

## EFFECTS OF AGE ON NEUROGENIC AND NON-NEUROGENIC ATP RELEASES FROM ISOLATED HUMAN BLADDER

### Aims of Study

It has been reported that bladder smooth muscle function changes with age. The main neurotransmitter in bladder contraction is acetylcholine released from cholinergic nerve endings. However, non-adrenergic and non-cholinergic (NANC) neurotransmitters may contribute to contractile responses in human bladder with pathological conditions or aged bladder (1). It has been reported that adenosine triphosphate (ATP) is the most important NANC neurotransmitter in bladder contraction. Furthermore, several reports suggest that urothelium releases ATP, which may be one of the stimulating factors for afferent nerves resulting to increase in micturition reflex (2). The present study was designed to evaluate the effects of age on ATP release in the isolated human bladder smooth muscles strips with or without urothelium.

### Methods

Human bladders were obtained from 18 patients (male; average 65.8 years old), who were undergoing cystectomy due to bladder carcinoma. Smooth muscle strips with or without urothelium were suspended in organ bath filled with Krebs-Henseleit solution, and tension developments were recorded. Microdialysis probe was inserted into the strip, Ringer solution was perfused into the probe at a constant flow rate of 2.0  $\mu$ l/min, and dialysate was collected every 10 min in polyethylene tube. The effects of age on non-neurogenic ATP releases were evaluated. Furthermore, the dialysate during electrical field stimulation (EFS: supramaximum voltage, 0.5 msec duration, 60 Hz and 3 sec train at 1 min interval for 10 min) was collected, and the effect of age on EFS-induced ATP releases (neurogenic ATP) is also evaluated. The amount of ATP released in the dialysate was measured by luciferine-lusiferase assay.

### Results

In human bladder strips before EFS, there were non-neurogenic ATP releases, which were not inhibited by treatment with tetrodotoxin (1.0  $\mu$ M). The non-neurogenic ACh releases from muscle strips with urothelium were significantly higher than that from strips without urothelium. The non-neurogenic ATP releases from smooth muscle strips with urothelium increased with age. The non-neurogenic ATP in older (over 65 years) patients was significantly higher than that in younger patients (under 65 years) (table). EFS caused significant increases in ATP releases in both human bladder smooth muscles strips with and without urothelium, which were significantly inhibited by treatment with tetrodotoxin (1.0  $\mu$ M). ATP releases induced by EFS (neurogenic ATP) increased with age, and there was a significant positive correlation between age and neurogenic ATP release. The neurogenic ATP in older patients was also significantly higher than that in younger patients (table).

Table - Effects of age on neurogenic and non-neurogenic ATP release from human bladder strips with urothelium

Age	Neurogenic ATP release pmol/g tissue	Non-neurogenic ATP release pmol/g tissue
Over 65 years (n=10)	0.83 $\pm$ 0.18*	0.12 $\pm$ 0.03*
Under 65 years (n=8)	0.48 $\pm$ 0.11	0.04 $\pm$ 0.01

P < 0.01; significantly different from comparable values under 65 years.

### **Conclusions**

The data demonstrate that there are age-related increases in neurogenic and non-neurogenic ATP releases in human bladder smooth muscles. It is suggested that bladder urothelium may play an important role on non-neurogenic ATP release, and that the age-related changes in neurogenic and non-neurogenic ATP releases may contribute to age-related changes in bladder function.

### **References**

1. Experimental Gerontology 36: 99, 2001
2. Annual Review of Pharmacology and Toxicology 41: 691, 2001