Neuroscience Letters 644 (2017) 18–23

Contents lists available at ScienceDirect

Neuroscience Letters

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

Research article

Descending facilitatory pain pathways mediate ongoing pain and tactile hypersensitivity in a rat model of trigeminal neuropathic pain

C.F.M. Nones [a], R.F. Claudino [a], L.E.N. Ferreira [a], R.C. Dos Reis [a], T. King [b], J.G. Chichorro [a,*]

[a] Department of Pharmacology, Federal University of Parana, Curitiba, PR, Brazil
[b] Department of Biomedical Science, University of New England, Biddeford, ME, USA

HIGHLIGHTS

- Constriction of the trigeminal nerve induces early heat and late tactile facial hypersensitivity.
- CCI-ION induced spontaneous pain emerges between 5 and 15 days post injury.
- Descending pain facilitatory pain pathways mediate evoked and spontaneous pain 15 but not 5 days post-CCI-ION.
- Targeting RVM facilitatory pathways may improve treatment of chronic trigeminal neuropathic pain.

ARTICLE INFO

Article history:
Received 19 December 2016
Received in revised form 1 February 2017
Accepted 18 February 2017
Available online 22 February 2017

Keywords:
Trigeminal neuropathic pain
RVM
Spontaneous pain
Hypersensitivity

ABSTRACT

The Chronic Constriction Injury of the Infraorbital Nerve (CCI-ION) is a well-established model to study facial sensory changes related to trigeminal neuropathic pain. CCI-ION induces heat hypersensitivity that resolves within 2–3 weeks and a delayed mechanical hypersensitivity that emerges during the second week post-injury. The role of descending facilitatory pain pathways from the rostro ventromedial medulla (RVM) in mediating the heat and tactile hypersensitivity was examined. CCI-ION induced heat hypersensitivity observed 5 days post-surgery was reversed by systemic, but not RVM lidocaine. CCI-ION-induced tactile hypersensitivity observed 15 days post-surgery was reversed by systemic lidocaine and attenuated by RVM lidocaine. CCI-ION-induced spontaneous pain was determined using conditioned place preference (CPP) to pain relief at each time-point. At day 5 post-CCI-ION, neither systemic nor RVM lidocaine induced CPP. However, at 15 days post-CCI-ION, CPP was observed to the chamber paired with RVM lidocaine, but not systemic lidocaine. These data indicate that CCI-ION induced heat hypersensitivity is not dependent on descending facilitatory pain pathways 5-days post-injury whereas descending facilitatory pain pathways mediate tactile allodynia and spontaneous pain 15 days post-CCI-ION. This suggests that CCI-ION induces early peripheral sensitization followed by development of central sensitization that mediates spontaneous pain and contributes to mechanical hypersensitivity.

1. Introduction

Trigeminal neuralgia (TN) is a form of neuropathic pain characterized by severe lancinating pain in orofacial regions innervated by the trigeminal nerve. It is thought that most cases of TN result from compression of a trigeminal nerve root by the superior cerebellar artery in the posterior cranial fossa [1–3]. A significant proportion of TN patients report spontaneous pain in addition to the pain attacks [4,5]. Treatment options currently available for TN fail to provide reliable and permanent pain relief in all patients. Studies that contribute to a better understanding of mechanisms underlying TN pain are warranted to improve therapeutic strategies for treatment of these patients with persistent facial pain.

CCI-ION has been developed and characterized as an experimental model that reproduces important aspects of TN. CCI-ION induces mechanical and thermal hypersensitivity that occur across different time courses [6–8]. Heat hyperalgesia emerges within 2 days post-injury and dissipates between 2 and 3 weeks post injury [6,9]. In contrast, tactile hypersensitivity does not emerge until approximately 12–15 days post-surgery [6,9]. The observation that mechanical and thermal hypersensitivity emerge at different times post-CCI-ION suggests that different mechanisms mediate the early and late facial sensory changes after CCI-ION.

* Corresponding author.
E-mail address: juliana.chichorro@ufpr.br (J.G. Chichorro).

http://dx.doi.org/10.1016/j.neulet.2017.02.047
0304-3940/

Multiple laboratories have proposed an imbalance of inhibitory and facilitatory input during chronic pain states with a net gain in descending pain facilitation [10–13]. The rostral ventromedial medulla (RVM) is a critical site for descending modulation of nociceptive transmission [14] and has been implicated both in inhibition and facilitation of pain [15]. Preclinical models using spinal nerve injury indicate that the role of the RVM in pain modulation seems to change following nerve injury [12]. Spinal nerve injury induced evoked hypersensitivity has an initiation stage across approximately the first week during which evoked hypersensitivity is independent of descending facilitatory pathways from the RVM [12]. This is followed by a maintenance stage emerging during the second week during which evoked hypersensitivity is dependent on descending facilitation [12]. Subsequent studies demonstrated that descending facilitatory pain pathways from the RVM also mediate nerve-injury induced spontaneous pain [16,17]. The role of descending projections from the RVM to maintain different aspects of trigeminal neuropathic pain has not been explored. The present study examined the hypothesis that RVM neurons mediates tactile hypersensitivity and spontaneous pain 15 days post-CCI-ION, but do not alter thermal hypersensitivity or spontaneous pain 5 days post-CCI-ION.

2. Material and methods

2.1. Animals

Conventional heterogenic adult male Wistar rats, an outbred rat strain, weighing 200–220 g were used. Animals were bred and housed by the animal care facility of UFPR. Rats were housed 5 per cage on wood shaver bedding, maintained in a climate-controlled room at 22 ± 2 °C on a 12-h light/dark cycle with laboratory chow and tap water *ad libitum*. Studies were performed during the light cycle between 7:00 a.m. and 7:00 p.m. All procedures were approved by UFPRís institutional Committee on the Ethical Use of Animals (authorization # 805) and performed in accordance with the recommendations of the International Association for Study of Pain [18] and Brazilian regulations on animal welfare. All efforts were made to improve welfare and minimize the number of animals used.

2.2. RVM cannulation

Bilateral cannulation of the RVM was performed as previously described [12]. Animals were anesthetized with ketamine/xylazine (90/10 mg/kg, respectively) and placed in a stereotaxic apparatus. The skull was exposed and two 26-gauge guide cannulas separated by 1.2 mm (Plastics One Inc, Roanoke, VA) were directed toward the lateral portions of the RVM (AP-11.0 mm from bregma, L ±0.6 mm, DV-8.5 mm from the base of skull). The guide cannulas were cemented in place and fixed by small stainless steel screws. Rats then received oxytetracyclin (480 mg/kg, i.m.) and were allowed to recover 7 days before any behavioral testing or surgery.

2.3. Chronic Constriction Injury of the Infraorbital Nerve – CCI-ION

One week after RVM cannulation, animals were subjected to CCI-ION or sham surgery with some modifications [19] of the method proposed by Vos et al. [8]. Rats were anesthetized with ketamine/xylazine (90/10 mg/kg, respectively). An incision was made in the skin of the snout, under the right eye, about 3 mm caudal to the mystacial pads to expose the rostral end of the infraorbital nerve as it emerged from the infraorbital fissure. Two silk 4-0 ligatures were tied loosely around the infraorbital nerve 2 mm apart. The wound was closed with silk sutures. Sham operated rats were treated identically, but no ligatures were applied to the infraorbital nerve. After surgery, all rats were maintained in a warm room until they recovered from anesthesia. Independent groups of CCI-ION or sham rats were used for each behavioral test.

2.4. Evaluation of heat hypersensitivity

Heat sensory thresholds on the ipsilateral side of the face were measured before and 5 days post-surgery as previously described by Almeida et al. [20], with some modifications [21]. Each animal was removed from its home cage and gently held by the experimenter. A radiant heat source maintained at 50°C was positioned 1 cm from the surface of the right vibrissal pad. The latency to display either head withdrawal or vigorous flicking of the snout was recorded. A 20 s cut-off time was used to prevent tissue damage. To examine the effects of systemic administration of lidocaine, rats received lidocaine (10 mg/kg, s c.) or equivolume saline by a subcutaneous injection into the loose skin over the neck. Response latencies were assessed 30, 60, 90, 120, 150 and 180 min post-injection. To evaluate effects of RVM lidocaine injections, separate groups of rats received lidocaine (4% w/v in 0.5 μl) or equivolume saline injections across 1 min through a 33-gauge injection cannula protruding an additional 1 mm into fresh brain tissue to prevent backflow of drug. Response latencies were assessed 30, 60, 90 and 120 min post-injection. Only the ipsilateral side of the face was tested in to avoid overstimulation of the animal during testing of the time-course of the lidocaine effects on heat hypersensitivity.

2.5. Evaluation of mechanical hypersensitivity

Facial mechanical hypersensitivity was assessed before and 15 days post-surgery. During each testing session, animals were habituated to individual cages (30 × 30 × 30 cm) for a minimum of 2 h as described [22]. Mechanical thresholds were measured using calibrated von Frey filaments ranging from 0.04 to 8 g. Each stimulation series began with the 0.4 g filament, applied 3 times near the center of the right vibrissal pad, and proceeded up to the filament that evoked one of the following nociceptive behaviors twice: brisk head withdrawal, escape or attack reactions or short-lasting facial grooming. Only rats that did not react to application of the 8 g filament at pre-surgery testing were used for subsequent testing. Systemic or intra RVM injections of lidocaine were performed in separate cohorts of rats as described above. Mechanical thresholds were assessed 30, 60, 90 and 120 min post-injection. Only the ipsilateral side of the face was tested in to avoid overstimulation of the animal during testing of the time-course of the lidocaine effects on mechanical hypersensitivity.

2.6. Conditioned place preference (CPP)

A single-trial conditioning procedure was performed as previously described [23]. CPP was performed either days 4–6 post-surgery or days 14–16 post-surgery. Separate groups of rats were used at each time-point.

On the first day (i.e. preconditioning), rats were placed in the neutral chamber with access to all chambers for 15 min. Time spent in each chamber was analyzed to verify absence of preconditioning chamber preference. For all CPP groups, animals were removed from the study if they spent more than 720 s or less than 180 s in a single chamber (about 8% of rats tested) as previously reported [23].

Single trial conditioning occurred the following day. To determine if systemic lidocaine produced CPP, chamber pairing occurring 30 min following saline or drug administration to coincide with peak anti-hypersensitivity effects of systemic lidocaine.