

CELLtainer technology supports the growth of a rat-anti-mouse IgG2a producing hybridoma

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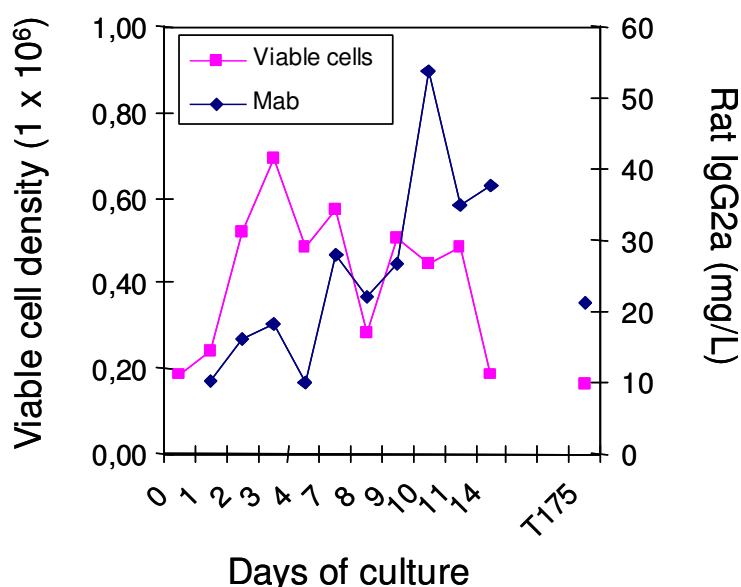
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The primary aim of the present study was to show that the CELLtainer technology is able to support the growth of a hybridoma cell-line producing a rat-anti-mouse IgG2a antibody. The secondary aim was to compare productivity in the CELLtainer to conventional stationary T175 cm² culture flasks.

Materials and Methods: Prior to the start of the culture in the CELLtainer, a pre-culture was performed of hybridoma cells in roller-bottles with medium IMDM, 5% FCS and 50 ug/ml gentamycin. The CELLtainer was inoculated at cell density of 0.2×10^6 cells/ml. At day 4 of the culture cells were supplemented with 7.5 L IMDM, 2% FCS and 50 ug/ml gentamycin. Two days later 5 L of culture supernatant was harvested and 5 L of fresh IMDM, 2% FCS and 50 ug/ml gentamycin was added to the culture. For direct comparison the cells also were grown under stationary conditions in T175 cm² culture flasks. Experiments were terminated when viability was < 50%.



days	%v	speed	
0	94	4	2,5 L IMDM + genta + 2% FCS
1	83	4	
2	89	4	
3	74	4	add 7,5 L IMDM
4	74	8	counts after adding IMDM
7	80	10	change medium -5L, add 5L
8	80		
8	69	10	
9	82	10	
10	75	10	
11	53	10	
14	16,5	10	
T175	31		

Results:

Cell growth and maximal cell densities were comparable under stationary culture conditions, both in roller-bottles and in the CELLtainer with population-doubling times of 20-24 hours. Maximal cell densities in all culture systems were $0.5-0.7 \times 10^6$ viable cells/ml. At day 14 the cultures (stationary T175 and CELLtainer) was terminated due to low viability. Rocking frequency in the CELLtainer was started at 4 rpm and increased to 8rpm subsequently to 10 rpm. During the 2 weeks of the experiment 15 L of supernatant was produced in the CELLtainer, while in the same period 250 ml was produced in the stationary flask. In the supernatant of the CELLtainer the final concentration of Mab was 60% higher compared to the concentration under stationary conditions.

Conclusion:

The CELLtainer provides a method to produce fast and efficiently large batches of Mab from a rat-anti-mouse hybridoma. During the same period of time 60 times more supernatant was produced in the CELLtainer when compared to T175 cm² culture flasks. Since the Mab concentration in the CELLtainer supernatant is 60% higher, this run has delivered an minimal the same amount of Mab that could have been produced in $60 \times 1,6 = 96$ culture flasks (T175cm²).