THE ROLE OF TISSUE BIOMECHANICS IN PAINFUL INJURIES: MECHANICS, CELLULAR RESPONSES AND PAIN BEHAVIORS

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INTRODUCTION

Pain affects as many as 50 million Americans and injuries leading to persistent pain syndromes make up a unique class of mechanisms which remain largely uncharacterized. A common painful injury results from mechanical loading of nerve roots, which occurs in both the low back and neck due to spinal injuries. Relationships have been demonstrated between tissue compression and behavioral hypersensitivity responses in animal models, with differential patterns of sensitivity depending on the nature of the mechanical insult [1,2]. Mechanical allodynia (MA) is an increased behavioral sensitivity to a non-noxious stimulus and is observed in the dermatome of the injured tissue. It can be measured by the frequency of paw withdrawals elicited by stimulation with normally non-noxious von Frey filaments [2]. Allodynia is a clinical measure of sensitivity and therefore provides a useful gauge of nociceptive responses.

A combination of neurologic, electrophysiological, biochemical, structural, and mechanical contributions produces persistent pain [3]. Edema and increased endoneurial pressure have been reported in compressed nerve roots [4,5]. Electrophysiologic responses are altered according to the magnitude of injury intensity [6], suggesting possible mechanisms by which differential clinical pain symptoms may be produced. However, while the relationships between mechanical injury and the aspects of the pain response are currently being delineated, no research has defined mechanical thresholds for the onset and maintenance of pain-related behaviors in these animal models.

A host of CNS neuroimmune changes contribute to persistent pain [7], including glial cell activation, cytokine and chemokine upregulation and release. The role of CNS neuroinflammatory and neuroimmune activation responses in persistent pain is well documented and has been proposed as a potential mechanism through which central sensitization can occur [7]. While neuroimmune and electrophysiologic responses of the CNS likely work together to affect behavioral hypersensitivity, it is hypothesized that local biomechanics at the injury site modulates nociceptive cascades leading to pain.

Therefore, this study examines injury biomechanics for pain-behaviors in a rat lumbar radiculopathy (nerve root injury) model and examines neuroimmune responses in the CNS in the context of pain behaviors (MA) and injury mechanics. Using assessments of allodynia, mechanical thresholds for pain initiation and persistence are defined for nerve root compression in this injury model.

METHODS

Male Holtzman rats were divided into a sham (n=4) group with only nerve root exposure or a ligation group (n=18) with ligation of the left L5 nerve roots using 6-0 silk suture. Using image analysis, in vivo nerve root compressive strains were calculated at injury [1], using the nerve root boundaries in the unloaded and loaded (injured) conditions. Following injury, behavioral hypersensitivity was measured by recording allodynia responses postoperatively on days 1,3,5,7,10,14, using a 12-gm von Frey filament.

Compressive thresholds for pain onset and maintenance were determined using total allodynia responses, for each animal postoperatively. The onset of pain was defined as the highest allodynia response of any sham animal. Each ligated rat was assigned a pain score of 1 or 0 based on having a total allodynia sum: (1) above this limit or (0) equal to or below it. Likewise, the mean total allodynia for the ligation group was used to define a behavioral persistent pain response. In this case, a persistent pain score was assigned 1 for those responses above the mean and 0 for those below it. Logistic regression was used to determine tissue strain threshold values for pain behaviors due to nerve root compression.

In a subset of rats, lumbar spinal cord tissue was harvested on postoperative days 1,3,7,14 (5 rats/time point) for immunohistochemical (IHC) analysis of glial activation using OX-42 for microglial activation and GFAP for astrocytic activation [2].
To provide mechanical context for the injury strains, L5 spinal nerve roots (n=12) were harvested from matched rats for ex vivo mechanical compression testing. Displacements were applied incrementally using a translation micrometer. Compressive load was recorded until a maximum force of 1.35 N was reached. Load, displacement and image data were acquired at each increment, following a 30 second relaxation time. Using specimen geometry, stress-strain responses were determined for each specimen and the no-load strain was calculated by a linear fit of these stress-strain data.

RESULTS

For all shams, the mean calculated strain from image analysis was 0.32 ± 0.79%, which is within the range of analysis errors, indicating no measured tissue injury in shams. The mean applied strain for the ligations was 19.7 ± 10.6%. All animals undergoing nerve root ligation exhibited allodynia following injury (Figure 1). The typical behavioral response exhibited a robust initial increase in MA which was maintained with only minor decreases over time. Allodynia following ligation injuries was elevated over shams (p<0.001). For the ligations, allodynia responses displayed significant correlation (p<0.05) with the degree of applied compression (Figure 2).

DISCUSSION

These results provide specific mechanical thresholds for initiating and maintaining persistent behavioral hypersensitivity as measured by allodynia. While values suggest tissue tolerances for eliciting MA for nerve root ligation, they represent only one aspect of the nociceptive responses of pain. For example, this work does not define those thresholds for loading above which specific physiologic responses are initiated; they do not indicate when production and/or release of spinal cytokines is elevated, when glial activation occurs, or when electrophysiologic changes occur. The IHC findings here, and previously reported relationships between compression and cytokine upregulation [1], point to a modulatory role for injury biomechanics in elements of the nociceptive neuroimmune cascade for persistent pain.

The pain thresholds for tissue deformation are less than one-half those of the no-load strain for the rat nerve root in compression, suggesting that the tissue deformations eliciting persistent pain may actually be much less than those deformations at which nerve roots sustain load. This, along with the physiologic findings, highlights the need for continued research focused on understanding this unique class of injuries where physiologic dysfunction may precede that of any mechanical failure.

REFERENCES


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