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ROLE OF SPINAL METABOTROPIC GLUTAMATE RECEPTORS IN THE REGULATION OF LOWER URINARY TRACT FUNCTION IN DECEREBRATE, UNANESTHETIZED RATS

Hypothesis / aims of study

Glutamate receptors consist of two major classes, the ionotropic receptors which form ligand-gated channels and metabotropic receptors (mGluRs) which are G-protein coupled receptors. The former which include *N*-methyl-D-aspartate (NMDA) and α-amino-3-hydroxy-5-methylisoxazole-4-propionic acid (AMPA), receptors have an essential role in the control of micturition [1]; however less is known about the function of mGluRs. Eight subtypes of mGluRs (1 to 8) have been divided into three groups (I to III). In the rat lumbosacral spinal cord mGluR5 are expressed in the preganglionic parasympathetic neurons and mGluR1 are in Onuf's nucleus [2]. The present study determined if mGluRs have a role in the micturition reflex in decerebrate, unanesthetized rats by testing an mGluR antagonist and agonist. Effects of mGluR antagonist were also evaluated in rats with chronically transected as well as intact spinal cord, to determine if spinal cord injury altered the responses.

Study design, materials and methods

Animal preparations: Experiments were performed on 19 female decerebrate unanesthetized Sprague-Dawley rats (240-280 g). All surgical procedures were performed under halothane anesthesia. Spinalization was performed in 7 rats by sectioning at the T8-9 level. The experiments on spinalized rats were performed 3 to 4 weeks post-spinalization. In spinalized rats, an i.t. catheterization (PE-10) was performed after a T11-12 laminectomy on the day of cystometric recording. In rats with intact spinal cord, an i.t. catheter was inserted through a slit in the atlanto-occipital membrane and passed caudally to the L6-spinal level. A precollicular decerebration was performed under halothane anesthesia using a blunt spatula. To examine the external urethral sphincter (EUS) EMG activity, epoxy-coated stainless steel wire (50 μm) EMG electrodes were placed percutaneously in the striated muscle of the EUS/pelvic floor. A transurethral bladder catheter connected to a pressure transducer was used to record bladder pressure during continuous infusion cystometry with physiological saline (0.21 ml/min). Experiments were performed 2-3 h after decerebration.

Drugs: The group I (mGluR1/5) antagonist, (+/-)-alpha-methyl-4-carboxyphenylglycine (MCPG) and the group I/II agonist (mGluR1/5 and 2/3) trans-(+/-)-1-amino-1,3-cyclopentanedicarboxylic acid (ACPD) was dissolved in phosphate buffered saline and DMSO, respectively.

Evaluations and statistics: The effects of drugs on bladder contraction amplitude (BCA) and frequency and on EUS EMG activity were evaluated. Graded doses of drugs were given in each animal at 30-50 min intervals to examine dose-response relationships. All values are expressed as mean +/- S.E.M. Paired *t* test was used and $P<0.05$ was considered significant.

Results

As shown in Fig.1, i.t. injection of MCPG (3-100 μg) dose-dependently increased EUS EMG activity, whereas it did not change BCA in spinal intact rats ($n=6$). In chronically spinalized rats ($n=7$), MCPG (3-100 μg) changed neither BCA nor EUS EMG activity.

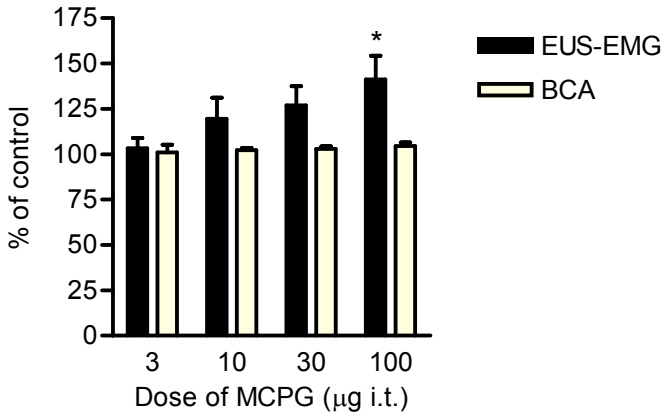

Fig.1: Effect of MCPG on BCA and EUS EMG activity in spinal intact rats. *P<0.05: statistical analysis of drug effects was made based on the raw data values. Each bar indicates % value 'after' the drug injection when 'before' as 100 %.

In spinal intact rats low doses of ACPD (1-3 μg) had no effect; whereas 10 μg transiently increased BCA. Higher doses of ACPD (30 and 100 μg) produced intense hindlimb movements and prominent activity in the EUS EMG (3 of 6 and all 5 rats, respectively) accompanied by an initial rise in baseline intravesical pressure followed by loss of large amplitude bladder contractions (also see reference [3]). After 20-30 min small amplitude (40% of control) high frequency bladder contractions (1/min) reappeared. The latter were associated with bursts of EUS EMG activity.

Interpretation of results

Blockade of mGluRs1/5 in the spinal cord by MCPG facilitated EUS EMG activity in spinal intact rats, but had no effect on BCA. MCPG was inactive in spinalized rats. Activation of group I/II mGluRs with large doses of ACPD suppressed the micturition reflex, induced intense activity of the EUS and limb muscles and inhibited efficient voiding.

Concluding message

In decerebrate, unanesthetized rats with an intact spinal cord glutamatergic mGluRs1/5 are involved in tonic inhibition of the excitatory pathway (eg, glutamatergic transmission *via* AMPA and/or NMDA receptors) to the EUS but not involved in the control of the bladder. However activation of silent mGluRs in bladder reflex pathways suppressed bladder activity. The lack of effect of MCPG on EUS activity in chronically spinalized rats indicates that an mGluR-mediated inhibitory control of the EUS was eliminated after spinal cord injury, raising the possibility that removal of the inhibitory mechanism may contribute to the uncoordinated activity of bladder and EUS (ie, detrusor-sphincter dyssynergia, DSD) that occurs after spinal injury. Thus, the mGluRs in the lumbosacral spinal cord may be useful targets for the pharmacologic treatment of lower urinary tract dysfunctions such as stress incontinence and DSD.

References

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