Received: 22 December 2017 | Accepted: 1 February 2018

DOI: 10.1002/rmb2.12094

REVIEW ARTICLE

WILEY Reproductive Medicine and Biology

Role of pituitary adenylate cyclase-activating polypeptide in modulating hypothalamic-pituitary system

Aki Oride | Haruhiko Kanasaki | Satoru Kyo

Department of Obstetrics and Gynecology, Faculty of Medicine, Shimane University, Izumo City, Japan

Correspondence
Aki Oride, Department of Obstetrics and Gynecology, Faculty of Medicine, Shimane University, Izumo City, Japan.
Email: oride@med.shimane-u.ac.jp

Abstract

Background: Pituitary adenylate cyclase-activating polypeptide (PACAP) is a multifunctional peptide that is isolated and identified from the ovine hypothalamus, whose effects and mechanisms have been elucidated in numerous studies. The PACAP and its receptor are widely expressed, not only in the hypothalamus but also in peripheral organs.

Methods: The studies on the role of PACAP in the hypothalamic-pituitary system, including those by the authors, were summarized.

Results: In the pituitary gonadotrophs, PACAP increases the gonadotrophin α-, luteinizing hormoneβ-, and follicle-stimulating hormone β-subunit expression and the expression of gonadotropin-releasing hormone (GnRH) receptor and its own receptor, PAC1R. Moreover, a low-frequency GnRH pulse increases the expression of PACAP and PAC1R more than a high-frequency GnRH pulse in the gonadotrophs. The PACAP stimulates prolactin synthesis and secretion and increases PAC1R in the lactotrophs. In the hypothalamus, PACAP increases the expression of the GnRH receptors, although it is unable to increase the expression of GnRH in the GnRH-producing neurons.

Conclusion: The PACAP not only acts directly in each hormone-producing cell, it possibly might regulate hormone synthesis via the expression of its own receptors or those of other hormones.

KEYWORDS

gonadotropin, gonadotropin-releasing hormone, hypothalamus, pituitary, pituitary adenylate cyclase-activating polypeptide

1 | INTRODUCTION

Pituitary adenylate cyclase-activating polypeptide (PACAP) was isolated from ovine hypothalamic extracts as a novel peptide hormone that could stimulate cyclic adenosine monophosphate (cAMP) synthesis in anterior pituitary cells.[1] Although PACAP was first discovered in the hypothalamic area, PACAP actually is widely distributed in various brain regions, including the cerebral cortex, substantia nigra, pineal gland, hippocampus, amygdala, cerebellum, and pons.[2,3] Thereafter, it was revealed that PACAP also is expressed broadly in the peripheral nervous system[4] and in many peripheral organs, including the pituitary gland,[5,6] adrenal gland,[7] pancreas,[8] gonads,[9,10] and placenta.[11] Furthermore, PACAP has been reported to act as a hormone, neurotransmitter, and neuromodulator.[12] For example, in the central and peripheral nervous systems, PACAP is involved in neuronal differentiation and activation of the neurosecretory system and plays roles in the differentiation of neural progenitor cells,[13] regulation of neuronal synaptic

plasticity,[14] glucose-dependent insulin secretion,[15] and many other physiological effects.

There are two functional isoforms, PACAP-38 and PACAP-27. Both are derived from the same precursor protein that is encoded by the *Adcyap1* gene. It shares 68% amino acid sequence homology with vasoactive intestinal polypeptide (VIP) and 37% homology with secretin and thus PACAP belongs to the VIP-secretin-glucagon peptide superfamily.[12] The primary structure of PACAP38 has been remarkably conserved among most mammals, suggesting that PACAP plays important roles in maintaining life.[16] The distribution ratio of PACAP27 and PACAP38 varies from tissue to tissue.[10]

The G protein-coupled PAC1, VPAC1, and VPAC2 receptors have been cloned in vertebrates as the PACAP receptor. Both VPAC1 and VPAC2 respond to VIP and PACAP with high affinity, whereas the PAC1 receptors are selective for PACAP.[17] The PACAP-specific PAC1 receptor (PAC1R) couples mainly to Gs protein and induces intracellular c production, which ultimately activates the adenylate cyclase/protein kinase A (PKA) pathway. Also, PACAP is able to couple Gq protein and activate the phospholipase C (PLC)/protein kinase C (PKC) pathway and elevate intracellular calcium.[18]

The importance of PACAP in reproductive function has been clarified by studies of PACAP and PAC-1-knockout and -overexpressing mice. The PACAP null female mice had lowered implantation rates and decreased prolactin and progesterone levels.[19] In another report, PACAP knockout female mice showed decreased mating frequency and reduced fertility (the number of parturitions relative to the number of pairings).[20] In contrast, the overexpression of PACAP in the anterior pituitary gland of transgenic male mice delayed puberty and suppressed luteinizing hormone (LH) and follicle-stimulating hormone (FSH) levels and gonadotropin-releasing hormone (GnRH) receptor expression.[21] Thus, PACAP plays important roles in the hypothalamic-pituitary system.

This review focuses on the modulatory role that is played by PACAP in the hypothalamic-pituitary system in reproduction. Interactions between PACAP and the principle regulators of the hypothalamic-pituitary system—kisspeptin, GnRH, gonadotropin, and prolactin—are discussed, based on the authors' observations using cell models.

2 | ROLE OF PITUITARY ADENYLATE CYCLASE-ACTIVATING POLYPEPTIDE IN THE ANTERIOR PITUITARY GLAND

Gonadotropins, namely LH and FSH, are secreted from the anterior pituitary gland and regulate puberty and reproductive function through the production of sex steroids in the gonads. The LH and FSH are controlled primarily by GnRH, which is released from the hypothalamus in a pulsatile manner.[22] However, several reports suggested that PACAP also modulates the secretion of gonadotropins as it is present in the hypothalamus with a dense PACAP-immunoreactive fiber network in the external and internal zone of the median eminence in close contact with the hypophyseal portal vein[23] and PACAP has been detected in portal blood at a higher concentration than in the peripheral blood.[24] Moreover, PACAP receptors are found in the anterior pituitary gland[23,25] and PACAP stimulates the anterior pituitary hormones, such as growth hormone, adrenocorticotropic hormone, prolactin, thyroid-stimulating hormone (TRH), and gonadotropins in normal pituitary cells.[1,26,27] In addition, in vivo, i.v., and intra-atrial injections of PACAP increase the LH level in male rat blood,[28,29] suggesting that PACAP might act as a hypothalamic factor via the pituitary portal vein, similar to GnRH. In addition, PACAP is expressed by gonadotrophs and folliculostellate cells[6,30] and is synthesized[10] and released[31] from the anterior pituitary gland. Moreover, i.v. injection of PACAP increases its own expression in the pituitary gland.[28] These results suggest that PACAP might act not only as a paracrine factor through the hypothalamus, but also as an autocrine factor in the pituitary gland.

3 | REGULATION OF THE GONADOTROPHS BY PITUITARY ADENYLATE CYCLASE-ACTIVATING POLYPEPTIDE IN THE ANTERIOR PITUITARY GLAND

With regard to the effects of PACAP on gonadotrophs, many studies have been conducted by using model cells. αT3-1 cells, which are immortalized cells that are derived from transgenic mice, synthesize and secrete the α-subunit gene even though they do not express the β-subunit and cannot synthesize LH or FSH.[32] The PACAP dose-dependently increases the cAMP accumulation and increases the basal levels of the α-subunit through the PAC1 receptors and has a synergistic effect on GnRH in the αT3-1 cells.[33,34]

The LβT2 cells are another gonadotropin-producing cell model. These cells, also immortalized cells that are derived from transgenic mice, contain α-, LHβ-, and FSHβ-subunits and can be induced to express each subunit by GnRH stimulation.[35,36] The PACAP increases the intracellular cAMP levels and stimulates the α-, LHβ-, and FSHβ-subunit promoter activities in the LβT2 cells, although the effects were more modest than those of GnRH. The effects of GnRH on gonadotropin expression were not altered by the presence of PACAP.[36]

However, continuous PACAP stimulates α-subunit messenger (m) RNA levels but decreases FSHβ-subunit mRNA without affecting the LHβ-subunit mRNA in rat primary pituitary culture.[37] Additionally, if PACAP is administered in a pulsatile manner, PACAP stimulates the α- and LHβ-subunit mRNA but has no effect on the FSHβ-subunit mRNA in rat pituitary cells.[38]

Continuous GnRH stimulation fails to restore sustained LH and FSH secretion, whereas gonadotropin secretion can be induced by pulsatile GnRH administration.[39] In addition, it is generally accepted that FSH is predominantly secreted after low-frequency GnRH pulses and LH is predominantly secreted after high-frequency GnRH pulses.[40] The GnRH pulse frequency-dependent regulation of gonadotropin production also was investigated in the gonadotroph-immortalized cell line, LβT2.[41,42] It is suggested that the effects of

GnRH on gonadotroph regulation are altered by the stimulation mode of PACAP, although it is unclear how PACAP is released from the hypothalamus to the anterior pituitary gland. As above, pulsatile stimulation with PACAP specifically induces the expression of gonadotropin subunits in gonadotrophs. More interestingly, stimulation with high-frequency PACAP pulses (pulse interval of 30 minutes) increases the LHβ-subunit mRNA more than the low-frequency PACAP pulses, whereas the low-frequency PACAP pulses (pulse interval of 120 minutes) increase the FSHβ-subunit mRNA in a preferential manner in the LβT2 cells.[43]

4 | PITUITARY ADENYLATE CYCLASE-ACTIVATING POLYPEPTIDE AND GONADOTROPIN-RELEASING HORMONE CROSS-TALK IN THE GONADOTROPHS

The PACAP receptors are present not only in the hypothalamus, but also in the anterior pituitary gland.[44] There are two types of receptors—PAC1R and VPAC2R—in the anterior pituitary gland and gonadotrophs.[45,46] The PACAP mainly acts via the PAC1R in rat gonadotrophs.[34] The GnRH promotes the expression of its own receptors in the gonadotrophs[41] and PACAP also regulates PAC1R expression.[43,47] Moreover, the stimulation of PACAP increases the expression of the GnRH receptor in LβT2 cells.[43] Interestingly, the expression of PAC1R is markedly increased by low-frequency PACAP stimulation in the LβT2 cells. In contrast, the GnRH receptor expression increases with high-frequency PACAP stimulation and does not increase with low-frequency PACAP stimulation.[43] The results of these studies suggest that PACAP regulates not only the expression of the gonadotropin subunits, but also that of its own receptors and GnRH receptors. Larger numbers of GnRH receptors tended to increase the expression of LHβ, but tended to decrease that of FSHβ.[39] However, the expression of both the LHβ- and FSHβ-subunit mRNA was promoted as the number of PAC1Rs increased in the LβT2 cells.[48] In addition, previous studies have shown that GnRH increases the expression of PACAP and PAC1R in the gonadotrophs and primary pituitary culture.[43,49] The authors also have observed that low-frequency GnRH pulses increase the expression of PACAP and PAC1R more than the high-frequency GnRH pulses. Moreover, the GnRH-induced expression of the FSHβ-subunit is significantly prevented by the PAC1R antagonist.[50] Thus, GnRH and PACAP each display receptor-mediated cross-talk and might be involved in gonadotropin production.

4.1 | Role of pituitary adenylate cyclase-activating polypeptide in prolactin synthesis and secretion in the pituitary gland

Prolactin is released from the lactotrophs and somatolactotrophs in the anterior pituitary gland and has a wide variety of functions. Prolactin plays important roles in reproduction and is involved in mammary gland development during pregnancy and breast milk synthesis and secretion.[51] The frequency and amplitude of LH pulses decrease in patients with hyperprolactinemia, resulting in menstrual cycle dysfunction and amenorrhea.[52] In addition, prolactin suppresses LH pulses in rats.[53] Prolactin-receptor null female mice fail embryonic implantation and have irregular cycles and reduced fertilization rates.[54]

Prolactin secretion is predominantly inhibited by dopamine from the tuberoinfundibular dopaminergic neurons.[55] This means that if prolactin increases in circulation, dopamine is secreted from the hypothalamus and prolactin production is suppressed in the anterior pituitary gland. However, other factors promote prolactin synthesis and secretion. One such factor is TRH. The structure of TRH has been elucidated and the hormone has been proven to induce the rapid release of pituitary thyrotropin.[56] Subsequently, TRH was shown to induce prolactin secretion from rat pituitary cell culture[57] and its i.v. administration has induced the release of prolactin in humans.[58] The TRH binds TRH receptor-coupled G protein on the lactotrophs and activates the PKC-related pathway.[59] Extracellular signal-regulated kinase (ERK) is also activated by TRH and activated ERK is inactivated by dual-specificity threonine/tyrosine mitogen-activated protein kinase (MAPK) phosphatase.[60,61] The activation of ERK signaling is important for TRH-induced prolactin expression.[62] However, it is not yet clear how TRH regulates the production and secretion of prolactin.

The PACAP receptors are expressed in most types of cells in the anterior pituitary gland and PAC1R mainly is expressed in the lactotrophs.[23] It was shown initially that PACAP promotes prolactin secretion from superfused rat pituitary cells.[1] Although there have been reports that PACAP does not affect prolactin secretion in sheep[63] or rat[64] pituitary cells, subsequent studies have observed that PACAP can increase prolactin release from rat pituitary cells.[65,66] Moreover, PACAP increases the plasma prolactin levels in hypothalamus-lesioned rats.[67] A previous study showed that PACAP stimulated prolactin gene expression in the prolactin-producing GH3 cell model, although the effect was mild, compared with TRH stimulation.[68] The GH3 cells are immortalized cells that are derived from rat pituitary tumor that synthesizes and secretes prolactin and growth hormone[69] and PACAP can stimulate prolactin and growth hormone release.[70] It increases prolactin gene expression in GH3 cells through the ERK and cAMP/PKA pathways.[71]

Although the effect of PACAP on prolactin gene expression in GH3 cells is limited, the degree of prolactin promoter activity is similar to that of TRH stimulation if there is sufficient PAC1R due to its overexpression. In addition, PACAP and TRH have a synergistic effect on the expression of the prolactin gene. Increased PAC1R expression via overexpression potentiates the effect of TRH on prolactin promoter activity.[72] This suggests that the presence of PAC1R itself might enhance the effect of TRH on prolactin expression. Moreover, PACAP itself increases PAC1R expression, although TRH cannot stimulate PAC1R expression in GH3 cells.

Desensitization occurs in the GnRH receptor due to prolonged continuous stimulation with GnRH, suppressing gonadotropin secretion.[73] Similar to the GnRH receptor, prolonged TRH stimulation

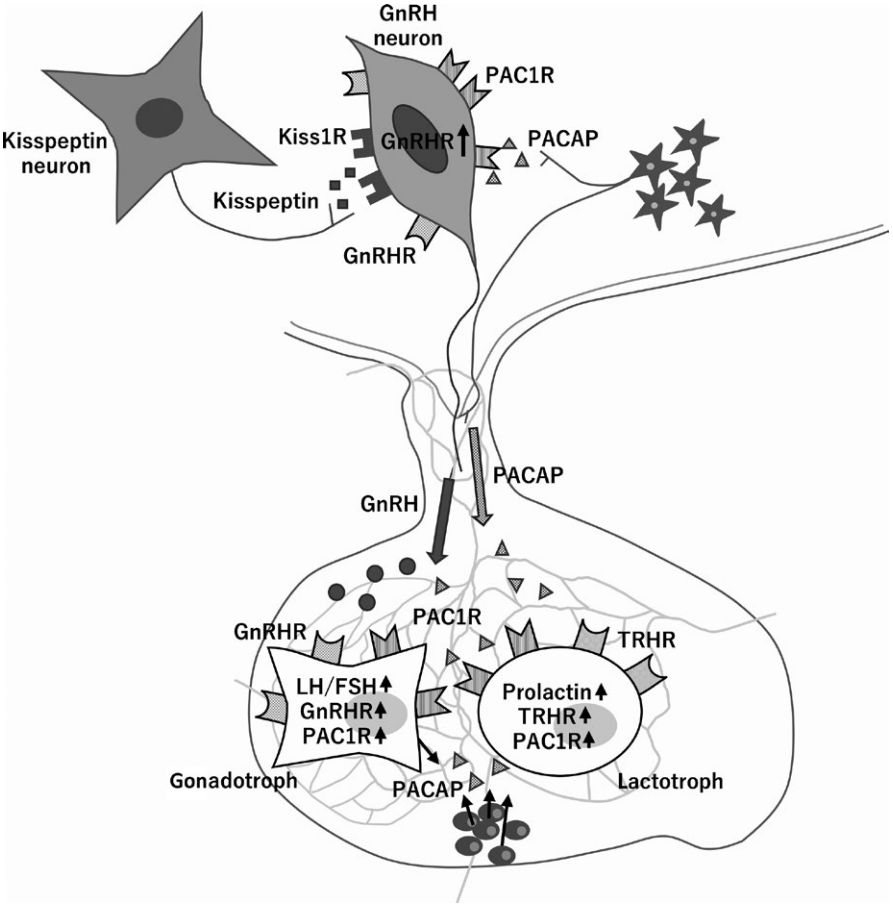

FIGURE 1 Schematic summary of the role of pituitary adenylate cyclase-activating polypeptide (PACAP) in reproductive function regulation in the hypothalamus-pituitary system. The PACAP is not only released from the hypothalamus but also expresses in gonadotrophs and folliculostellate cells in the anterior pituitary gland, in which PACAP type1 receptors (PAC1R) exist in the gonadotrophs and PACAP increases the expression of the gonadotropin α-, luteinizing hormone (LH)β-, and follicle-stimulating hormone (FSH) β-subunits. The PACAP increases the expression of the gonadotropin-releasing hormone (GnRH) receptor (GnRHR) and its own PAC1R in the gonadotrophs. The PAC1R receptor is also present in the lactotrophs and PACAP increases the expression of prolactin and PAC1R. In addition, PACAP enhances thyrotropin-releasing hormone receptor (TRHR)-induced prolactin production. In the hypothalamus, both PACAP and kisspeptin increase GnRHR expression. The GnRHR expression by kisspeptin stimulation is potentiated in the presence of PACAP

decreases the number of TRH receptors in the pituitary cells.[74] In the authors' work, the response of prolactin promoter activity to TRH and PACAP was diminished with prolonged TRH stimulation in the GH3 cells. In contrast, prolonged PACAP stimulation counteracted the response of prolactin promoter activity, not only to PACAP but also to TRH.[75] This means that PACAP and TRH desensitize their own and each other's receptor in prolactin-producing cells.

5 | PITUITARY ADENYLATE CYCLASE-ACTIVATING POLYPEPTIDE AND PITUITARY ADENYLATE CYCLASE-ACTIVATING POLYPEPTIDE RECEPTOR IN THE HYPOTHALAMUS

High levels of PACAP are found in the hypothalamus of different species by radioimmunoassay.[10] Thereafter, there have been several reports on the localization of PACAP in the hypothalamus and it was clear that PACAP is present in the supraoptic nuclei, paraventricular nuclei,[76,77] arcuate nucleus, and in a wide area in the hypothalamus.[78] The PACAP receptors are distributed in the arcuate nucleus, supraoptic nuclei,[79] paraventricular nuclei, suprachiasmatic nucleus,[80] and preoptic area.[81] The presence of PACAP and the PACAP receptors in the hypothalamus suggests that PACAP acts as a paracrine and autocrine factor.

5.1 | Role of pituitary adenylate cyclase-activating polypeptide in the hypothalamus

As GnRH is synthesized and secreted by parvocellular neurons that extend from the preoptic area[82] and PACAP is also present in this area, it is possible that PACAP regulates gonadotropin synthesis and secretion via the control of GnRH neurons. The release of LH and ovulation are inhibited in female rats by the intracerebroventricular administration of PACAP[83] and the injection of PACAP into the medial basal hypothalamus reduces the LH concentration, LH pulse

frequency, and pulse amplitude in ovariectomized ewes.[84] Moreover, the intracerebroventricular injection of PACAP increases the GnRH mRNA levels in male rats.[85] These results suggest that PACAP has an effect on gonadotropin secretion at the hypothalamus.

6 | ROLE OF PITUITARY ADENYLATE CYCLASE-ACTIVATING POLYPEPTIDE IN GONADOTROPIN-RELEASING HORMONE-PRODUCING NEURONS

In studies of the response of GnRH neurons, GT1-7 cells, which are mouse hypothalamic immortalized cells, are widely used as a model. The PACAP receptors are expressed in the GT1-7 cells and cAMP accumulation is dose-dependently increased by PACAP stimulation.[86]

The GnRH had been thought to be the primary regulator of the hypothalamus-pituitary-sex axis, but it has become clear that kisspeptin is positioned upstream of GnRH and controls GnRH secretion from the hypothalamus. The kisspeptin neuron fibers are located close to the GnRH neuronal cell bodies and processes and make synaptic contacts with the GnRH neurons.[87,88] Kisspeptin causes depolarization of the GnRH neurons and increases their firing frequency.[89] These results show that kisspeptin directly activates GnRH neurons. Indeed, GnRH is released after stimulation by kisspeptin from the hypothalamic explants of male rats.[90]

In a study using Chinese hamster ovary cells, kisspeptin receptor (Kiss1R) coupled to Gq protein and stimulated intracellular calcium mobilization through the activation of the PLC pathway. Kisspeptin also strongly phosphorylated ERK and p38 MAPK.[91] The authors previously reported that kisspeptin activates ERK- and cAMP/PKA-mediated pathways in GT1-7 cells.[92]

Several studies have reported that Kiss1R is expressed in GT1-7 cells and that kisspeptin is able to stimulate GnRH synthesis and secretion from the GT1-7 cells.[93,94] However, in the authors' experiments, the effect of kisspeptin stimulation on GnRH expression in the GT1-7 cells was unable to be confirmed, although the cells did express Kiss1R. However, it was observed that kisspeptin increased the expression of the GnRH receptors in Kiss1R-overexpressing GT1-7 cells.[92]

It is unclear how PACAP directly affects GnRH-producing neurons. In GT1-7 cells, PACAP also increased the expression of the GnRH receptor, similar to kisspeptin. The increase in the GnRH receptor expression by kisspeptin stimulation was potentiated by PACAP.[95] Pulsatile GnRH secretion is dependent on an autocrine interaction between GnRH and its receptors that are expressed in GnRH-producing neurons.[96] These results suggest that PACAP could regulate GnRH expression by modulating GnRH receptor expression.

7 | CONCLUSION

The PACAP is a widely expressed pleiotropic peptide. It is involved in reproductive function and affects the hypothalamus-pituitary system. This review summarized the roles of PACAP in the hypothalamus-pituitary system and reproductive function, focusing on experiments using cell models (Figure 1). Regarding gonadotropin production, GnRH is the major regulator, but PACAP also increases the expression of the gonadotropin subunit gene and further stimulates GnRH receptor expression in gonadotrophs. The PACAP is also a prolactin synthesis-stimulating factor and the presence of PAC1R itself promotes TRH-induced prolactin production in lactotrophs. In the GnRH-producing neurons, PACAP is involved in GnRH receptor expression. It is suggested that PACAP not only acts directly in each hormone-producing cell, but also influences the production of each hormone by regulating the expression of its own receptors or those of other hormones.

DISCLOSURES

Conflict of interest: The authors declare no conflict of interest. *Human Rights Statement and Animal studies*: This study has been approved by a suitably constituted Ethics Committee. This article does not contain any studies with human and animal participants that have been performed by the any of the authors.

ORCID

Aki Oride http://orcid.org/0000-0001-6609-2487
Haruhiko Kanasaki http://orcid.org/0000-0001-7011-8109

REFERENCES

1. Miyata A, Arimura A, Dahl RR, et al. Isolation of a novel 38 residue-hypothalamic polypeptide which stimulates adenylate cyclase in pituitary cells. *Biochem Biophys Res Commun*. 1989;164:567-574.
2. Ghatei MA, Takahashi K, Suzuki Y, Gardiner J, Jones PM, Bloom SR. Distribution, molecular characterization of pituitary adenylate cyclase-activating polypeptide and its precursor encoding messenger RNA in human and rat tissues. *J Endocrinol*. 1993;136:159-166.
3. Hannibal J. Pituitary adenylate cyclase-activating peptide in the rat central nervous system: an immunohistochemical and in situ hybridization study. *J Comp Neurol*. 2002;453:389-417.
4. Mulder H, Uddman R, Moller K, et al. Pituitary adenylate cyclase activating polypeptide expression in sensory neurons. *Neuroscience*. 1994;63:307-312.
5. Vigh S, Arimura A, Gottschall PE, Kitada C, Somogyvari-Vigh A, Childs GV. Cytochemical characterization of anterior pituitary target cells for the neuropeptide, pituitary adenylate cyclase activating polypeptide (PACAP), using biotinylated ligands. *Peptides*. 1993;14:59-65.
6. Koves K, Kantor O, Scammell JG, Arimura A. PACAP colocalizes with luteinizing and follicle-stimulating hormone immunoreactivities in the anterior lobe of the pituitary gland. *Peptides*. 1998;19:1069-1072.
7. Frodin M, Hannibal J, Wulff BS, Gammeltoft S, Fahrenkrug J. Neuronal localization of pituitary adenylate cyclase-activating polypeptide 38 in the adrenal medulla and growth-inhibitory effect on chromaffin cells. *Neuroscience*. 1995;65:599-608.
8. Filipsson K, Sundler F, Hannibal J, Ahren B. PACAP and PACAP receptors in insulin producing tissues: localization and effects. *Regul Pept*. 1998;74:167-175.

9. Fahrenkrug J, Hannibal J. Pituitary adenylate cyclase activating polypeptide innervation of the rat female reproductive tract and the associated paracervical ganglia: effect of capsaicin. *Neuroscience*. 1996;73:1049-1060.
10. Arimura A, Somogyvari-Vigh A, Miyata A, Mizuno K, Coy DH, Kitada C. Tissue distribution of PACAP as determined by RIA: highly abundant in the rat brain and testes. *Endocrinology*. 1991;129: 2787-2789.
11. Koh PO, Kwak SD, Kim HJ, et al. Expression patterns of pituitary adenylate cyclase activating polypeptide and its type I receptor mRNAs in the rat placenta. *Mol Reprod Dev*. 2003;64:27-31.
12. Sherwood NM, Krueckl SL, McRory JE. The origin and function of the pituitary adenylate cyclase-activating polypeptide (PACAP)/ glucagon superfamily. *Endocr Rev*. 2000;21:619-670.
13. Yan Y, Zhou X, Pan Z, Ma J, Waschek JA, DiCicco-Bloom E. Pro- and anti-mitogenic actions of pituitary adenylate cyclase-activating polypeptide in developing cerebral cortex: potential mediation by developmental switch of PAC1 receptor mRNA isoforms. *J Neurosci*. 2013;33:3865-3878.
14. Starr ER, Margiotta JF. Pituitary adenylate cyclase activating polypeptide induces long-term, transcription-dependent plasticity and remodeling at autonomic synapses. *Mol Cell Neurosci*. 2017;85:170-182.
15. Filipsson K, Tornoe K, Holst J, Ahren B. Pituitary adenylate cyclase-activating polypeptide stimulates insulin and glucagon secretion in humans. *J Clin Endocrinol Metab*. 1997;82:3093-3098.
16. Arimura A. Perspectives on pituitary adenylate cyclase activating polypeptide (PACAP) in the neuroendocrine, endocrine, and nervous systems. *Jpn J Physiol*. 1998;48:301-331.
17. Harmar AJ, Fahrenkrug J, Gozes I, et al. Pharmacology and functions of receptors for vasoactive intestinal peptide and pituitary adenylate cyclase-activating polypeptide: IUPHAR review 1. *Br J Pharmacol*. 2012;166:4-17.
18. Vaudry D, Falluel-Morel A, Bourgault S, et al. Pituitary adenylate cyclase-activating polypeptide and its receptors: 20 years after the discovery. *Pharmacol Rev*. 2009;61:283-357.
19. Isaac ER, Sherwood NM. Pituitary adenylate cyclase-activating polypeptide (PACAP) is important for embryo implantation in mice. *Mol Cell Endocrinol*. 2008;280:13-19.
20. Shintani N, Mori W, Hashimoto H, et al. Defects in reproductive functions in PACAP-deficient female mice. *Regul Pept*. 2002;109:45-48.
21. Moore JP Jr, Yang RQ, Winters SJ. Targeted pituitary overexpression of pituitary adenylate-cyclase activating polypeptide alters postnatal sexual maturation in male mice. *Endocrinology*. 2012;153:1421-1434.
22. Gharib SD, Wierman ME, Shupnik MA, Chin WW. Molecular biology of the pituitary gonadotropins. *Endocr Rev*. 1990;11:177-199.
23. Rawlings SR, Hezareh M. Pituitary adenylate cyclase-activating polypeptide (PACAP) and PACAP/vasoactive intestinal polypeptide receptors: actions on the anterior pituitary gland. *Endocr Rev*. 1996;17:4-29.
24. Dow RC, Bennie J, Fink G. Pituitary adenylate cyclase-activating peptide-38 (PACAP)-38 is released into hypophysial portal blood in the normal male and female rat. *J Endocrinol*. 1994;142:R1-R4.
25. Hashimoto H, Nogi H, Mori K, et al. Distribution of the mRNA for a pituitary adenylate cyclase-activating polypeptide receptor in the rat brain: an in situ hybridization study. *J Comp Neurol*. 1996;371:567-577.
26. Okada R, Yamamoto K, Ito Y, et al. VIP and PACAP stimulate TSH release from the bullfrog pituitary. *Peptides*. 2007;28:1784-1789.
27. Culler MD, Paschall CS. Pituitary adenylate cyclase-activating polypeptide (PACAP) potentiates the gonadotropin-releasing activity of luteinizing hormone-releasing hormone. *Endocrinology*. 1991;129:2260-2262.
28. Radleff-Schlimme A, Leonhardt S, Wuttke W, Jarry H. Evidence for PACAP to be an autocrine factor on gonadotrope cells. *Ann N Y Acad Sci*. 1998;865:486-491.
29. Osuga Y, Mitsuhashi N, Mizuno M. In vivo effect of pituitary adenylate cyclase activating polypeptide 38 (PACAP 38) on the secretion of luteinizing hormone (LH) in male rats. *Endocrinol Jpn*. 1992;39:153-156.
30. Jin L, Tsumanuma I, Ruebel KH, Bayliss JM, Lloyd RV. Analysis of homogeneous populations of anterior pituitary folliculostellate cells by laser capture microdissection and reverse transcription-polymerase chain reaction. *Endocrinology*. 2001;142:1703-1709.
31. Szabo F, Horvath J, Heinzlmann A, Arimura A, Koves K. Neonatal PACAP administration in rats delays puberty through the influence of the LHRH neuronal system. *Regul Pept*. 2002;109:49-55.
32. Windle JJ, Weiner RI, Mellon PL. Cell lines of the pituitary gonadotrope lineage derived by targeted oncogenesis in transgenic mice. *Mol Endocrinol*. 1990;4:597-603.
33. Schomerus E, Poch A, Bunting R, Mason WT, McArdle CA. Effects of pituitary adenylate cyclase-activating polypeptide in the pituitary: activation of two signal transduction pathways in the gonadotrope-derived alpha T3-1 cell line. *Endocrinology*. 1994;134:315-323.
34. Tsujii T, Attardi B, Winters SJ. Regulation of alpha-subunit mRNA transcripts by pituitary adenylate cyclase-activating polypeptide (PACAP) in pituitary cell cultures and alpha T3-1 cells. *Mol Cell Endocrinol*. 1995;113:123-130.
35. Mutiara S, Kanasaki H, Harada T, Miyazaki K. Dopamine D(2) receptor expression and regulation of gonadotropin alpha-subunit gene in clonal gonadotroph LbetaT2 cells. *Mol Cell Endocrinol*. 2006;259:22-29.
36. Harada T, Kanasaki H, Mutiara S, Oride A, Miyazaki K. Cyclic adenosine 3',5'monophosphate/protein kinase A and mitogen-activated protein kinase 3/1 pathways are involved in adenylate cyclase-activating polypeptide 1-induced common alpha-glycoprotein subunit gene (Cga) expression in mouse pituitary gonadotroph LbetaT2 cells. *Biol Reprod*. 2007;77:707-716.
37. Tsujii T, Ishizaka K, Winters SJ. Effects of pituitary adenylate cyclase-activating polypeptide on gonadotropin secretion and subunit messenger ribonucleic acids in perifused rat pituitary cells. *Endocrinology*. 1994;135:826-833.
38. Tsujii T, Winters SJ. Effects of pulsatile pituitary adenylate cyclase activating polypeptide (PACAP) on gonadotropin secretion and subunit mRNA levels in perifused rat pituitary cells. *Life Sci*. 1995;56:1103-1111.
39. Belchetz PE, Plant TM, Nakai Y, Keogh EJ, Knobil E. Hypophysial responses to continuous and intermittent delivery of hypopthalamic gonadotropin-releasing hormone. *Science*. 1978;202:631-633.
40. Wildt L, Hausler A, Marshall G, et al. Frequency and amplitude of gonadotropin-releasing hormone stimulation and gonadotropin secretion in the rhesus monkey. *Endocrinology*. 1981;109:376-385.
41. Bedecarrats GY, Kaiser UB. Differential regulation of gonadotropin subunit gene promoter activity by pulsatile gonadotropin-releasing hormone (GnRH) in perifused L beta T2 cells: role of GnRH receptor concentration. *Endocrinology*. 2003;144:1802-1811.
42. Kanasaki H, Bedecarrats GY, Kam KY, Xu S, Kaiser UB. Gonadotropin-releasing hormone pulse frequency-dependent activation of extracellular signal-regulated kinase pathways in perifused LbetaT2 cells. *Endocrinology*. 2005;146:5503-5513.
43. Kanasaki H, Mutiara S, Oride A, Purwana IN, Miyazaki K. Pulse frequency-dependent gonadotropin gene expression by adenylate cyclase-activating polypeptide 1 in perifused mouse pituitary gonadotroph LbetaT2 cells. *Biol Reprod*. 2009;81:465-472.
44. Gottschall PE, Tatsuno I, Miyata A, Arimura A. Characterization and distribution of binding sites for the hypothalamic peptide, pituitary adenylate cyclase-activating polypeptide. *Endocrinology*. 1990;127:272-277.

45. Rawlings SR, Piuz I, Schlegel W, Bockaert J, Journot L. Differential expression of pituitary adenylate cyclase-activating polypeptide/vasoactive intestinal polypeptide receptor subtypes in clonal pituitary somatotrophs and gonadotrophs. *Endocrinology*. 1995;136:2088-2098.
46. Hezareh M, Journot L, Bepoldin L, Schlegel W, Rawlings SR. PACAP/VIP receptor subtypes, signal transducers, and effectors in pituitary cells. *Ann N Y Acad Sci* 1996;805:315-327; discussion 27-28.
47. Pincas H, Laverriere JN, Counis R. Pituitary adenylate cyclase-activating polypeptide and cyclic adenosine 3',5'-monophosphate stimulate the promoter activity of the rat gonadotropin-releasing hormone receptor gene via a bipartite response element in gonadotrope-derived cells. *J Biol Chem*. 2001;276:23562-23571.
48. Purwana IN, Kanasaki H, Oride A, et al. GnRH-induced PACAP and PAC1 receptor expression in pituitary gonadotrophs: a possible role in the regulation of gonadotropin subunit gene expression. *Peptides*. 2010;31:1748-1755.
49. Grafer CM, Thomas R, Lambrakos L, Montoya I, White S, Halvorson LM. GnRH stimulates expression of PACAP in the pituitary gonadotropes via both the PKA and PKC signaling systems. *Mol Endocrinol*. 2009;23:1022-1032.
50. Kanasaki H, Purwana IN, Mijiddorj T, Oride A, Miyazaki K. Possible involvement of PACAP and PACAP type 1 receptor in GnRH-induced FSH beta-subunit gene expression. *Regul Pept*. 2011;167:227-232.
51. Brisken C, Kaur S, Chavarria TE, et al. Prolactin controls mammary gland development via direct and indirect mechanisms. *Dev Biol*. 1999;210:96-106.
52. Matsuzaki T, Azuma K, Irahara M, Yasui T, Aono T. Mechanism of anovulation in hyperprolactinemic amenorrhea determined by pulsatile gonadotropin-releasing hormone injection combined with human chorionic gonadotropin. *Fertil Steril*. 1994;62:1143-1149.
53. Park SK, Keenan MW, Selmanoff M. Graded hyperprolactinemia first suppresses LH pulse frequency and then pulse amplitude in castrated male rats. *Neuroendocrinology*. 1993;58:448-453.
54. Ormandy CJ, Camus A, Barra J, et al. Null mutation of the prolactin receptor gene produces multiple reproductive defects in the mouse. *Genes Dev*. 1997;11:167-178.
55. Ben-Jonathan N, Hnasko R. Dopamine as a prolactin (PRL) inhibitor. *Endocr Rev*. 2001;22:724-763.
56. Folkers K, Enzmann F, Boler J, Bowers CY, Schally AV. Discovery of modification of the synthetic tripeptide-sequence of the thyrotropin releasing hormone having activity. *Biochem Biophys Res Commun*. 1969;37:123-126.
57. Tashjian AH Jr, Barowsky NJ, Jensen DK. Thyrotropin releasing hormone: direct evidence for stimulation of prolactin production by pituitary cells in culture. *Biochem Biophys Res Commun*. 1971;43:516-523.
58. Jacobs LS, Snyder PJ, Wilber JF, Utiger RD, Daughaday WH. Increased serum prolactin after administration of synthetic thyrotropin releasing hormone (TRH) in man. *J Clin Endocrinol Metab*. 1971;33:996-998.
59. Gershengorn MC. Mechanism of thyrotropin releasing hormone stimulation of pituitary hormone secretion. *Annu Rev Physiol*. 1986;48:515-526.
60. Ryser S, Tortola S, van Haasteren G, Muda M, Li S, Schlegel W. MAP kinase phosphatase-1 gene transcription in rat neuroendocrine cells is modulated by a calcium-sensitive block to elongation in the first exon. *J Biol Chem*. 2001;276:33319-33327.
61. Oride A, Kanasaki H, Mutiara S, Purwana IN, Miyazaki K. Activation of extracellular signal-regulated kinase (ERK) and induction of mitogen-activated protein kinase phosphatase 1 (MKP-1) by perifused thyrotropin-releasing hormone (TRH) stimulation in rat pituitary GH3 cells. *Mol Cell Endocrinol*. 2008;296:78-86.
62. Kanasaki H, Yonehara T, Yamamoto H, et al. Differential regulation of pituitary hormone secretion and gene expression by thyrotropin-releasing hormone. A role for mitogen-activated protein kinase signaling cascade in rat pituitary GH3 cells. *Biol Reprod*. 2002;67:107-113.
63. Sawangjaroen K, Anderson ST, Curlewis JD. Effects of pituitary adenylate cyclase-activating polypeptide (PACAP) and vasoactive intestinal polypeptide (VIP) on hormone secretion from sheep pituitary cells in vitro. *J Neuroendocrinol*. 1997;9:279-286.
64. Hart GR, Gowing H, Burrin JM. Effects of a novel hypothalamic peptide, pituitary adenylate cyclase-activating polypeptide, on pituitary hormone release in rats. *J Endocrinol*. 1992;134:33-41.
65. Arbogast LA, Voogt JL. Pituitary adenylate cyclase-activating polypeptide (PACAP) increases prolactin release and tuberoinfundibular dopaminergic neuronal activity. *Brain Res*. 1994;655:17-24.
66. Benter S, Leonhardt S, Wuttke W, Jarry H. Paracrine cell to cell interactions determine the effects of pituitary adenylate cyclase activating polypeptide (PACAP) on in vitro prolactin release from rat pituitary cells. *Exp Clin Endocrinol Diabetes*. 1995;103:386-390.
67. Jarry H, Leonhardt S, Schmidt WE, Creutzfeldt W, Wuttke W. Contrasting effects of pituitary adenylate cyclase activating polypeptide (PACAP) on in vivo and in vitro prolactin and growth hormone release in male rats. *Life Sci*. 1992;51:823-830.
68. Kanasaki H, Fukunaga K, Takahashi K, Miyazaki K, Miyamoto E. Mitogen-activated protein kinase activation by stimulation with thyrotropin-releasing hormone in rat pituitary GH3 cells. *Biol Reprod*. 1999;61:319-325.
69. Tashjian AH Jr, Yasumura Y, Levine L, Sato GH, Parker ML. Establishment of clonal strains of rat pituitary tumor cells that secrete growth hormone. *Endocrinology*. 1968;82:342-352.
70. Propato-Mussafiri R, Kanse SM, Ghatei MA, Bloom SR. Pituitary adenylate cyclase-activating polypeptide releases 7B2, adrenocorticotrophin, growth hormone and prolactin from the mouse and rat clonal pituitary cell lines AtT-20 and GH3. *J Endocrinol*. 1992;132:107-113.
71. Yonehara T, Kanasaki H, Yamamoto H, Fukunaga K, Miyazaki K, Miyamoto E. Involvement of mitogen-activated protein kinase in cyclic adenosine 3',5'-monophosphate-induced hormone gene expression in rat pituitary GH(3) cells. *Endocrinology*. 2001;142:2811-2819.
72. Mijiddorj T, Kanasaki H, Purwana IN, Oride A, Miyazaki K. Stimulatory effect of pituitary adenylate-cyclase activating polypeptide (PACAP) and its PACAP type I receptor (PAC1R) on prolactin synthesis in rat pituitary somatolactotroph GH3 cells. *Mol Cell Endocrinol*. 2011;339:172-179.
73. Knobil E. Patterns of hypophysiotropic signals and gonadotropin secretion in the rhesus monkey. *Biol Reprod*. 1981;24:44-49.
74. Hinkle PM, Tashjian AH Jr. Thyrotropin-releasing hormone regulates the number of its own receptors in the GH3 strain of pituitary cells in culture. *Biochemistry*. 1975;14:3845-3851.
75. Mijiddorj T, Kanasaki H, Unurjargal S, Oride A, Purwana I, Miyazaki K. Prolonged stimulation with thyrotropin-releasing hormone and pituitary adenylate cyclase-activating polypeptide desensitize their receptor functions in prolactin-producing GH3 cells. *Mol Cell Endocrinol*. 2013;365:139-145.
76. Koves K, Arimura A, Somogyvari-Vigh A, Vigh S, Miller J. Immunohistochemical demonstration of a novel hypothalamic peptide, pituitary adenylate cyclase-activating polypeptide, in the ovine hypothalamus. *Endocrinology*. 1990;127:264-271.
77. Koves K, Arimura A, Gorcs TG, Somogyvari-Vigh A. Comparative distribution of immunoreactive pituitary adenylate cyclase activating polypeptide and vasoactive intestinal polypeptide in rat forebrain. *Neuroendocrinology*. 1991;54:159-169.
78. Koves K, Vereczki V, Kausz M, et al. PACAP and VIP in the photoneuroendocrine system (PNES). *Med Sci Monit*. 2002;8:SR5-SR20.
79. Masuo Y, Ohtaki T, Masuda Y, Tsuda M, Fujino M. Binding sites for pituitary adenylate cyclase activating polypeptide (PACAP):

comparison with vasoactive intestinal polypeptide (VIP) binding site localization in rat brain sections. *Brain Res*. 1992;575:113-123.
80. Hashizume T, Soliman EB, Kanematsu S. Effects of pituitary adenylate cyclase-activating polypeptide (PACAP), prostaglandin E2 (PGE2) and growth hormone releasing factor (GRF) on the release of growth hormone from cultured bovine anterior pituitary cells in vitro. *Domest Anim Endocrinol*. 1994;11:331-337.
81. Ha CM, Kang JH, Choi EJ, et al. Progesterone increases mRNA levels of pituitary adenylate cyclase-activating polypeptide (PACAP) and type I PACAP receptor (PAC(1)) in the rat hypothalamus. *Brain Res Mol Brain Res*. 2000;78:59-68.
82. Maggi R, Cariboni AM, Marelli MM, et al. GnRH and GnRH receptors in the pathophysiology of the human female reproductive system. *Hum Reprod Update*. 2016;22: pii: dmv059.
83. Koves K, Kantor O, Molnar J, et al. The role of PACAP in gonadotropic hormone secretion at hypothalamic and pituitary levels. *J Mol Neurosci*. 2003;20:141-152.
84. Anderson ST, Sawangjaroen K, Curlewis JD. Pituitary adenylate cyclase-activating polypeptide acts within the medial basal hypothalamus to inhibit prolactin and luteinizing hormone secretion. *Endocrinology*. 1996;137:3424-3429.
85. Li S, Grinevich V, Fournier A, Pelletier G. Effects of pituitary adenylate cyclase-activating polypeptide (PACAP) on gonadotropin-releasing hormone and somatostatin gene expression in the rat brain. *Brain Res Mol Brain Res*. 1996;41:157-162.
86. Olcese J, McArdle CA, Middendorff R, Greenland K. Pituitary adenylate cyclase-activating peptide and vasoactive intestinal peptide receptor expression in immortalized LHRH neurons. *J Neuroendocrinol*. 1997;9:937-943.
87. Clarkson J, Herbison AE. Postnatal development of kisspeptin neurons in mouse hypothalamus; sexual dimorphism and projections to gonadotropin-releasing hormone neurons. *Endocrinology*. 2006;147:5817-5825.
88. Clarkson J, d'Anglemont de Tassigny X, Colledge WH, Caraty A, Herbison AE. Distribution of kisspeptin neurones in the adult female mouse brain. *J Neuroendocrinol*. 2009;21:673-682.
89. Zhang C, Roepke TA, Kelly MJ, Ronnekleiv OK. Kisspeptin depolarizes gonadotropin-releasing hormone neurons through activation of TRPC-like cationic channels. *J Neurosci*. 2008;28:4423-4434.
90. Thompson EL, Patterson M, Murphy KG, et al. Central and peripheral administration of kisspeptin-10 stimulates the hypothalamic-pituitary-gonadal axis. *J Neuroendocrinol*. 2004;16:850-858.
91. Kotani M, Detheux M, Vandenbogaerde A, et al. The metastasis suppressor gene KiSS-1 encodes kisspeptins, the natural ligands of the orphan G protein-coupled receptor GPR54. *J Biol Chem*. 2001;276:34631-34636.
92. Sukhbaatar U, Kanasaki H, Mijiddorj T, Oride A, Miyazaki K. Kisspeptin induces expression of gonadotropin-releasing hormone receptor in GnRH-producing GT1-7 cells overexpressing G protein-coupled receptor 54. *Gen Comp Endocrinol*. 2013;194:94-101.
93. Novaira HJ, Ng Y, Wolfe A, Radovick S. Kisspeptin increases GnRH mRNA expression and secretion in GnRH secreting neuronal cell lines. *Mol Cell Endocrinol*. 2009;311:126-134.
94. Ozcan M, Alcin E, Ayar A, Yilmaz B, Sandal S, Kelestimur H. Kisspeptin-10 elicits triphasic cytosolic calcium responses in immortalized GT1-7 GnRH neurones. *Neurosci Lett*. 2011;492:55-58.
95. Kanasaki H, Mijiddorj T, Sukhbaatar U, Oride A, Miyazaki K. Pituitary adenylate cyclase-activating polypeptide (PACAP) increases expression of the gonadotropin-releasing hormone (GnRH) receptor in GnRH-producing GT1-7 cells overexpressing PACAP type I receptor. *Gen Comp Endocrinol*. 2013;193:95-102.
96. Martinez-Fuentes AJ, Hu L, Krsmanovic LZ, Catt KJ. Gonadotropin-releasing hormone (GnRH) receptor expression and membrane signaling in early embryonic GnRH neurons: role in pulsatile neurosecretion. *Mol Endocrinol*. 2004;18:1808-1817.

How to cite this article: Oride A, Kanasaki H, Kyo S. Role of pituitary adenylate cyclase-activating polypeptide in modulating hypothalamic-pituitary system. *Reprod Med Biol*. 2018;17:234–241. https://doi.org/10.1002/rmb2.12094