increased from 3 day to 7 day in RVM as compared with sham operated animals. However, the expression of GFAP (an astrocyte marker) was not change in mirror image animal as compared with sham control. Intra-RVM pretreatment of minocycline (microglia inhibitor) or lidocaine exclusively inhibited contralateral allodynia rather than ipsilateral allodynia. Moreover, same effective dose of minocycline or lidocaine at mirror image pain did not modify the mechanical allodynia in ipsilateral pain only animals.
Conclusions: These results suggest that contralateral mechanical allodynia induced by SNL is associated with specific activation of neuron and glial cell in RVM.

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THE EFFECT OF GLIAL ACTIVATION ON CONTRALATERAL ALLODYNIA FOLLOWING SPINAL NERVE INJURY IN RATS

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Introduction: Unilateral nerve injury has been sometimes shown to induce contralateral mechanical allodynia in a rat model, and this phenomenon has been called "mirror-image pain". However, the mechanisms of mirror-image pain have not been well documented.
Objectives: Because spinal glial cells play an important role in the initiation and maintenance of neuropathic pain, we investigated the possible involvement of glial cells in mirror image pain.
Methods: Neuropathic pain was evoked by L5 spinal nerve ligation (SNL) in rats.
Results: From 3 day to 7 days after SNL, mirror image pain was evident about 30% of surgery rats. In mirror image pain animals, the expression of CD11b (a microglia marker) and GFAP (an astrocyte marker) was significantly increased from 3 day to 7 day in the contralateral spinal dorsal horn as well as ipsilateral spinal cord as compared with sham operated animals. Although intrathecal pretreatment of minocycline (microglia inhibitor) completely removed bilateral spinal CD11b elevation, it exclusively inhibited contralateral allodynia at 3 day and 7 day after SNL rather than ipsilateral allodynia. Moreover, intrathecal injection of fluorocitrate (astrocyte inhibitor) at 3 day after SNL failed to decrease contralateral allodynia as well as ipsilateral allodynia, but it showed significant antinoceptive effect only in contralateral allodynia at 7 days.
Conclusions: These results suggest that contralateral mechanical allodynia induced by SNL is associated with specific activaiton of glial cells in the contralateral spinal cord.

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TRANSMEMBRANE TNFA-MEDIATED SIGNALING IN MICROGLIA: IMPLICATIONS FOR NEUROPATHIC PAIN

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Introduction: States of chronic neuropathic pain caused are characterized by activation of microglia in the dorsal horn of spinal cord and enhanced expression of transmembrane tumor necrosis factor alpha (mTNFα).
Objectives: To determine the mechanisms responsible for expression of mTNFα in microglia and the role of mTNFα signaling in neuropathic pain.
Methods: We carried out a series of studies with immortalized microglial (HAPI) cells in vitro.
Results: We found that:

1. HAPI cells express the full-length NK1 receptor, as well as TNFR1 and TNFR2;
2. HAPI cells are activated by LPS in induce expression of TNFα and TACE, and rlease sTNFα and MCP-1 in a manner identical to primary microglia;
3. Exposure to substance P increases expression of mTNF through activation of PI3K-AKT but does not induce TACE or increase release of sTNF;
4. TACE is requred for the release of sTNFα, and knocking down TACE by siRNA enhances accumulation of mTNFα, as does treatment with the TACE inhibitor TAPI-2;
5. mTNF α expressing cells activate microglia through cell-cell contact, measured by increased expression of OX-42 and release of MCP-1;
6. Recombinant mTNFα protein increases TNF but not TACE in microglia;
7. Cells that overepxress mTNFα in the membrane assume a stellate morphology.

Conclusions: SP increases expression of mTNFα in microglia, and mTNFα activates microglia though cell-cell contact. Taken together these results suggest the existence of a feed-forward mechanism trigered by SP release that may be important in the transition from acute to chronic pain.

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THE EFFECT OF BOTULINUM NEUROTOXIN A ON CCI-INDUCED NEUROIMMUNOLOGICAL CHANGES IN RAT DRG AND SPINAL CORD

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Introduction: Botulinum neurotoxin serotype A (BoNT/A) relieves neuropathic pain symptoms, however the mechanism still remains unclear.
Objectives: Descovering the potential mechanism of action of BoNT/A after chronic constriction injury (CCI) of the sciatic nerve in rat.
Methods: We measured neuropathic pain using behavioral tests and mRNA levels of neuropeptides, synaptosomal-associated protein-25 (SNAP-25), neuronal (NOS1) and inducible (NOS2) nitric oxide synthases as well as microglial (C1q) and astroglial (GFAP) markers using competitive RT-PCR – according to IASP rules.
Results: In the lumbar spinal cord of CCI-exposed rats the ipsilateral upregulation of prodynorphin, SNAP-25 and C1q and no changes in NOS1, NOS2, GFAP, proenkephalin and pronociceptin mRNAs were observed. In DRG the ipsilateral upregulation of prodynorphin, pronociceptin, NOS1, NOS2, C1q and GFAP mRNAs and, in contrast, downregulation of proenkephalin and no changes in SNAP-25 mRNAs were observed. A single intraplantar administration of BoNT/A (75 pg/paw) induced long-lasting antiallodynic/antihyperalgesic effects, which are paralleled by reduced prodynorphin, pronociceptin, NOS1 mRNAs upregulation in DRG together with significant decrease of C1q-positive cells and SNAP-25 mRNA upregulation, both in spinal cord and DRG. No changes were observed for GFAP mRNA both in spinal cord and DRG.
Conclusions: We provide evidence that BoNT/A not only relieves neuropathic pain behavior, but also attenuates microglial/macrophage and SNAP-25 activation in the spinal cord and DRG and NOS1, prodynorphin and pronociceptin mRNAs level increase in DRG. These results indicate the possible mechanism of long-lasting effects of BoNT/A in neuropathic pain.
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