

Product Instruction

SF003 MDCK Serum-free Medium

Product Type: SF003

Product Description

MDCK serum-free medium is a serum-free, non animal-derived base medium with clear chemical composition, which is independently developed, researched and produced by Shanghai BioEngine Sci-Tech Co., Ltd. This medium is suitable for the high-efficiency expression of the products under the MDCK suspension culture process.

Product Formula

The intellectual property rights of MDCK formula are owned by Shanghai BioEngine Sci-Tech Co., Ltd.

Product Ingredient

The medium contains carbohydrates, amino acids, vitamins, metal ions and other nutritional components.

This product does not contain components of animal origin, genetically modified plant origin or raw material with mad cow virus origin.

Product Preservation

- Store in a dark environment at 2-8°C.
- This product is vulnerable to water damage. Please use immediately after opening. If it needs to be stored, please sealed by heat sealing and sealing clips, avoiding damp and being ineffective.
- Do not recommend to use, when the product is beyond expiration date.

Instruction for the preparation of MDCK medium

Do the preparation of MDCK medium as per the one Table 1 shows

Component	Concentration
Dry powder of MDCK medium	27.21g/L

Sodium bicarbonate 2.00 g/L

Table 1 Formula table of MDCK medium

(1) Weigh the final medium preparation volume of water into the medium preparation container. When preparing, ultrapure water or water for injection and above standard water should be used, and the water temperature should be controlled at 28-32°C. Turn on the mixing system of the medium preparation container, stir thoroughly, and avoid the generation of air bubbles during stirring.

(2) Accurately weigh 27.21 g/L dry powder of MDCK medium, and add them into the preparation container of (1), and stir thoroughly for 18-22 min.

(3) Add 5mol/l sodium hydroxide solution according to the dosage of 0.25 ± 0.01 g/L. (if 1L is prepared, the addition amount should be 1.25ml) to the above solution, and stir thoroughly for 20 ± 2 min.

(4) Slowly add sodium bicarbonate powder to the above solution according to the dosage of 2.00 ± 0.05 g/L, and stir thoroughly for 18-22 min.

(5) It is recommended to use a pulse pump or compressed air (3-15 psi) to sterile filter the MDCK medium solution through a sterile filter membrane with 0.22 μm pore size.

(6) The prepared medium liquid should be stored in a dark environment at 2~8°C.

(7) The reference parameters of product

Indicator	Reference Standard
Product initial pH value	4.50-4.90
Osmolality	300-350 mOsm/Kg
Product turbidity	<4.00NTU

Notes:

(1) The above units of "g/L" are volume concentration (solute weight/solution volume).

(2) The above preparation parameters (such as stirring time, etc.) are for the reference of small-scale preparation in research and development. When in production, please set appropriate preparation parameters according to the stirring capacity of the preparation vessel.

(3) The product belongs to carbon dioxide buffer system. The product final pH value may rise when vigorous stirring or long-time stirring, which is a normal phenomenon and does not affect the use of the product.

Medium usage

Subculture

- MDCK cells that have been suspended in other serum-free medium can be directly replaced with Xeno-S001S medium.

- The seeding density should be controlled at $0.8-1.1 \times 10^6$ cells/ml during serum-free suspension subculture, and subculture should be carried out every 48 hours.
- Use the shake flask with a vented cap and place it in a 37°C, 5% CO₂ environment. The recommended shaker speed is 110-130 rpm.

Cryopreservation

Select the cells that are in good condition in the logarithmic growth phase for cryopreservation. The cryopreservation density is $2.5-3.5 \times 10^7$ cells/ml/tube. The ratio of cryopreservation solution is 93% fresh medium + 7% DMSO. Centrifuge 175 g cells for 5 min, discard the supernatant, resuspend the mixed freezing solution, and dispense the cells into freezing tube, 1 ml/vial. Then, place them in a programmed cooling box at -80°C overnight, and transfer to liquid nitrogen save

Recovery

Rotate the freezing tubes in the same direction in a 37°C water bath to quickly melt the cryopreservation solution, and take them out to a clean bench when only small ice crystals remain. Add 10 ml of culture medium and 175 g cell to the centrifuge tube, centrifuge for 5 min, and wash away the DMSO. Use 20-30 ml of medium to resuspend the cells into a 125 ml shake flask with a vented cap, and control the seeding density at $0.8-1.1 \times 10^6$ cells/ml.

