Anesthesiology
SCHOOL OF MEDICINE
UNIVERSITY OF COLORADO
ANSCHUTZ MEDICAL CAMPUS

Novel Neuroactive Steroid Attenuates Mechanical Hypersensitivity and Weight-Bearing Deficits in a Murine Model of Chronic Pain

Matheus Ribeiro[1], Benjamin Volvovitz[1], Christin Tran[2], Mingxing Qian[3], Douglas F Covey[3,4], Slobodan M. Todorovic [1,5], Vesna Jevtovic-Todorovic [1,6]

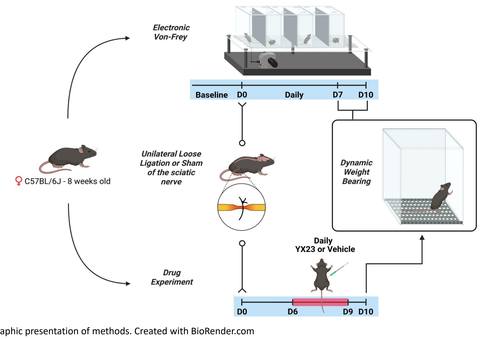
Scan me for full paper.

Background

We developed a novel neuroactive steroid (**YX23** - (3β,5α,17β)-3-Methoxyestran-17-ol) that is an effective **blocker of CaV3.2 channels** with diminished sedative properties.

Our research aims to assess the effects of the novel drug in a chronic pain model. Therefore, using the **unilateral sciatic nerve chronic constriction injury (CCI) model of neuropathic pain in mice**, we performed **the e-Von Frey Mechanical Sensitivity Test** and **the Dynamic Weight Bearing (DWB) Test** to assess **evoked** and **spontaneous pain**, respectively.

Methods



Graphic presentation of methods. Created with BioRender.com

8-week-old C57BL/6J female mice subjected to either unilateral loose ligation of the sciatic nerve or sham surgery.

CCI is performed under anesthesia by exposing the sciatic nerve through a skin incision and muscle dissection, then a chromic ligature is tied loosely around it without arresting epineural blood flow.

Mechanical sensitivity was assessed using an electronic Von Frey apparatus (Ugo Basile), measuring the minimum force (grams) required to elicit a withdrawal response. Testing was performed at baseline and on 10 consecutive days post-surgery. **Spontaneous pain** was assessed on day 10 post-surgery using the DWB device (Bioseb), quantifying the weight-bearing percentage of each hind paw.

After establishing the chronic pain model, behavioral tests were repeated following daily intraperitoneal injections of either **YX23 (100mg/kg) or cyclodextrin (vehicle) from day 6-9 post-surgery.**

Statistical analysis of the e-Von Frey data was performed using two-way analysis of variance (ANOVA) with Tukey's multiple comparisons test, while a paired t-test was used to analyze the DWB data. Experiments were approved by the Institutional Animal Care and Use Committee at the University of Colorado.

Figure 1

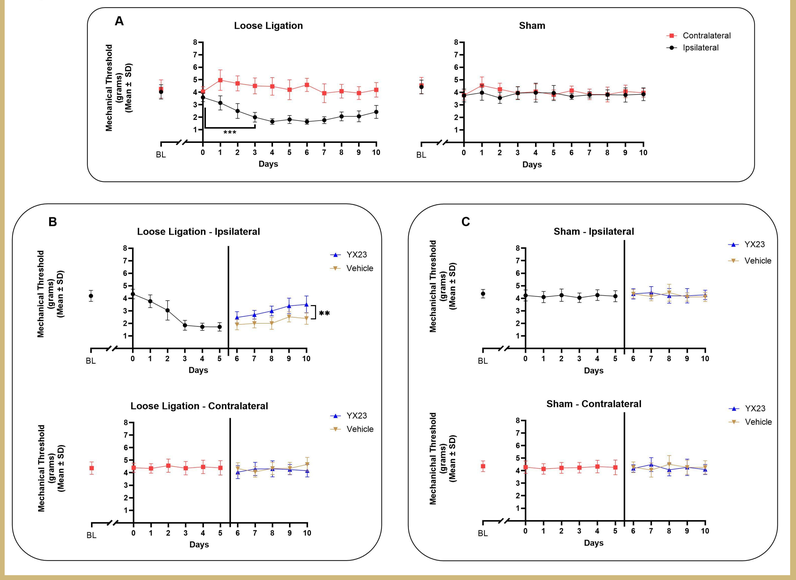
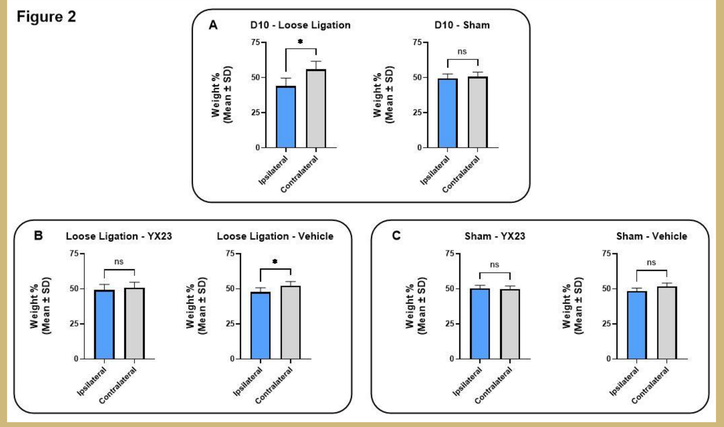


Figure 2





Results

YX23 attenuated mechanical hypersensitivity in the ipsilateral paw, evidenced by a significant increase in mechanical threshold compared to vehicle-treated CCI animals (p=0.0034 at day 10 post-surgery).

Furthermore, treatment reversed the hind-paw weight-bearing deficit, as indicated by the lack of a statistically significant difference between ipsilateral and contralateral weight-bearing measurements.

Conclusions

The literature establishes that the analgesic potential of neuroactive steroids (NAS) is mediated largely by targeting CaV3.2 subtype of voltage-gated calcium channels, as evidenced by the lack of effect in CaV3.2 KO mice. However, current NASs hold a significant hypnotic/sedative properties, hindering their clinical use[1,2,3].

Notably, a previous study demonstrated that YX23 (100mg/kg) exhibits minimal sedative properties, using Open Field behavioral testing [3]. These findings suggest YX23 as a promising analgesic with reduced sedation.

References

1. Joksimovic SL, Joksimovic SM, Manzella FM, et al. Novel neuroactive steroid with hypnotic and T-type calcium channel blocking properties exerts effective analgesia in a rodent model of post-surgical pain. Br J Pharmacol. 2020;177(8):1735-1753. doi:10.1111/bph.14930
2. Jevtovic-Todorovic V, Covey DF, Todorovic SM. Are neuroactive steroids promising therapeutic agents in the management of acute and chronic pain?. Psychoneuroendocrinology. 2009;34 Suppl 1:S178-S185. doi:10.1016/j.psyneuen.2009.06.001
3. Volvovitz B, Miah R, Park K, et al. Novel Neuroactive Steroid Analogs and Voltage-Dependent Blockers of CaV3.2 Currents, B372 and YX23, Are Effective Anti-Nociceptive with Diminished Sedative Properties in Intact Female Mice. Biomolecules. 2025;15(8):1175. Published 2025 Aug 16. doi:10.3390/biom15081175

Disclosures/Acknowledgements

I have no conflicts of interest to disclose. I would like to thank the Vesna Jevtovic-Todorovic Lab members, specially to Benjamin Volvovitz for collecting the behavior data.