locates in the eyestalk of decapod crustaceans. Its functions are still not firmly established. Some researcher found ciliary-types structure with basal body, suggesting that it is a sensory organ (Fingerman, 1992).

Vertebrate-type Steroid Hormones

A numbers of vertebrate steroid hormones have been identified in crustaceans, including **pregnenolone, 17ß-estadiol, 17α-progesterone** and **progesterone.** The levels of these hormones in the ovaries, hepatopancreas and hemolymph are closely related to the stage of ovarian development (Okumura, 2004).

Invertebrate Ecosanoids

Fatty acid derivatives presents in various crustacean tissues including in the ovary. **Prostaglandin E-2, F-2α** and **H2** have been reported in ovarian tissues of crayfish and the giant freshwater prawn, *Macrobrachium rosenbergii*, suggesting that may have a role in the endocrine regulation of crustacean reproduction (Van Herp and Soyez, 1997; Wongprasert et al., 2006).

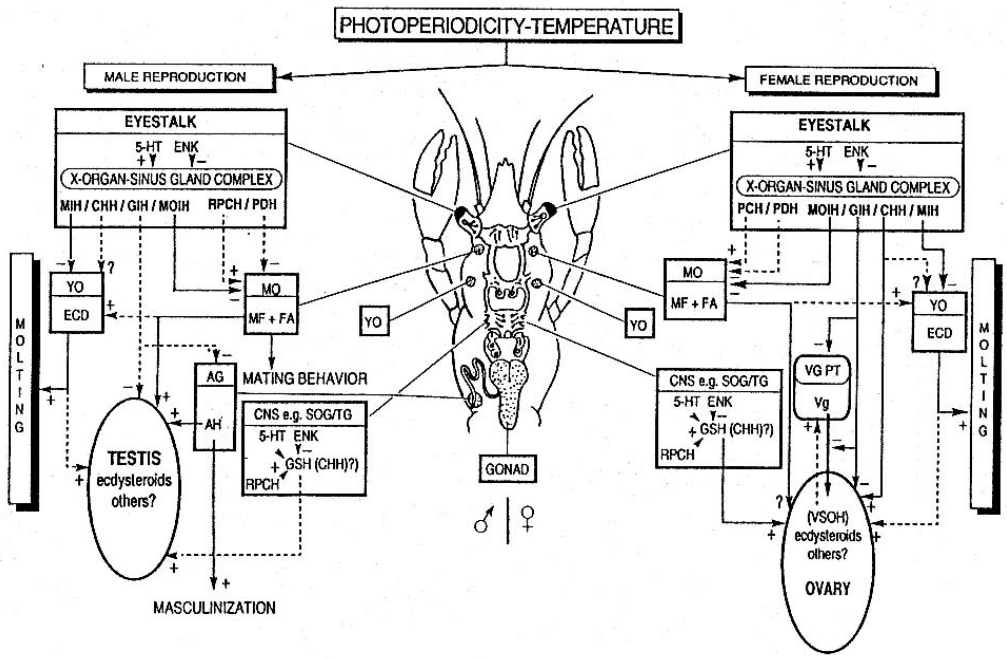

Figure 10 Hormonal regulation of reproduction in male and female crustaceans: Schemiv representation of the neuroendocrine and endocrine organs and factors involved in the controlling pathways. AG, androgenc gland; AH, androgenic hormone; CHH, crustacean hyperglycemic hormone; CNS, central nervous system; ECD, 20-hydroxyecdysone; ENK, enkephalin; FA, farnesoic acid; GIH, gonad inhibiting hormone; GSH, gonad stimulating hormone; 5-HT, 5-hydroxy tryptamine; MF, methyl farnesoate; MIH, molt inhibiting hormone; MO, mandibular organ; MOIH, mandibular organ inhibiting hormone; RPCH, red pigment concentrating hormone; PCH, pigment concentrating hormone; PDH, pigment dispersing hormone; SOG, supraesophageal ganglion; TG, thoracic ganglion; VG, vitelloghenin; VGPT, vitellogenin producing tissue; VSOH, vitellogenin stimulating ovarian hormone; YO, Y-organ (from Van Herp and Soyez, 1997)

Conclusions

Shrimp aquaculture in the world has developed remarkably; however, to enable further development, new technological advances in hormonal manipulation of shrimp reproduction are increasingly important for effective stock enhancement. To develop hormonal manipulation techniques, progress in shrimp endocrinology is necessary (Okumura, 2004).

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