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CENTRAL INNERVATION OF THE PROSTATE GLAND AS REVEALED BY THE TRANSNEURONAL TRANSPORT OF PSEUDORABIES VIRUS

Aim of the Study: The prostate gland is involved in different functional pathologies of the lower urinary tract (LUT). Because the central innervation plays a key role in LUT function the neuroanatomy of the prostate was explored. Recent progress in neuroscience methodology allows now a transneuronal tracing by using a self amplifying virus tracer – Pseudorabiesvirus (PRV).

Methods: 44 individual adult male Sprague-Dawley rats were used for retrograde transneuronal mapping of the spinal cord and brain stem. A PRV-tracer (5µl, $1x10^8$pfu/ ml) was injected into the prostate. After a survival time of 72, 96 or 120 hours the animals were sacrificed. The brain and spinal cord were harvested via a dorsal laminectomy. After cutting on a freezing microtome the tissue was immunostained for PRV.

Results: PRV-positive cells were found within the sacral (S1-S2) and the thoracolumbar (T12-L2) spinal cord. At the supraspinal level positive cells were found within the following regions: nucleus raphe, lateral reticular formation, nucleus gigantocellularis, A5 noradrenergic cell region, locus coeruleus, pontine micturition center, hypothalamus, medial preoptic region and periaquaductal gray.

Conclusion: There is a broad central representation of neurons involved in the control of the prostate gland. It's obvious, comparing data from the literature, that there is a broad overlap in the innervation of pelvic visceral organs (bladder, rectum, urethra). The appreciation of this neuroanatomical circumstances allow a deeper understanding of common urological pathologies within the pelvis (pelvic pain, prostatism and BPH, neurogenic bladder and bowel dysfunction).

(supported by: Deutsche Forschungsgemeinschaft DFG Ze 415/1-1)

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ULTRASTRUCTURAL EVIDENCE FOR A DIRECT PATHWAY FROM THE LUMBOSACRAL SPINAL CORD TO THE PONTINE MICTURITION CENTER OF RATS

AIMES OF STUDY

The behavioral pattern of micturition and urinary continence differs strongly among species. For example, cats and humans urinate primarily in a safe environment (**guarded urination**), while voiding in rats is more reflexively (**reflex urination**).
The synergic action between the detrusor muscle of the bladder and the external urethral sphincter during micturition is coordinated by a group of neurons in the dorsolateral pontine tegmentum, called M-region or pontine micturition center (PMC). This study in adult male rats investigates the existence of direct lumbosacral cord projections to the pontine micturition center (PMC), which is absent in cats. Electron microscopical analysis is necessary to determine whether sacral afferents are in direct contact with spinally projecting PMC neurons.

METHODS

In three adult male rats injections with the retrograde and anterograde neuronal tracer wheat germ agglutinin horseradish peroxidase

(WGA-HRP) were injected in the caudal lumbar and rostral sacral cord, bilaterally, in order to retrogradely label spinally projecting neurons in the PMC and to anterogradely label terminals on these neurons. After a survival time of 18 hours the animals were perfused with a fixative containing 1% paraformaldehyde and 2 % glutaraldehyde in a 0.1 M phosphate buffer. The tissue was processed with standard light and electron microscopical techniques.

RESULTS

Light microscopy showed that both anterogradely and retrogradely labeled profiles were present in the PMC after WGA-HRP injections in the caudal lumbar and rostral sacral cord. Ultrastructural analysis demonstrated that many anterogradely labeled terminals were present in the area of the PMC, which were filled with round, pleiomorphic and flat vesicles. In about 15% the terminals contacted retrogradely labeled dendrites. The majority contained asymmetric synaptic clefts, but also symmetric clefts (25%) were found.

CONCLUSIONS

The results provide evidence for a direct lumbosacral-PMC pathway in the rat. This implicates that sensory neurons involved in processing bladder filling information have direct access to the PMC, which is not the case in cats. The results provide an explanation for the differences in micturition behavior between rats, and cats and humans. Thus, species dependent differences in neuronal pathways involved in micturition reflect differences in their micturition pattern.

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A RAT MODEL FOR INVESTIGATION OF BLADDER DYSFUNCTION ASSOCIATED WITH MULTIPLE SCLEROSIS-LIKE DEMYELINATING DISEASE

AIMS OF STUDY

Myelin basic protein (MBP) can be used as antigen for inducing experimental allergic encephalomyelitis (EAE). Various studies have been reported using EAE animals as an experimental model of demyelinating diseases such as multiple sclerosis. However, no study on the bladder dysfunction in EAE animals has been reported. The aim of this study was to determine whether an EAE rat induced by MBP is useful for the investigation of bladder dysfunction associated with demyelinating disease.

METHODS

Female Lewis rats (weighing 175-245 g) were used.

Study 1) Time course of behavioral and cystometric changes

MBP injection : Fifty μg (10 μl) of MBP was injected subcutaneously in the right hindpad of 13 rats and saline as the vehicle in 5 control rats.

Behavioral investigations : Behavioral changes were investigated at least once a day during for 21 consecutive days after injection of MBP or saline.

Cystometric investigations : A polyethylene catheter was implanted into the bladder through the dome 3 days after injection. Cystometric investigations were performed without any anesthesia for 12 consecutive days from 7 days after injection. Intravesical pressure and micturition volumes were recorded continuously on a polygraph. Detrusor hyperactivity (DH) was defined by the following criteria: micturition volume less than 0.3 ml and residual volume less than 0.1 ml.

Study 2) Correlation between behavioral, cystometric and histological abnormalities

MBP injection : Separately from the Study 1, 50 μg of MBP was given to 20 other rats in the same manner as described above.

On the basis of the results of Study 1 described below, behavioral, cystometric and histological abnormalities were investigated on the day 12.

Behavioral investigations : The degree of paralysis was classified : grade 0 ; no paralysis, grade 1 ; paralysis only of the tail, grade 2 ; incomplete paralysis of the hind limbs, grade 3 ; complete paralysis of the hind limbs.

Cystometric investigations : Bladder catheter implantation was made on the day 9. Continuous cystometry was performed without any anesthesia.

Histological investigation : Immediately after behavioral and cystometric investigations, the rats were perfused transcardially with 80 ml of 4% paraformaldehyde in 0.1 M phosphate buffer (PB, pH 7.4) under general anesthesia, and the brain and spinal cord were removed, then kept overnight in the same fixation solution at room temperature. Investigations were performed mainly of the spinal cord at the level of L6-S1. The brain, and the spinal cord levels C5, Th4 and L2 were also investigated in some animals. Histological findings were evaluated with a double blind method by two investigators, and inflammation observed was scored as the following three degrees : Mild (1) ; no parenchymal infiltration / meningeal mononuclear cells infiltration, Moderate (2) ; mononuclear cells almost localized to focal area of neuronal parenchyma and Severe (3) ; panmyelitis involving both white matter and gray matter. The inflammation score was defined as the sum of the points (from 1 to 3) of the two investigators.

Wilcoxon signed rank test was used for statistical analysis.

RESULTS

Study 1) Time course of behavioral and cystometric changes

Behavioral investigations : Hind paralysis was observed in all the rats but one given MBP. The onset of the hind paralysis was at the day 10-14 (mean 12.2) after MBP injection, and the duration of the paralysis was for 1-7 (mean 3.5) days. Then all rats recovered and looked normal.