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The physiological role of orexins

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Abstract

Orexins/hypocretins are recently discovered neuropeptides synthetized mainly by neurons located in the posterolateral hypothalamus. Hypocretin-1 and -2 are the same peptides as orexin-A and orexin-B. Orexin A is a 33 amino acid peptide with N-terminal pyroglutamyl residue and two intrachain disulphide bonds. Orexin B is a linear peptide of 28 amino acids. These two peptides are potent agonists at both the orexin-1 (OxR1) and orexin-2 (OxR2) receptors. Orexin-A is selective ligand for OxR1 and OX2 binds both orexins. The structure of orexins and their receptors is highly conservative in mammals. Orexin A sequence is identical in several mammalian species (human, mouse, rat, bovine and porcine). Intracerebroventricular administered orexin-A stimulates food intake and energy expenditure. Orexins are also involved in the regulation of neurohormones and pituitary hormones secretion as well as in the control of cardiovascular and sleep-wake function. Orexins also play a role in the pathogenesis of narcolepsy. Mutation in the gene coding preproorexin or OxR2 receptor gene results in narcolepsy in mice and canine. In patients with narcolepsy orexin neurotransmition was altered and orexin level in cerebrospinal fluid was undetectable.

REVIEW ARTICLE

Introduction

Orexins/hypocretins are recently discovered neuropeptides synthetized by neurons located in the posterolateral hypothalamus. Hypocretin-1 and -2 are essentially the same peptides as orexin A and B. Orexins/hypocretins are derived from 131 amino acid human (130 aa in the rat) precursor prepro-orexin/hypocretin.

Orexin A/hypocretin 1 is 33 an amino acids peptide with N-terminal pyroglutamyl residue and two intrachain disulphide bonds. The human orexin A/hypocretin 1 sequence is identical to the mouse, rat, bovine and porcine orexin/hypocretin.

Orexin B is a linear peptide of 28 amino acids. There is 46% amino acid sequence identity between orexin B (OxB) and orexin A (OxA) [1]. It has been reported that Ox A, but not Ox B crosses the blood-brain barrier [2].

Orexins (Oxs) action is mediated via G protein-coupled receptors of two different subtypes (OxR1 and OxR2). OxR2 binds both orexin A and B and OxR1 is highly selective for orexin A [3]. Orexin immunoreactivity is widely distributed in the central nervous system (in the hypothalamus, septum, thalamus, brainstem and spinal cord [4, 5]. Orexin fibres innervating neurons in the locus coeruleus

represented the great peptidergic input in the pontine region [6].

The role of orexins in the control of food intake and energy expenditure

Immunohistochemical methods showed interaction between orexin axon terminales and neuropeptide Y (NPY) and proopiomelanocortin (POMC) –neurons in the arcuate nucleus (ARC) of the rats [7, 8, 9]. Muroya et al. [7] demonstrated that orexins directly regulated NPY, POMC and glucose-responsive neurons in the ARC and ventromedial hypothalamus (VMH) and these results may indicate on the orexigenic action of orexins.

It has been known that the lateral hypothalamus has classically been considered to be the "feeding" centre whereas the VMH is known as the "satiety" centre. Orexin neurons in lateral hypothalamus have connections with NPY and agouti-releated peptide (AgRP)-neurons in the ARC [6, 8]. The anatomical location of orexins and interaction with orexigenic peptides may indicate that the orexins play an important role in the control of food intake.

Orexins are neuropeptides present in hypothalamic nuclei involved in feeding, sleep-wakefulness, neuroendocrine homeostasis and autonomic regulation [10].

Orexins receptors are also expressed in peripheral tissues. The expression of orexin receptors as well as their biological role in the hypothalamus-pituitary axis, gastrointestinal tract, endocrine pancreas, gonadal tissues and other peripheral tissues was revealed [10, 11, 12].

There is some evidence that both NPY- and POMC-containing neurons are regulated simultaneously by both leptin and orexins [13]. The stimulatory effects of ghrelin and orexin and inhibitory effect of leptin play a role in the regulation of the activity of NPY neurons in feeding behavior [14]. However, melanin-concentrated hormone (MCH) effects on feeding is independent of NPY and orexins [9].

Neurons expressing orexins are stimulated by starvation and by hypoglycemia. Orexin neurons are activated by low level of glucose but inhibited by visceral feeding system mediated via vagal sensory pathway and the nucleus of the solitary tract [15].

It has been published that intracerebroventricular (icv) administration of orexin stimulate food intake in a dose-depended manner in young rats [16]. However, no effects were observed in old rats. The lack of response to orexin administration in old rats may be connected with the decrease in the OXR1 protein level in the hypothalamus in old rats [16].

Sweet et al [17] suggested that functional opioid pathways are necessary for orexin A-induced feeding.

Orexins play an important role in the regulation of metabolic rate [18]. The orexins like galanin are "fat-responsive" peptides that respond to circulating lipid [19].

Dalal et al [20] and Arihara et al [21] showed for the first time that orexin A can be detected in human plasma. Adam et al [22] showed that orexin levels were decreased in obese subjects and they correlated negatively with leptin plasma level. Matsumura et al [23] and Tomasik et al [24] demonstrated that orexin A concentration significantly correlated with body mass index (BMI) in normal subjects. Our previous data indicated that plasma orexin A were significantly lower, however plasma leptin and NPY were significantly higher in obese patients as compared with the control group of lean people [25]. Plasma orexin and leptin concentrations inversely change during fasting and significantly correlate with energy metabolism in non obese subjects [26].

It has been reported that orexins are not only involved in the regulation of energy balance but also play an important role in the control of pituitary hormones release.

The role of orexins in the regulation of hormones release

Orexin fibers innervated hypothalamic regions important for regulation of pituitary hormones release. Hagan et al [27] showed that orexin A activates neurons in the locus coeruleus. Orexin A releases neuropeptides from hypothalamic explants *in vitro* [28]. Intracerebroventricular administration of orexin A inhibited prolactin (PRL) release and this effect is partially dependent of dopamine [28]. A decrease of preproorexin expression during pregnancy and lactation is involved in the adaptation of homeostatic mechanism and in mechanism of elevated of PRL levels in these states [29].

On the other hand the high hypothalamic orexin concentration may contribute to the LH and PRL surges in the proestrus [30].

Centrally administred orexin A altered adrenocorticotropin (ACTH) and corticosterone release. Orexin A activates pituitary-adrenal axis [31]. It has been demonstrated increased c-fos mRNA in the paraventricular nucleus (PVN) of hypothalamus where corticotropin releasing hormone (CRH) neurons are located [27, 31]. Intracerebroventricular injection of orexin A increased ACTH and corticosterone and significantly increased CRH mRNA expression in PVN in normal rats but the response of pituitary-adrenal axis to orexin injection was inhibited in pregnant rats [32]. These results may indicate that orexin is involved in an anabolic adaptation in pregnancy. The existence of both OX1 and OX2 receptor subtypes in the adrenal cortex has been described [33]. This finding suggests that orexins may also affect directly on glucocorticoids release.

Lopez et al [34] in experiments "in situ hybridisation" showed that growth hormone releasing hormone (GHRH) mRNA in PVN is decreased after orexin A treatment. The effects of icv injection of OxA on hypothalamic somatostatin and GHRH mRNA levels demonstrated interaction among the systems involved in the control of food intake and GH secretion [34]. Chen and Xu [35] showed the increase of GH release *in vitro* in response to alone injection of OxB and to co-administration of OxA and GHRH. In experiments

in vivo icv injection of OxA inhibited GH release, and GH responses to ghrelin. However, orexin A failed to modify *in vivo* GH response to GHRH [36]. Orexin plays an inhibitory role in GH secretion and may be involved in regulatory mechanism nutritional status and GH release.

Campbell et al [37] found that 75–85% of GnRH neurons were contacted by orexin fibers and about 85% of GnRH neurons were co-localized with OxR1 and OxR2 – expressing GnRH neurons. These authors showed co-expression of NPY Y4 receptor and orexin fibers in relation to GnRH neurons. Stimulation of Y4 receptors leads to increase of LH release. These results may suggest that orexin modulates GnRH neurons directly via OxR1 or by stimulation Y4 receptor. Orexin A stimulates GnRH release from hypothalamic explants in vitro. Orexin stimulates LH secretion in steroid-primed ovariectomized (OVX) female rats and suppresses LH secretion in non-primed OVX rats [38].

The results of Kok et al [39] indicate that orexins are involved in the regulation of hypothalamo-pituitary-gonadal (HPG) axis in humans. Reduction of basal LH levels and normal LH release to GnRH stimulation in narcoleptic men suggests that orexin may play a role in the GnRH secretion [39]. Russell et al [40] demonstrated in the rat OxA interaction with hypothalamo-pituitary-gonadal axis. An interaction between orexin and NPY was confirmed because a specific NPY Y1 antagonist abolished *in vitro* release GnRH by orexin A [40].

The long form of leptin receptor is also found in orexin cells [41] and leptin treatment regulated orexin mRNA level [42].

Orexins – the role in the pathogenesis of narcolepsy

It has been observed that orexins deficiency play a role in the pathogenesis of narcolepsy [43]. Narcolepsy is the neurological disease, characterized by excessive daytime sleepiness, catalepsy, hypnagogic hallucinations and sleep paralysis. Deficiencies of orexins neurotransmission may be the cause of narcolepsy. Mutation in OxR2 in canine model and mice with a targeted deletion of preproorexin gene display a phenotype similar to that of human narcolepsy [44].

Orexins produced by the neuronal cell body located in the lateral hypothalamus and parafornical nucleus are important regulators of wakefulness, autonomic nervous system tone, neuroendocrinal secretion, feeding behavior and energy expenditure [8]. Plasma leptin concentration and its circadian pulsatility are altered in narcoleptic humans. In patients with narcolepsy orexin neurotransmission is altered and orexin level in cerebrospinal fluid is undetectable [46]. Genetic studies in autosonal recessive canine model and in gene-targeted mice have identified the hypothalamic orexin system is very important to evaluation of human narcolepsy [47, 48, 49].

Orexins – other physiological effects

Sakurai et al [50] observed very low orexin A concentration in patients with obstructive sleep apnea hypopnea syndrome (OSAHS). These results suggest that orexin may play a role in the pathogenesis of this syndrome.

Orexins are involved in the regulation of cardiovascular and autonomic functions. Samson et al [51] found that OxA and B injected into lateral cerebral ventricle stimulated the sympathetic function and resulted in an increase in blood pressure and heart rate [51]. Orexin containing fibres and receptors are associated with regulation of cardiovascular and autonomic function [52].

Orexin A may play a role in the thermoregulation [53] and hypothermic effects of OxA are mediated by NPY [53]. The role of OXR1 in mediating appoptosis was also demonstrated [54].

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