472

Shibata T. [1], Duffy P. G. [2], Copp A. J. [3]
1. Dept. of urology and renal surgery, Hokkaido University, 2. Dept. of urology, Great Ormond Street Hospital, Lo, 3. Neural development unit, ICH, UCL, London, UK

DEVELOPMENT OF THE BLADDER WALL IN MUTANT MICE WITH SPINA BIFIDA

Aims of Study: It is reported that the smooth muscle of the bladder in humans with spina bifida does not differentiate or develop normally because of abnormal bladder innervation. Here, we used a mouse genetic model of spina bifida to investigate this topic experimentally. Specifically, we asked whether the poorly compliant bladder in individuals with spina bifida is the inevitable result of intrinsic abnormal innervation, or whether it results from potentially avoidable secondary changes caused by functional outlet obstruction and high bladder pressure during storage and voiding of urine.

Methods: Mice developing spina bifida as a result of an interaction between the *loop-tail* and *curly tail* mutations (*Lp/ct*), or in homozygotes for the *splotch* (Sp^{2H}) mutation, were examined in comparison with normal littermates. The bladder was removed before fixation from embryos at embryonic days (E) 18 (the day before birth), and then fixed and processed for immunohistochemistry for nerves (PGP9.5, VAChT, and L1) and smooth muscle (alpha SM actin). Elastic van Gieson (EVG) staining was used to visualize the extracellular matrix (ECM). This setting allows us to observe the distended bladder wall in natural and tension-free condition, comparable with the non-distended bladder wall. In addition, embryos at E14 and E16 were collected and processed for immunohistochemistry for alpha SM actin and EVG staining to see the developmental changes of the bladder wall during the late gestational period.

Results: At E14, no urine was seen in the bladder of any embryo, whereas, at E16, the bladder showed physiological extension in all cases. At E18, no differences were detected in nerve, smooth muscle or ECM staining between spina bifida and normal control fetuses from *Lp/ct* matings. In *splotch* fetuses with spina bifida, however, the bladder was abnormally distended and nerve staining was very scanty. Bladder smooth muscle and ECM staining did not differ between spina bifida and control fetuses in *splotch* litters.

Conclusions: Early development of the bladder wall in mice with spina bifida appears normal even if the innervation of the bladder is abnormal, as in *splotch* fetuses. These findings suggest that the poorly compliant bladder in human spina bifida is a secondary change, perhaps resulting from functional obstruction of the lower urinary tract, which could be avoided if treated from an early stage.

References:

1. J Urol 145: 1024-1029, 1991
2. Neurourol Urodyn 20: 377, 2001