



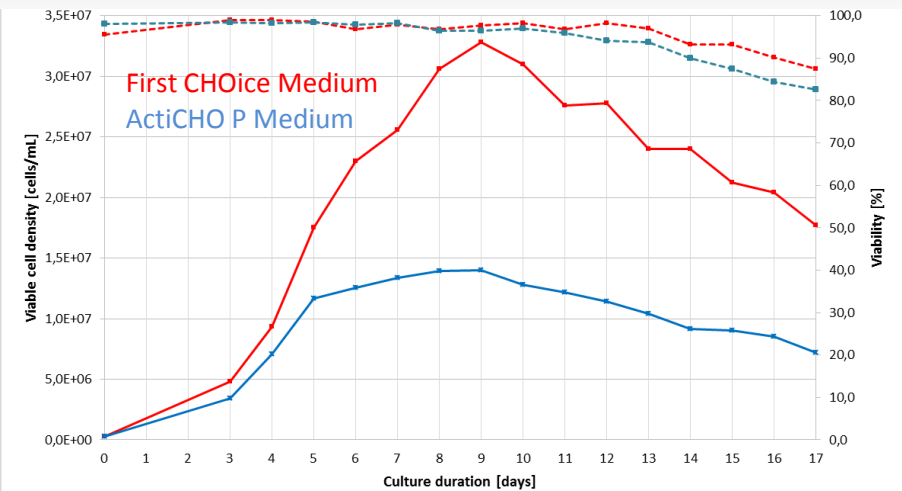
Biosimilar Development Services & Products

The start has to be right!

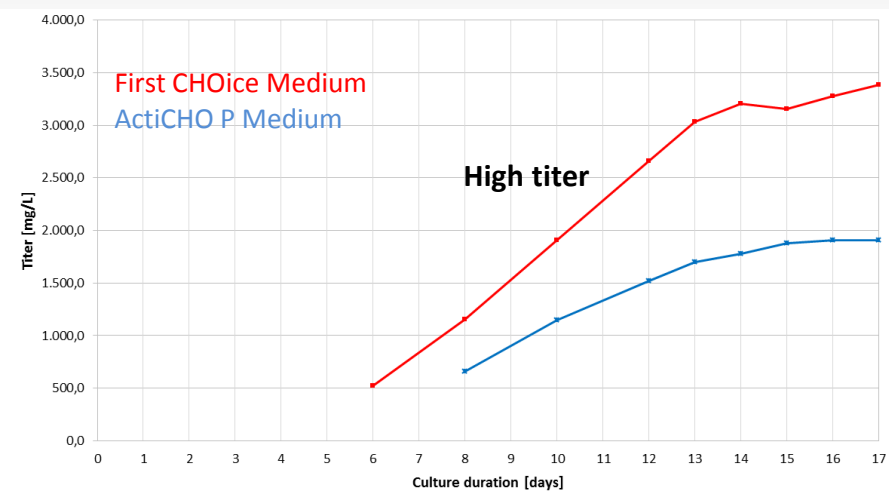
Presentation of First CHOice Medium

- Very good performance for CHO cells
- For high titer and good profile

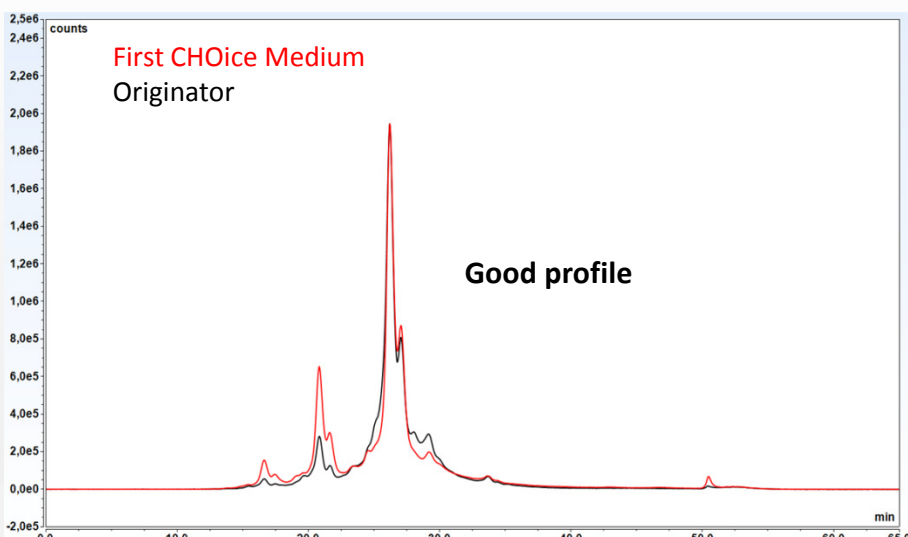
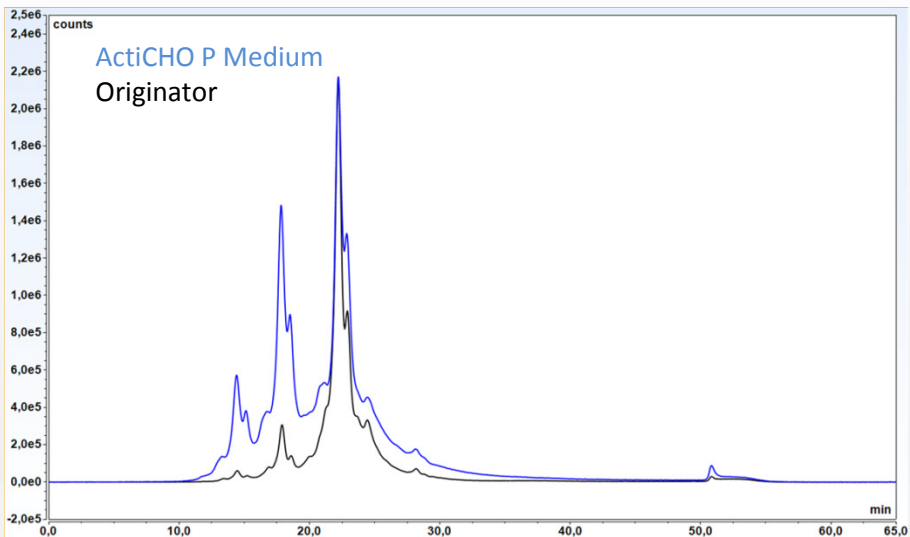
Biosimilar 1, stable transfected CHO cells



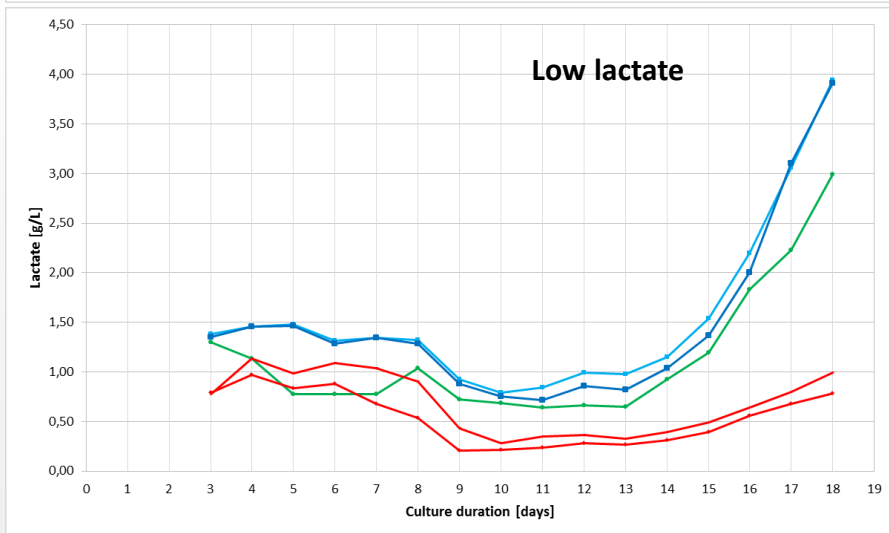
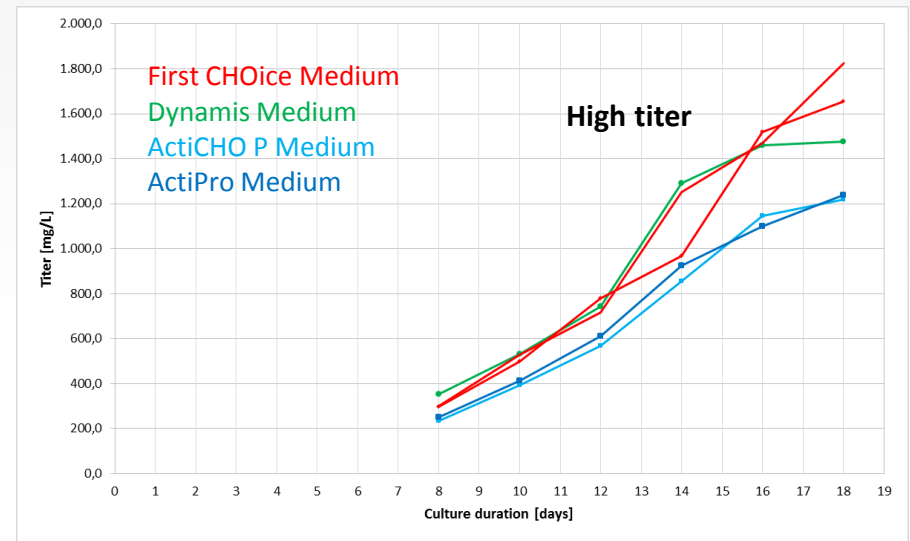
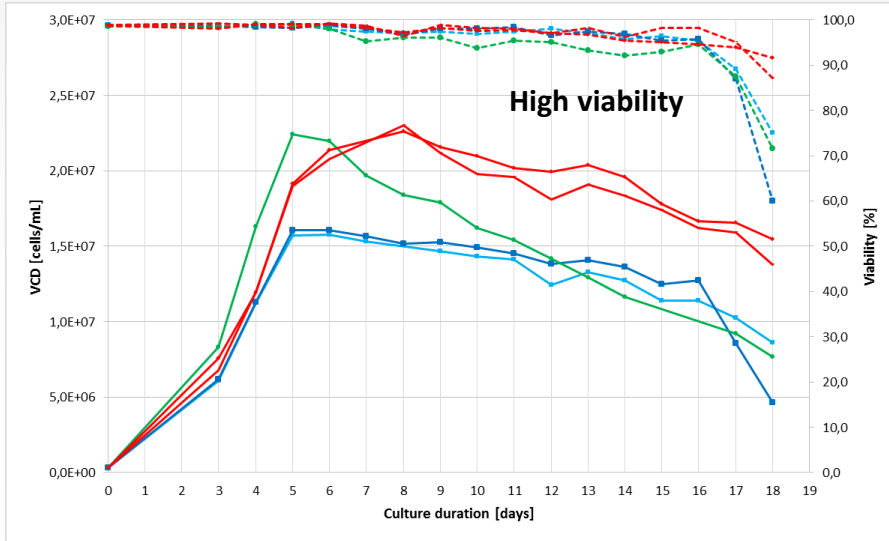
Fed-Batch in 5 L stirred bioreactor



Charge variant analysis (SAX)



Biosimilar 2, stable transfected CHO cells



First CHOice® Medium - User Guide

Last revision: 01 June 2017

First CHOice® Medium is a cultivation and production medium optimized for CHO platform technology. The completely chemically defined and protein free medium has been developed in close cooperation with customers using CHO-S, DG44 and CHO-K1 cell lines to boost cell growth and to increase protein expression and quality in continuous bioprocessing. The medium enhances performance by minimising the production of stress related metabolites such as lactate and ammonia and thereby conserves high viabilities in all process phases.



Storage conditions: First CHOice® Medium is best stored at 2° to 8°C and protected from light.

Supplementation: Before use, First CHOice® Medium needs to be supplemented with L-Glutamine. It is recommended to use 6 mM final concentration as a standard condition. For GS-selection based cells, lower concentrations may prove sufficient. First CHOice® Medium has been designed as a ready-to-use medium to meet the needs of most CHO cells and support high growth and productivity without commonly used supplements (e.g. serum, insulin, and transferrin).

Adaptation to First CHOice® Medium and routine cultivation: For most CHO clones, it is possible to transfer cells into First CHOice® Medium directly after thawing. Time consuming stepwise adaptation is usually not required. Instead, cells habitually recover from thawing without a significant change in viability and can be used for batch and fed batch experiments after two to three splits.

For adaptation of your protocols, the following recommendations are listed below:

Standard protocol for cell thaw: Thaw vials at 37 °C and transfer the content of the vial immediately into a conical tube with 9 mL pre-warmed First CHOice® Medium. After centrifugation at 200-300 g for 3-5 min, the cell pellet should be resuspended in the required working volume of First CHOice® Medium supplemented with 6 mM L-glutamine to achieve a cell density of $3\text{-}5 \cdot 10^5$ cells/mL.

Standard protocol for routine cultivation: After thawing cultivate cells at 37°C in a humidified atmosphere containing 5-10% CO₂ in vented Erlenmeyer flasks (150 rpm, 50 mm orbit). Maintain cells always in the logarithmic growth phase. Therefore, split cells depended on cell growth every 2-4 days into fresh First CHOice® Medium supplemented with 6 mM L-glutamine. The recommended seeding density is $3\text{-}5 \cdot 10^5$ cells/mL.

Standard protocol for freezing of cells: To freeze $1 \cdot 10^7$ cells/mL in 1 mL aliquots, transfer the required volume of cell suspension into conical tubes and centrifuge at 200-300 g for 3-5 min. Remove the supernatant and resuspend the cell pellet in pre-cooled (2-8°C) First CHOice Medium supplemented with 10% DMSO. Immediately distribute 1 mL/vial of the suspension into cryogenic vials and transfer them into a suitable freezing container with a cooling rate of about 1°C/min. Place the freezing container for 24 hours at -80°C. Afterwards transfer vials into a nitrogen tank for long term storage.

Standard protocol for fed batch: Transfer cells into vented Erlenmeyer flasks filled with First CHOice® Medium with 6 mM L-glutamine. Split cells depended on cell growth every 2-4 days into fresh medium. The recommended seeding density is $3\text{-}5 \cdot 10^5$ cells/mL. Cultivate these cultures at 37°C in a humidified atmosphere containing 5-10% CO₂. Repeat this procedure until cells are fully adapted to the First CHOice® Medium.

Afterwards, inoculate vented Erlenmeyer flasks or bioreactors with a seeding density of $3\text{-}5 \cdot 10^5$ cells/mL. Cultivate these cultures for 72 hours. Afterwards add every day the recommended feed portion (for further information see quick guide below). In order to avoid glucose limitations, it is recommended to feed an extra glucose solution to ensure that the glucose concentration remains above 2 g/L throughout the fed batch process.

Quick guide:

Cultivation in Erlenmeyer flasks:

37°C, 8% CO₂, agitation speed 150 rpm, orbit 50 mm

Cultivation in Stirred bioreactors:

37°C, pH 7.1, 40-60% pO₂

Feeding strategy:

It is recommended to use a fixed feeding rate of 2-3% Acti CHO Feed A with 0.2-0.3% Acti CHO Feed B or 2-3% Cell boost 7a with 0.2-0.3% Cell boost 7b.

In order to avoid glucose limitations, it is recommended to feed glucose solution to ensure that the concentration remains above 2 g/L throughout the process.

Coming soon:**First CHOice® Feeds** will be commercially available very soon!**Customer feedback:**

We are looking forward to your reviews of First CHOice® Medium. It is appreciated very much to see your data and get feedback on how First CHOice® Medium performs with cell lines and applications other than CHO platform technology.

Exclusive sourcing channel:

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