

# 4Cell<sup>®</sup> NutriVero<sup>™</sup> Flex 10 Medium

Next generation chemically defined medium designed to support the growth of Vero cells and virus production

## Benefits

Developed together with Intravacc, an R&D organization for translational vaccinology, this chemically defined, serum-free, animal component-free medium will give you consistent results and maximum control over your virus production process:

- **Production Efficacy:** High cell growth and virus titer in a chemically defined format
- **Operational Efficiency:** Simplify your downstream purification and filtration processes while increasing virus productivity
- **Safety & Regulatory-friendly:** Reduced process variability and contamination risks for optimal safety

## Product Information

4Cell<sup>®</sup> NutriVero<sup>™</sup> Flex 10 is a chemically defined, serum-free, animal component-free medium, newly developed to support Vero cell growth in 2D monolayers as well as in 3D microcarriers suspension culture systems and optimized for the production of viruses.

The chemically defined 4Cell<sup>®</sup> NutriVero<sup>™</sup> Flex 10 is a robust medium which contains solely recombinant components and does not contain any plant extract (hydrolysates), therefore providing consistent results.

## Applications

Isolated from the kidney of the African green monkey by Yasumura and Kawakita in Japan (1962), the Vero cell line is used for various purposes, most importantly for the production of cell culture-based viral vaccines. Reasons for the extensive use of the Vero cell line are the consistent high viral yields and relatively easy adaptation for growth in bioreactors on microcarriers, thus allowing greater vaccine purity as well as quantity.

## Features of the Standard Product



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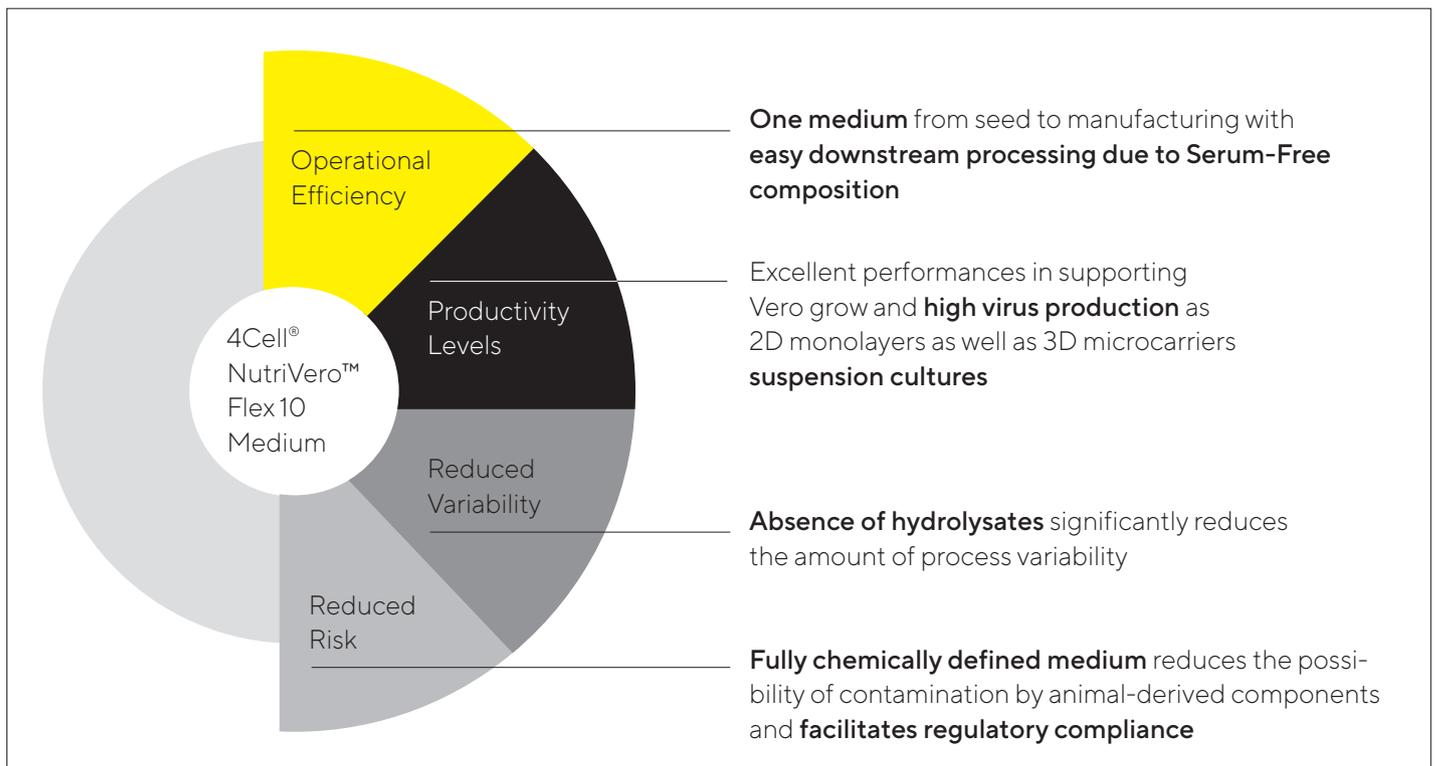
- CD** **Chemically Defined:** The exact concentration and size of every component is known
- NAO** **Non-Animal Origin:** The formulation is entirely made from non-animal -human origin components
- RP** **Recombinant Protein:** The formulation contains very low concentration of recombinant factors
- FFM** For research or for further manufacturing use
-  Product available in liquid format

# Technical Data

## Specifications

Media Type	Chemically defined medium: does <b>not</b> contain serum or animal origin components
Easy-to-use	Available in liquid or as powder with supplement
Cell Line	Developed for adherent Vero cells in monolayers and microcarriers
Storage Condition	2 – 8°C, protect from light

## Your Benefits at a Glance



4Cell® NutriVero™ Flex 10 Medium performance have been evaluated on Vero cells' growth in adherent 2D monolayers (Fig. 1A) and 3D microcarriers beads (Fig. 1B). In both systems 4Cell® NutriVero™ Flex 10 Medium showed equivalent performance as reference medium containing undefined extracts (hydrolysates).

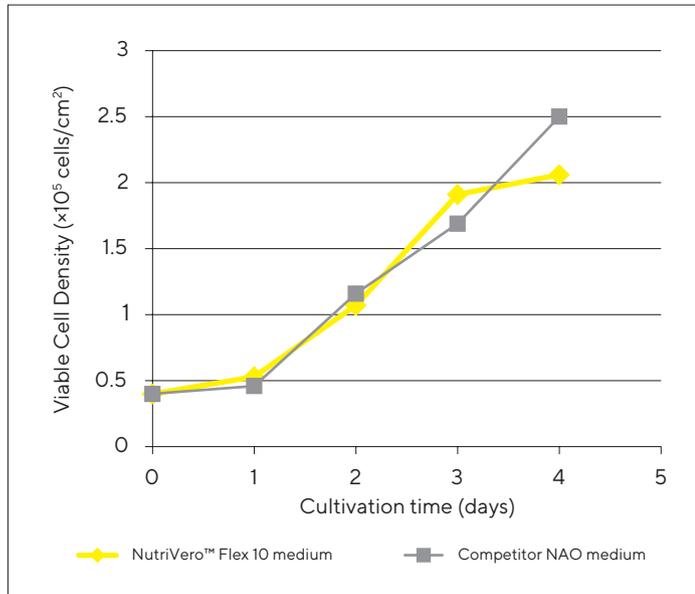


Figure 1A. 4Cell® NutriVero™ Flex 10 performance in Vero cell 2D culturing system.

Vero cells were seeded in T25 flasks at a cell density of 40,000 cells/cm<sup>2</sup> and incubated at 37°C in a humidified atmosphere and 5% CO<sub>2</sub> with 4Cell® NutriVero™ Flex 10 or reference medium.

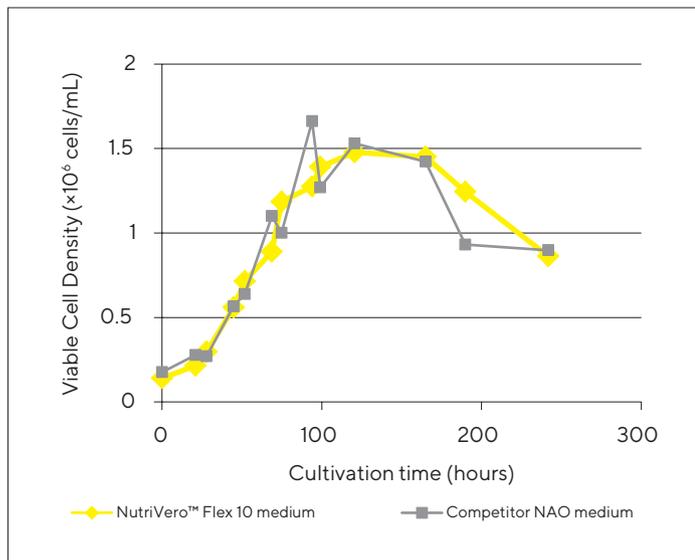


Figure 1B. Vero cell growth density in a 3D culture system.

Two parallel bioreactors were filled up to 2 L of working volume of chemically defined 4Cell® NutriVero™ Flex 10 and undefined reference medium. Stirring speed was set between 70 and 130 rpm, temperature set to 37°C and pH controlled to 7.2. The bioreactors were seeded with  $0.15 \times 10^9$  cells/L and 3 g/L of microcarriers

Virus quantification was performed by calculating the TCID<sub>50</sub> (50% Tissue Culture Infective Dose) index. 4Cell® NutriVero™ Flex 10 Medium showed high abilities in supporting Vero infection and virus productivity in 2D monolayers (Fig. 2A) and suspension in microcarriers beads (Fig. 2B) compared to reference medium containing undefined extracts (hydrolysates).

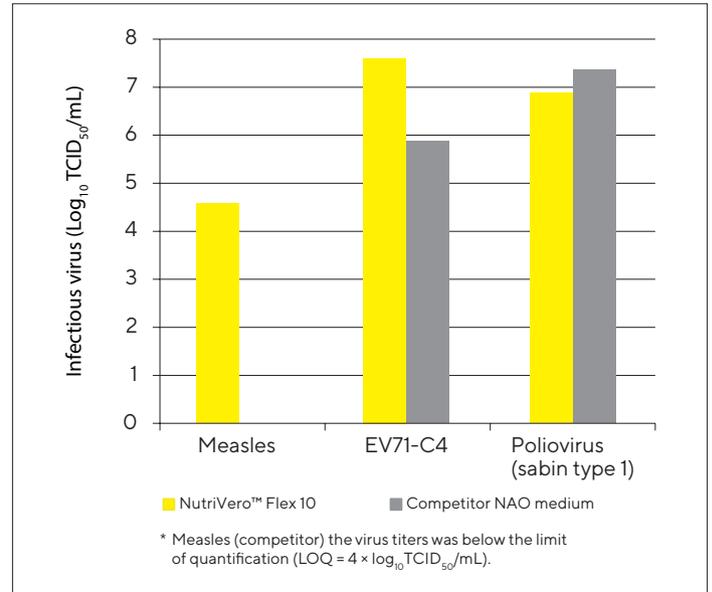


Figure 2A. Initial viral production capacity of defined 4Cell® NutriVero™ Flex 10 Medium.

Vero cells were seeded in 6-well plates at a cell density of 30,000 cells/cm<sup>2</sup> of culture, the cultures were infected with various viruses: Measles, Sabin poliovirus type 1 and EV71-C4. Following 7 days the cultures showed a positive CPE and the supernatant was harvested and analyzed for the amount of infectious particles by means of a virus titration procedure.

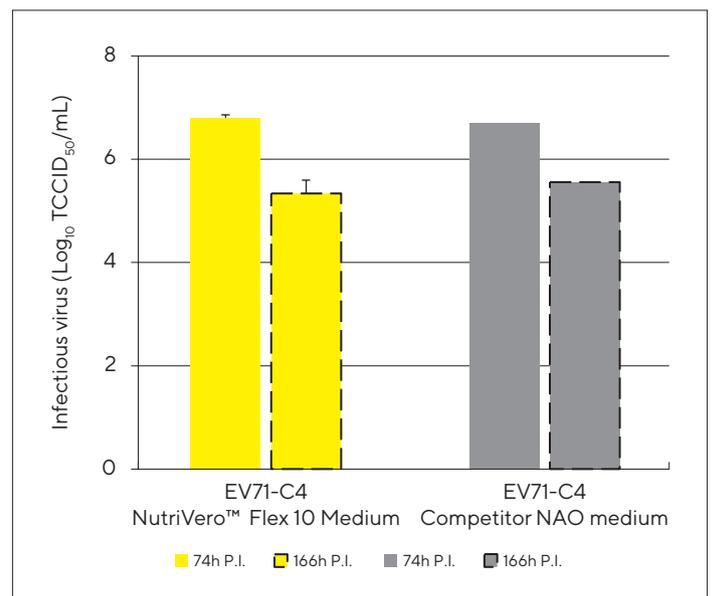


Figure 2B. Enterovirus 71-C4 virus production in a 3D microcarriers culture system.

Vero cells were cultured in 1L bioreactors at  $0.12 \pm 0.03 \times 10^6$  cells/ml for 3 days, up to a concentration of  $0.9 \pm 0.2 \times 10^6$  cells/ml. At 66 hours post seeding, the cells were infected with EV71-C4 at a titer of 7.55 log<sub>10</sub> TCID<sub>50</sub>/ml. Samples were taken at indicated time points post infection, and analyzed for virus titer.

# Ordering Information

Description	Form*	Package	Volume	Order Code
4Cell® NutriVero™ Flex 10 Medium	Liquid	Bottle	0.5 L	CFV3FA4009
4Cell® NutriVero™ Flex 10 Medium	Liquid	Bottle	1 L	CFV3FA4010

\* Other sizes are available on request

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