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INTRATHECAL ONABOTULINUMTOXINA IMPROVES BLADDER FUNCTION AFTER SPINAL CORD INJURY AND EFFICIENTLY SUPRESSES NEUROGENIC DETRUSOR OVERACTIVITY

Hypothesis / aims of study

Spinal cord injury (SCI) is a devastating event that often leads to the loss of motor, sensitive and/or autonomic function. In most patients, SCI results in chronic loss of voluntary control of bladder function. Injury is followed by the spinal shock, during which the bladder is unable to contract. Areflexia is substituted by neurogenic detrusor overactivity (NDO), that results from sprouting of bladder afferents at the lumbosacral spinal cord and consequent reorganization of neuronal pathways. NDO results in urinary incontinence, the principal cause of social withdrawal and diminished quality of life of SCI patients. Surveys clearly indicate that, after improving motor function, regaining bladder control is the highest priority for those patients (1).

Detrusor injection of onabotulinumtoxinA (OnabotA) has been demonstrated to improve bladder function in SCI patients, being an effective therapeutic option to manage chronic NDO (2). However, this route of administration requires repeated interventions, which leads to a high rate of urinary infections. Recently it was shown that intrathecal administration of OnabotA improves bladder function and reduces pain caused by chronic cystitis by a direct effect on sensory afferents (3). Here, we investigated if the same route of administration is effective in NDO suppression.

Study design, materials and methods

Rats (n=10) were subjected to a complete transection of the spinal cord at the T4 level. A silicone catheter was implanted intrathecally (IT) at L5/L6 spinal segments. Sham-operated rats (n=4) were used as control. Four weeks later, rats received IT OnabotA (2U in 50 µL of saline) (n=5) or saline (n=5). Bladder reflex activity was recorded by cystometry 48h after treatment. At the end of the experiments, L5 and L6 segments and dorsal root ganglia (DRG) were collected and processed for immunohistochemistry (IHC) against cleaved SNAP-25 (cSNAP-25), CGRP, VAChT (spinal cord sections) and against CGRP and ATF3 (DRG sections).

Results

Four weeks after SCI saline-treated animals presented high basal intravesical pressures and frequent detrusor contractions during bladder filling. IT administration of OnabotA significantly ($p<0.05$) reduced detrusor basal pressure and frequency of bladder reflex contractions and significantly ($p<0.05$) increased intercontraction intervals. Urinary retention was not observed in IT OnabotA-treated rats.

Consistent with the effects of OnabotA on bladder function, we found strong cSNAP-25 immunoreactivity in L5 and L6 spinal segments of rats that were injected with the toxin. SCI lead to an increase of spinal cord CGRP immunoreactivity, in comparison with sham-manipulated animals. Spinal CGRP immunolabelling was not affected by IT saline but was significantly reduced ($p<0.05$) after IT OnabotA. OnabotA did not alter spinal VAChT labelling.

In L5 and L6 DRG sections from sham-manipulated animals and SCI rats receiving IT saline ATF3 expression was very low and undetected in some sections. In contrast, the expression of this lesion marker was increased after IT administration of OnabotA. This was observed, but not restricted to, in CGRP-positive neurons.

Interpretation of results

Four weeks after SCI, animals treated with saline presented evident signs of NDO, as indicated by high basal intravesical pressures and frequent detrusor contractions during bladder filling. Intrathecal administration of OnabotA 4 weeks after SCI produced a significant decrease in detrusor basal pressure and in bladder reflex contractions and an increase in intercontraction intervals and in voiding amplitude. The positive effects of OnabotA on bladder function were confirmed by the strong presence of cSNAP-25, an established marker of this neurotoxin's action, in spinal sections from L5 and L6 segments. Also in the spinal cord, we found that the SCI-induced upregulation of CGRP expression was reduced by Onabot/A. VAChT immunoreactivity was not changed. Taken together, these data are indicative of an effect of the toxin on sensory afferents. Consistent with these results, we found increased ATF3 expression, a marker of neuronal damage, in L5-L6 DRG sections.

Overall, these results suggest that impairment of bladder sensory afferents by OnabotA is sufficient to restore bladder function, decreasing the number of episodes of urinary incontinence and efficiently protecting the upper urinary tract in chronic SCI rats. This work supports the intrathecal route for toxin administration as an effective alternative to detrusor injections.

Concluding message

IT OnabotA impaired bladder sensory afferents and significantly improved bladder function after SCI. This route of administration should be considered for SCI patients in order to diminish the episodes of urinary incontinence and infections and the possibility to develop potentially dangerous upper urinary tract complications.

References

1. Simpson LA, Eng JJ, Hsieh JT, Wolfe DL. The health and life priorities of individuals with spinal cord injury: a systematic review. J Neurotrauma. 2012 May 20: 29:1548-55

2. Cruz F, Herschorn S, Aliotta P, et al. Efficacy and safety of onabotulinumtoxinA in patients with urinary incontinence due to neurogenic detrusor overactivity: a randomised, double-blind, placebo-controlled trial. Eur Urol. 2011 Oct: 60:742-50
3. Coelho A, Oliveira R, Rossetto O, Cruz CD, Cruz F, Avelino A. Intrathecal administration of botulinum toxin type A improves urinary bladder function and reduces pain in rats with cystitis. Eur J Pain. 2014 Nov: 18:1480-9

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