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ROLE OF GONADOTROPIN- INHIBITORY HORMONE (GNIH) IN REPRODUCTION OF ANIMALS

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ABSTRACT
In 2000 Tsutsui discovered a newly neuropeptide Gonadotropin- inhibitory hormone. It is a key hormone for reproduction in vertebrate. There are many studies indicating the GnIH directly inhibits the gonadotropin synthesis and release in birds and mammals. We are already know about Gonadotropin Releasing hormone, it is regulate the function of reproduction but recently known GnIH inhibits sex hormones and counteracting neuropeptide to regulate the reproductive function. These two (GnRH and GnIH) neuropeptide seem to be pivotal for the regulation of gonadal activity in response to internal and external cues. GnIH axons and terminal are present in hypothalamic area in avian brain, and its receptor is expressed on GnRH neurons. GnIH have many orthologous peptides are present in mammals and other vertebrate, this is reported by previous study. There are now several authentications that GnIH and its orthologs directly inhibits gonadotropin releasing and synthesis in vertebrate. This short review discussed about the recent study of GnIH and highlights the localization of GnIH and its function to reproduction in avian and other vertebrate. The GnIH discovered not only neuro-physiological study but also even more focused on reproduction. This review works based on previous laboratory and field studies.

KEYWORDS: Gonadotropin-Inhibitory hormone (GnIH), reproduction, avian, mammals.

GENERAL INTRODUCTION

Reproduction is one of the important physiological processes in almost every organism which leads to variations and evolution for better adaptation. Reproduction can be considered as continuous process or seasonal process. Certain organisms breed throughout the year and considered as continuous breeder like human and certain organisms like birds are recognized as opportunistic breeders or seasonal breeders and depends upon variety of favorable conditions suitable for breeding. Sexual interests and behaviors are active and accepted only during this time. Reproduction is affected by variety of environmental components used to set the timing of reproductive functions. It includes photoperiod, rainfall, humidity, temperature and availability of food and water in addition to other intrinsic mechanism(s) which may have hormonal or neural regulation.[26] Circadian rhythmicity in hormonal secretion, certain neurotransmitters and hypothalamic factors in vertebrates playing a vital role in explaining the different physiology. Even variation on daily or seasonal basis changes the physiology like reproduction to a greater extent. Hypothalamus is the prime controller of reproductive activity which defines optimum time for mating. Hypothalamus produces a hormone known as gonadotropin releasing hormone (GnRH) which is a decapeptide first found to be isolated from mammals[20],[7] and subsequently from birds[17],[23] and vertebrates which promotes stimulation of the pituitary to release its secretion from both anterior and posterior lobe. Anterior secretion includes two important pituitary gonadotropin hormones known as follicle-stimulating hormone (FSH) and luteinizing hormone (LH). These hormones promote gonadal growth and development. FSH acts on sperm-producing structures in the testes in male and egg producing structure in female, while LH acts on the interstitial cells of the testes causing them to secrete the steroid hormone testosterone. Thus, levels of GnRH specify the seasonality of reproduction.

Recently a new hypothalamic dodecapeptide (SIKPSAYLPLRF-NH2) known as gonadotropin inhibitory hormone was reported from anterior lobe of quail which inhibits the gonadotropin secretion.[32] This was the first report in any vertebrate for the presence of neuropeptide inhibiting gonadotropin secretion. Various studies Reported on birds[32],[37],[34],[25],[11],[35], GnIH as an important neuropeptide for the regulation of avian reproduction. In addition, different GnIH homologs were reported from the brains of other vertebrates, such as mammals, amphibians, and fish.[37],[13] GnIH neuropeptide found to possess a LPXRF-amide (X

represents L or Q) motif at the C termini in all reported animals. As per the studies GnIH cultured from quail pituitary showed the decreased level of LH and FSH[32] and reduced gonadal growth and development in gonads of mail quail bird[35] by decreasing the gonadotropin synthesis and release.[34] This study carried out certain studies to see the physiological and biological significance of GnIH. This study includes the observation of various developmental changes during expression of GnIH in embryonic and posthatch stages in quail diencephalon including the PVN and ME by using competitive PCR and ELISA. This review highlights the discovery of GnIH in the vertebrate and the function of GnIH neuron in the hypothalamo-hypophysial system related to reproduction of birds and other vertebrates.

THE NEW BRAIN HORMONE GONADOTROPIN INHIBITORY HORMONE DISCOVERY AND LOCALIZATION ON ANIMALS

Upon studies on avian reproduction, a new peptide with the sequence Ser-Ile-Lys-Pro-Ser-Ala-Tyr-Leu-Pro-Leu-Arg-Phe-NH2 was isolated from avian brain & Immunohistochemical studies showed the localization of this peptide in the paraventricular nucleus. This peptide cultured from anterior pituitary of quail is considered to have inhibitory effect on gonadotropin release in a dose dependent manner. The first GnIH hormone was discovered in the Japanese quail brain. It has 12 amino acid sequences with RFa peptide at the C-terminus. This novel RFa peptide was localized in the diencephalon. These peptide abundant immunoreactive cell bodies were localized in PVN and ME in the hypothalamus.[32] [25] The presence the presence of GnIH in white crowned sparrow by identification and characterization of a cDNA encoding sparrow GnIH& found the localization of GnIH mRNA in the hypothalamus by the methods like in situ hybridization. They even explained in vivo anti gonadotropic effect on the breeding time of sparrow in northern Alaska and compared the effect of quail GnIH injections in both white crowned sparrow and song sparrow.[42] characterized the GnIH receptor showing specific binding to peptides having C-terminal LPXRF-amide motif as well as to GnIH and GnIH-RPs. Similar studies the studies of[32], [34], [37] the expression of GnIH receptor mRNA is limited to pituitary. Thus the mode of action of GnIH is found directly to pituitary thereby inhibiting the gonadotropin release. The cell bodies and terminal containing GnIH is localized in the paraventricular nucleus and median eminence.[36] This study their studies in European starling and investigated the interaction between GnIH and GnIH-1, GnIH-2 neuron in starling brain. They isolated the GnIH cDNA from the cloned starling and found to possess three peptides having characteristic sequence of LPXRF-amide (X=L or Q) motif at the C termini. By the use of mass spectrometry combined with immunoaffinity purification & characterize the sequence as SIKPFANLPLRF-NH2.

GnIH directly act on the neurons regulating GnRH.[36], [11],[4] Melatonin receptor is present in GnIH neuron [34] and shows the GnIH release in quail through melatonin pathway.[10] [5] The relative the relative distribution of GnIH and GnRH in the neuron and found *in vivo* effect of exogenous administration of GnIH in song sparrow and house sparrow. They even reported the presence of GnIH in species of two families of song sparrow and determined the gonadotropin inhibitory activity and neuroanatomical relation between GnIH and GnRH.[33] reported the structure, history and role of GnIH neuropeptide in physiology and showed its mode of action at the anterior pituitary level & found homologs of GnIH peptide in hypothalamus of other vertebrates including mammals, reptiles, amphibians and teleost.

In European starlings the direct effect of melatonin on reproduction and gonadal neuropeptide expression. This study has been demonstrated that GnIH is regulated seasonally, probably through a mechanism involving melatonin.[22] The studies have been provided to active participation of hypothalamic GnIH and Kisspeptin in the regulation of reproductive system according to metabolic cues. *KISS1* expression, the gene encoding of KP, is reduced while RERP (NPVF) expression, the gene encoding of GnIH, is enhance in condition of metabolic deficiency.[40] This study was to show appetite-associated GnIH effect in chicks from lines selected low and high body weight, and are anorexic or become fat, respectively. Central administration of GnIH increased intake of food in both lines with a similar magnitude of response. Effect on water intake was not there. GnIH mRNA of Hypothalamic was greater in the low than high weight lines and was greater in the fasted than fed of chicks.[21] [39] This study reported that characterized a cDNA encoding of GnIH, its orthologs have a general C-terminal LPXRFamide (X = L or Q) motif in the brain of the red-eared slider turtle. The presumed precursor protein consisted of 205 amino-acid residues, encoded by three putative peptide sequences that contained the LPXRF amide motif at their C-termini**.**

THE FUNCTIONAL ROLE OF GONADOTROPIN INHIBITORY HORMONE IN AVIAN REPRODUTION

In 2000, the discovery of GnIH neuropeptide and it is acts as a reproductive ‘control button’ momentarily inhibiting the activity of the reproduction in animal. According to[19] chicken ovary has characterized the expression of GnIH mRNA and inhibitory effect of GnIH on ovarian follicular development. The ovarian GnIHR is probably to be observed in development of ovarian follicles. The abundance of ovarian GnIHR mRNA is decreased due to sexual maturation or estradiol and progesterone treatment would involve an inhibitory function for GnIHR in development of ovarian follicular.[22] studied on direct effect of melatonin in gonadal neuropeptide and sex steroids secretions in European starling, it was reported the presence of Gonadotropin inhibitory hormone (GnIH) and its receptor (GnIHR)and melatonin receptors 1B(mel 1B) and 1C(mel 1C) through PCR method. This study

explained the involvement of melatonin in the regulation of GnIH seasonally and also effected reproduction.[35] This study the effect of GnIH on gonadal activity and maintenance in male quail during reproduction. When GnIH continuously administered in mature birds via osmotic pumps for about 2 weeks and found the decreased expression of Gonadotropin common α and LHβ subunit mRNA. Plasma LH and testosterone concentration were also decreased and these activities were dose dependent. The GnIH injected to mature birds induced apoptosis of testis and showed decreased spermatogenic activity. Daily injections of GnIH in mature birds for about 2 weeks suppressed the testicular growth and rose in plasma testosterone. Changes in the ratio of expression of two receptors GnRHR-2/GnIHR in pituitary glands of chicken during sexual maturation shows the hypothalamic control of gonadotropin secretions from inhibitory to stimulatory level thus expressing high level of GnRH induced cAMP. Autocrine/paracrine activity between GnIH and its receptor within gonads interfere with levels of LH & FSH influencing the ovarian follicular development as well as spermatogenesis.[3][8] This study that significant less numbers of GnIH peptide-producing cells in birds, it is outcompeted others for nest boxes ('winners') than those without nest boxes ('losers') and their changes of relationship with breeding stage. GnRH content, corticosterone and testosterone did not differ with ownership of nest box. Therefore, the birds showed reproductively competent across treatments, this data showed that GnIH may provide as modulator of reproductive behaviours reacts to social environment in European starlings.

This study was the localization of peptides and GnRH-I and GnIH mRNA in the adult male spotted munia (*Lonchura punctulata*), a circannual breeder. The hypothalamus present of GnRH-I and GnIH mRNA, it is analysis by RT-PCR 37. The GnRH cell bodies and GnIH fibres in close proximity to each other advised that the one fibre must be interacting with the other peptide cell bodies and vice versa. This purposed that GnIH function in modulating GnRH and after all controlling the reproduction in spotted munia.[31] When the given of melatonin injection, it seen that raised GnIH release on the dosage dependent mode from explants of hypothalamic *in vitro*. During the dark period GnIH mRNA expression and GnIH release were greater than those explants from quail exposed to long day photoperiods during the light period. Conversely, during the dark period decreased the concentration of plasma luteinizing hormone (LH). This review sum up that melatonin emerges to act on GnIH neurons in exciting not only synthesis of GnIH but also its release, thus inhibiting concentration of plasma LH in birds.[9] Similar study[19] reported that the ovarian GnIHR is probably to be engrossed in development of ovarian follicules, the abundance of GnIHR mRNA owing to decreased sexual maturation. GnIH may affect maturation of follicles by decreasing the feasibility of prehierarchial follicular granulosa cells through binding to GnIHR.[10] The role of the role of melatonin upon the release of GnIH. Photoperiodicity effects the secretion of GnIH. Thus photoperiodic control of melatonin affects the neural function of GnIH. Melatonin given in a specific dose can increase or decrease the levels of GnIH and thereby correlating LSH release in quail. In Birds higher corticosterone had higher testicular expression of GnIH and inferior testosterone. Expression of StAR and LHR were inferior in the fasted bird's testes than controls. Thus, the testosterone decrease was not likely mediated by GnIH of hypothalamic, but rather by direct acts of rapiding and/or corticosterone on the testes, showing that the testes can react to directly stress cue. Such local testosterone inhibition synthesis may allow for fast and reversible changes in behaviour and physiology when circumstances are in suitable for breeding.[18] This experimental strongly suggested that the inhibition of GnIH actions of gonadotropin-releasing hormones on secretion of gonadotropin, behaviour, and it has direct inhibited to formation of steroidogenesis in testis. During breeding activities, it is lead to the ovulation may be involve environmental signal releasing and inhibition on the hypothalamo-pituitary–gonad axis.[41] This study suggested that although GnIH responsiveness to stress emerges to be preserved across species, specific tissue react and direction of GnIH regulation is not. The GnIH are response to stress between species might be the effect of ecological adaptations or differences of other species in the response of the GnIH system to stress[12]

THE FUNCTION OF GONADOTROPIN INHIBITORY HORMONE ON REPRODUCTION IN OTHER VERTEBRATES

[24] This study that RFRP-3 does not affect LH secretion via the GnRH release and it's directly functions to the pituitary to suppress GnRH-stimulated and secretion of LH in female rats. The gene of the mammalian RFamide-related peptides (RFRP) is orthologous to the GnIH gene. Three orthologous of gene for gnihr was also indentified in zebrafish. It is detected to all embryonic stages of zebra and also showed in early development after hatching. The teleost biological action gnih on LH release was additional examined in *in vivo* of goldfish. The mature zebra fish gnih peptide (LPXRFa peptide-3) intraperitoneal injection could significantly decrease the basal level of serum LH in goldfish. These results were provided the first confirmation that plays of gnih a significant function in the negative regulation of LH release in teleost.[43] The treatment of GnIH *in vivo* also revealed the decreased insulin receptor protein expression in the testis, which possibly responsible for the decreased activity of testis in the mice. These findings thus propose that GnIH increases the uptake of glucose and TGs in the adipose tissue, to result in increased accumulation of fat, while simultaneously in the testis, GnIH suppressed the GLUT8-mediated glucose uptake, which in turn may be responsible for decreased testosterone synthesis. This study showed GnIH as increases mediator adiposity and impaired

testicular activity in mice.[1] [27] These studies the inverse relationship between expression of RFRP-3 neurones and gonadal activity. They found high levels of immune reactive RFRP-3 in immature and 8-hrs mice which stimulate gonadal suppression & low levels in mature and 12-hrs mice which stimulate gonadal stimulation.[28] reported this short communication demonstrates that the gonadal quiescence in sexually immature mice and a decline in reproductive performance in aging mice are inversely correlated with increased and increasing expression of immune-reactive RFamide-related peptide-3 in the dorso-median region of the hypothalamus. Further, it is also quite possible that, decreased level of this peptide may lead to reproductive development and increased gonadal activity, although further experimental study is required to identify such mechanism of action involving both GnRH and GnIH like activity in relation to age-related gonadal development in mammals. RF-amide-related peptide-3 (RFRP-3), a mammalian neuropeptide, may be functionally similar and genetically related to this avian GnIH. Similar study show that although RFRP-3 has similar effects on LH as examined with GnIH in avian species, RFRP-3 has additional roles in regulating feeding and growth in the rat.[16] These results suggested that during the development the changes of hypothalamic RFRP system. An ovarian steroid, E2, may stimulate RFRP mRNA expression in the prepubertal period when the basal concentration of E2 is low.[14] These results suggested that the grass puffer PQRF a peptide may have many functions in the reproduction control that are dependent on the reproductive stages.[29] This study reported that hypothalamic, pituitary and testicular GnIH levels and its receptors mRNA significantly altered on 30 and 90 postnatal day. The GnIH proteins immunoreactive were mainly positioned to the spermatogenic cells, sustentacular cells and interstitial cells of the testis all through sexual development. The molecular and morphological data on the regulation of GnIH on reproductive development in male pigs.[44] These results suggested that social segregation in early-life consequences in lower levels of serotonin, which decrease the activity of GnIH neurone and may lead to reproductive failure.[30] These results supported that the regulation of kisspeptin as an important stimulatory for breeding season in deer, as in other species, but lack involvement of GnIH in the reproductive seasonality in deer. In appetite regulation case, the females exhibited pattern for NPY and c-MSH was as anticipated for breeding and non-breeding seasons, these peptides in sheep and the seasonal cycle of appetite reported for various species of deer, it is already reported. An opposite result in male deer most likely reflects the rejoinder of appetite regulating cells to balance of negative energy during the mating season.[2] [6]first time reported that appear to GnIH immunoreactivity in the brain during development of Indian major carp, Labeo rohita and assimilates it with the GnRH localization. GnRH and GnIH immunoreactivities were examined from the olfactory system to spinal cord through development. In the central nervous system and pituitary, both neuropeptides were localized in the diencephalon, telencephalon including the preoptic area and rhombencephalon. In addition, the GnRH and GnIH presence in several other brain regions including the olfactory system proposes their participation in the regulation of other function of physiological**.** The activation of RFRP neuronal positively correlated with delays to display several aggressive behaviours within winners. These outcomes were not examined in the anteroventral periventricular (AVPV) nucleus kisspeptin cell population. Together, these experiments indicate potential neuromodulatory role for RFRP in offensive behaviour and in disinhibiting the reproductive axis to make easy an increase in T in rejoinder to social challenge in mice [15].

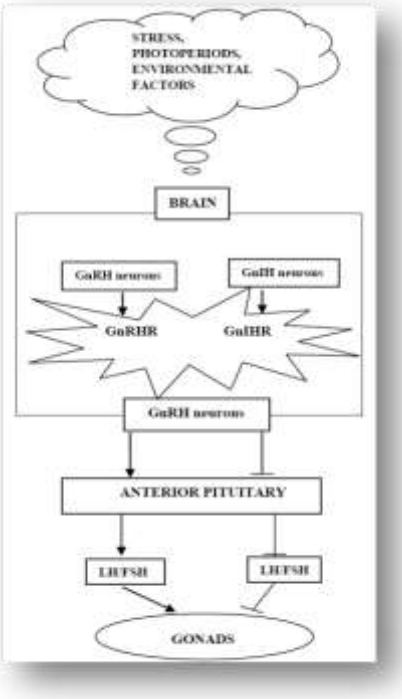

Figure.1: Reproduction is affected by environmental signal such as stress, photoperiods and environmental factors. GnRH and GnIH regulate of reproductive system. The hypothalamic GnRH neurons provide a positive stimulatory drive to the neuroendocrine reproductive axis while GnIH neurons produce an inhibitory drive to the reproductive axis. The expressions of GnRHR and GnIHR have been already reported on the GnRH – containing neurons of hypothalamus (modified from Wahab et al., 2015).

CONCLUSION

It is concluded that the discovery of GnIH has essentially modified our understanding of function and regulation of reproduction, although it remains to be verified whether GnIH is present in every vertebrate. In general the distribution of GnIH related to GnRH and its mode of action on LH/FSH release appear to be preserved between avian and mammals. This novel neuropeptide participate not only in reproductive functions but also in

social behaviour and autonomic mechanism. The further researches are needed to disclose any possible involvement of GnIH in vertebrates and human being. Proceeding studies will give an extra complete understanding of the GnIH physiological functions.

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