7th Malta Medical School Conference Nov 2009.

An imaging study of the neuronal subsets in the GFP-M line of transgenic mice

Mario Valentino[1], Richard Muscat[2], Claude Bajada[3], Bridget Ellul[1].
Dept.of Pathology, University of Malta[1], Dept.of Physiology & Biochemistry, University of Malta[2], Medical School, Mater Dei Hospital[3].

The aim of this study was to characterise the expression of green fluorescent protein in different neuronal subsets of the Thy1-GFP-M line in transgenic mice in order to establish which populations of neurons, in an identifiable brain structure, can be studied using this line.

Coronal cerebral and cerebellar slices (400µm), sagittal cerebellar slices including brainstem and spinal cord (400µm) and horizontal retinal slices (250µm) were prepared from the Thy1-GFP-M mice. Care was taken to retrieve as many slices as possible so as to preserve the histology of the prepared sections. The slices were fixed for 1hr and cryoprotected overnight. 60µm slices were then prepared by a vibroslicer in cold PBS, pH7.2 and visualized at 488nm using a confocal microscope (Bio-Rad MRC1024). Images were acquired under both low and high magnification using 10x and 40x air Nikon lenses. Maximum intensity projections were acquired through a Z-stack set at an interval of 1µm using a 60x oil immersion Nikon lens.

The M-line shows good expression of the GFP in the mossy fibres of the cerebellum and in spinal cord axons. There was very sparse, yet intense expression that allowed visualisation of dendritic arborisation including spines in pyramidal cells of layer 5 of the cerebral cortex. There was also some neuronal expression in the hippocampus and in retinal ganglion cells. There was minimal GFP expression in the corpus callosum.

Since there is no record in the literature as to whether there is expression of the GFP in the corpus callosum, our observation clearly shows that this line does not express the protein in the corpus callosum and therefore it is not ideal as a model for the study of white matter injury in the brain.

Due to the heterogeneous distribution of GFP together with high intensity expression in individual neurons, Thy1-GFP-M mice are suitable for anatomical and functional studies of dendritic spines in experimental models of neuronal plasticity, cellular pharmacology and learning and behaviour.

This line also has the potential to be used in animal models of spinal cord injury and in regeneration experiments involving axonal sprouting and path finding.