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Peer Review

# Review of: "Phosphatidylserine (PS)-Targeting Chimeric Interferon (IFN) Fusion Proteins for Anti-Tumour Applications"

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First, although the authors demonstrate activation of interferon signaling pathways, additional mechanistic characterization would improve the study. Inclusion of quantitative downstream immune signaling analyses such as interferon-stimulated gene profiling, cytokine secretion assays, or immune cell infiltration profiling within tumor tissues would strengthen the immunological interpretation of the results.

Second, while the in vivo tumor suppression results are promising, additional safety and toxicity assessments would be valuable, particularly considering the known adverse effects of interferon-based therapies. Systematic evaluation of hematological parameters, inflammatory cytokine levels, and long-term toxicity would enhance confidence in translational applicability.

Third, the manuscript would benefit from a more detailed quantitative evaluation of PS-binding specificity and affinity. Comparative binding assays or kinetic studies against established PS-targeting agents such as Annexin V or anti-PS antibodies would further validate the targeting strategy and emphasize its novelty.

Fourth, several methodological descriptions require further clarity. Expanded details regarding vector construction, fusion protein quantification, transfection stability, and animal experimental design would improve reproducibility. Additionally, reporting precise protein concentrations rather than relative dilutions in functional assays would strengthen data interpretation.

Finally, the discussion could be enhanced by providing a more balanced comparison between this platform and existing PS-targeting or interferon-based therapeutic approaches, including their relative



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advantages and potential limitations.

Overall, this study represents a novel and promising therapeutic concept with strong experimental support and translational potential. With additional mechanistic validation, improved methodological clarity, and expanded safety evaluation, the manuscript would make a valuable contribution to the fields of cancer immunotherapy and targeted biologic drug development.

## Declarations

**Potential competing interests:** No potential competing interests to declare.