

## MEASUREMENT OF NORADRENALINE RELEASE FROM RAT AND HUMAN BLADDER USING MICRODIALYSIS PROCEDURE

### Hypothesis / aims of study

It is generally accepted that the micturition cycles are divided into the storage and voiding phases. It has been suggested that in the filling phase, the inhibition of bladder contraction was mediated by noradrenaline (NA) through the beta-adrenoceptors. Several reports presented the functional evidence of an important role for  $\beta_3$ -adrenoceptor in urine storage in anesthetized rats and selective  $\beta_3$ -adrenoceptor agonist reduced bladder tone and increased bladder capacity significantly. However, there were few reports about the NA release from urinary bladder in the storage phase. In the present study, using microdialysis procedure, we attempted to measure the NA release from rat urinary bladder by bladder distension. In addition, we have evaluated the stretch-induced NA release from isolated human bladder strips.

### Study design, materials and methods

Female wistar rats were anesthetized with 0.9 mg/kg urethane. The lower abdominal cavity was opened, and a 20G cannula was inserted into the bladder transurethraly and physiological saline was infused into the urinary bladder at a 0.1 ml/min constantly. The microdialysis probe was inserted into the bladder wall, and Ringer solution was constantly perfused. The dialyzate was collected in a microtube, when the physiological saline was infused 0.2 ml, 0.5 and 1.0 ml into the bladder. Furthermore, human bladders were obtained from 8 patients who were performed bladder resection due to malignant tumour. After insertion of microdialysis probe, each bladder strips with urothelium was suspended in organ bath filled with Krebs-Henseleit solution, connected to a force displacement transducer and an isometric force was recorded. Ringer solution was continuously perfused into the microdialysis probe at a rate of 2  $\mu$ l/min. The dialysate was collected in a micro tube every 10 minutes. The effects of elevation of the resting tension (0 to 40 mN) on the NA release were evaluated in the human bladder strips. The collected dialyzates in both experiments were injected into HPLC-ECD system for measurement for NA.

### Results

NA release from rat urinary bladder was determined at 0 ml in bladder volume (basal release). According to the increase in bladder volume, the amounts of NA release from rat urinary bladder were gradually increased. The amount of NA release at 0.5 and 1.0 ml in bladder volume were  $0.062 \pm 0.010$  and  $0.080 \pm 0.012$  pmol/injection, respectively, which were significantly higher than that of 0 ml in bladder volume ( $0.039 \pm 0.011$  pmol/injection). In human bladder strips, Significant NA release was observed at 0 mN resting tension ( $0.224 \pm 0.058$  pmol/g tissue weight; basal release). The basal release was not inhibited by treatment with 1  $\mu$ M tetrodotoxin. There was stretch-induced increase in NA release from human bladder strip. The maximum % increase in NA release was  $334 \pm 48$  % in 40 mN resting tension (table)

Table: Effects of stretch on NA release from human bladder strips

Tension; mN	0	10	20	30	40
NA; pmol/g (SEM)	0.224 (0.058)	0.407 (0.187)	0.634 (0.201)	0.872 (0.24)	0.970 (0.28)
% increase; % (SEM)		80.0 (14)	183 (28)	290 (35)	334 (48)

### Interpretation of results

The present results demonstrate that there are basal NA releases in rat and human bladder. The failure of inhibitory effect of tetrodotoxin on basal NA release in human bladder implies that basal NA is non nerve-evoked release. Distension- or stretch-induced increases in NA

release in rat and human bladder may suggest that the increased NA release may contribute to the regulation of bladder tone in the storage phase.

Concluding message

In rat and human bladder, there are non-nerve evoked NA releases. It may be suggested that the increased non-nerve-evoked NA release during bladder distension may have a significant role on bladder function in the storage phase.