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Pain and bladder dysfunction in an animal model of multiple sclerosis

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Aim: Here, we investigated if MS-induced pain and bladder dysfunction can be attenuated by TRPV1 desensitization with RTX.

Introduction: Multiple sclerosis (MS) is the most prevalent neurological disorder in young people, causing irreversible disability and producing substantial economic and social impact. Among the most incapacitating symptoms, neuropathic pain and bladder dysfunction are reported by the majority of patients. The transient receptor potential vanilloid 1 (TRPV1) is a receptor described to have an important role in neuropathic pain, bladder dysfunction and inflammation. TRPV1 desensitization with agonists, such as resiniferatoxin (RTX), has been shown to improve bladder function and reduce behavioural signs of pain in various animal models of disease. In the context of MS, a recent study showed that TRPV1 knockout mice were protected from disease progressions, presenting delayed disease onset, myelin preservation and reduced clinical scores.

Methods: Experimental Auto-immune Encephalitis (EAE) was induced by a single injection in the flank of a solution of myelin basic protein (MBP) in Complete Freund’s adjuvant (CFA). Behavioural tests were performed to evaluate symptoms. One month after MS-induction, animals were anesthetized and cystometries performed. Two other groups of MS animals received intrathecal RTX or vehicle and also submitted to behavioural tests and cystometries. At end of experiments, tissue was collected and processed.

Results: EAE rats developed neuropathic pain, as shown by the presence of mechanical allodynia and hypersensitivity to thermal stimuli. Cystometries performed at this time point showed signs of neurogenic detrusor overactivity. These clinical signs were accompanied by decreased spinal expression of MBP and increased activity of astrocytes and microglia. Preliminary observations suggest that intrathecal RTX improved cutaneous hypersensitivity and bladder function. These results suggest that TRPV1 might be involved in pain bladder dysfunction accompanying MS and that its modulation could have therapeutic relevance.

Conclusion: These results suggest that TRPV1 might be involved in pain bladder dysfunction accompanying MS and that its modulation could have therapeutic relevance.

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PS227

Neurogenesis in a rat model of sporadic Alzheimer’s disease

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Aim: Characterize adult hippocampal neurogenesis in a rat model of the initial stages of sporadic Alzheimer’s disease (AD).

Introduction: Sporadic late-onset AD is the most common cause of dementia, that can be characterized by a progressive cognitive decline, with a noteworthy episodic long-term memory impairment at early stages, accompanied by an excess accumulation of amyloid beta (Aβ) peptide in the brain. Present treatment options are very limited, so understanding AD pathophysiology is essential for exploring efficient therapies. Adult hippocampal neurogenesis is thought to play a crucial role in hippocampus-dependent cognitive abilities, namely learning/memory, although how this process is modulated in AD remains unclear.

Methods: An Aβ1–42 peptide solution was intracerebroventricularly injected into the rats’ lateral ventricle (the same volume of vehicle was injected to controls). Moreover, rats were injected with 5-bromo-2’-deoxyuridine (BrdU) intraperitoneally to study cell proliferation and differentiation. Two weeks after Aβ1–42 injection, the open field (OF) test and the novel object recognition (NOR) test were performed. Further behaviour tests are currently being performed, including the elevated plus maze (EPM), the Y-maze forced alternation test, and the Morris water maze (MWM) test. Focusing on the dentate gyrus, immunohistochemical analysis is presently being performed to investigate cell proliferation, neuronal differentiation and neuroblast/neuron morphology. Additionally, the presence of Aβ1–42 monomers and oligomers will be assessed by western-blots and the eventual occurrence of Aβ1–42 aggregates by histology.

Results: Our results show that the Aβ1–42 injection did not affect locomotor activity, as assessed by the OF test. Furthermore, this injection did not affect exploratory drive or episodic long-term memory performance, as indicated by the NOR test.

Conclusion: Since the NOR test is dependent from several brain regions besides the hippocampus that might not be affected in our model, additional behaviour tests as well as cellular and molecular analysis are needed to further characterize this model.

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PS086

Hydroalcoholic extract of Dorema aucheri leaves prevents weakening of the brain antioxidant defense system and inhibits oxidative damage in rat model of ischemic stroke

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Aim: To investigate the effect of hydroalcoholic extract of Dorema aucheri leaves on the antioxidant defense system and oxidative damage in rat model of ischemic stroke.

Introduction: Dorema aucheri is a native plant in many countries and its extracts have been traditionally used as a treatment for different pathological conditions. Ischemic stroke is a major cause of disability and death worldwide and the current treatment options are limited. Therefore, the search for new and effective therapeutic agents is essential.

Methods: Male Wistar rats were divided into four groups: Control, Ischemic, Ischemic + Dorema extract and Ischemic + Dexamethasone. The rats were subjected to 60 min of bilateral carotid artery occlusion followed by 24 h of reperfusion. The brain samples were collected for further analysis.

Results: The results showed that the hydroalcoholic extract of Dorema aucheri leaves prevents the weakening of the brain antioxidant defense system and inhibits oxidative damage in rat model of ischemic stroke.

Conclusion: The hydroalcoholic extract of Dorema aucheri leaves has a potential protective effect against the oxidative damage in rat model of ischemic stroke.

PS150

The Levator Auris Longus (LAL) muscle as an accessible system to study the effects of Botulinum Toxins in vivo

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Aim: In the present work, we aimed to find a reproducible model to study the effects of Botulinum neurotoxins (BoNTs), that allowed a widespread visualization of the intoxicated nerve terminals.

Introduction: Despite the well-established successful use of BoNTs to treat a variety of human conditions, their mechanism of action is still not fully understood. Thus, there is an emergent need of new and accurate models to study the effects of BoNTs. However, considering their potential lethality, it is challenging to find reproducible models to study the local application of BoNTs in living animals that allow a widespread visualization of the intoxicated nerve terminals.

In these work, we studied the innervation pattern and the effect of BoNTs in a group of small subcutaneous cranial muscles that are responsible for moving the pinna in rodents. Although all are easily accessible and manipulated, we focused on the levator auris longus (LAL).

Methods: Animals were injected subcutaneously with the indicated doses of BoNT/A, in the cranial muscles area. Muscles were then dissected and prepared for wholemount staining for Synapsin-I, cleaved SNAP-25 (synaptosome-associated protein of 25 kDa) and β3-tubulin.

Results: Detection of cleaved SNAP-25, the end-product of the catalytic action of BoNT/A, was possible even with injections as low as 0.1 ng. Mapping of the injected muscle showed the effect of BoNT/A in the majority of the endplate population. Also, seven days after BoNT/A injection, a sprouting process was evident, a landmark of regeneration.

Conclusion: BoNTs delivery to the LAL is a sensitive, simple and reproducible model to study the mechanisms of action of these toxins as it allowed the evaluation of BoNT/A effects throughout the entire muscle, without sampling bias. Thus, we forward that the LAL manipulation may constitute an excellent model to clarify the mechanisms of action of BoNTs in the neuromuscular system.

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