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Thalamic activity and biochemical changes in individuals with neuropathic pain after spinal cord injury

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ABSTRACT

There is increasing evidence relating thalamic changes to the generation and/or maintenance of neuropathic pain. We have recently reported that neuropathic orofacial pain is associated with altered thalamic anatomy, biochemistry, and activity, which may result in disturbed thalamocortical oscillatory circuits. Despite this evidence, it is possible that these thalamic changes are not responsible for the presence of pain per se, but result as a consequence of the injury. To clarify this subject, we compared brain activity and biochemistry in 12 people with below-level neuropathic pain after complete thoracic spinal cord injury with 11 people with similar injuries and no neuropathic pain and 21 age- and gender-matched healthy control subjects. Quantitative arterial spinal labelling was used to measure thalamic activity, and magnetic resonance spectroscopy was used to determine changes in neuronal variability quantifying N-acetylaspartate and alterations in inhibitory function quantifying gamma amino butyric acid. This study revealed that the presence of neuropathic pain is associated with significant changes in thalamic biochemistry and neuronal activity. More specifically, the presence of neuropathic pain after spinal cord injury is associated with significant reductions in thalamic N-acetylaspartate, gamma amino butyric acid content, and blood flow in the region of the thalamic reticular nucleus. Spinal cord injury on its own did not account for these changes. These findings support the hypothesis that neuropathic pain is associated with altered thalamic structure and function, which may disturb central processing and play a key role in the experience of neuropathic pain.

1. Introduction

Pain is a common and debilitating consequence of spinal cord injury (SCI) [53]. Although several types of pain can develop after SCI, neuropathic pain is one of the most intractable and the most prevalent, occurring in almost 50% of individuals after SCI and may be due to damage to the spinal cord or nerve roots [42]. Although the mechanisms responsible for generating SCI neuropathic pain remain unknown, some have suggested that activity within the spinal cord itself is important. For example, it has been hypothesized that SCI neuropathic pain results from ongoing activity in intact residual spinothalamic tract pathways [51], or from an irritated focus immediately above the injury [22]. Although such factors may help maintain neuropathic pain, they do not seem essential because complete sensory blockade or verified surgical removal of several spinal cord segments immediately above the injury fail to relieve the pain in a considerable proportion of SCI subjects [4,24]. These observations strongly suggest that neuropathic SCI pain may ultimately result from supraspinal alterations.

There is increasing evidence for a critical role of the thalamus in the generation and/or maintenance of neuropathic pain. For example, neuropathic pain is associated with altered thalamic anatomy

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[10,12], altered thalamic biochemistry [12,29] and decreased thalamic perfusion [12,26,28]. Furthermore, there is increasing evidence that neuropathic pain may be associated with thalamocortical dysrhythmia [21,40,49], which may result from altered inhibitory output from the thalamic reticular nucleus (TRN) [18,32]. Consistent with the hypothesis, we have recently shown decreased thalamic gamma amino butyric acid (GABA) content and decreased perfusion in the region of the TRN in individuals with orofacial neuropathic pain [12].

Despite this evidence, it is possible that these thalamic changes (decreased GABA, perfusion, gray matter volume, and neuronal viability) are not responsible for the presence of pain per se, but result as a consequence of deafferentation. Indeed, there is evidence that although thalamic ventroposterior lateral (VPL) neurons in spinally injured rats show increased burst firing, this dysrhythmia is linked to spinal injury and not to the presence of behaviors suggesting pain in animal models [9,20]. Furthermore, we have previously reported that VPL thalamic anatomy was significantly altered in SCI subjects both with and without neuropathic pain, although a significantly greater change occurred in those with pain [10]. This raises the prospect that in neuropathic pain subjects, the injury itself may result in altered thalamic activity and biochemistry. Determining this distinction is important because future improvements in treatment are likely to hinge on our understanding of the neural mechanisms responsible for neuropathic pain.

To resolve this issue, we measured thalamic perfusion and biochemistry in both complete thoracic SCI individuals with and without neuropathic pain and in healthy control subjects. We hypothesized that individuals with SCI neuropathic pain would have reduced thalamic neuronal density (VP), gamma-aminobutyric acid (GABA) content, and regional perfusion (TRN). Further, we hypothesized that in individuals without pain, both those with SCI and healthy control subjects, no significant change in any of these parameters would occur.

2. Methods

2.1. Subjects

Twenty-two subjects with established complete thoracic SCI (16 men; age [mean ± SEM], 54 ± 3 years) and 21 age- and gender-matched control subjects without pain or spinal cord injury (13 men; mean age 51 ± 2 years) were recruited for the study. For the SCI subjects, the neurological level of injury was determined using the American Spinal Injury Association (ASIA) Impairment Scale (AIS) [23]. Each subject was examined by a clinician (P.W.) in the research group. The standardized AIS examination protocol was used to determine the most caudal level of the spinal cord with normal sensory and motor function on both sides of the body. An injury was termed complete when there was an absence of sensory and motor function in the lowest sacral segments. Twelve (8 men; mean age 57 ± 4 years) of the 22 SCI subjects experienced persistent neuropathic pain. All 12 SCI subjects had below-level neuropathic pain as defined by the International Association for the Study of Pain SCI pain taxonomy [43]. These subjects experienced continuous burning pain with intermittent stabbing and electric shock–like sensations in the region of sensory loss at least 3 segments below the neurological level of injury (Fig. 1). To assess the intensity of their pain, each subject completed a pain diary in which they indicated, with a vertical pencil stroke on a 10-cm horizontal line, the intensity of their pain (0 cm = no pain to 10 cm = maximum imaginable pain) 3 times per day for the week before the scanning session. These pain diary values were averaged to provide an indication of each subject's chronic pain rating. Each subject also rated their ongoing pain intensity during the magnetic resonance imaging (MRI) scanning session (Table 1). The remaining 10 SCI subjects (8 men; mean age 50 ± 4 years) did not have chronic neuropathic pain (Fig. 2). The frequency of nonneuropathic pain did not differ between the 2 SCI groups. No significant differences in age ($P = .17$) was found between SCI with pain and SCI without pain subjects. Both SCI groups were not significantly different in age compared with healthy control subjects (SCI with pain vs control subjects: $P = .18$; SCI without pain vs control subjects: $P = .69$). Informed written consent was obtained for all procedures, and the study was approved by the institutional human research ethics committees.

2.2. MRI acquisition

2.2.1. Quantitative arterial spin labelling

Subjects lay supine head-first on the bed of a 3-T MRI scanner (Intera, Philips, Best, The Netherlands) with their head immobilized in a tight-fitting 8-channel head coil. An isotropic 3-dimensional scan covering the whole brain was acquired (turbo field echo; echo time: 2.5 ms, repetition time: 5600 ms, flip angle: 8°, voxel size: 0.8 × 0.8 × 0.8 mm). Whole-brain cerebral blood flow (CBF) maps were then acquired using quantitative arterial spin labelling. The pulsed qASL version called QUASAR was used for this study, and the sequence and subsequent CBF quantification has been described in details elsewhere [30,31]. In short, the sequence is a multislice, multiple-time-points capable sequence based on pulsed qASL and a Look-Locker readout. In addition, the sequence acquires data with and without vascular crushers (gradients) (velocity cutoff = 4 cm/s), which allows the estimation of the arterial input function on a regional level. CBF is subsequently estimated by model free deconvolution, similar to what is done for gadolinium perfusion scans [30]. By adding a few dynamics at a lower flip angle, correct B1, T1, and M0 of tissue can be obtained, which are subsequently used for CBF quantification [31]. General scan parameters were: repetition time (TR): 4000 ms, echo time (TE): 23 ms, ΔTI: 300 ms, TI1: 40 ms, 13 inversion times (40 to 3640 ms), 64 × 64 matrix, 7 slices, slice thickness: 6 mm, 2 mm gap, field of view (FOV): 240 × 240, flip angle: 35/11.7°, SENSE: 2.5, 84 averages (48 with crusher: 4 cm/s, 24 without crushers, 12 low flip angle, all implemented in a single sequence with a scan duration of 5 minutes, 52 seconds.

2.2.2. Functional MRI (fMRI)

To accurately place the proton magnetic resonance spectroscopy (MRS) voxel in the somatosensory thalamus, we functionally defined the VPL thalamic nucleus in a group of 20 healthy subjects (14 men, age [mean ± SEM], 43 ± 3 years). A continuous series of 130 gradient echo, echo-planar fMRI image volumes using blood oxygen level–dependent contrast were collected. Each image volume contained 43 axial slices covering the entire brain (voxel 1.95 × 1.95 × 3.00 mm thick, repetition time 3000 ms; echo time 40 ms). During each fMRI series, the right big toe pad was brushed innocuously using a plastic brush at approximately 2 strokes/s. Each of these stimulation paradigms were performed for a period of 10 fMRI volumes (30 seconds) after a baseline period of 10 fMRI volumes (30 seconds). This was repeated a further 5 times for a total of 6 stimulation and 7 baseline periods.

2.2.3. Proton MRS

2.2.3.1. GABA-edited Mescher-Garwood Point Resolved Spectroscopy (MEGA PRESS). Multiplanar (axial, sagittal, coronal) reformats were used for voxel placement. GABA-edited MEGA-PRESS spectra [6,25] were acquired from a voxel (20 × 20 × 20 mm^3), which was centered in each subject in the thalamus. Because 11 of 12 subjects with pain had bilateral pain and the remaining subject had pain on the left side only, we collected MRS data from the right thalamus only. The MEGA-PRESS sequence parameters were as follows: TR: