THE ABUNDANCE AND DISTRIBUTION OF MELANOPSIN (OPN4) PROTEIN IN THE HUMAN BRAIN

NISSILA J1, MÄNTTÄRI S1, TUOMINEN H2, SÄRKIOJA T3, TAKALA T3, TIMONEN M4 & SAARELA S1

1University of Oulu, Department of Biology, Box 3000, 90014, Oulu, Finland
2University of Oulu, Institute of Diagnostics, Box 5000, 90014, Oulu, Finland
3 Oulu Deaconess Institute, P.O. Box 365, 90101, Oulu, Finland
4University of Oulu, Institute of Health Sciences, Box 5000, 90014, Oulu, Finland

INTRODUCTION
OPN4 (melanopsin) is a photoreceptor sensing light outside of the visual system in the spectral area of blue color (maximum sensitivity between 470-480 nm). According to earlier research [1], OPN4 has been found in the retina but not in samples of the whole brain, several neuroanatomical sites, or periphery. The existing main hypothesis is that phototransduction via OPN4 molecules in intrinsically photosensitive retinal ganglion cells (IPRGCs) and the retinohypothalamic track (RHT) mediate light induced neural signals further into the suprachiasmatic nucleus (SCN) and other thalamical areas to maintain circadian rhythmicity and generate change in cognitive parameters and psychogenic effects [2, 3].

OBJECTIVES
To investigate whether OPN4 protein is expressed also outside of the RHT in functionally central areas of the human brain.

METHODS
Samples from a total of ten cadavers were taken during forensic examination, and the samples were prepared for assessment. Brain tissue was sampled according to the protocol used in the neuropathological assessment of neurodegenerative diseases. In addition, samples from pineal and pituitary glands, as well as from testicular/ovarian tissue and the spinal cord were included in the study. OPN4 protein content was measured using SDS-PAGE and Western blotting. The localization of OPN4 protein in the human brain and peripheral tissues was assessed by immunohistochemical staining using polyclonal antibody against OPN4. Samples were cut into sections and stained with a fluorescent dye labelled antibody before confocal laser scanning microscopy. The specificity of labelling and immunoreaction was validated by primary antibody omitting and immunizing peptide blocking.

RESULTS
We found OPN4 protein abundant in the neurons of all seventeen examined sites of the human brain, testes and spinal cord (Fig. 1). Neuronal OPN4 was present in granular pattern in numerous cerebral cortical areas, cerebellar cortex, and several nuclei in phylogenetically old regions (Fig. 2). Immunoreaction took place mostly in neuronal soma, but not in nuclei.

CONCLUSIONS
Hitherto OPN4 has been known to be present only in the non-image forming visual system (NIF) [1, 2 & 3] mediating circadian pace-making properties of light [2 & 3]. Worth of note is the abundance of OPN4 receptor protein in human deep brain nuclei responsible for monoamine production and functionality (mesencephalon / substantia nigra & pons / locus coeruleus), and central areas for overall homeostasis and hormonal control (hypothalamsus, thalamus and pituitary). However, our findings of OPN4 protein in the neurons of functionally central areas in the human brain raises the need for closer study of the effects of direct extraretinal photic stimuli upon the brain.

REFERENCES