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Author's reply to commentary on 'altered in vivo early neurogenesis traits in patients with depression: Evidence from neuron-derived extracellular vesicles and electroconvulsive therapy' by Dr. Yakovlev

Dear Editors

In our previous paper [1], Dr. Yakovlev addresses a critical point [2] about CD81 and CD63 Western blot bands appearing at approximately 50 kDa, instead of at 25 kDa and 30 kDa, respectively. Given that small extracellular vesicles (EVs) may contain IgG, we cannot definitively conclude that there is no heavy chain contamination in the CD81 and CD63 blots from our previous paper [1].

To address this concern, we conducted additional Western blot tests for another three EV markers, CD9 (25 kDa, Abcam, Catalog# ab263019), Alix (96 kDa, Proteintech, Catalog #67715-1-Ig), and HSP70 (70 kDa, Elabscience, Catalog# E-AB-22005), which do not have target molecular weights around 50 kDa, to minimize the risk of potential contamination from heavy chains. In addition, although our earlier paper [1] showed NCAM1 bands at the expected molecular weight (93 kDa), we also tested another neuronal marker, SNAP25 (25 kDa, Invitrogen, Catalog # MA5-17610), which do not have target molecular weights around 50 kDa, neither. The results, presented in the supplementary file, showed bands at the expected molecular weights.

For clarity, it's important to note that in our prior work [1], western blotting was used alongside nanoparticle tracking analysis (NTA) and transmission electron microscopy (TEM) solely to verify the presence of neuron-derived EVs (NDEVs). The primary analysis was relied on quantitative data from enzyme-linked immunosorbent assay kits, not on western blotting images.

Finally, although the NDEVs method [3] were widely used [4–7], we are grateful for Dr. Yakovlev's insightful critique, which has contributed to enhancing the reliability of our research.

CRediT authorship contribution statement

Shu-xian Xu: Investigation, Writing – original draft. **Zhongchun Liu:** Funding acquisition, Supervision, Writing – review & editing.

Declaration of competing interest

The authors declare that there is no conflict of interest in this work.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.brs.2024.05.001.

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