GENAXXON bioscience
fon: +49(0)731 – 3608 123
fax: +49(0)731 – 3608 962
E-Mail: info@genaxxon.com

Serum Replacement DC

Defined Serum Substitute for Adherent Cells

	Cat#	C4367.0050	C4367.0100	C4367.0500
Component				
Serum Replacement DC chemically defined serum substitute for adherent and non-adherent cells		50mL	100mL	500mL

Product Description
Serum Replacement DC is a ready-to-use, chemically defined serum substitute for the cultivation of adherent and non-adherent cells under serum-free conditions or to significantly reduce the amount of serum in cell culture. The fully synthetic serum replacement DC only contains chemically defined components. It supports the growth of many cell types in an optimum manner without any extra handling compared to serum.

Composition
Serum Replacement DC contains recombinant proteins, lipids, salts, amino acids, trace elements, hormones and a special substrate release system for an optimized formulation. It contains no growth factors, nor undefined hydrolysates or peptones.

Special advantages
Serum Replacement DC is designed to replace or to reduce serum in the cell culture in a very simple manner. In most cases there is no need to change the basal medium. As Serum Replacement DC is fully defined and contains no peptones or hydrolysates, lot testing is no more necessary.

- It also allows high reproducibility and a simplified downstream process.
- Serum Replacement DC contains no growth factors and enables defined proliferation and differentiation of stem cells.
- Characterization studies of growth factors will obtain more reproducible and clearer results.

Suitability
Serum Replacement DC is suitable for the cultivation of a variety of adherent and non-adherent cells under serum-free culture conditions (please see figure 1) or to reduce the necessary FBS amount in cell culture.

Effect of Serum Replacement DC supplement on different cell lines

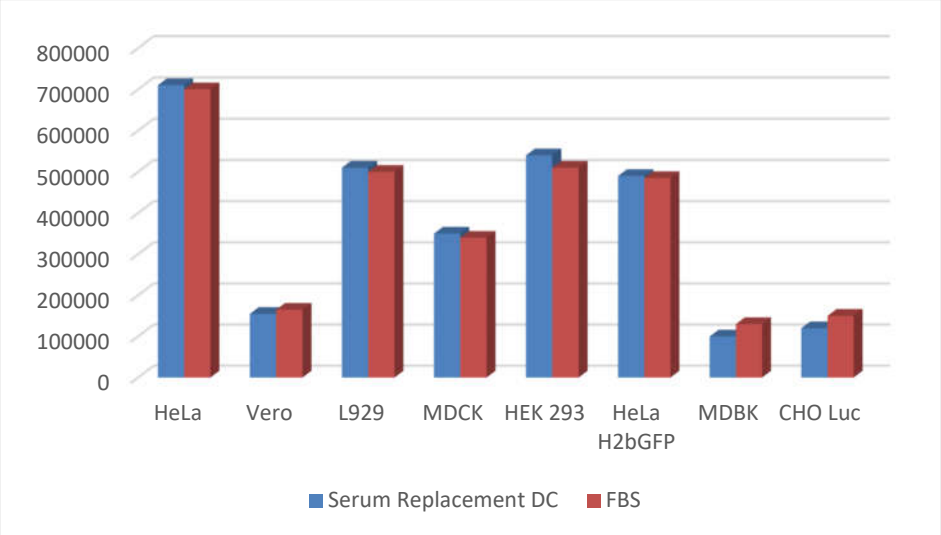

Fig. 1: Efficiency and growth stimulation of Serum Replacement DC compared to FBS (10% each in DMEM) for different cell types. Tests were performed at 37°C, 5% CO2. Cells were seeded with 10000 cells/cm².

GENAXXON bioscience GmbH
Söflinger Str. 70 ● 89077 Ulm ● Geschäftsführer: Dr. Norbert Tröndle
Amtsgericht: 89014 Ulm ● HRB 641623
U.St.-Id.Nr. DE 220 603 767 ● Deutsche St.-Nr. 88002/33658

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Storage conditions
Storage: -20°C (in the dark)
Stability: 2 years from date of production

Special advantages

Serum Replacement DC is designed to replace or to reduce serum in cell culture in a very simple manner. Compared to other serum replacements, it contains only chemically defined components. Due to its defined composition, lot testing is not necessary anymore. It also allows high reproducibility and simplified downstream processing.

Serum Replacement DC contains no growth factors and enables defined proliferation and differentiation of stem cells. Characterization studies of growth factors will obtain more reproducible and clearer results.

Serum Replacement DC is also useful to develop sensitive cell-based in vitro tests and coculture procedures. For cell lines which require specific growth factors these should be added in a concentration as previously used.

Instructions for use:
Serum Replacement DC can be stored and used in the same manner as serum.

- **Thaw Serum Replacement DC** at maximum 37°C. **Please avoid repeated freeze-thaw cycles!**
- **To replace serum:** Use the same basal medium and the same concentration of Serum Replacement DC as FBS. The performance can be further improved by optimizing the concentration of Serum Replacement DC or modifying/changing the basal medium (As a basal medium, standard media such as RPMI 1640, DMEM (high or low glucose), DMEM/F12, etc. can be used. Make sure that L-Glutamine is present in sufficient quantity (supplement L-Glutamine as needed)).
- **To reduce serum concentration:** Use the same basal medium and add the same amount of Serum Replacement DC as the reduced amount of serum, until the minimal necessary concentration of FBS is found (1 to 2.5% in most cases). The performance can be further improved by optimizing the concentration of Serum Replacement DC or modifying/changing the basal medium (also see adaptation instruction).
- **Recommended inoculation** cell density: 50000 – 100000 cells/cm^2 for non-adherent cells or 5000 to 20000 cells/cm^2 for adherent cells.
- If working with adherent cells: **Solve cells as usual** from the cell culture vessel (e.g., Trypsin 0.25%/EDTA 0.02% in PBS, or Accutase®). Once the cells have become round and detach from the surface inactivate trypsin with trypsin inhibitor: Simply resuspend cells in about 1mL trypsin inhibitor solution for every mL of trypsin solution used for dissociation**. Note** that Accutase® does not need to be inhibited.

Depending on the cell type, some differences in morphology or proliferation rate may be observed with varying standard media. Most applications were performed with RPMI 1640 for non-adherent cells and DMEM and DMEM/F12 for adherent cells. With these combinations very good growth stimulation was achieved in a range of 5-15% Serum Replacement DC.

Please note: For more demanding cells (e.g., primary cells) an adaptation to Serum Replacement DC may be necessary.

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Adaptation instructions for Serum Replacement DC if 10% FBS was used in the original protocol
Precondition for a successful transition are vital cells (trypan blue exclusion staining), which should be harvested in the logarithmic growth phase.

Step 1: 7.5% FBS + 2.5% Serum Replacement DC
- Seed cells at 5 x 10^4 – 10 x 10^4 cells/mL (non-adherent cells) or at 5 x 10^3 – 20 x 10^3 cells/cm^2 (adherent cells).
- Observe cells under a microscope, at about 90 % confluence passage the cells for another 2-3 passages.

If normal growth is obtained transfer cells into:

Step 2: 5% FBS + 5% Serum Replacement DC
- Seed cells at 5 x 10^4 – 10 x 10^4 cells/mL (non-adherent cells) or at 5 x 10^3 – 20 x 10^3 cells/cm^2 (adherent cells).
- Observe cells under a microscope, at about 90% confluence passage the cells for another 2-3 passages.

If normal growth is obtained transfer cells into:

Step 3: 2.5% FBS + 7.5% Serum Replacement DC
- Seed cells at 5 x 10^4– 10 x 10^4 cells/mL (non-adherent cells) or at 5 x 10^3 – 20 x 10^3 cells/cm^2 (adherent cells).
- Observe cells under a microscope, at about 90% confluence passage the cells for another 2-3 passages.

If normal growth is obtained transfer cells into:

Step 4: 1% FBS + 9% Serum Replacement DC
- Seed cells at 5 x 10^4 – 10 x 10^4 cells/mL (non-adherent cells) or at 5 x 10^3 – 20 x 10^3 cells/cm^2 (adherent cells).
- Observe cells under a microscope, at about 90% confluence passage the cells for another 2-3 passages.

If normal growth is obtained transfer cells into:

Step 5: 10% Serum Replacement DC
- Seed cells at 5 x 10^4 – 10 x 10^4 cells/mL (non-adherent cells) or at 5 x 10^3 – 20 x 10^3 cells/cm^2 (adherent cells).
- Observe cells under a microscope.

For some cells an adaptation to serum-free conditions is difficult to reach or even impossible. The following measures may help to facilitate a successful adaptation:

- Reseeding with a higher cell amount (about 2x to 4x of the usual cell density).
- Addition of growth factors (if known, which factors have a positive effect on the relevant cells).
- Coating the culture dishes or flasks with attachment factors (e.g., fibronectin, laminin, collagen, gelatine, etc.).
- Change the basal medium. **Note:** A change of the basal medium to a richer or more complex formulation may be all that is needed to achieve growth in serum free condition.