Scale-up of Protein Production By Flow Electroporation With A Variety of CHO Cell Lines

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Abstract

Pharmaceutical, biopharmaceutical, and biotechnology companies must efficiently identify, develop, and quickly bring to market candidates with the highest level of efficacy at the lowest cost. Being able to work in the cell line of choice during discovery helps to meet these requirements. MaxCyte flow electroporation technology offers a universal means of flexible, fully scalable, high efficiency transient gene expression and is capable of producing multiple grams of antibodies following a single CHO transient transfection. In this poster, data will be presented demonstrating the ability of MaxCyte flow electroporation to produce high antibody titers in a variety of different CHO cell lines. Data also will be presented showing the use of MaxCyte electroporation to scale up protein production with consistent cell viability, growth kinetics, and antibody titers in CHO cells.

MaxCyte STX®
5E5 Cells in Seconds
Up to 2E10 Cells in <30 Min

MaxCyte VLX®
Up to 2E11 Cells in <30 Min

The MaxCyte STX® and MaxCyte VLX® Transfection Systems use fully scalable flow electroporation for rapid, highly efficient transfection with very high cell viability post transfection. Transfected cells support multi-gram scale production of antibodies and proteins for efficient biotherapeutic development.

CHO-S Cells: Multi-Gram Antibody Production

2.7 g/L Achieved in <3 Weeks

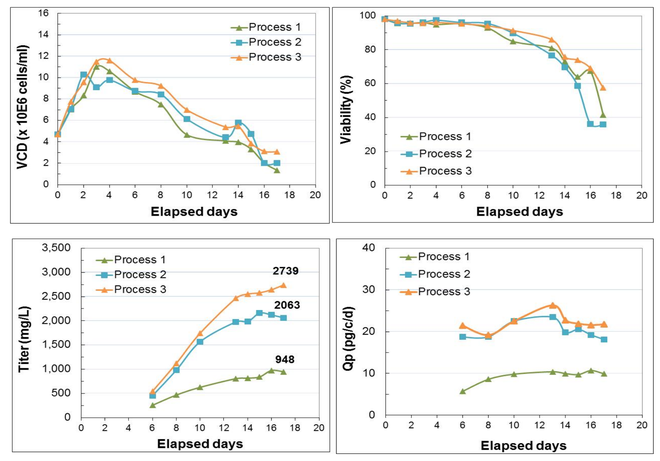

Figure 1. Transient Expression of hIgG1 antibody in MaxCyte EP Transfected CHO-S cells. The same transfected cells were in different production processes. Further optimized process (process 3) can reach 2.7 g/L as a fed batch. Titer was verified by both ELISA and Protein A capture assays.

High Titers With Different CHO Cell Lines

CHO EBNA and 293 EBNA: Superior Expression With STX

High Level Protein Expression

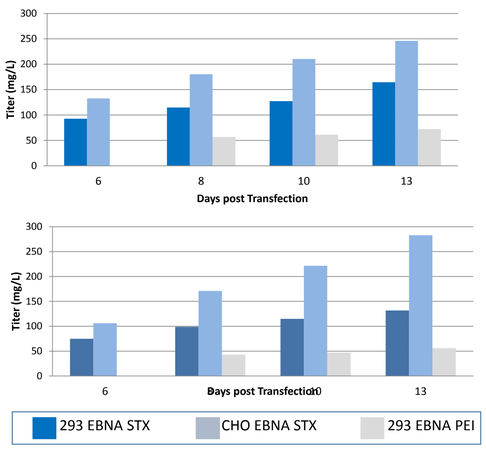

Figure 2. High Titer mAb Expression in CHO EBNA and 293 EBNA Cells. CHO EBNA and 293 EBNA cells were transfected with an IgG expression plasmid via static electroporation (6E7-8E7 cells per condition) and cultured in 125 mL shake flasks for 13 days. Secreted antibody titers in both STX-transfected cell lines greatly exceeded titers generated by an optimized PEI transfection method in 293 EBNA cells.

CHO-K1 Cells: Robust Protein Expression

3-24 Fold Higher than Other Methods

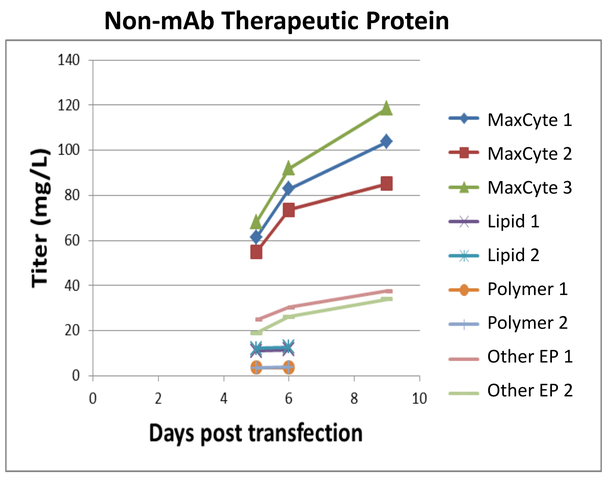

Figure 3. Superior Therapeutic Protein Production in CHO-K1 Cells with the MaxCyte STX Compared to Other Transfection Methods. CHO-K1 cells were transfected with a plasmid encoding a recombinant protein and cultured in 125 mL shake flasks for up to 10 days. Titers in three sets of cells transfected with the STX were significantly higher than titers generated by cells transfected with lipids, polymers, and a research-scale electroporation instrument,

CHO-K1SV Cells: High Titer Expression

Rapid Production of Antibodies

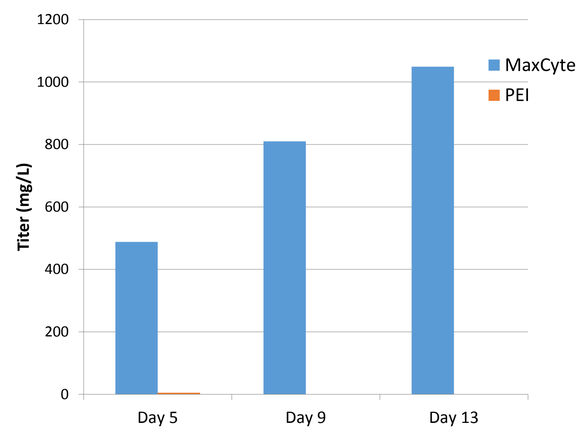

Figure 4. High Titer Antibody Production in CHO-K1SV Cells via MaxCyte EP. CHO-K1SV cells were transfected via static EP or PEI with an IgG expression plasmid and cultured for 13 days or 5 days, respectively. Titer was assayed in the transfected cells on days 5, 9, and 13 post EP; titer was measured in the PEI transfected cells on day 5. The day 5 titer data indicated clear superiority of MaxCyte EP vs PEI, and the titer data on day 13 revealed productivity exceeding 1 g/L in STX-transfected CHO-K1SV cells.

Consistent, Reliable Scale Up With CHO Cells

Flow Electroporation on MaxCyte STX & VLX Instruments

Consistent Viability, VCD, and Antibody Titers

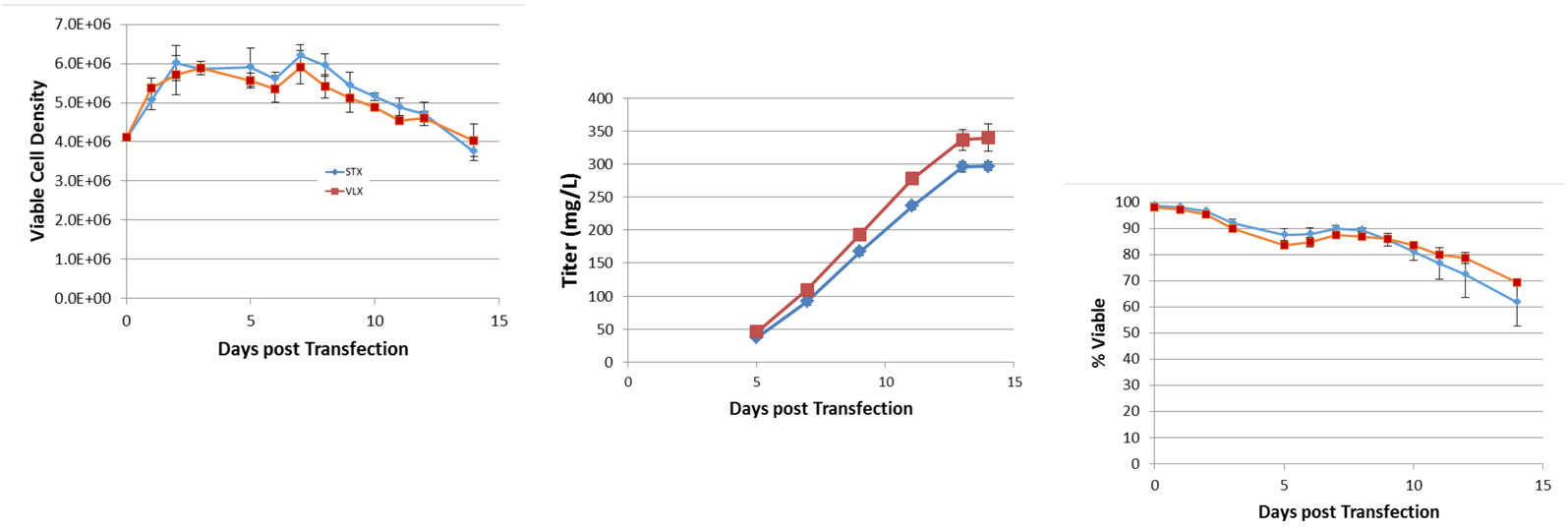

Figure 5. CHO-S Cells Transfected via Flow EP using the STX and VLX Instruments Exhibit Consistent Protein Titers, Cell Viabilities, and Growth Kinetics. Duplicate sets o f2E10 CHO-S cells were transfected with an hIgG1 expression plasmid (1 µg DNA/1E6 cells) via flow EP with the STX and the VLX. Transfected cells were seeded into 1-liter cultures at a starting density of 4E6 cells/mL, and fed batch cultures were maintained for 15 days. Secreted protein titers were monitored by ELISA; cells counts and viabilities were measured via trypan blue staining. Error bars denote standard deviations from three replicate shake flasks.

Transient Transfection for Bioproduction

Seamless MaxCyte STX to VLX Transition

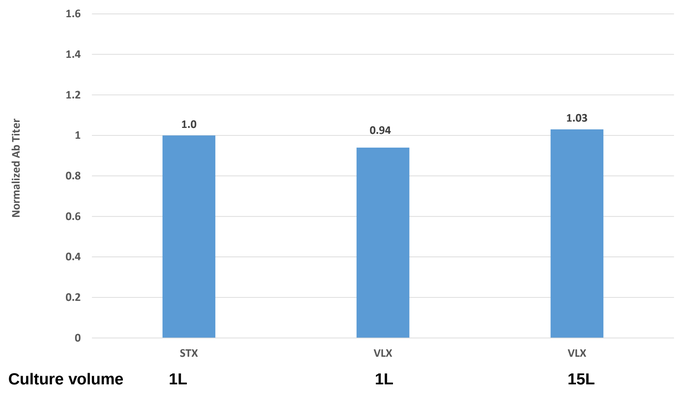

Figure 6. Scale up of CHO-S Cells from Small Scale to Large Scale Using the MaxCyte Platform. Two sets of 2E10 CHO-S cells were transfected by flow electroporation with an hIgG expression plasmid using the MaxCyte STX and VLX instruments. Following EP, cells were seeded into 1-L shake flasks. Another transfection was performed on the VLX with 2E11 cells, and those cells were seeded into a 15-L WAVE bag at the same density as the cells in the shake flasks. Relative titers in the three sets of cells measured two weeks post EP demonstrate reproducibility and scalability of the MaxCyte transfection process.

Summary

- MaxCyte offers a flow electroporation-based platform that is fully scalable from 5E5 cells to 2E11 cells, allowing for production of milligram to multi-gram quantities of proteins..
- MaxCyte transient transfection of CHO cells can produce secreted antibody titers **over 2.7 g/L** with optimization of post transfection culture conditions.
- A variety of CHO cell lines including CHO-K1, CHO EBNA, CHO-K1SV, can be transfected using the MaxCyte platform with resulting titers higher than chemical-based reagents.
- CHO cells transfected at small scale and at large scale show consistent cell viability, VCD, and titers.
- Production scale-up from the MaxCyte STX to the MaxCyte VLX is seamless – transfection performance is maintained without reoptimization.

Benefits of MaxCyte Flow Electroporation

- MaxCyte electroporation enables the use of any mammalian cell line, without requiring specialized constructs, media additives, or chemical reagents, allowing any cell line to be used.
- The ability to transfect a large number of cells (2E11) allows for both the generation of antibodies for screening along with clonal selection from the same cell population, saving time and resources and improving consistency.
- Electroporation and scale-up protocols are optimized and computer controlled, with reproducible results from day-to-day and operator-to-operator. There is no need for reoptimization of reagents or upstream/downstream scale changes, saving time and improving productivity by allowing more campaigns with the same amount of resources.

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