Compatibility of the OPM-293 Transient Expression Platform with Expi293F and Expi293 Pro Cells Supports Enhanced Protein Production

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1. Introduction

Transient transfection-based protein expression in mammalian cells is widely used for rapid recombinant protein production in research and early-stage development[1]. The overall performance of a transient expression workflow depends not only on transfection efficiency, but also on post-transfection cell growth, culture robustness, and sustained protein productivity[1,2].

The OPM-293 transient expression platform is developed to support high-yield protein expression in HEK293-derived suspension cultures. However, because different Expi293-based host cells can behave differently under the same expression workflow, it is important to assess whether the platform performs consistently across commonly used HEK293-derived cell lines.

In this study, we evaluated the compatibility of the OPM-293 transient expression system with both Expi293F and Expi293 Pro cells using two expression plasmids encoding representative biologic formats: an Fc-based fusion protein and a human IgG. We further compared productivity profiles over time and total protein output to determine whether Expi293 Pro cells provide an advantage under the OPM system.

2. Materials & Methods

Category	Description
Cell Lines	Expi293F and Expi293 Pro Cells
Expression System	OPM-293 CD05 Medium, OPM-293 ProFeed, and CarpTrans Transfection Reagent (TR)
Protein Models	Fc-based fusion protein and human IgG
Seeding Density	Expi293F (n=1): 3 × 10^6 cells/mL; Expi293 Pro (n=2): 3 × 10^6 and 5 × 10^6 cells/mL
Culture Conditions	37 °C, 8% CO_2, orbital shaking; cells passaged 1 day prior to transfection
Transfection Conditions	Cells viability confirmed prior to use; plasmid DNA added at 1 µg/mL culture; DNA:TR ratio adjusted based on system
Feeding Strategy	OPM-293 ProFeed added 20h post-transfection
Analytical readouts	VCD (viable cell density), viability, titer, glucose and lactate

3. Results & Discussion

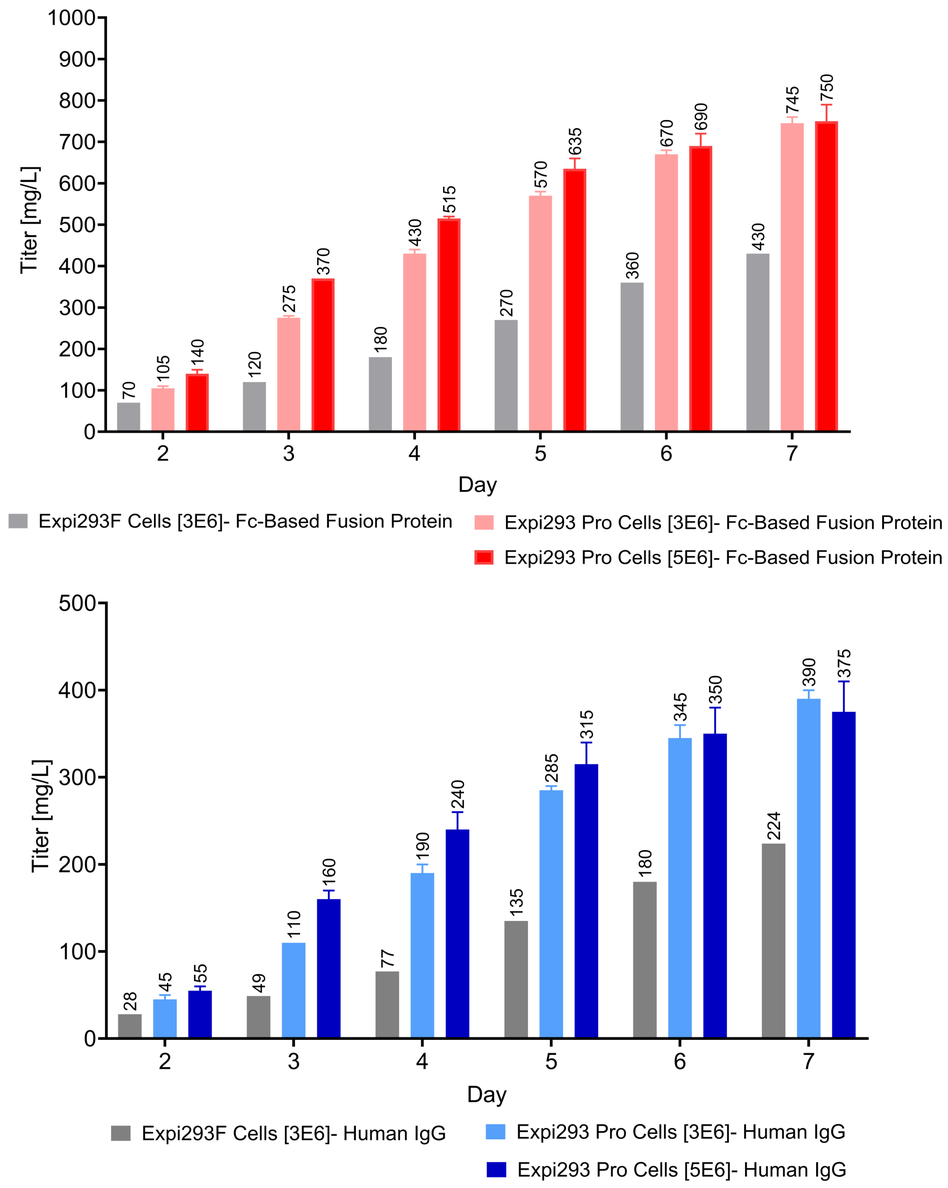

Figure 1. OPM-293 expression system supports transient expression in both Expi293F and Expi293 Pro cells, with enhanced productivity in Expi293 Pro cells.
The OPM-293 system supported protein expression in both Expi293F and Expi293 Pro cells for both recombinant protein models tested, demonstrating compatibility across Expi293-based host cells. Under the OPM workflow, Expi293 Pro cells consistently produced higher titers than Expi293F cells for both Fc-based fusion protein and human IgG. This productivity advantage became more pronounced over time, suggesting improved post-transfection performance in Expi293 Pro cells. These results indicate that while the OPM-293 system is compatible with both host cell types, Expi293 Pro cells provide enhanced productivity under the tested conditions.

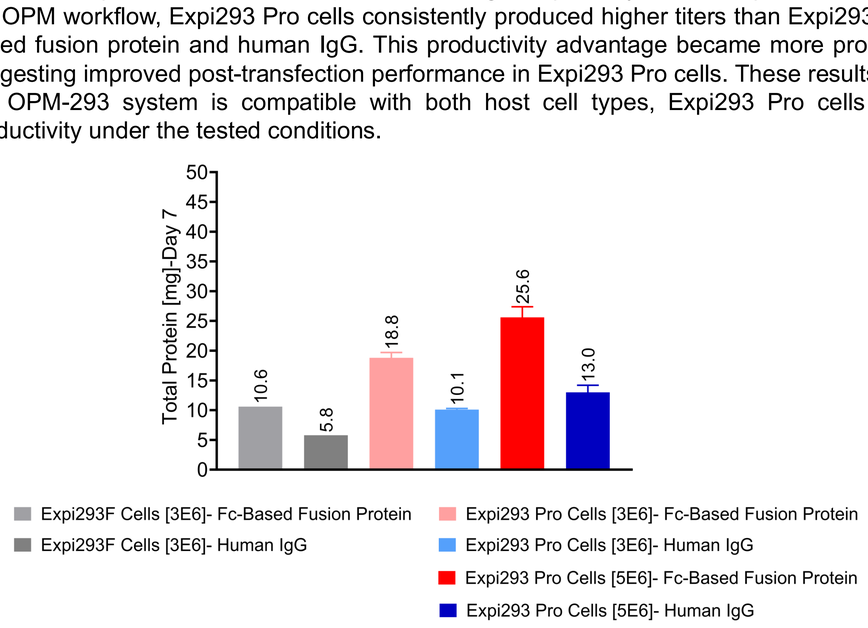

Figure 2. Day 7 total protein confirmed the productivity advantage of Expi293 Pro cells.
Since culture volumes differed between the 3 × 10^6 and 5 × 10^6 seeding density conditions, Day 7 total protein was used as a more accurate measure of productivity than titer alone. This analysis confirmed the same trend observed in the titer data:
- Expi293 Pro cells generated more total protein than Expi293F cells
- This trend was consistent across both protein models tested

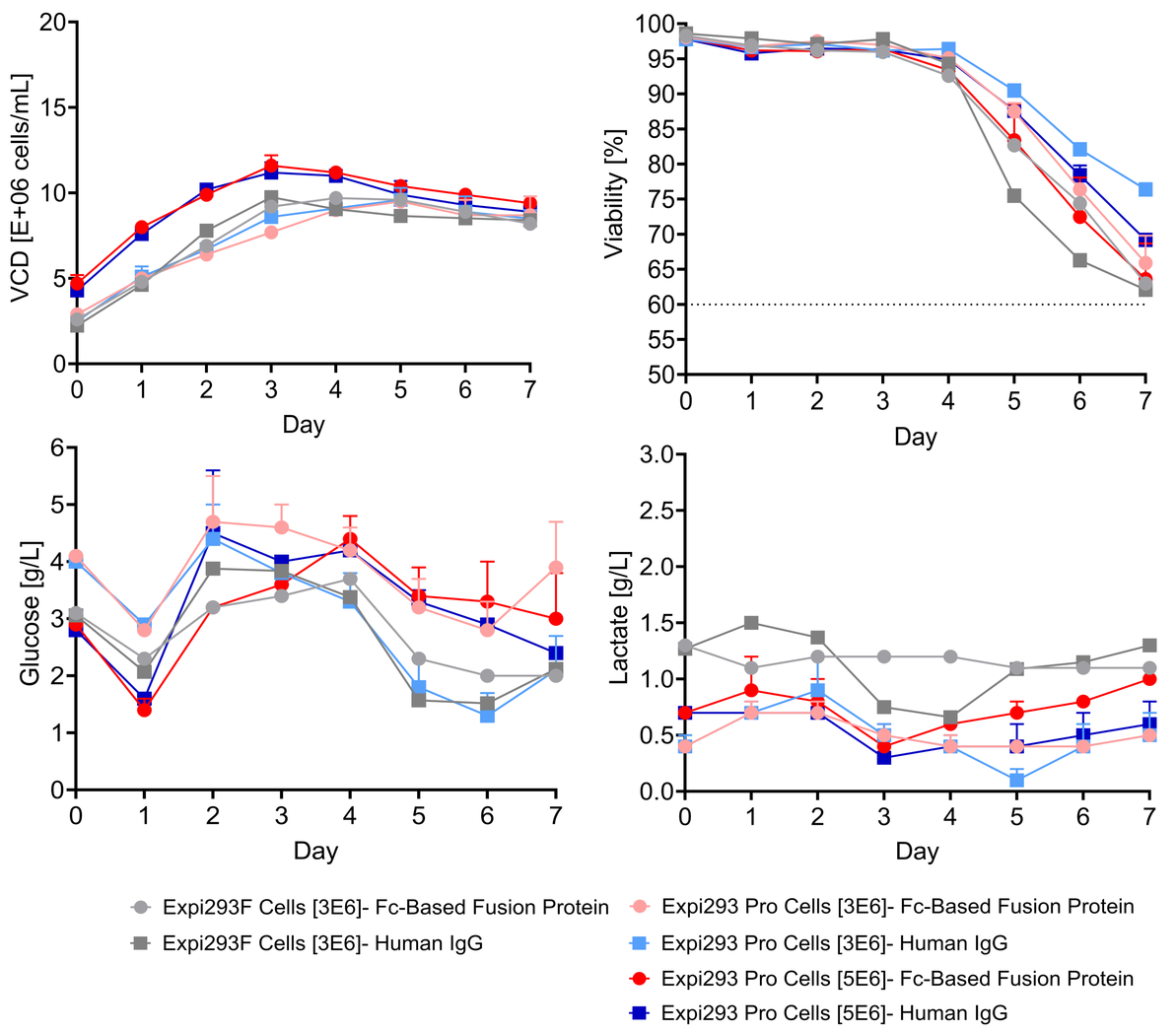

Figure 3. Growth, viability, glucose, and lactate profiles demonstrate robust post-transfection performance in both host cells.
All conditions exhibited similar growth and viability trends. In Expi293 Pro cells, viability remained >85% through Day 5 across all conditions. Glucose was monitored daily and supplemented when levels fell below 2 g/L. Lactate levels remained low, indicating controlled metabolic activity.

4. Conclusions & Future Work

The OPM-293 transient expression system supported efficient transient protein production in both Expi293F and Expi293 Pro cells, demonstrating broad compatibility across Expi293-based host cells. Under the tested OPM workflow:
- Expi293 Pro cells consistently achieved higher titers
- Expi293 Pro cells generated greater total protein by Day 7
- This productivity advantage was observed for both tested expression plasmids, encoding an Fc-based fusion protein and a human IgG

Together, these findings indicate that while the OPM-293 system is compatible with both host cell types, Expi293 Pro cells provide enhanced post-transfection productivity and may represent a more favorable host for high-yield transient protein expression workflows.

Future studies will focus on further optimizing the OPM-293 transient expression workflow to improve productivity, particularly for high-density Expi293 Pro processes. Future optimization areas include:
- Optimization of transfection reagent performance (e.g. DNA to transfection reagent ratio)
- Refinement of feed timing and supplementation strategy
- Evaluation of culture conditions to support sustained post-transfection cell growth and productivity

These efforts will help further define the operating window of the OPM-293 system and support its application in scalable transient protein expression workflows for recombinant protein and biologics research.

Acknowledgements

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References

[1] Fu, Yushun et al. "Improvement strategies for transient gene expression in mammalian cells." *Applied microbiology and biotechnology* vol. 108,1 480. 4 Oct. 2024.
[2] Baldi, Lucia et al. "Recombinant protein production by large-scale transient gene expression in mammalian cells: state of the art and future perspectives." *Biotechnology letters* vol. 29,5 (2007): 677-84.

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