

## Commentary

# Inhibitory Control of the Brain-Pituitary Reproductive Axis of Male European Sea Bass: Role of Gonadotropin Inhibitory Hormone

Hongwei Yan<sup>1</sup>

College of Fisheries and Life Science; Center for Marine Ranching Engineering Science Research of Liaoning, Dalian Ocean University, Dalian, China

Reproduction in vertebrates is controlled by a hierarchically organized endocrine system called the hypothalamic-pituitary-gonadal (HPG) axis. In the early of 1970s, two teams, Schally's and Guillemin's, discovered gonadotropin-releasing hormone (GnRH), a hypothalamic neuropeptide that stimulates the release of both luteinizing hormone (LH) and follicle-stimulating hormone (FSH) from gonadotropes in the anterior pituitary of mammals [1–3]. Subsequently, several GnRHs have been identified in other vertebrates [4–6], and it is generally accepted that GnRH is the only hypothalamic neuropeptide that regulates the release of gonadotropin in mammals and other vertebrates. Despite the divergent reproductive strategies and behaviors within this taxon, this endocrine network is remarkably well conserved across vertebrates. In response to hypothalamic GnRH, gonadotropins are secreted from the pituitary and stimulate the gonads, where they induce the synthesis and release of sex steroid hormones, which in turn elicit growth and maturation of the gonads.

However, control of pituitary function by a single neuroendocrine mechanism is not always the rule in vertebrates in which dual (positive and negative) hypothalamic control has often been observed in somatotropes, thyrotropes, and lactotropes. Several investigations have challenged the dogma of single control of gonadotropes by GnRH. Peter et al. [7–10] were the first to establish that a dopaminergic inhibitor can control reproduction in goldfish *Carrassius auratus*. After that, the role of dopamine (DA) as a gonadotropin release-inhibiting factor was confirmed not only in teleosts, but also in chondrosteian fish [11–15]. The discovery of DA inhibition led to the development of a new method to induce spawning in aquaculture, using a combined treatment with a GnRH agonist and a DA-D2 type receptor antagonist. This method, called LinPe, is now widely applied in aquaculture as an alternative means of inducing spawning because it is more reliable and physiologically relevant than treatments involving pituitary extracts. However, wide variations in the intensity of DA inhibition have been noted among the species expressing the DA inhibition, indicating a major role for DA in some species, such as the cyprinids, and a minor one in others, such as the salmonids. Interestingly, some species (e.g. the Atlantic croaker *Micropogonias undulatus*, gilthead seabream *Sparus aurata*, striped bass *Morone saxatilis*, sea bass *Dicentrarchus labrax*, and red seabream *Pagrus major*) do not exhibit a dopaminergic inhibition of gonadotropin [16–20]. These

variations suggest that DA neuroendocrine inhibition may have been differentially conserved and expressed among teleosts in relation to the diversity of their reproductive cycles and their dependency upon various environmental factors. However, it is also possible that other inhibitory factors may be involved in the reproduction neuroendocrine systems in fish.

In 2000, a hypothalamic neuropeptide, referred to as gonadotropin inhibitory hormone (GnIH), was first discovered in birds. This neuropeptide actively inhibits the release of gonadotropin [21]. Subsequently, GnIH homologous peptides have also been identified in the hypothalamuses of other vertebrates from agnathans to humans. Moreover, GnIH and its homologous peptide inhibit the release of gonadotropin in higher vertebrates. In a search for novel factors regulating reproduction of fish using the *D. labrax* as a model [22], Paullada-Salmerón and colleagues isolated a GnIH ortholog precursor containing two putative GnIH peptides (sbGnIH-1 and sbGnIH-2). However, studies in fish have shown both stimulatory and inhibitory roles of GnIH in the reproductive axis by using pituitary cultures and intraperitoneal injection.

In the current issue of *Biology of Reproduction*, Paullada-Salmerón and colleagues demonstrate an inhibitory role of GnIH in the reproductive axis of the male sea bass [23]. Unlike earlier studies, the present study by Paullada-Salmerón et al. introduced GnIH by intracerebroventricular (icv) injection. Using icv injection of sbGnIH-1 and sbGnIH-2, Paullada-Salmerón et al. examined their effects on the brain and pituitary expression of reproductive hormone genes (*gnrh1*, *gnrh2*, *gnrh3*, *kiss1*, *kiss2*, *gnih*, *lhbeta*, *fshbeta*) and their receptors (*gnrhr II-1a*, *gnrhr II-2b*, *kiss1r*, *kiss2r*, and *gnih*) as well as on the plasma FSH and LH levels [23]. In addition, they determined the effects of GnIH on pituitary somatotropin (GH) expression. The results showed the distinct actions of sbGnIH-1 and sbGnIH-2 on the reproductive axis of sea bass. The icv administration of sbGnIH-1 was found to induce a decrease in *gnrh1* mRNA levels in the brain and provoked a significant decrease in plasma LH levels. In turn, sbGnIH-2 suppressed *gnrh2*, *gnih*, *gnih*r, *kiss1*, *kiss2*, and *kiss1* receptor mRNA levels in the brain, *lhβ*, *fshβ*, and *gh* mRNA levels in pituitary, and LH levels in plasma. Considering the previous observations, especially the evidence from other vertebrates and morphological characteristics of the peptide interactions in sea bass, several conclusions can be drawn. First, some of the actions of GnIH on the reproductive axis that inhibit the release of gonadotropin might be centrally mediated through indirect action of sbGnIH-1 at the level of the preoptic/hypothalamic GnRH-1 neurons. The reduction in plasma LH levels could be correlated with the inhibition of brain *gnrh1* expression

<sup>1</sup>Correspondence: College of Fisheries and Life Science; Center for Marine Ranching Engineering Science Research of Liaoning, Dalian Ocean University, 52 Heishijiao Street, Dalian, Liaoning province 116023. E-mail: yanhongwei@dlo.edu.cn

observed in fish treated with sbGnIH-1. Second, the inhibitory effects of GnIH-2 on *gnrh2* transcription levels might relate to the sexual behavior or the mediation of photoperiodic control of reproduction in this species. Third, sbGnIH might modulate the reproductive axis of sea bass through direct central action not only on GnRH but also on kiss2 neurons. Fourth, sbGnIH-2 could regulate the release of gonadotropin by acting directly on the adenohypophyseal cells in which the sbGnIH-2-immunoreactive fibers are in close proximity to FSH and LH cells. It is also possible that the action of sbGnIH-2 on gonadotropin synthesis and/or release could be mediated, at least in part, by its modulation of pituitary GnRH signaling. Finally, the sbGnIH-2 form exerts its main autocrine regulation through a negative feedback on the brain GnIH system.

In summary, GnIH might participate not only in neuroendocrine functions but also in behavioral and photoperiodic control of reproduction in this species with the main inhibitory role being mediated by sbGnIH-2. This work, for the first time in teleosts, examined the effects of GnIH on the whole HPG axis. There is no doubt it not only makes a significant contribution to understanding of the regulatory network controlling the HPG axis in fish but also provides new evidence for the role of GnIH inhibition in vertebrate reproduction. However, there are still many open questions to be answered, such as whether 1) GnIH is a substitutor or coregulator of dopamine in the reproductive axis in fish, 2) GnIH signaling is involved in the regulation of sexual behavior and mediation of the photoperiodic control of reproduction in teleosts and do sbGnIH/Kiss1/pineal interactions participate in the sexual maturation of sea bass in relation to photoperiods, and 3) the activity of GnIH are similar in female sea bass and in animals at different reproductive stages. Addressing these matters will require an intensive research effort, but it holds the promise of generating more exciting discoveries thereby providing a more complete picture of the regulatory network that control reproduction in this species.

## REFERENCES

- Matsuo H, Baba Y, Nair RM, Arimura A, Schally AV. Structure of the porcine LH- and FSH-releasing hormone. I. The proposed amino acid sequence. *Biochem Biophys Res Commun* 1971; 43:1334–1339.
- Burgus R, Butcher M, Ling N, Monahan M, Rivier J, Fellows R, Amoss M, Blackwell R, Vale W, Guillemin R. Molecular structure of the hypothalamic factor (LRF) of ovine origin monitoring the secretion of pituitary gonadotropic hormone of luteinization (LH). *C R Acad Sci Hebd Seances Acad Sci D* 1971; 273:1611–1613.
- Guillemin R. Hypothalamic hormones a.k.a. hypothalamic releasing factors. *J Endocrinol* 2005; 184:11–28.
- Miyamoto K, Hasegawa Y, Nomura M, Igarashi M, Kangawa K, Matsuo H. Identification of the second gonadotropin-releasing hormone in chicken hypothalamus: evidence that gonadotropin secretion is probably controlled by two distinct gonadotropin-releasing hormones in avian species. *Proc Natl Acad Sci U S A* 1984; 81:3874–3878.
- Powell JF, Reska-Skinner SM, Prakash MO, Fischer WH, Park M, Rivier JE, Craig AG, Mackie GO, Sherwood NM. Two new forms of gonadotropin-releasing hormone in a protochordate and the evolutionary implications. *Proc Natl Acad Sci U S A* 1996; 93:10461–10464.
- Sherwood N, Eiden L, Brownstein M, Spiess J, Rivier J, Vale W. Characterization of a teleost gonadotropin-releasing hormone. *Proc Natl Acad Sci U S A* 1983; 80:2794–2798.
- Peter RE, Paulencu CR. Involvement of the preoptic region in the gonadotropin release-inhibition in the goldfish. *Neuroendocrinology* 1980; 31:133–141.
- Peter RE, Crim LW, Goos HJTh, Crim JW. Lesioning studies on the gravid female goldfish: neuroendocrine regulation of ovulation. *Gen Com Endocrinol* 1978; 35:391–401.
- Peter RE, Chang JP, Nahorniak CS, Omeljaniuk RJ, Sokolowska M, Shih SH, Billard R. Interactions of catecholamines and GnRH in regulation of gonadotropin secretion in teleost fish. *Recent Pro Horm Res* 1986; 42: 513–548.
- Peter RE, Trudeau V, Sloley BD, Peng C, Nahorniak CS. Actions of catecholamines, peptides and sex steroids in regulation of gonadotropin-II in the goldfish. In: Scott AP, Sumpter JP, Kime DE, Rolfe MS (eds.), *Reproductive Physiology of Fish*. Sheffield, United Kingdom; Department of Biological Sciences, University of Sheffield; 1991:30–34.
- Lin HR, Karaak GVD, Zhou XJ, Liang JY, Peter RE, Rivier JE, Vale WW. Effects of [D-Arg6, Trp7, Leu8, Pro9NET]-luteinizing hormone-releasing hormone (sGnRH-a) and [D-Ala6, Pro9NET]-luteinizing hormone-releasing hormone (LHRH-a), in combination with pimozone or domperidone, on gonadotropin release and ovulation in the Chinese loach and common carp. *Gen Comp Endocrinol* 1988; 69:31–40.
- de Leeuw R, Goos HJ, van Oordt PG. The dopaminergic inhibition of the gonadotropin-releasing hormone-induced gonadotropin release: an in vitro study with fragments and cell suspensions from pituitaries of the African catfish, *Clarias gariepinus* (Burchell). *Gen Comp Endocrinol* 1986; 63: 171–177.
- Saligaut C, Linard B, Breton B, Anglade I, Baihache T, Kah O, Jégo P. Brain aminergic systems in salmonids and other teleosts in relation to steroid feedback and gonadotropin release. *Aquaculture* 1999; 177:13–20.
- Yaron Z, Gur G, Melamed P, Rosenfeld H, Elizur A, Levavi-Sivan B. Regulation of fish gonadotropins. *Int Rev Cytol* 2003; 225:131–185.
- Pavlick RJ Jr, Moberg GP. Dopaminergic influence on gonadotropin secretion in white sturgeon. *Fish Physiol Biochem* 1997; 16:35–43.
- Copeland PA, Thomas P. Control of gonadotropin release in the Atlantic croaker (*Micropogonias undulatus*): evidence for lack of dopaminergic inhibition. *Gen Comp Endocrinology* 1989; 74:474–483.
- Zohar Y, Harel M, Hassin S, Tandler A. Broodstock management and manipulation of spawning in the gilthead seabream, *Sparus aurata*. In: Bromage NR, Roberts RJ (eds.), *Broodstock Management and Egg and Larval Quality*. London, United Kingdom: Blackwell Press; 1994:94–114.
- Holland MC, Hassin S, Zohar Y. Effects of long-term testosterone, gonadotropin-releasing hormone agonist, and pimozone treatments on gonadotropin II levels and ovarian development in juvenile female striped bass (*Morone saxatilis*). *Biol Reprod* 1998; 59:1153–1162.
- Prat F, Zanuy S, Carrillo M. Effect of gonadotropin-releasing hormone analogue GnRH<sub>a</sub> and pimozone on plasma levels of sex steroids and ovarian development in sea bass *Dicentrarchus labrax* L. *Aquaculture* 2001; 198:325–338.
- Kumakura N, Okuzawa K, Gen K, Kagawa H. Effects of gonadotropin-releasing hormone agonist and dopamine antagonist on hypothalamus-pituitary-gonadal axis of prepubertal female red seabream (*Pagrus major*). *Gen Comp Endocrinol* 2003; 131:264–273.
- Tsutsui K, Saigoh E, Ukena K, Teranishi H, Fujisawa Y, Kikuchi M, Ishii S, Sharp PJ. A novel avian hypothalamic peptide inhibiting gonadotropin release. *Biochem Biophys Res Commun* 2000; 275:661–667.
- Paullada-Salmerón JA, Cowan M, Aliaga-Guerrero M, Gómez A, Zanuy S, Mañanos E, Muñoz-Cueto JA. LPXRFa peptide system in the European sea bass: a molecular and immunohistochemical approach. *J Comp Neurol* 2016; 524:176–198.
- Paullada-Salmerón JA, Cowan M, Aliaga-Guerrero M, Morano F, Zanuy S, Muñoz-Cueto JA. Gonadotropin inhibitory hormone down-regulates the brain-pituitary reproductive axis of male European sea bass (*Dicentrarchus labrax*). *Biol Reprod* 2016; 94:121.