Journal of Pharmacological and Toxicological Methods 80 (2016) 19–25

Contents lists available at ScienceDirect

Journal of Pharmacological and Toxicological Methods

journal homepage: www.elsevier.com/locate/jpharmtox

Research article

An animal model of detrusor overactivity induced by depression

Andrzej Wróbel *, Tomasz Rechberger

Second Department of Gynecology, Medical University of Lublin, Jaczewskiego 8, PL 20-090 Lublin, Poland

ARTICLE INFO

Article history:
Received 22 January 2016
Received in revised form 18 March 2016
Accepted 1 April 2016
Available online 2 April 2016

Keywords:
Cystometry
Depression
Detrusor overactivity
Methods
Rats
13-*cis*-Retinoic acid

ABSTRACT

Introduction: Depression is frequently found in patients suffering from overactive bladder. The aim of the study was to verify whether the 13-*cis*-retinoic acid, a synthetic retinoid used in the treatment of acne, which was proven to induce depressive changes in both humans and animals, can cause detrusor overactivity symptoms in conscious rats.
Methods: In order to assess the 13-*cis*-retinoic acid impact on the behavioural parameters, after 6 weeks of intraperitoneal administration of retinoid in a dose of 1 mg/kg/day, a forced swim test and cystometry were performed, and the locomotor activity of animals was assessed. The control group received a mixture of DMSO and physiological saline at a 1:1 ratio.
Results: 13-*cis*-retinoic acid caused cystometric parameter changes analogous to those observed in people with a urodynamic diagnosis of detrusor overactivity. The retinoid caused also an extension of the immobility time in the forced swim test which is consistent with increased depression-related behaviour, with no impact on the locomotor activity of rats. The intravenous administration of solifenacin succinate in a single dose of 0.03 mg/kg turned out to reverse changes in the cystometric parameters modified by 13-*cis*-retinoic acid treatment. The histopathological analysis of bladders did not show any lesions in the upper layer of the umbrella cells, urothelium or muscles. The retinoid concentration level achieved in the animals tested turned out to be identical to that occurring in humans.
Discussion: 13-*cis*-retinoic acid can induce detrusor overactivity symptoms that are reversed by solifenacin succinate.

1. Introduction

A range of mental disorders are more frequently found in patients suffering from overactive bladder (OAB) (Freeman, McPherson, & Baxby, 1985). The most frequently observed disorders include depression, which is one of the affective disorders (Saltiel & Silvershein, 2015). The connection between OAB and depression was described for the first time in 1964 (Dugan et al., 2000). In the medical literature we can very often see the informal term "uropsychiatry", highlighting the close connection between the aforementioned diseases, which has been corroborated in numerous epidemiological studies. Women suffering from wet or dry OAB achieve substantially lower scores on the Short Form Health Survey (SF-36) quality of life scale. It was also demonstrated that the aforementioned group had worse results on the Centre of Epidemiologic Studies Depression (CES-D) and quality of sleep scales (Dugan et al., 2000; Stewart et al., 2003). In turn, research by Nuotio et al. proved that depression is one of the risk factors of OAB (Nuotio et al., 2009). In other research conducted on subjects aged 65 or older it was demonstrated that depression measured using the Geriatric Depression Scale (GDS) is an independent risk factor of OAB, in the first year following the diagnosis (Hirayama et al., 2012). The impact of the lack of urinary incontinence therapy on the risk of developing depression was also assessed. In OAB patients, giving the therapy proved to be an independent risk factor for this affective disorder. In turn, non-treatment in the group of people with SUI did not appear to increase the risk of depression. This proves the direct correlation between depression and OAB or MUI, but not SUI (Shumaker et al., 1994).

It remains unknown whether depression exacerbates the existing symptoms of OAB or whether persistent symptoms of OAB are a factor inducing depression. According to other researchers, OAB is merely a manifestation of psychosomatic disorders. The presence of neurochemical disorders, which can underlie the etiopathogenesis of both depression and OAB, also should not be ruled out. Indeed, in pre-clinical and clinical studies it was demonstrated that 5-HT, noradrenaline (NA) and corticotropin-releasing factor (CRF) neurotransmission disorders can play an important role in the pathophysiology of both diseases (Lee, Na, & Dean-McKinney, 2003). It was established that a decrease in serotonergic transmission leads to the emergence of detrusor overactivity symptoms (Sweeney & Chancellor, 2005). It was demonstrated that duloxetine – an antidepressant acting as a selective serotonin–norepinephrine reuptake inhibitor – had a positive therapeutic effect, not only on SUI treatment, but also should be considered a drug of choice for multiple sclerosis patients displaying the symptoms of depression

* Corresponding author at: 20-954 Lublin, Jaczewskiego 8, Poland.
E-mail address: wrobelandrzej@yahoo.com (A. Wróbel).

http://dx.doi.org/10.1016/j.vascn.2016.04.001
1056-8719/

and OAB (Di Rezze et al., 2012). Its efficiency was verified in the treatment of both OAB-wet and OAB-dry patients (Steers et al., 2007). Tricyclic antidepressants – imipramine and amitriptyline – which increase the level of NA by inhibiting its reuptake, are used as second-line (off label) therapy for individuals with antimuscarinic-drug-resistant OAB (Hunsballe & Djurhuus, 2001).

13-*cis*-retinoic acid (13-*cis*-RA) is an extremely efficient drug used in severe acne pharmacotherapy. It is a synthetic retinoid and the only non-psychotropic drug most commonly reported to be associated with depression. Studies in human patients have shown that 13-*cis*-RA can induce depressive behaviours. A connection between treatment with 13-*cis*-RA and an outbreak of depression symptoms and suicidal ideation has also been identified (Bremner & McCaffery, 2008). Significant changes in the metabolic activity in the orbitofrontal cortex, in the cerebral structure, which intermediates in depression symptoms, have also been found in patients receiving 13-*cis*-RA (Bremner et al., 2005). In turn, people with diagnosed depression have been found to display up-regulation of the retinoic acid receptor α-immunoreactive neurons in the hypothalamic paraventricular nucleus (Chen et al., 2009).

These findings were confirmed in the studies conducted on animals, in which the chronic application of 13-*cis*-RA led to extending the immobility time in the forced swim test, which is consistent with increased depression-related behaviour (Cai et al., 2010 and Cai, Li, & Zhoub, 2015). Hippocampal neurogenesis inhibition, identified in connection with this retinoid, is a widely recognised factor causing depression (Crandall et al., 2004; O'Reilly et al., 2006 and Hu et al., 2013).

The aim of the study was to verify whether the 13-*cis*-RA administered in doses identical to those used in the treatment of human patients, which was proven to induce depressive changes in both humans and animals, can cause detrusor overactivity (DO) symptoms in conscious rats.

2. Materials and methods

All procedures were conducted in line with the NIH Animal Care and Use Committee guidelines, and approved by the Ethics Committee of the Medical University of Lublin. Female Wistar rats were used and randomly assigned to the treatment groups of 15 rats each. All animals were 4 weeks old at the start of treatment. A natural light/dark cycle, temperature + 22 °C and humidity 60% were maintained. Each rat was placed in a metabolic cage (3700M071,Tecniplast, USA) with free access to food and water. The animals were weighed on the day commencing the experiment. Their body weight was checked at one-week intervals until the completion of the experiment.

All the surgical procedures were performed under anaesthesia with an intraperitoneal injection of 75 mg/kg of ketamine hydrochloride (Ketanest, Pfizer) and 15 mg/kg of xylazine (Sedazin, Biowet). Rats were placed supine on a warming mattress (37 °C). Ketamine is often used in animal studies because it causes a dissociative type anaesthesia with minimal cardiac and respiratory depression. It is reported that ketamine in combination with xylazine does not eliminate the micturition reflex in female rats (Cannon & Damaser, 2001). The lack of spontaneous movement and withdrawal response to noxious toe pinch was viewed as indicating an adequate depth of anaesthesia.

2.1. Surgical procedures

The shaved and cleaned abdominal wall was opened through an approximately 10 mm vertical midline incision. The bladder was gently freed from adhering tissues. A double lumen polyethylene catheter (i.d. 0.28 and o.d. 0.61 mm.; BD, USA) filled with physiological saline with a cuff at the end was inserted through a small incision into the apex of the bladder dome and fixed with 6-0 Vicryl suture. In the same session the right femoral vein was catheterised through an inguinal approach. A polyethylene catheter (i.d. 0.28 and o.d. 0.61 mm; BD, USA) filled with 40 IU/ml heparinised physiological saline for infusion of solifenacin succinate or vehicle into the bloodstream was inserted into the vessel and advanced proximally until the tip of the catheter reached the abdominal aortic bifurcation. The catheters were tunnelled subcutaneously and exteriorised in the retroscapular area, where they were connected with a plastic adapter, to avoid the risk of removal by the animal. The chronically implanted catheters ensured stress-free conditions during the experiment. Finally, Healon (Pharmacia A.B.) in the dose of 0.85 ml was applied around the urinary bladder to avoid adhesions. The abdomen was closed in multiple layers. Anatomic layers were closed using 4/0 catgut sutures. The free ends of catheters were sealed with silk ligatures. The animals were injected subcutaneously with 100 mg of cefazolin sodium hydrate (Biofazolin, Sandoz) to prevent urinary tract infection.

2.2. Conscious cystometry

Cystometric investigations were performed in conscious unrestrained rats 3 days after the surgical procedures and after 6 weeks of 13-*cis*-RA treatment. A bladder catheter was connected via a three-way stopcock to a pressure transducer (FT03, Grass Instruments) situated at the level of the bladder and to a microinjection pump (CMA 100, Microject, Solna, Sweden) for recording intravesical pressure and for infusing physiological saline into the bladder. Conscious cystometry was performed by slowly filling the bladder with physiological saline (at a constant rate 0.05 ml/min, i.e. 3 ml/h) at a room temperature of 22 °C to elicit repetitive voiding. The analogue signal obtained from the pressure transducer was amplified and digitised using the Polyview system (Grass Instruments). Micturition volumes were measured by means of a fluid collector attached to a force displacement transducer (FT03C, Grass Instruments). Both transducers were connected to a polygraph (7 DAG, Grass Instruments). Cystometry profiles and micturition volumes were recorded continuously on a Grass polygraph (Model 7E, Grass Instrument) and were determined graphically. The data were analysed using a sampling rate of 10 samples/s. The body temperature of the rats was maintained at 37 °C with a heating pad throughout the study. The measurements in each animal represented the average of 5 bladder micturition cycles after obtaining repetitive voiding. Data on reproducible micturition cycles (5 per rat) were analysed and a mean of ± SEM for each animal in each condition was calculated. The mean values from all animals in each condition were averaged to create pooled data for each condition. The data is presented as the mean and SEM of the pooled data.

The following cystometric parameters were recorded: basal pressure (BP, cm H2O), threshold pressure (TP, cm H2O), micturition voiding pressure (MVP, cm H2O), voided volume (VV, ml), post-void residual (PVR, ml), volume threshold (VT, ml), intercontraction interval (ICI, sec), bladder compliance (BC, ml/cm H2O), the detrusor overactivity index (DOI, cm H2O/ml) — depicted as the quotient of the sum of amplitudes of all detrusor contractions during the filling phase and functional bladder capacity (Abrams, 2003; Juszczak et al., 2010; Wróbel, Łańcut & Rechberger, 2015; Wróbel & Rechberger, 2015), the non-voiding contraction amplitude (ANVC, cm H2O), the non-voiding contraction frequency (FNVC, times/filling phase), the volume threshold to elicit NVC (VTNVC, %) (Wróbel et al., 2015; Wróbel & Rechberger, 2015).

2.3. Locomotor activity

The locomotor activity of animals was assessed using a Digiscan apparatus — an Optical Animal Activity Monitoring System (Omnitech Electronics, Inc., Columbus, Ohio, USA). Activity chambers consisting of clear acrylic open field boxes were located in a room lit by a dim red light. The Digiscan system monitored animal locomotor activity via a grid of invisible infrared light beams. A number of equally spaced beams transversed the animal cage. The body of the animal placed in