Spinal NR2B-Regulated EMR-1-BAF-LEMD2 Complex Drives EZH2-Mediated Mu-Opioid Receptor Downregulation in Neuropathic Pain

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Abstract:

Neuropathic pain is a debilitating condition with limited treatment options, partly driven by spinal epigenetic mechanisms. This study highlights the critical role of NR2B-driven Barrier-to-Autointegration Factor (BAF) signaling in mediating mu-opioid receptor (Oprm1) downregulation in the dorsal horn after spinal nerve ligation (SNL). Following nerve injury, SNL induced a significant increase in BAF levels in the ipsilateral dorsal horn, peaking on postoperative Day 7, with males showing greater elevations than females. Immunostaining localized the upregulation of BAF to neurons, correlating with mechanical allodynia. Intrathecal siRNA knockdown of BAF reduced BAF expression, restored withdrawal thresholds, and alleviated allodynia, demonstrating its pivotal role in neuropathic pain. Further investigations revealed that NR2B glutamate receptor activation triggered the upregulation of BAF and its interacting partner LEMD2 and EMR-1. Immunostaining and biochemical analyses showed colocalization of BAF, LEMD2, and EZH2, a histone methyltransferase responsible for H3K27 trimethylation. Chromatin immunoprecipitation demonstrated that EMR-1/BAF/LEMD2 signaling facilitated EZH2 recruitment to the Oprm1 promoter, leading to H3K27me3 enrichment and subsequent mu-opioid receptor silencing. AlphaFold 3 structural modeling provided additional insights, uncovering conserved domains critical for the interactions between EMR-1-BAF, BAF-LEMD2 and LEMD2-EZH2 complexes. Blocking NR2B receptors disrupted BAF-related pathways and restored mu-opioid receptor expression, linking glutamatergic input to the observed epigenetic modifications. These findings establish a novel pathway in which NR2B-driven glutamatergic signaling activates EMR-1BAF/LEMD2, contributing to epigenetic repression of the Oprm1 promoter. This pathway offers promising therapeutic targets for neuropathic pain management.

Keywords:

pNR2B, BAF, LEMD2, EZH2, H3K27me3, mu-opioid receptor, dorsal horn, neuropathic pain.