CELL-tainer® single use bioreactor supports the growth of a rat-anti-mouse IgG2a producing hybridoma

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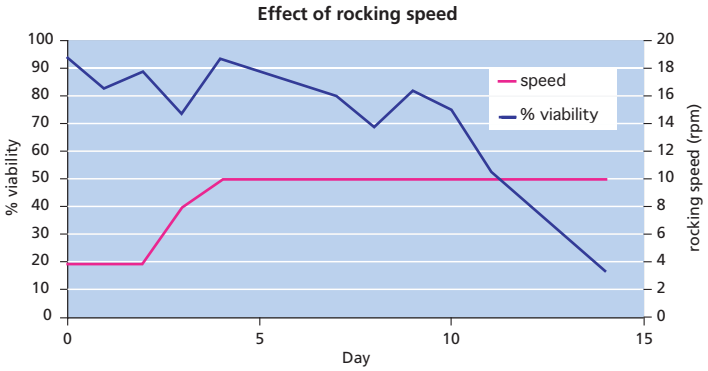
The CELL-tainer® single use bioreactor is using a 2-D rocking motion to improve mixing and gas-liquid mass transfer. In this study the suitability of this technology to support the growth of a hybridoma cell-line producing a rat-anti-mouse IgG2a antibody is proven. The productivity in the CELL-tainer® bioreactor is compared to conventional stationary T175 cm² culture flasks.

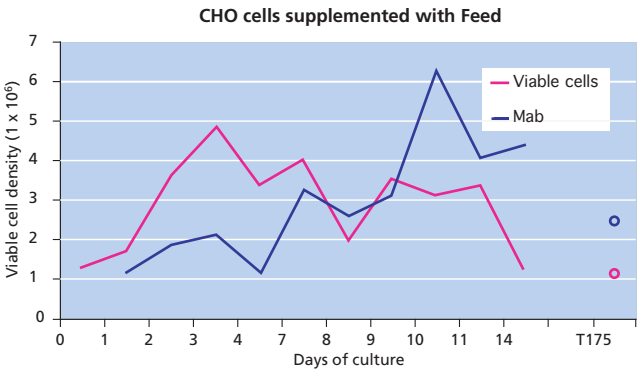
Materials and Methods

Prior to the start of the CELL-tainer® a pre-culture of hybridoma cells in roller-bottles have been performed in IMDM, 5% FCS and 50 µg/ml gentamycin. Cells were inoculated at cell density of 0.2 x 106 cells/ml in 2.5 L IMDM, 2% FCS and 50 µg/ml gentamycin. At day 4 of the culture cells were supplemented with 7.5 L IMDM, 2% FCS and 50 µg/ml gentamycin. Two days later 5 L of culture supernatant was harvested and 5 L of fresh IMDM, 2% FCS and 50 µg/ml gentamycin was provided to the culture again. For direct comparison the cells were also grown under stationary conditions in a T175 cm² culture flasks. Experiments were terminated when viability were < 50%.

Results

Cell growth and maximal cell densities were – in the first days –comparable under stationary culture conditions, in roller-bottles and in the CELL-tainer® with population-doubling times of 20-24 hours. Maximal cell densities in all culture systems were 0.5-0.7 x 106 viable cells/ml. At day 14 the cultures (stationary T175 and CELL-tainer®) were terminated due to low viability. The rocking speed in the CELLtainer was started at 4 rev/min, but gradually increased to 10 rev/min. During the 2 weeks of the experiment 15 L of supernatant was produced in the CELLtainer, while in the stationary flask 250 ml was produced in the same period. Final concentration of Mab was 60% higher in the CELL-tainer® supernatant compared to the stationary conditions





Conclusion

The CELL-tainer® provides a method to produce fast and efficient large batches of Mab from a rat-anti-mouse hybridoma and the 2-D motion does not harm the viability of the hybridoma cells. During the same period of time 60 times more supernatant was produced in the CELL-tainer® when compared to T175 cm² culture flasks. Since the Mab concentration is 60% higher in the CELL-tainer® supernatant, this run has delivered comparable amount of Mab than could have been produced in 60 x 1,6 = 96 culture flasks (T175cm²). The CELL-tainer® single bioreactor is a simple instrument to produce IgG antibodies using hybridoma cells in large quantities.

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