5

Authors: Takashi Shibata(1), Andrew J Copp(1), Dorothea Stiefel(1), Patrick G Duffy(2)
Institution: (1) Neural Development Unit, Institute of Chidl Health, University College Londo, (2) Urology, Great Ormond Street Hospital for Children, NHS Trust
Title: MORPHOLOGICAL ANALYSIS OF SPINAL CORD PATHOLOGY IN MICE WITH SPINA BIFIDA, WITH PARTICULAR REFERENCE TO THE SPINAL MICTURITION CENTER

Introduction and Objectives:

Myelomeningocele caused by the failure of neural tube closure is the most common cause of neurogenic bowel and voiding dysfunction in children. However, the specific morphological features of the spinal cord responsible for innervating the pelvic organs have not been characterized previously because of paucity of human material. To elucidate the pathology of spinal cord in myelomeningocele, we studied a mutant mouse strain with neural tube defects (NTD) as a model for human myelomeningocele. The process of neural tube closure in the mouse is closely similar to the sequence of events in the human embryo, making the mouse a useful model system.

Methods:

Loop tail / curly tail mutant mice at postcoital days 14 to 18 (E14-18, E18: the day before birth) were processed for morphological analysis including microdissection of the spinal cord, H&E staining, immunohistochemistry for neurofilament and caspase3, and neuronal labeling using tracer dye DiI to label afferent fibers, preganglionic neurons (PGN) and somatomotor neurons (SMN) in the L6-S1 lumbosacral spinal cord.

Results:

Microdissection showed a spinal tethering phenomenon in mice with NTD similar to that reported in humans with myelomeningocele. Tethering of the NTD lesion to surrounding tissues leads to axial elongation of the spinal cord region immediately rostral to the lesion. Neurofilament staining showed that the tethering starts at E15. By E17, H&E staining showed that the open spinal cord is beginning to degenerate, with the result that little spinal cord tissue remains at E18. Caspase 3 positive neurons were dramatically increased from E17, suggesting programmed cell death is the main cause for the degeneration. DiI labeling shows that the L6-S1 nerve roots terminate at the open spinal cord or at the narrowed abnormal part above the NTD lesion in most mice with NTD except for those with very small NTD. Fluorescence microscopy shows that afferent fibers and both PGN and SMN motor neurons are clearly labeled by E16, while afferent fibers are not labeled at E17 and no afferent fibers nor motor neurons can be labeled at E18.

Conclusions:

The central nervous system region innervating the pelvic organs, within the lumbosacral spinal cord, presents a pathological appearance from E15 in NTD mutant mice, and degenerates rapidly in the late gestational period. Hence, these mutant mice are useful for the analysis and understanding of the mechanisms of neural degeneration in the open spinal cord of myelomeningocele and may provide insight into the likely effectiveness of therapeutic challenges such as intrauterine closure of NTD in humans.