ISSN: 2279 - 0594

Journal of Biomedical and Pharmaceutical Research

Available Online at www.jbpr.in

CODEN: - JBPRAU (Source: - American Chemical Society)

Volume 5, Issue 3: May-June: 2016, 40-45

Review Article

Interaction of tau proteins and orexin receptors in dementia and allied syndromes

Manoj G Tyagi

Department of Biotechnology, Vellore Institute of Technology, Vellore, Tamilnadu

Received 26 May 2016; Accepted 15 June 2016

ABSTRACT

The neuronal and neurobiological basis of cognitive dysfunction accompanying neurodegenerative processes forms an important focal point of research interest in neuroscience. The so-called, mild cognitive impairment refers to a syndrome of neuropsychological deficits, which exceed normal cognitive effects of aging, but actually are subclinical in nature. The syndrome is associated with an elevated tau protein level. Neurodegeneration markers, such as tau proteins and ß-amyloids, but also hypocretins (orexins), in the liquor cerebrospinalis have gained a great significance for the detection and clinical monitoring of diseases like Alzheimer's disease and narcolepsy as well possibly in other forms of cortical dementia. The objective of this study is a better understanding of the functional interrelations between orexin and tau in transmitter regulation and in cognitive impairment and dementia.

Keywords: Orexin,Tau, dementia, neurodegeneration, neuropeptides, alzheimer's disease

INTRODUCTION:

Orexin (OX) or hypocretin are neuropeptides which function largely to regulate the sleep-wake cycle and feeding behaviour and possibly in neural regulation[1]. OXs are products of the cleavage of a 130 amino acid precursor protein also known as prepro-orexin (PPO) to form two distinct proteins from one precursor molecule. Initially the 33 amino acid N-terminal is cleaved, forming pro-orexin; this is then cleaved by pro-hormone 22 convertases giving rise to one molecule each of orexin-A (OXA) and orexin-B (OXB) [2]. Sleep disorders are reported in Parkinson's disease, and dementia with Lewy Bodies and Alzheimer' Disease. Whilst the pathophysiology of sleep disorders in these diseases remains unclear, it has been linked with reduced hypocretin levels in other sleep disorders such as narcolepsy. On the other hand in 1983, it was discovered that tau could be phosphorylated at multiple sites by various protein kinases, including cyclic-AMP-dependent protein kinases and casein kinase type-1. Further studies showed that tau is a phosphoprotein and that phosphorylation negatively regulates its ability to stimulate MT assembly and are implicated in the senile dementia of the Alzheimer's. Both the orexin and tau proteins could be potential neurodegenerative markers. This review article evaluates the role of tau proteins and orexin in dementia *per se* and allied syndromes.

Orexin receptors and affinities: OXA and OXB peptides bind to two different receptors: orexin receptor 1 (OX1R) and orexin receptor 2 (OX2R); both of which are GPCRs. OX1R is 425 amino acids and OX2R is 444 amino acids, sharing 66 % sequence identity. Both receptors are strongly conserved amongst mammals and have almost 94% homology between human and rats[3]. OXs have different affinities for the receptors; OX1R binds OXA with very high affinity (IC50: 20nM in a competitive binding assay) but it exhibits a much lower affinity for OXB of 420nM [4]. OX2R shows less discrimination between the OXs and they both have similarly high affinities of IC50 38nM (OXA) and 36nM (OXB)[5,6].

Expression and distribution of orexin receptors: The most intense areas of OX producing neurons are around the paraventricular Nucleus (PVN), arcuate nucleus and tuberomammillary nucleus. OX2R is mostly expressed in the PVN, cerebral cortex and nucleus accumbens. OX1R is highly expressed in the hippocampal formation, dorsal raphe and locus cereuleus[7]. They are co-expressed in the ventromedial hypothalamic nucleus, posterior hypothalamus, dorsomedial nucleus,

*Corresponding author: Dr. Manoj G Tyagi | E-mail: tyagi243@yahoo.co.in

hippocampal formation, thalamic nuclei and subthalamic Nuclei[8]. These specific areas of dense orexinergic neurons fire rapidly during wakefulness, at a much slower rate during non-rapid eye movement (REM) and not at all in REM[9]. It is believed that OX exerts its effects on wakefulness by acting upon histaminergic neurons through OX2R. When injected in rats OX experience prolonged periods of wakefulness and this is less pronounced when the histamine H_1 receptor antagonist pyrilamine is also administered [10,11]. Other areas of OXR expression include the testes and adrenal glands, however only small amounts of OXR expression are found here[12].

Orexin and dementia: Recent results are consistent with studies showing reduced cerebrospinal fluid levels of hypocretin in Parkinson's disease cases[13,14] and similar findings in experimental animal models [15,16]. It has been shown that a reduction of neocortical hypocretin-immunoreactive fibers and neurons in Dementia with Lewy Bodies cases compared to Alzheimer's disease and control. As most of the hypocretin is produced in the lateral hypothalamus [17-19], these hypocretin-immunoreactive neurons in the neocortex may represent uptake from projecting fibers. Alternatively, the presence of hypocretin in these neurons may indicate it is being produced by them, this is consistent with a study reporting the presence of pre-pro hypocretin mRNA in pyramidal neurons in the hippocampus under basal conditions and during epilepsy[20] suggesting that hypocretin may be made in cell populations other than the lateral hypothalamus.

Tau phosphorylation and alzheimer's disease:
Tau (tubulin-associated unit) proteins are isolated from porcine brain extracts as a heat-stable, highly soluble protein essential for microtubule (MT) assembly. Following the initial discovery of tau, two research studies reported the process of tau purification and its physical and chemical properties, including the ability of tau to become phosphorylated[21]. Fig.2 depicts the importance of Tau propogation in aging induced dementia as well as in the dementia of Alzheimer's type. This abnormal behavior is enhanced by conformational changes and misfoldings in the normal structure of tau that leads to its aberrant aggregation into fibrillary structures inside the neurons of demented individuals [22,23]. Thus, most of the altered pools of tau protein in the disease are redistributed and aggregated in both the somato-dendritic compartment and isolated processes of affected neurons. Alterations in the amount or the structure of tau protein can affect stabilization of microtubules and other processes related to this protein.

Orexin receptors and phospholipase C mediated pathway:

OXRs are linked to the phospholipid breakdown mediated signal transduction. OXRs initiate increases in intracellular calcium in many cell types including CHO, HEK and neuro-2a [24,25].This activation occurs through activation of PLC potentially through Gq coupling. PLC is a family of cytosolic phosphoinositide specific enzymes whose main target is PIP_2. Cleavage by PLC generates DAG and IP_3, IP_3 then binds endoplasmic reticulum based IP_3 receptors and facilitates the release of calcium into the cytosol [26,27].This depletion of calcium causes the membrane to allow a calcium influx to replace the diminished stores and this can regulate calcium responsive ion channels, enzymes and proteins. This depolarisation also activates voltage gated calcium channels. OX1R and OX2R strongly activate PLC in several different cell types including CHO, HEK and the neuronal neuro-2a cell line [28]. However it is not well understood whether the downstream effects of this elevated PLC is to increase calcium or to elevate DAG for potential PKC elevation.

Interaction of tau proteins and orexin in neurodegenerative disorders: Human studies of CSF samples collected via lumbar catheters in young healthy volunteers have revealed peak CSF Aβ concentrations in the evening and lower concentrations overnight [29]. A slight correlation between $A\beta_{42}$ and hypocretin-1 levels was reported in 6 patients with AD and 6 healthy controls, with no between-group difference in CSF hypocretin-1 levels and its circadian amplitude. In contrast, no association between $A\beta_{42}$ and hypocretin-1 was reported in either control subjects or patients with AD in a larger study, but with low hypocretin-1 levels in 9 female patients with LBD[30-31]. Higher CSF t-tau protein levels mark the AD neurodegeneration. Increased t-tau levels represent a sign of rapid cognitive decline because

they have been faster more pronounced neuronal degeneration, supporting the transition from early to more advanced disease stages[32-33]. CSF hypocretin levels were directly correlated with t-tau protein levels in AD[34-35]. This finding suggests that higher hypocretin levels may be related to rapid tau-mediated degeneration in AD. The pathogenesis of AD may therefore involve dysregulation of the hypocretins system, with over expression of hypocretins output and function, and Hypocretin (orexin) pathology in Alzheimer's disease. Evidence that CSF hypocretin-1 may be related to CSF Tau and pTau.These associations might be independent of AD, but It can be speculated that high hypocretin-1 concentrations contribute to symptoms and pathophysiology of AD. Clearly, these data need replication in larger patient samples characterized with regard to sleep and in-depth neuropsychological assessment. In healthy control subjects and in prodromal AD[36]. In addition, further research regarding signalling pathways induced by hypocretin-1 may elucidate the molecular mechanisms the relationship between hypocretin-1 and Tau/pTau. Considering preclinical data [37], mediating hypocretin receptors could be potential drug target for the prevention or treatment of tau pathology.

Sandwich ELISA technique for estimation of Tau protein phosphorylation at serine 199:

Tau phosporylation at multiple brain sites was initially studied in a well designed by Pierre and Nunez [38]. In recent years both ELISA and non – ELISA based assay for Tau have been described. A monoclonal capture antibody specific for TAU has been coated onto the wells of the 96-well plate provided. The thermo fisher scientific ELISA kit is ideal for this estimation. During the first incubation, standards of known TAU [pS199] content and unknown samples are pipetted into the wells and the TAU antigen binds to the immobilized antibody. After washing, a rabbit antibody specific for TAU phosphorylated at serine 199 is added to the wells. During the second incubation, this antibody serves as a detection antibody by binding to the immobilized TAU protein captured during the first incubation. After washing, a horseradish peroxidase labeled anti-rabbit IgG is added. This binds to the detection antibody to complete the four member sandwich. After a third incubation and washing to remove all the unbound enzyme, a substrate solution tetramethylbenzidine (TMB) is added, which is acted upon by the bound enzyme to produce color. The intensity of this colored product is directly proportional to the concentration of TAU [pS199] present in the original specimen and the optical density can be read on a standard microplate reader. The total assay incubation time is only 4 hours (Refer Fig.1).

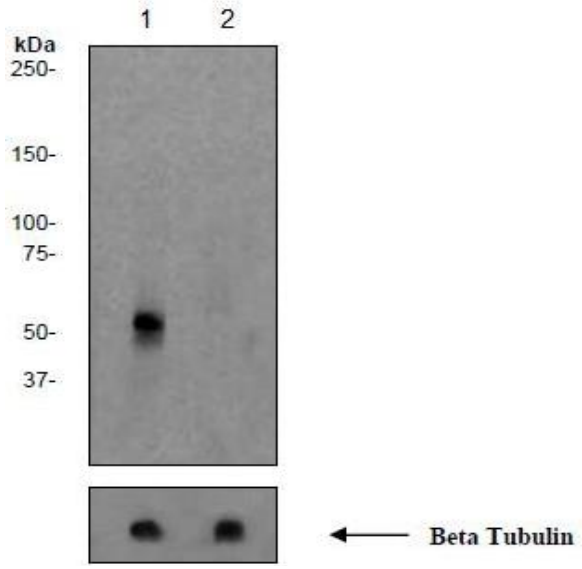

Figure 1: Western blotting with anti-tau antibody (S-199). Picture courtesy Abcam biotechnology company, USA.

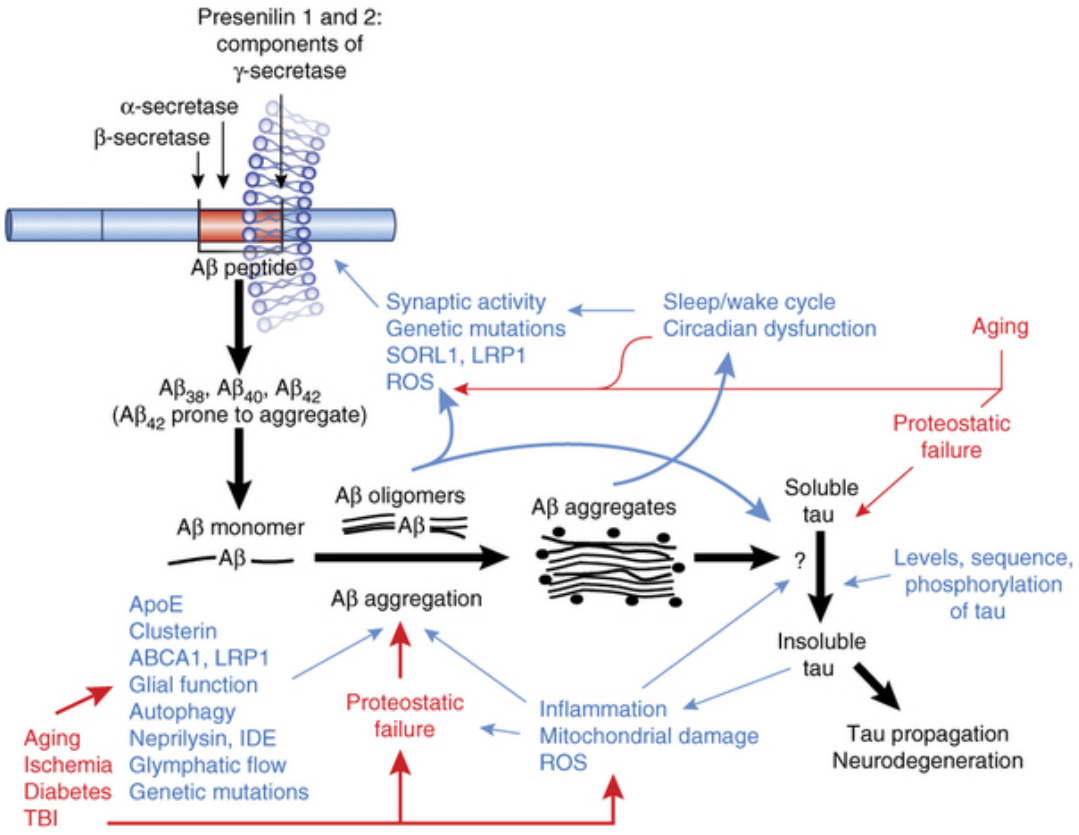

Figure 2: Tau propogation and neurodegeneration. (Fig.reprinted Courtesy: Erik S Musiek & David M Holtzman.Three dimensions of the amyloid hypothesis: time, space and 'wingmen' Nature Neuroscience. 800–806 (2015).

Conclusions:

It is now largely accepted that clinical manifestations of dementia are due to the neuronal loss occurring in those areas of the brain associated with cognitive functions of the patients. Fibrillary inclusions are reported to be the main cause for cell death. It appears that both orexin receptor down regulation and degradation of the hypocretin appears to contribute to the pathology of dementia. Extrapolation of recent studies to the real onset of the disease in humans is still considered accurate for some researchers. In this regard, few of reports analyzing the brain of AD patients come to an agreement that fibrillary aggregation of tau is the best co-relator with the onset and progression of dementia. It is mostly accepted that abnormal posttranslational modifications, that is, hyperphosphorylation, acetylation, glycation, nitration, truncation, and others, are responsible for altered tau structure and orexin peptides in AD and cortical forms of dementia. Moreover, accurate determination of both altered tau protein and hypocretin in the cerebrospinal fluid and other body fluids may provide better expectation to predict the onset and evolution of dementia.

Acknowledgements:

The author thanks Dr.Jacob Peedicayil, MD, CMC Vellore for useful scientific discussions

References:

1. Jöhren O, Neidert SJ, Kummer M, Dendorfer A, Dominiak P. Prepro-orexin and orexin receptor mRNAs are differentially expressed in peripheral tissues of male and female rats.Endocrinology. 2001 Aug;142(8):3324-31.
2. Spinazzi R, Andreis PG, Rossi GP, Nussdorfer GG.Prepro-orexin and orexin receptor mRNAs are differentially expressed in peripheral tissues of male and female rats. Pharmacol Rev. 2006 Mar;58(1):46-57
3. Sakurai T, Amemiya A, Ishii M, Matsuzaki I, Chemelli RM, Tanaka H, Williams SC, Richardson JA, Kozlowski GP, Wilson S, Arch JR, Buckingham RE, Haynes AC, Carr SA, Annan RS, McNulty DE, Liu WS, Terrett JA, Elshourbagy

NA, Bergsma DJ, Yanagisawa M.Cell. 1998 Feb 20;92(4):573-85.

4. Scammell TE, Winrow CJ. Orexin receptors: pharmacology and therapeutic opportunities.Annu Rev Pharmacol Toxicol. 2011;51:243-66
5. Trivedi P., Yu H., MacNeil D.J., Van der Ploeg L.H. and Guan X.M. (1998) Distribution of orexin receptor mRNA in the rat brain. FEBS Lett., 438,71-75.
6. Lu X.Y., Bagnol D., Burke S., Akil H. and Watson S.J. (2000) Differential distribution and regulation of OX1 and OX2 orexin/hypocretin receptor messenger RNA in the brain upon fasting. Horm. Behav., 37,335-344.
7. Lee M.G., Hassani O.K. and Jones B.E. (2005) Discharge of identified orexin/hypocretin neurons across the sleep-waking cycle. J. Neurosci., 25,6716-6720.
8. Sakurai T. (2007) The neural circuit of orexin (hypocretin): Maintaining sleep and wakefulness. Nat. Rev. Neurosci., 8,171-181.
9. Hasegawa E, Yanagisawa M, Sakurai T, Mieda M.Orexin neurons suppress narcolepsy via 2 distinct efferent pathways. J Clin Invest. 2014 Feb;124(2):604-16.
10. Yamanaka A., Tsujino N., Funahashi H., Honda K., Guan J.L., Wang Q.P., Tominaga M., Goto K., Shioda S. And Sakurai T. (2002) Orexins activate histaminergic neurons via the orexin 2 receptor. Biochem. Biophys. Res. Commun., 290, 1237-1245.
11. Karteris E., Machado R.J., Chen J., Zervou S., Hillhouse E.W. and Randeva H.S. (2005) Food deprivation differentially modulates orexin receptor expression and signaling in rat hypothalamus and adrenal cortex. Am. J. Physiol. Endocrinol. Metab., 288, E1089-100.
12. Blanco M, García-Caballero T, Fraga M, Gallego R, Cuevas J, Forteza J, Beiras A, Diéguez C.Cellular localization of orexin receptors in human adrenal gland, adrenocortical adenomas and pheochromocytomas. Regul Pept. 2002 Mar 15;104(1-3):161-5.
13. Fronczek R, Overeem S, Lee SY, Hegeman IM, van Pelt J, van Duinen SG, et al. Hypocretin (orexin) loss in Parkinson's disease. Brain. 2007;130:1577–1585.
14. Asai H, Hirano M, Furiya Y, Udaka F, Morikawa M, Kanbayashi T, et al. Cerebrospinal fluid-orexin levels and sleep attacks in four patients with Parkinson's disease. Clin Neurol Neurosurg. 2009;111:341–344.
15. Compta Y, Santamaria J, Ratti L, Tolosa E, Iranzo A, Munoz E, et al. Cerebrospinal hypocretin, daytime sleepiness and sleep architecture in Parkinson's disease dementia. Brain. 2009;132:3308–3317.
16. Gerashchenko D, Murillo-Rodriguez E, Lin L, Xu M, Hallett L, Nishino S, et al. Relationship between CSF hypocretin levels and hypocretin neuronal loss. Exp Neurol. 2003;184:1010–1016.
17. Ohno K, Sakurai T.Orexin neuronal circuitry: role in the regulation of sleep and wakefulness.Front Neuroendocrinol. 2008 Jan;29(1):70-87.
18. Morales A, Bonnet C, Bourgoin N, Touvier T, Nadam J, Laglaine A, et al. Unexpected expression of orexin-B in basal conditions and increased levels in the adult rat hippocampus during pilocarpine-induced epileptogenesis. Brain Res. 2006;1109:164–175.
19. Boeve BF. REM sleep behavior disorder: Updated review of the core features, the REM sleep behavior disorder-neurodegenerative disease association, evolving concepts, controversies, and future directions. Ann N Y Acad Sci. 2010;1184:15–54.
20. Chokroverty S. Sleep and neurodegenerative diseases. Semin Neurol. 2009;29:446–467.
21. Weingarten, M.D.; Lockwood, A.H.; Hwo, S.Y.; Kirschner, M.W. A protein factor essential for microtubule assembly. Proc. Natl. Acad. Sci. USA 1975, 72, 1858–1862.
22. Cleveland, D.W.; Hwo, S.Y.; Kirschner, M.W. Purification of tau, a microtubule-associated protein that induces assembly of microtubules from purified tubulin. J. Mol. Biol. **1977**, 116, 207–225.
23. Cleveland, D.W.; Hwo, S.-Y.; Kirschner, M.W. Physical and chemical properties of purified tau factor and the role of tau in microtubule assembly. J. Mol. Biol. **1977**, 116, 227–247.
24. Lund PE, Shariatmadari R, Uustare A, Detheux M, Parmentier M, Kukkonen JP *et al.* (2000). The orexin OX1 receptor activates a novel Ca^{2+} influx pathway necessary for coupling to phospholipase C. J Biol Chem 275: 30806–30812.
25. Johansson, L., Ekholm, M.E., and Kukkonen, J.P. (2008). Multiple phospholipase activation

by OX(1) orexin/hypocretin receptors. Cell Mol Life Sci.65,1948-1956.

26. Nasman, J., Bart, G., Larsson, K., Louhivuori, L., Peltonen, H., and Akerman, K.E. (2006). The orexin OX1 receptor regulates Ca2+ entry via diacylglycerol-activated channels in differentiated neuroblastoma cells. J Neurosci.26, 10658-10666.

27. Nilssen, L.S., Dajani, O., Christoffersen, T., and Sandnes, D.(2005). Sustained diacylglycerol accumulation resulting from prolonged G protein-coupled receptor agonist-induced phosphoinositide breakdown in hepatocytes. J Cell Biochem.94, 389-402.

28. Bateman R. J., Wen G., Morris J. C., Holtzman D. M. (2007). Fluctuations of CSF amyloid-beta levels: implications for a diagnostic and therapeutic biomarker. Neurology 68, 666–669

29. Wennstrom M., Londos E., Minthon L., Nielsen H. M. (2012). Altered CSF orexin and alpha-synuclein levels in dementia patients. J. Alzheimers Dis. 29, 125–132 10.3233/JAD-2012-111655

30. J.E. Kang, M.M.Lim, R.J.Bateman,J.J.Lee,L.P. Smyth,J.R. Cirrito, N. Fujiki,S. Nishino, D.M.Holtzman, Amyloid-beta dynamics Are regulated By orexin And the sleep–wake cycle, Science 326 (2009) 1005–1007

31. F. García-Sierra, N. Ghoshal, B. Quinn, R. W. Berry, and L. I. Bínder, "Conformational changes and truncation of tau protein during tangle evolution in Alzheimer's disease," Journal of Alzheimer's Disease, vol. 5, no. 2, pp. 65–77, 2003.

32. Michael Deuschle, Claudia Schilling, Markus Leweke, Frank Enning, Thomas Pollmächer, Hermann Esselmann, Jens Wiltfang, Lutz Frölich, Isabella Heuser. Hypocretin in cerebrospinal fluid is positively correlated with Tau and pTau. Neuroscience Letters 561 (2014) 41– 45

33. N. Ghoshal, F. García-Sierra, Y. Fu et al., "Tau-66: evidence for a novel tau conformation in alzheimer's disease," Journal of Neurochemistry, vol. 77, no. 5, pp. 1372–1385, 2001.

34. R. W. Carrell and B. Gooptu, "Conformational changes and disease–serpins, prions and Alzheimer's," Current Opinion in Structural Biology, vol. 8, no. 6, pp. 799–809, 1998.

35. N. Fox, R. J. Harvey, and M. N. Rossor, "Protein folding, nucleation phenomena and delayed neurodegeneration in Alzheimer's disease," Reviews in the Neurosciences, vol. 7, no. 1, pp. 21–28, 1996.

36. Sasaki K, Suzuki M, Mieda M, Tsujino N, Roth B, Sakurai T.Pharmacogenetic modulation of orexin neurons alters sleep/wakefulness states in mice.PLoS One. 2011;6(5):e20360

37. Thomas C Thannickal. Hypocretin (orexin) pathology in Alzheimer's disease .World J Neurol. Sep 28, 2015; 5(3): 64-67

38. Pierre, M.; Nunez, J. Multisite phosphorylation of tau proteins from rat brain. Biochem. Biophys. Res. Commun.1983, 115, 212–219.