Astrocyte reaction in the lateral habenula contributes differently to spontaneous and evoked pain in paclitaxel-induced peripheral neuropathy

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Introduction

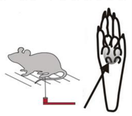
The painful neuropathy induced by paclitaxel treatment negatively influences the quality of life for patients with cancers and thus hinders its clinical usage. The contribution of reactive astrocytes to chronic pain during chemotherapy has gained increasing attention in recent several years but less is understood regarding their regulation of pain related responses at a supraspinal level. The lateral habenula was extensively studied in stress related conditions like chronic pain, however, whether astrocytes in the lateral habenula are responsible for pain signaling regulation is not yet understood. In the current study, we demonstrated the presence of astrocyte reaction in the lateral habenula and illustrated its distinct contributions to the sensory and affective components of paclitaxel induced painful neuropathy during the late phase.

Methods

❖Evoked pain evaluation

➢ *von Frey test*

Calibrated von Frey filaments were applied perpendicular to the mid-plantar surface of the left hind paw of mice to assess mechanical hyperalgesia which was demonstrated as reduced paw withdrawal threshold.

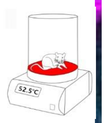


➢ *Hargreaves test*

A radiant heat stimulation was applied to the mid-plantar surface of the left hind paw to assess thermal hyperalgesia which was demonstrated as reduced paw latency to the heat stimulus.

➢ *Hot plate test*

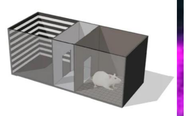
Mice were placed on the plate with the temperature set at 52.5 ℃ and the latency to the first hind paw withdraw was recorded as a positive response.



❖Affective pain evaluation

➢ *Conditioned place preference*

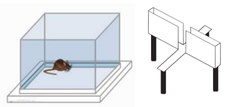
A single-trial conditioned place preference protocol was used to evaluate affective and spontaneous pain.



❖Pain associated anxiety evaluation

➢ *Elevated plus maze and open filed test*

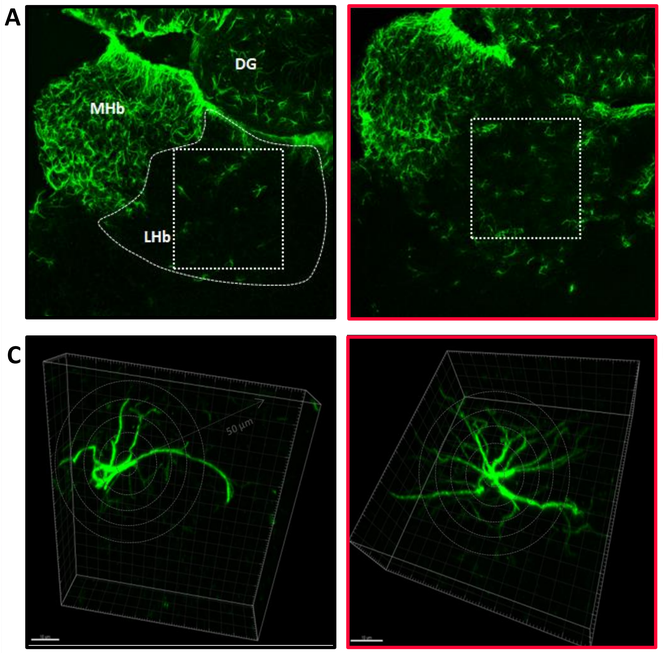
Elevated plus maze test and open field test were used to evaluate the anxiety-like behavior which was interpreted as less time spent in the open arms and the center area, respectively.



Results

➢ **Astrocyte reaction and hypertrophy in the lateral habenula (LHb) during the late phase of PIPN.**

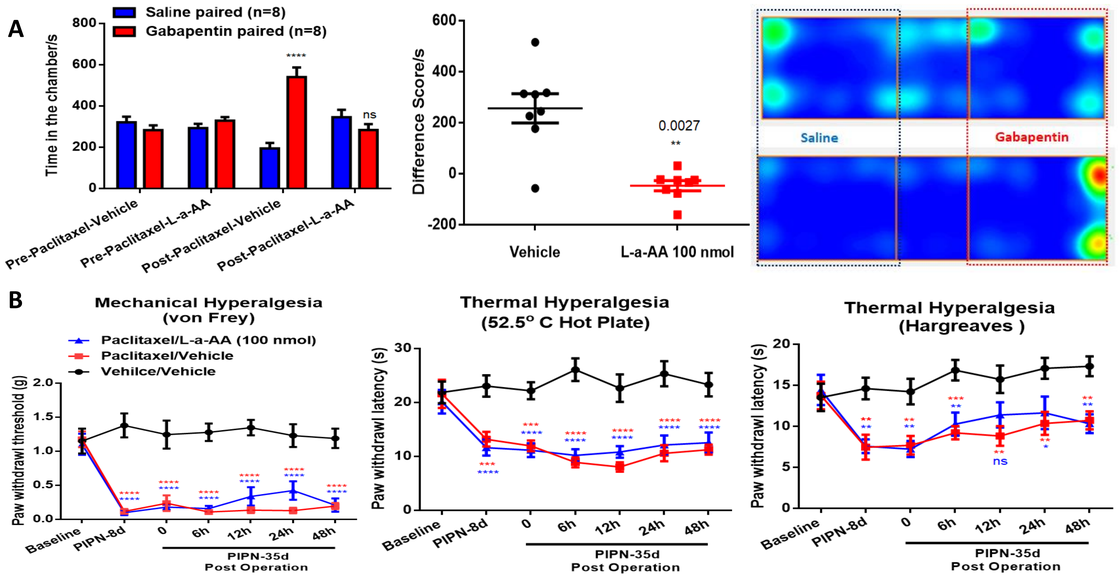
Vehicle *Paclitaxel*



Overexpression of GFAP in LHb and cellular hypertrophy during the late phase of PIPN.

(A) Representative images of GFAP staining in LHb. (**B**) Western blot analysis showed increased GFAP expression at day 35 following repeated paclitaxel compared to vehicle treatment. **(C)** 3D animation representative astrocytes from each group. **(D)** Sholl profile of astrocytes in each group: significant hypertrophy appeared at day 35 following paclitaxel compared to vehicle treatment group.

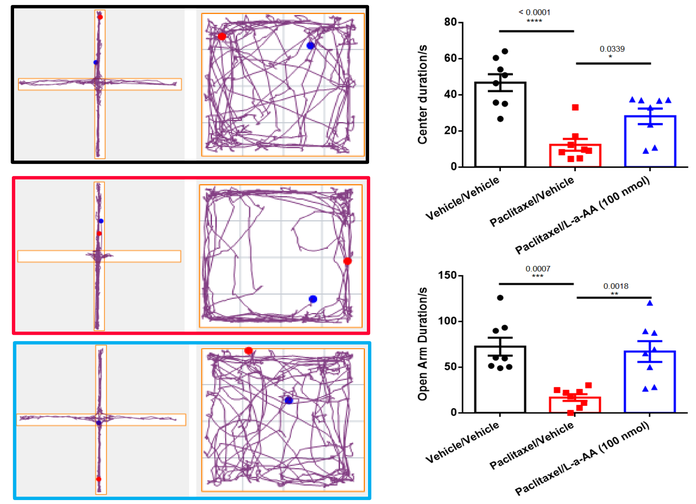
➢ **Activated astrocytes in LHB contributes differently to evoked and spontaneous pain behaviors**



Unilateral microinjection of L-α-AA in right LHb attenuated spontaneous (A) but not evoked pain (B) during the late phase of PIPN.

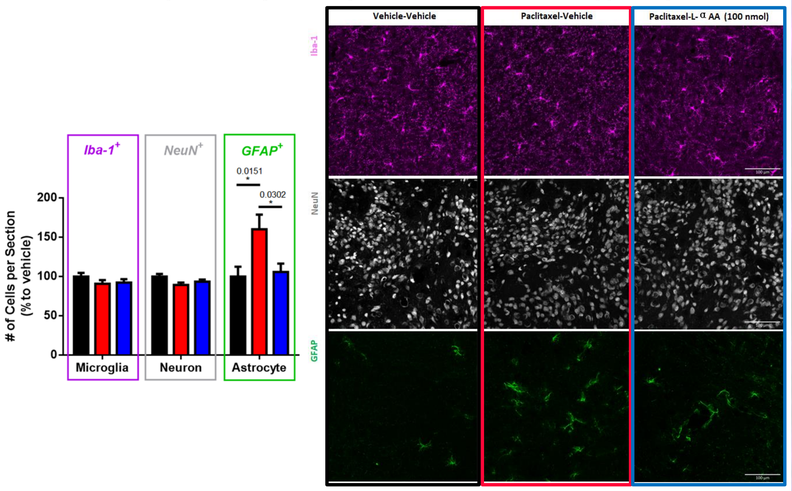
Results

➢ **Activated astrocytes in LHB contribute to PIPN associated anxiety behaviors**



Unilateral microinjection of L-α-AA in right LHb attenuated PIPN associated anxiety-like behaviors

➢ ***Selective depletion of astrocyte but not other cell types by L-α-AA***



Astroglial toxin (L-α-AA) specifically induced astrocyte depletion without affecting neurons or microglia in LHb

A remarkable decrease was seen in the number of astrocytes but not microglia or neurons in the paclitaxel/L-α-AA group, compared to the paclitaxel/vehicle group.

Conclusion

1.Astrocyte reaction occurred in LHb during the late phase of PIPN.

2.Astrocyte reaction contributes differently to sensory and affective pain aspects.