Enhanced Post-Electroporation Cell Recovery Using a High-Density Cell Respirator

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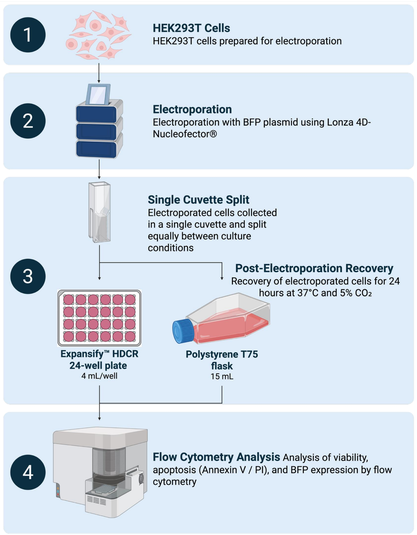
BACKGROUND

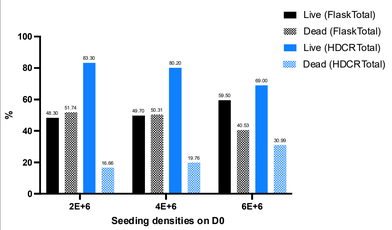
Electroporation is a widely used non-viral approach for delivering nucleic acids and gene-editing cargo into cells, but it imposes substantial cellular stress that can reduce viability, recovery, and functional output. As cell and gene therapy workflows increasingly demand scalable manufacturing with high engineered cell yield, optimization of the post-electroporation recovery environment has become increasingly important. Expansify™ high-density cell respirator (HDCR) cultureware utilizes a gas-permeable silicone membrane designed to enhance oxygen transfer while maintaining a static, low-shear culture environment. Here, we investigated whether the Expansify™ microenvironment could improve post-electroporation recovery and transgene expression relative to conventional polystyrene culture systems.

WORKFLOW



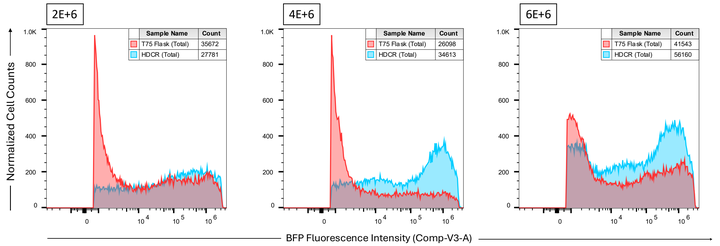
IMPROVED POST-ELECTROPORATION RECOVERY AND FUNCTIONAL OUTPUT IN EXPANSIFY™

1. Enhanced Post-Electroporation Viability

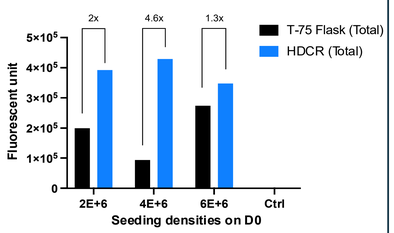


Flow cytometric viability analysis at 24 hours post-electroporation demonstrates improved recovery of HEK293T cells in Expansify™ HDCR cultureware relative to polystyrene T75 flasks across seeding densities. Enhanced viability was most pronounced at lower cell densities, suggesting density-dependent recovery effects in conventional culture systems.

2. Increased Functional BFP Expression Post-Electroporation

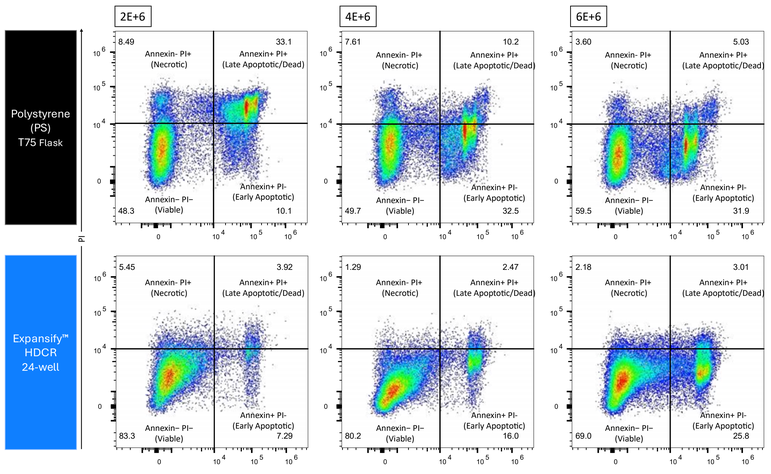


Representative flow cytometry histograms acquired 24 hours post-electroporation demonstrate increased BFP-positive cell populations in Expansify™ HDCR cultures relative to conventional polystyrene T75 flasks across all tested seeding densities. Enhanced fluorescence distributions in HDCR cultures are consistent with improved functional recovery and transgene expression following electroporation-induced cellular stress.



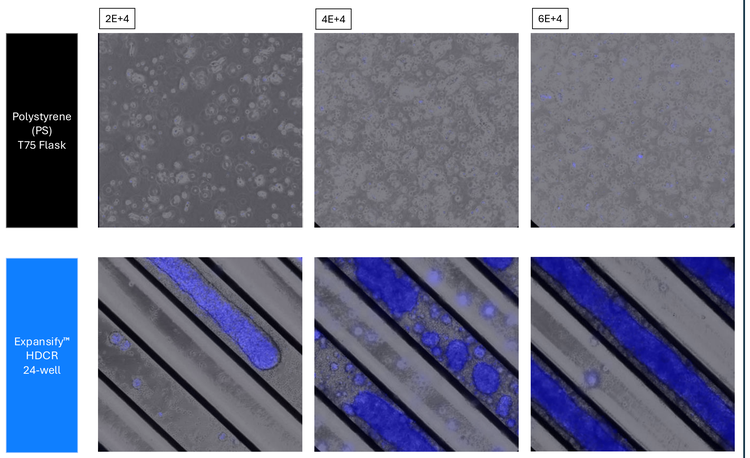
Quantification of BFP mean fluorescence intensity (MFI) at 24 hours post-electroporation demonstrates enhanced transgene expression in Expansify™ HDCR cultures relative to conventional polystyrene cultureware across seeding densities, consistent with improved functional recovery of engineered cells.

3. Annexin V / PI Recovery Profiles (24h)



Representative Annexin V / PI pseudocolor plots at 24 hours post-electroporation demonstrate reduced apoptotic and non-viable cell populations in Expansify™ HDCR cultures relative to polystyrene controls across seeding densities. Analysis was performed on the total recovered population, including both adherent/live cells and cells collected from the culture supernatant. Quadrants represent viable (Annexin−/PI−), early apoptotic (Annexin+/PI−), late apoptotic/dead (Annexin+/PI+), and necrotic (Annexin−/PI+) populations.

4. Representative Recovery Morphology and BFP Expression (24h)



Representative brightfield/BFP overlay images acquired 24 hours post-electroporation demonstrate marked differences in post-electroporation recovery between conventional polystyrene cultureware and Expansify™ HDCR cultureware across seeding densities. Cells recovered in Expansify™ HDCR wells exhibited increased cell retention, higher localized cell density, and stronger BFP-associated fluorescence relative to polystyrene controls. Enhanced fluorescence signal and improved cellular organization in HDCR cultures are consistent with improved survival and functional recovery following electroporation. Images were acquired under matched imaging conditions for qualitative comparison across culture systems and seeding densities.

KEY TAKEAWAYS

1. Expansify™ HDCR cultureware enhances post-electroporation cell recovery relative to conventional polystyrene systems.
2. Improved recovery was most pronounced at lower seeding densities, where conventional culture conditions showed reduced viability.
3. Enhanced BFP fluorescence intensity in HDCR cultures indicates improved functional transgene expression following electroporation.
4. Annexin V / PI analysis demonstrated reduced apoptotic and non-viable cell populations in HDCR recovery conditions.
5. Improved recovery and functional output support the potential of HDCR systems for scalable non-viral cell engineering and CGT manufacturing workflows.

CONCLUSION

Expansify™ HDCR cultureware improved post-electroporation viability and functional BFP expression relative to conventional polystyrene culture systems, particularly at lower seeding densities. Reduced apoptotic populations and enhanced recovery profiles suggest that the HDCR microenvironment supports improved recovery of stressed cells following electroporation. These findings support further evaluation of HDCR systems as a scalable recovery platform for non-viral cell engineering and CGT manufacturing workflows.

DISCLOSURES

This work was sponsored by XDemics Corporation. Aishwarya Rajendra Churi, Austin R. Santiago, and Colin Cook are employees of XDemics Corporation. Austin R. Santiago and Colin Cook hold equity in XDemics Corporation. Alison W. Ashbrook received sponsored research support from XDemics Corporation but is not employed by the company.

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EXPLORE THE PLATFORM