

Growth of HeLa Cells

Objective

HeLa is one of the most well-known cell lines. It has the capability of growing both in suspension and as anchorage dependent. This application shows a convenient method for continuous harvesting of large numbers of HeLa cells.

Culture conditions

Vessels: 50 ml spinners(Technie).

Microcarrier: 1 g/l CultiSpher-G prepared according to instructions.

Cell line: HeLa(Institute of Microbiology, University of Lund, Sweden)

Agitation speed: 45 RPM.

Media: DME supplemented with 10% FBS, penicillin G(100 U/ml) and streptomycin(100 µg/ml).

pH was controlled through CO₂ atmosphere for both media. Media volume was varied according to the following scheme; day 0: 30 ml, day 1-6: 50 ml and day 7-10: 60 ml.

Inoculation: Culture was inoculated at 100,000 cells/ml in a reduced volume(30 ml).

Results

After 10 days the density of cells growing on CultiSpher-G reached $5.2 \cdot 10^6$ cells/ml, which corresponds to a yield of $60 \cdot 10^8$ cells/g dry CultiSpher-G. In the exponential growth phase the cell doubling time was 19 hours. During the later stages of culture a large number of cells appeared in the suspension. Concentration of this population varied considerable, showing a maximum of $3.5 \cdot 10^6$ cells/ml. As 80% of the media was exchanged daily the majority of these cells were withdrawn from the culture.

Discussion

HeLa cells growing in suspension have a doubling time of 23 hours and the maximum cell densities is approximate $3 \cdot 4 \cdot 10^5$ cells/ml(Jacobson and Ryan: Tissue and Cell 14, 69-83, 1982).

During the last 5 days of culture an average of $70 \cdot 10^6$ HeLa cells were collected each day.

In order to produce this amount of cells with suspension culture it would have been necessary to have a culture volume of 400 ml and harvest 50% of the culture every day. As culture volume with CultiSpher-G was 60 ml, production of HeLa cells is more than 6 times as effective with CultiSpher-G than for suspension culture.

When anchorage-dependent cells are dividing they will have a more rounded morphology which makes them prone to detach from the microcarriers. It is very likely that the majority of collected cells are synchronized in their cell cycles and consist mostly of newly divided cells.

