

Thermo Scientific Nunc PETG Roller Bottles

Growth of Vero cells in 1.2X Thermo Scientific Nunc PETG Roller Bottles using 5% Fetal Bovine Serum

Vero cells (CRL 1587), derived from the kidney of the African green monkey, are a hypodiploid cell line that has been used in the production of viruses for vaccines and basic research including detection of viruses in meat¹, herpes², Dengue³, Norwalk, and Influenza A and B5. This cell line is highly susceptible to a large number of viruses making it ideally suited for viral research and virus production.

The ability of Thermo Scientific™ Nunc™ PETG Roller Bottles to support sustained Vero cell growth using 5% Fetal Bovine Serum (FBS) was evaluated by passing Vero cells in 1.2X PETG Nunc Roller Bottles and a competitive Polystyrene (PS) Roller Bottle four times.

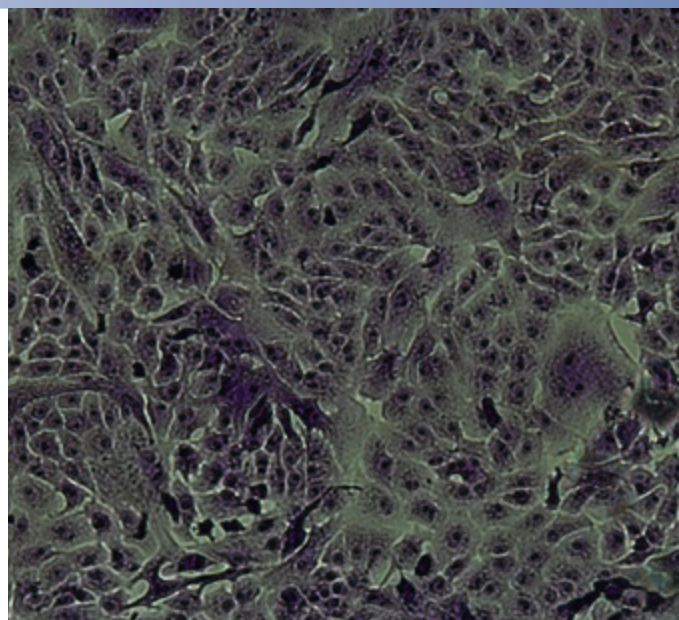
The results suggest

- Adaptation of the cells to PETG from PS flasks is not necessary to achieve high cell yields and viability.
- Higher cell densities (per cm²) and greater total cell yield were achieved on Nunc PETG Roller Bottles compared to a leading competitor's polystyrene roller bottles while cell viability approached 100%.
- Nunc PETG Roller Bottles may be a superior platform compared to polystyrene bottles for production of Vero cells when using 5% FBS in the growth medium.

Methods and Materials

Cell Line and Culture Conditions

Vero 76 cells, obtained from ATCC, were trypsinized (0.25% gamma irradiated porcine trypsin and seeded (n=5 per brand per passage). The 1.2X 1050 cm² PETG Nunc Roller Bottles were seeded at a density of 2.3×10^4 cells/cm². The competitive 850 cm² PS roller bottles were seeded at a density of 2.3×10^4 cells/cm². The 20% cell plant density was to accommodate the difference in surface growth area (1050 cm² vs. 850 cm²). Vero cells used were grown in DMEM/H containing 5% fetal bovine serum, 30 mg/mL Gentamicin sulfate (RXV) and 2 mm L-glutamine.



Vero Cell on a 1.2 PETG Nunc Roller Bottle

Passaging and Counting

Cells were passed every five days. On the fifth day, viable and total cell counts were performed on each of five bottles per brand. This process was repeated through passage 4.

Discussion

Two brands of roller bottles were compared for their ability to grow Vero cells. Nunc Roller Bottles, made from PETG and with a surface area of approximately 1050 cm², and competitive roller bottles, made from polystyrene, with a surface area of approximately 850 cm². PETG has an advantage over polystyrene in that it is stronger and more durable. As a result, the Roller Bottles are suitable for frozen applications such as cell release free/thaw methods and are far more resistant to breakage during normal usage or from accidental impact.

On the other hand, since most tissue culture flasks are polystyrene it was thought that cells may need to be acclimated to PETG when seeded from polystyrene flasks.

In order to address the issue of acclimation, cells were passed four times through both types of bottles while measuring cell yield, density, and viability. Cell Density (Fig. 1) was found to be comparable between PETG and PS bottles throughout the experiment. In spite of a non-significant decline at passage 2, by passage 4 the density of cells on PETG bottles actually exceeded that of PS bottles. Since the PETG bottles have greater surface area, this naturally translated to higher cell yields, especially in later passages.

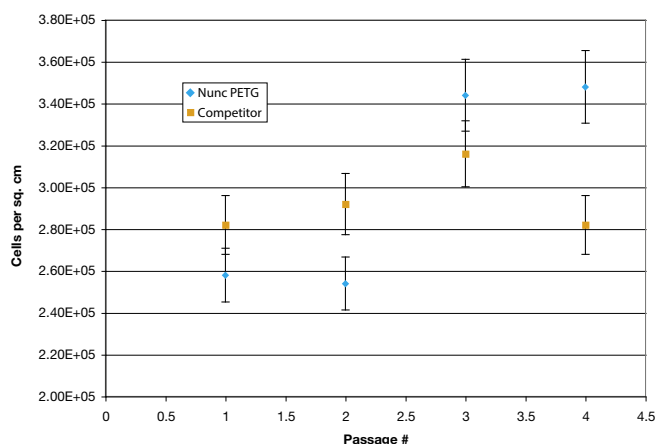


Fig. 1: Vero Cell Density (per cm²) Through 4 Passages

Conclusions

- Nunc PETG Roller Bottles produced higher cell density and yields and are therefore superior to PS bottles for the growth of Vero cells.
- No adaptation was required for Vero cells to grow successfully on PETG when transferred from a PS cell culture flask.
- Vero cells will grow successfully in reduced 5% FBS.

References

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