Prevention of Neuropathic Pain by A Selective Cannabinoid Type 2 Receptor (CB2) agonist (MDA7) In an Animal Model of Complex Regional Pain Syndrome Type 1

Authors: Jijun Xu, MD, PhD, Yu-Ying Tang, MD, Jiang Wu, MD, Bihua Bie, MD, PhD, and Mohamed Naguib, MD *Department of General Anesthesia, Anesthesiology Institute, Cleveland Clinic*

Background: Complex regional pain syndrome type 1 (CRPS-I) remains one of the most clinically challenging neuropathic pain syndromes with unclear mechanisms. In the spinal cord, microglial appears to be an upstream initiator of allodynia in neuropathic conditions, and activated microglia express CB2 receptors. Chemokine fractalkine receptor (CX3CR1) is primarily located in the microglia and is essential for neuroinflammation. The role of CX3CR1 and CB2 in CRPS-I remains unknown**.** Currently, there is no effective symptomatic treatment for CRPS-I. Cannabinoid receptor 2 (CB2) agonists have emerged as promising therapy for many neuropathic pain syndromes. MDA7 is a novel selective CB2 agonist. We hypothesize that the CB2 receptor functions in a negative-feedback loop and that early MDA7 administration can blunt the neuroinflammatory response and prevent mechanical allodynia induced by chronic post-ischemic pain (CPIP) through interference with specific signaling pathways in CRPS-I.

Methods: CPIP is used as the animal model of CRPS-I. CPIP is developed using an ischemia-reperfusion injury of the rodent hind paw. A tourniquet (a tight fitting O-ring) was placed on the right hindlimb of an anesthetized rat just proximal to the ankle joint for 3 h, and was removed prior to termination of the anesthesia to allow reperfusion. Sham rats were anesthetized but a cut O-ring was placed on the right hindlimb (no ischemia was induced). Additional groups of rats were treated with IP MDA7 (15 mg/kg) 30 min prior to and daily after CPIP induction for 14 days or IP AM630 (5 mg/kg, a CB2 antagonist) 15 min prior to MDA7 administration. Limb hyperemia, edema and spontaneous pain behaviors were noted. Mechanical allodynia was measured using von Frey filaments with logarithmic incremental stiffness.

Results: Rats in the CPIP group exhibited hyperemia and edema/plasma extravasation of the ischemic hindpaw, spontaneous pain behaviors (hindpaw shaking, licking and favoring), and spread of hyperalgesia/allodynia to the uninjured contralateral hindpaw. MDA7 prevented mechanical allodynia induced by CPIP (**Fig.1a**). MDA7 treatment was found to interfere with early events in the CRPS-I neuroinflammatory response as evidenced by reduced microglial activation and CB2 expression (**Fig.1b-d**), reduced expression of CX3CR1 (**Fig.1e-g**), and maintenance of intraepidermal nerve fiber architecture (**Fig. 1h,i**). MDA7's neuroprotective effect was blocked by a CB2 antagonist, AM630.

Conclusions: MDA7 is a drug candidate under study for its effects on neuroinflammation in several diseases. Our findings suggest MDA7 may offer an innovative therapeutic approach for treatment of allodynia induced by CRPS-I in the setting of traumatic ischemic or nerve injury.

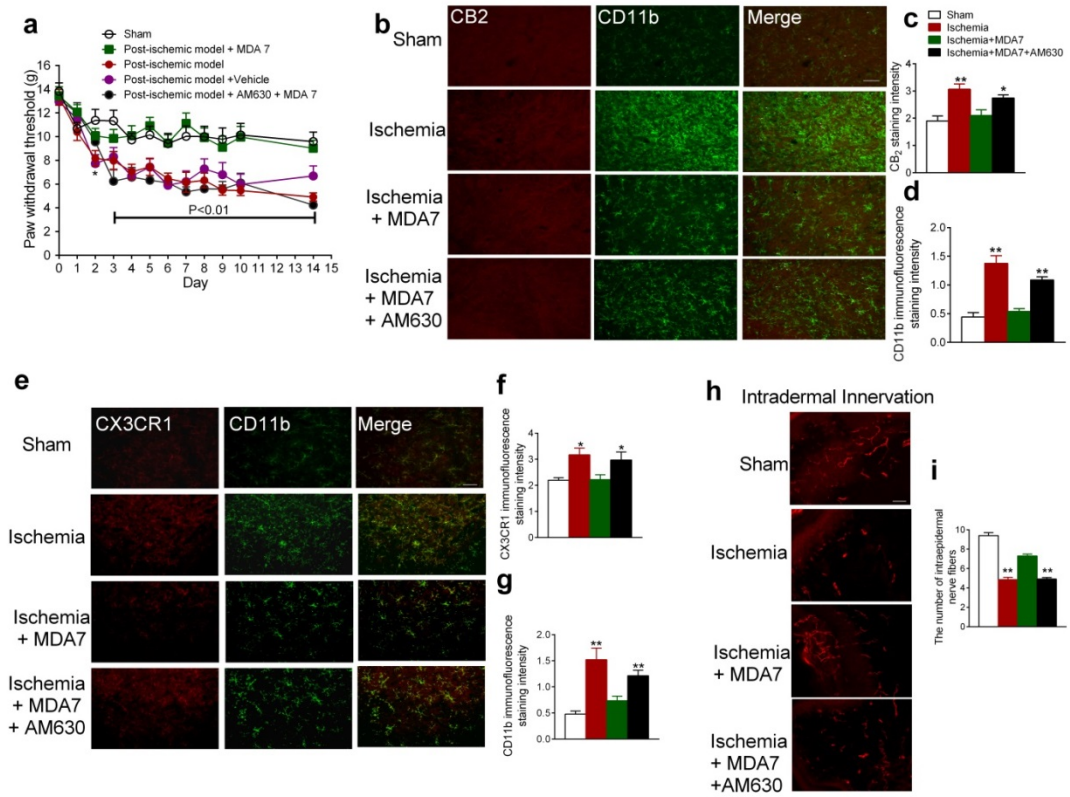

Figure. 1. Administration MDA7 significantly attenuated the upregulation of microglia, and expression of CB2 receptors and CX3CR1, and maintained intraepidermal nerve fiber architecture in a rat model of CRPS-1. Data represent mean ± SEM (n = 5-10 per group). Scale bar = 50 μm. *P<0.05, **P<0.01 *versus* sham and ischemia + MDA7 groups (ANOVA followed by Student-Newman-Keuls multiple range test).