



DISTRIBUTION AND EXPRESSION OF APELIN/APLNR SYSTEM IN THE MAMMALIAN BODY

Martin Ivanov^{1,2*} and Stoyan Pavlov^{1,2}

¹*Department of Anatomy and Cell Biology, Medical University, Varna, Bulgaria*

²*Department of Stem Cell Biology and Advanced Computational Bioimaging, Research Institute, Medical University, Varna, Bulgaria*

*Apelin is an endogenous ligand for apelin receptor (APLNR), a G protein-coupled receptor (GPCR). This peptide is derived from a 77aa long pre-proapelin. Upon cleavage by different peptidases, several isoforms that bind to the receptor with different strength and downstream effects are created. Many cellular processes that are regulated by the apelinergic system have been discovered – neural and immune functions, blood pressure regulation, fluid homeostasis, hormonal release, cellular proliferation, etc. Additional research has been also made to elucidate the role of the system in pathological conditions affecting the brain and the peripheral organs. In this review, we summarise the distribution of the apelinergic system components in the mammalian organism. We also include a comparative analysis between three different species - human (Homo sapiens), mouse (Mus musculus), and rat (Rattus norvegicus). **Biomed Rev 2023; 34: 61-68***

Keywords: apelin, APLN, APLNR, CNS, peripheral organs, expression

INTRODUCTION

Apelin receptor (APLNR) and its ligand apelin are widely expressed in the mammalian body. Apelin is a bioactive peptide with different isoforms that was first isolated from bovine stomach extracts (1). It was identified as a ligand for the human APLNR (Apelin Receptor), an orphan G protein-coupled receptor (GPCR) (2).

Monitoring the extracellular acidification rate of CHO (Chinese Hamster ovary) cell expressing APLNR, the group

led by Fujino was able to isolate the human, bovine, mouse, and rat cDNA responsible for the synthesis of apelin ligand. All those cDNAs encode for a 77 aa (amino acid) residue pre-propeptide named pre-proapelin (1,3,4).

The human apelin gene is mapped to chromosome Xq25-26.1 (5). It is synthesized as a pre-proapelin with a length of 77aa residues. Upon cleavage by different proteases that target basic amino acid residues like lysine and arginine, pro-apelin is converted into shorter bioactive isoforms. A

Received 28 August 2023, revised received 17 October 2023, accepted 18 October 2023.

*Correspondence to: Dr Martin Ivanov, Department of Anatomy and Cell Biology, Medical University, BG-9002 Varna, Bulgaria
Tel.: +359 890598282; E-mail: Martin.Ivanov@mu-varna.bg

36 amino-acids long peptide, named apelin-36 is derived from the C-terminal region of pre-proapelin-77. Apelin-13 and apelin-17 are created by cleavage of apelin-36. Those shorter forms exhibit much stronger affinity and binding capabilities to the APLNR receptor compared to the longer 36aa peptide (1,3,6).

Even simple modifications of the apelin ligand structure cause significant change in the peptide ligand activation. Substitution of the Leu2, Arg2, Lys8, or Phe13 with Ala results in a decrease of the activation activity of the ligand when binding to APLNR (7). Pyroglutamination of Glu at the N-terminus of the peptide modifies Apelin-13 into pyroglutaminated form - (Pyr¹) apelin-13, that is more stable (3).

The gene for APLNR in humans is located on q12 of chromosome 11 and encodes a protein with a length of 377aa. APLNR is a class A g-group of G-protein-coupled receptors (GPCRs) members and like all GPCR, APLNR has 7 hydrophobic transmembrane (TM) regions (2,8). Recently, the atomic-resolution structure of the receptor-bound with synthetic mimetic agonist has been presented (9).

Interestingly, the level of homology of pre-proapelin be-

tween rat, human, bovine, chimpanzees and monkey (*Macaca mulatta* and *Cercopithecus aethiops*) is ~80%, while the homology of the mature isoforms of apelin and the APLNR is 100% (2,4,6,8). APLNR is most closely related to the angiotensin II receptor (30–40% identity in amino acid sequence), although angiotensin II does not interact with the APLNR when expressed in Chinese hamster ovary (CHO) cells or in fibroblasts (1, 2).

EXPRESSION STUDIES OF APELIN AND APLNR IN ORGANS IN MAMMALIAN TISSUE

Studies using many techniques including in situ hybridization, immunohistochemistry, Western and Northern blot, and reverse transcriptase-polymerase chain reaction (RT-PCR) for identification of pre-proapelin, apelin ligand or APLNR in the organism, have provided a large body of data on the ubiquitous expression of this GPCR and its ligand in mammalian tissues (Table 1). Here, we review recent data about the expression of the apelinergic system components in different regions of the mammalian organism.

Table 1. Literature evidence for apelin and APLNR expression in human, rat, and mouse tissues

System	Tissue	Human		Rat		Mouse	
		Apelin	APLNR	Apelin	APLNR	Apelin	APLNR
	Whole Brain	(10),(11)	(7), (11)		(7), (4), (12)		(7), (12)
	Cortex (overall)					(13)	(13),(14)
CNS Telencephalon	Frontal Cortex		(7),(15)	(5)	(7), (5)		
	Temporal Cortex	(16)		(16)	(7)		
	Parietal Cortex				(7), (5)		
	Occipital Cortex			(5)	(7)		
	Entorhinal Cortex				(7), (17)		
	Piriform cortex				(17)		
	Rostro-Splenial Cortex				(7)		
	Striatum		(15)		(7), (4), (8)	11	(13)
	Putamen		(7)				
	n.Caudatus		(7)				
	Basal ganglia						
	n. Accumbens		(7)		(7)		
	Corpus callosum		(7), (15)				
	Hippocampus (GD+ CA)	(16)	(7), (15)	(16)	(7), (4), (17), (8), (5),(18)	(13)	(13)
	Amygdala		(7),(15)		(7)		

		Human		Rat		Mouse	
System	Tissue	Apelin	APLNR	Apelin	APLNR	Apelin	APLNR
Diencephalon	Thalamus		(7), (15)		(7)		
	Hypothalamus (Overall)		(7)		(7), (5)	11	(12), (13)
	-SON			(17)	(17), (8), (12)	(19)	
	-PVN magnocellular			(17)	(17),(8), (12), (5)	(19)	
	PVN parvocellular				(17),(8), (12)		
	Suprachiasmatic nucleus					(19)	
	Accessory neurosecretory nuclei					(19)	
	Preoptic area -Medial -Lateral -Median			(17) (17)			
	Pineal gland				(17), (5)		
Mesencephalon	S. nigra		(7), (15)		(7), (4), (17)		
Rhombencephalon	Cerebellum		(7)		(7), (4)(5)	(13)	(13)
	Medulla oblongata		(15)	(17)	(7), (4)	11	(13)
	Pons			(17)	(7), (5)	11	(13)
	Reticular Formation						
Others CNS	Spinal cord		(7), (15)	(20)	(7),(21)	(13)	(13)
	Dorsal-root ganglion				(7)		
	Dura/Pia/Arachnoid mater	(10)			(8)		
Endocrine System	Pituitary gland -Anterior lobe -Intermedial lobe -Posterior Lobe		(7)	(20) (12) (12) (12)	(17), (12) (17), (12) (12)	(19) (19) (19)	(12) (12) (12)
	Thyroid gland				(4)		
	Adrenal glands	(10)		(12), (20)	(12),(4)		(12)
Cardiovascular system	Heart	(10)	(7)	(12), (20), (3)	(12), (4), (8)		(7),(12)
Respiratory system	Lung	(10)	(7)	(12), (20),(3)	(7), (12),(4), (8)		(7),(12)
	Trachea				(4)		
Skeleto-muscular system	Skeletal muscle	(10)			(7), (4), (8)		(7)
	Bones				(4)		
	Cartilage	(10)			(4)		

		Human		Rat		Mouse	
System	Tissue	Apelin	APLNR	Apelin	APLNR	Apelin	APLNR
Gastrointestinal system	Stomach		(7)	(12), (20)	(12),(4)		(12)
	Small intestine		(7)	(12), (20)	(12), (4)		(12)
	Large intestine			(12), (20)	(12),(4)		(12)
	Pancreas	(10)	(7)	(20)	(12)		
	Liver	(10)	(7)	(12), (20)	(8)		(7)
	Submandibular gland				(4)		
Urinary system	Kidney	(10)	(7)	(12), (20)	(7), (12), (4), (8)		(7), (12)
	Urinary bladder				(12)		(7), (12)
Male/Female reproductive system	Testis			(12), (20), (3)			(7)
	Prostate						
	Placenta	(10)	(7)		(4)		
	Uterus		(7)	(12), (20)	(7), (12), (4), (8)		(7), (12)
	Ovary			(12), (20), (3)	(7), (12),(4), (8)		(7), (12)
	Placenta		(7)		(4)		
	Mamillary gland			(12), (20), (3)	(7),(12),(4)		(12)
Skin		(10)			(4)		
Adipose tissue		(10)		(20), (3)	(7), (4)		
Immune system	Spleen	(10)	(7)	(20)	(4)		(7), (12)
	Thymus	(10)			(4)		(7), (12)

EXPRESSION OF THE APLNR IN HUMAN, RAT AND MOUSE BRAINS

The overall expression of mRNA^{APLNR} in the whole brain in humans and mice is low while in rats it is relatively high. Expression of the APLNR is observed in different parts of the human telencephalon. Data shows that the presence of the receptor is detected in the spinal cord and corpus callosum as well. Interestingly the spinal cord exhibits very high levels of APLNR (4,7,13,15,21).

Expression is not limited to those regions and can be also observed in the temporal cortex, putamen, and nucleus caudatus (3,5–7,15,22). APLNR is also present in areas of the brain which are part of the limbic system such as the hippocampus (both DG and all of the CA), *amygdala* and *nucleus accumbens*,

in the olfactory system with areas such as the olfactory bulb, olfactory tubercle, nucleus of the lateral olfactory tract and cortical regions related to olfaction (entorhinal and piriform cortex) (3,5–7,15,22).

A recent article by our team showed that APLNR is expressed in the subventricular region located in the caudate nucleus, an area related to neurogenesis (22). Our unpublished data, suggest that APLNR is also expressed in the SGZ (subgranular zone) of the hippocampal formation, another region related to adult neurogenesis.

APLNR is detected throughout other regions of the brain including thalamus and hypothalamus (diencephalon), substantia nigra (mesencephalon), cerebellum (7), and medulla oblongata (rhombencephalon) (4).

In the rat brain the expression of APLNR is wider and

additionally to the regions described in human it includes the entorhinal, occipital, rostro-splenial, parietal and frontal cortex. It was also observed in parts of the rhombencephalon, including pons and medulla oblongata. Interestingly, stronger expression is observed in spinal cord and *nucleus accumbens* when compared to the human data (7). Compared to mice and humans, the presence of mRNA^{APLNR} is seen in the dorsal root ganglion only in rat specimens (7). Of note, the expression of mRNA^{APLNR} in the brain and the periphery in neonates is much higher than in adults (4).

In the hypothalamus, abundant expression is seen in regions related to drinking behaviour such as SON (supraoptic nucleus) and PVN (paraventricular nucleus) which are important structures related to the release of posterior pituitary hormones (5,6,8,12,17). Both parvocellular and magnocellular parts of the PVN of the hypothalamus express APLNR mRNA. Double labelling of the cell utilizing immunohistochemistry and RNA in-situ hybridization shows co-expression of vasopressin (AVP) and APLNR in SON (5,17). This leads to the conclusion that APLNR is involved in the modulation of the vasopressin activity. A proposed mechanism is that the apelin ligand may be synthesized locally or by the SON neurons themselves which might exert an inhibitory action on the APLNR expressing vasopressin neurons *via* a paracrine or autocrine process (17).

Both neuronal and glial subpopulations express APLNR. GFAP expressing astrocytes are shown positive for APLNR and can be observed in the cortex, the white matter of the spinal cord and caudate nucleus (7,15,22). Neuronal cells, positive for NeuN, also express APLNR in the grey matter of the spinal cord. Those neurons represent a cholinergic population of cells, given the fact that they co-express choline acetyltransferase (ChAT). Some studies point out also that microglia do not express APLNR (13).

EXPRESSION OF APLNR IN THE PERIPHERAL TISSUES

Respiratory system

APLNR transcripts can be found in most of the internal organs. Some of the highest levels of expression are observed in the respiratory system. There is a strong presence of APLNR in the lung parenchyma, while the vasculature associated with the respiratory system and the airways do not express the receptor (8,12). Detectable levels of the protein can be also observed in the foetal and neonatal lung parenchyma (2,4,5,20). Given the apelin ligand chemotactic effect (4), some authors hypothesise the role of the apelinergic system in the morphogenetic

process (20). Comparison between the expression of APLNR in humans, mice, and rats shows higher levels in mice and rats in contrast with the human lung tissue.

Cardiovascular system

In the circulatory system, the APLNR is found in the heart, between cardiomyocytes of the atria and the ventricles. In contrast, APLNR is present in neither the blood vessels nor the pericardium (4,8,15). Side by side comparison shows stronger expression in rats and mice compared to human cardiovascular system (7).

Endocrine system

APLNR can be found in parts of the pituitary gland - anterior and posterior hypophysis, and the intermediate lobe (7,12,17). An expression can be found also in the thyroid and adrenal glands (4,12).

Gastrointestinal system

APLNR is present in both the upper and lower gastrointestinal (GI) system. Expression is seen in the stomach (mucosa and glandular epithelium), the small and large intestine, and in the associated with GI glands- pancreas the liver and submandibular gland (4,7,8,10,12,20,23).

Urinary system

The overall expression in the mammalian renal parenchyma is low to moderate. The presence of APLNR transcripts is detected in the kidneys. Part of the cortex and the medulla are positive, including the cortical glomeruli, and the inner and outer medulla. The distal and proximal tubules, the collecting ducts, macula densa, and vasculature of the kidney parenchyma do not express the receptor (7,8,12).

Male and female reproductive system

In the female reproductive system, transcripts were found in the ovary and the uterus. In the ovary, a pronounced expression was observed only in the corpus luteum (8,12). The expression in the uterus was observed in stromal cells in the endometrium while the myometrium is not labelled (7,8,12). By contrast, in the male reproductive system, the APLNR expression is restricted to the mouse testis. In the rat and human testes, the levels were very low to undetectable (7,8,12).

Lymphatic system

In human, mRNA^{APLNR} is expressed in the spleen, in comparison with rat, where no expression can be observed (7).

Musculoskeletal system

APLNR is observed throughout the entire locomotor system: in the bones, cartilage, and muscles (4,7,8).

APLNR is also expressed in the skin and adipose tissue of rats (4, 7).

Expression of apelin in human, rat and mouse brain

The apelin ligand has been found expressed in the brain of rodents and humans (3,5,7,20,24). Apelin and its precursor are quite ubiquitous the brain tissues and can be detected in different regions of the CNS, including the cortex, cerebellum, thalamus, hypothalamus, hippocampus, basal ganglia, medulla, pons, and spinal cord (3,5,7,13,17,20,24). Many thalamic nuclei contain the peptide, including anterodorsal, mediodorsal, anteromedial, ventroposterior, ventromedial and medial habenular(5), paraventricular nuclei (17). Apelin is predominantly detected in different hypothalamic nuclei including the medial and lateral preoptic nuclei, paraventricular, periventricular, supraoptic arcuate, ventromedial, and dorsomedial nuclei (5,7,17,19,24).

Cells expressing apelin in the PVN, SO and accessory neurosecretory nuclei (Acc) project their processes towards the medial eminence, passing through the infundibulum toward the posterior lobe of the hypophysis. Cells located in those regions of the hypothalamus are known to be associated with oxytocin and vasopressin production. Interestingly neurons that contain oxytocin are associated with neurophysin-I and colocalize with apelin. This shows that APN is present in oxytocin secreting neurons. Apelin presence, but neurophysin-I absence is also possible. In the latter case, the neurons are shown to be vasopressinergic. In both cases, APLN is a major constituent of both vasopressin and oxytocin-containing neuronal vesicles (19). Intravenous or intraperitoneal administration of apelin causes an increase in the water intake (5,17).

In the midbrain, the ligand is observed in the interpeduncular nucleus, inferior and superior colliculus, periaqueductal grey matter, and the nucleus of Darkschewitsch. Expression is also present in the anterior pretectal area of the midbrain and the pineal body of the diencephalon, two structures that are related to circadian rhythms (5).

In the hippocampus, apelin can be observed in the dentate gyrus and CA regions of Ammon's horn (5). Interestingly, in patients with refractory epilepsy and in an epilepsy rat model, a significant elevation of apelin is observed in the hippocampus and the temporal cortex (16).

In the brainstem, the spinal nucleus of the trigeminal nerve

and the parabrachial nuclei are positive for apelin. Data indicates that apelin is expressed in some neurons and glial cells (10). Evidence from other researchers, however, shows that the cells that express apelin in the cortex and the hippocampus have phenotypic characteristics of neurons, but not of glial cells (16).

EXPRESSION OF APELIN IN HUMANS, RATS, AND MICE IN THE PERIPHERAL TISSUES

Respiratory system

In the lungs of rodents and humans, the apelin ligand and its precursor protein are detected in very high amounts (3,7,10,12,20). IHC staining of the respiratory organs labels specifically the ciliated epithelium, the smooth muscles cells, the secretory ducts and the acinar cells of the bronchial glands (10).

Cardiovascular system

In the heart, apelin is expressed at high levels in the connective tissue and in the cardiomyocyte's cytoplasm(3,10,12,20).

Endocrine system

Apelin can be found in both the anterior and the posterior pituitary (7,8,12,20). In the adrenal gland, Apelin is present in the medulla and all three layers of the adrenal cortex of the gland, with highest amounts in the fasciculate zone(10,12,20). Presence in the adeno- and neurohypophysis, and the adrenal glands suggests that the APLNR/APLN system plays a role in the hypothalamic-pituitary-adrenal axis (HPA).

Gastrointestinal system

Apelin can be detected in different parts of the GI tract: the stomach, small intestine, and large intestine (5). From the glands associated with the gastrointestinal system, the peptide is present in the pancreas and the liver (5,20). In humans, the liver expression is localised to the hepatocyte's cytoplasm and the histiocytes. In the same species, both the endocrine and exocrine pancreas are positive for the peptide (10).

Urinary system

APLN transcripts are observed in the kidney glomeruli and the collecting tubules (5,10).

Skin and Adipose tissue

In the skin, the IHC study shows high levels of apelin labelling in all layers of the epidermis. The staining was not restricted

to the epidermis and was present also in the skin glands (10). Detectable amounts of mRNA^{apelin} and its precursor are found in the white adipose tissue (3,10,20) .

Male/Female Reproductive tissue

In the female reproductive system, high levels of apelin are detected in the mammary gland (3,20). Basal level of expression of the peptide is found in mammary glands of non-pregnant animals, but interestingly during pregnancy, the ligand levels increase with a peak around parturition, and then gradually return to basal levels. Apelin is actively secreted from the mammary gland and it can be found in the colostrum and milk during lactation (3). The protein can be also observed in the ovary (3,20). Additionally, in the placenta, the syncytiotrophoblast and the cytotrophoblast are also positive for apelin (10). In the male reproductive system, the ligand is detected at moderate levels in the testis (3,5,20).

Lymphatic system/Immune system

Apelin is shown to be capable of modulating the immune system response. It suppresses the production of IL-2, IL-4, and IFN-gamma (3). Transcripts are seen in the spleen (5) In the human spleen, apelin can be observed in both the white and the red pulp. High expression is also evident in the thymus' Hassall's corpuscles and in thymocytes (10).

Locomotor system

In the human, apelin is detected in the skeletal muscle fibers and in the chondrocytes (10).

Conclusion and future directions

We have provided a comprehensive review of the expression of the APLNR/APLN system across the mammalian organism. Since APLNR/APLN is expressed in a wide variety of tissues and organs, a big number of studies have shown that the system participates in the regulation of diverse physiological processes. It has been also shown that the apelinergic system is involved in the pathophysiology of multiple neurological and non-neurological diseases. In the brain and the peripheral tissues, apelin exhibits protective effects including inhibition of caspase-3 and ROS, and attenuation of endoplasmic reticulum stress. APLNR is a co-receptor along with CD4 for human (HIV) and simian (SIV) immunodeficiency virus entry (25). Thus, apelin ligand receptor activation can block the entry of the virus in NP-2/CD4 cells (26). Finally, emerging evidence for apelin analogues with stronger receptor binding affinity and more potent effects improve our understanding

of the apelinergic system and pave the way for translational and clinical trials.

CONFLICT OF INTEREST

There authors declare no conflict of interests exists.

ACKNOWLEDGEMENTS

This work was supported by intramural grants of the Medical University, Varna, Bulgaria (19028/2019 and 22020/2022)

REFERENCES

1. Tatemoto K, Hosoya M, Habata Y, *et al.* Isolation and Characterization of a Novel Endogenous Peptide Ligand for the Human APLNR Receptor. *Biochem Biophys Res Commun* 1998; 251:471–6. DOI:10.1006/bbrc.1998.9489
2. O'Dowd BF, Heiber M, Chan A, *et al.* A human gene that shows identity with the gene encoding the angiotensin receptor is located on chromosome 11. *Gene* 1993; 136:355–60. DOI:10.1016/0378-1119(93)90495-O
3. Habata Y, Fujii R, Hosoya M, *et al.* Apelin, the natural ligand of the orphan receptor APLNR, is abundantly secreted in the colostrum. *Biochim Biophys Acta - Mol Cell Res* 1999; 1452:25–35. DOI:10.1016/S0167-4889(99)00114-7
4. Hosoya M, Kawamata Y, Fukusumi S, *et al.* Molecular and Functional Characteristics of APLNR. *J Biol Chem* 2000; 275:21061–7. DOI:10.1074/jbc.M908417199
5. Lee DK, Cheng R, Nguyen T, *et al.* Characterization of Apelin, the Ligand for the APLNR Receptor. *J Neurochem* 2001; 74:34–41. DOI:10.1046/j.1471-4159.2000.0740034.x
6. De Mota N, Lenkei Z, Llorens-Cortès C. Cloning, pharmacological characterization and brain distribution of the rat apelin receptor. *Neuroendocrinology* 2000; 72:400–7. DOI:10.1159/000054609
7. Medhurst AD, Jennings CA, Robbins MJ, *et al.* Pharmacological and immunohistochemical characterization of the APLNR receptor and its endogenous ligand apelin. *J Neurochem* 2003; 84:1162–72. DOI:10.1046/j.1471-4159.2003.01587.x
8. O'Carroll AM, Selby TL, Palkovits M, Lolait SJ: Distribution of mRNA encoding B78/APLNR, the rat homologue of the human APLNR receptor, and its endogenous ligand apelin in brain and peripheral tissues. *Biochim Biophys Acta - Gene Struct Expr* 2000; 1492:72–80. DOI:10.1016/S0167-4781(00)00072-5

9. Ma Y, Yue Y, Ma Y, *et al.*: Structural Basis for Apelin Control of the Human Apelin Receptor. *Structure*. 2017; 25:858-866.e4. DOI:10.1016/j.str.2017.04.008
10. De Falco M, De Luca L, Onori N, *et al.*: Apelin expression in normal human tissues. *Vivo Athens Greece*. 2002; 16:333–6.
11. Chen H, Wang L, Wang W, *et al.*: ELABELA and an ELABELA fragment protect against AKI. *J Am Soc Nephrol*. 2017; 28:2694–707. DOI:10.1681/ASN.2016111210
12. Pope GR, Roberts EM, Lolait SJ, O'Carroll AM: Central and peripheral apelin receptor distribution in the mouse: Species differences with rat. *Peptides*. 2012; 33:139–48. DOI:10.1016/j.peptides.2011.12.005
13. Kasai A, Kinjo T, Ishihara R, *et al.*: Apelin deficiency accelerates the progression of amyotrophic lateral sclerosis. *PLoS ONE*; 2011 DOI:10.1371/journal.pone.0023968
14. Zeng XJ, Yu SP, Zhang L, Wei L: Neuroprotective effect of the endogenous neural peptide apelin in cultured mouse cortical neurons. *Exp Cell Res* 2010; 316:1773–83. DOI:10.1016/j.yexcr.2010.02.005
15. Matsumoto M, Hidaka K, Akiho H, Tada S, Okada M, Yamaguchi T: Low stringency hybridization study of the dopamine D4 receptor revealed D4-like mRNA distribution of the orphan seven-transmembrane receptor, APLNR, in human brain. *Neurosci Lett* 1996; 219:119–22. DOI:10.1016/S0304-3940(96)13198-0
16. Zhang X, Peng X, Fang M, *et al.*: Up-regulation of apelin in brain tissue of patients with epilepsy and an epileptic rat model. *Peptides* 2011; 32:1793–9. DOI:10.1016/j.peptides.2011.08.006
17. Reaux A, De Mota N, Skultetyova I, *et al.*: Physiological role of a novel neuropeptide, apelin, and its receptor in the rat brain. *J Neurochem* 2001; 77:1085–96. DOI:10.1046/j.1471-4159.2001.00320.x
18. O'Donnell LA, Agrawal A, Sabnekar P, Dichter MA, Lynch DR, Kolson DL: Apelin, an endogenous neuronal peptide, protects hippocampal neurons against excitotoxic injury. *J Neurochem* 2007; 102:1905–17. DOI:10.1111/j.1471-4159.2007.04645.x
19. Brailoiu GC, Dun SL, Yang J, Ohsawa M, Chang JK, Dun NJ: Apelin-immunoreactivity in the rat hypothalamus and pituitary. *Neurosci Lett* 2002; 327:193–7. DOI:10.1016/S0304-3940(02)00411-1
20. Kawamata Y, Habata Y, Fukusumi S, *et al.*: Molecular properties of apelin: tissue distribution and receptor binding. *Biochim Biophys Acta BBA - Mol Cell Res* 2001; 1538:162–71. DOI:10.1016/S0167-4889(00)00143-9
21. Xiong Q, He W, Wang H, *et al.*: Effect of the spinal apelin-APLNR system on the pathogenesis of chronic constriction injury-induced neuropathic pain in rats. *Mol Med Rep* 2017; 16:1223–31. DOI:10.3892/mmr.2017.6734
22. Chongtham MC, Wang H, Thaller C, *et al.*: Transcriptome Response and Spatial Pattern of Gene Expression in the Primate Subventricular Zone Neurogenic Niche After Cerebral Ischemia. *Front Cell Dev Biol* 2020; 8:1–17. DOI:10.3389/fcell.2020.584314
23. Wang G, Anini Y, Wei W, *et al.*: Apelin, a New Enteric Peptide: Localization in the Gastrointestinal Tract, Ontogeny, and Stimulation of Gastric Cell Proliferation and of Cholecystokinin Secretion. *Endocrinology* 2004; 145:1342–8. DOI:10.1210/en.2003-1116
24. Reaux A, Gallatz K, Palkovits M, Llorens-Cortes C: Distribution of apelin-synthesizing neurons in the adult rat brain. *Neuroscience* 2002; 113:653–62. DOI:10.1016/S0306-4522(02)00192-6
25. Edinger AL, Hoffman TL, Sharron M, *et al.*: An Orphan Seven-Transmembrane Domain Receptor Expressed Widely in the Brain Functions as a Coreceptor for Human Immunodeficiency Virus Type 1 and Simian Immunodeficiency Virus. *J Virol* 1998; 72:7934–40. DOI:10.1128/jvi.72.10.7934-7940.1998
26. Zou M-X, Liu H-Y, Haraguchi Y, Soda Y, Tatemoto K, Hoshino H: Apelin peptides block the entry of human immunodeficiency virus (HIV). *FEBS Lett* 2000; 473:15–8. DOI:10.1016/S0014-5793(00)01487-3