Product Instruction

MDBK Cell Serum-free Medium

Product Type: Bofit-S001S

Product Description

Bofit-S001S is a serum-free medium developed by Shanghai BioEngine Sci-Tech Co., Ltd., which is designed based on the growth and metabolism characteristics of MDBK cells. This medium is suitable for high-density suspension culture of MDBK and high-efficiency production of bovine viruses.

Product Formula

The intellectual property rights of MDBK cell serum-free medium 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℃.
- The product is highly hygroscopic, so it should be used immediately after opening. If it is necessary to keep it, it is recommended to use heat sealing, sealing clips or other means to strictly seal the opening. Keep it away from moisture.
- It is not recommended to use beyond the shelf life.

Reconstitution of Bofit-S001S powder medium

Reconstitute of MDBK cell serum-free medium as Table 1 shows.

Component	Concentration
Bofit-S001S powder medium	23.35 g/L
Sodium bicarbonate	2.00 g/L

Table 1 Preparation of MDBK cell serum-free medium

(1) Add water at 100% of final preparation volume into the medium preparation container. Use ultrapure water, water for injection or other cell culture-grade water, and the water temperature should be controlled at 20-30℃.

(2) Turn on the mixing system to stir the water. Avoid air bubbles during stirring.

(3) Add 23.35 g/L of Bofit-S001S powder medium. Mix for 20-30 min or until dissolved.

(4) Slowly add sodium hydroxide particles or solution; adjust the pH value to 6.0-6.5. Mix for 10-20 min. The recommended addition amount of sodium hydroxide is 0.24 g/L.

(5) Add 2.00 g/L sodium bicarbonate, mix for 10-20 min. Check pH, the expected pH value is 7.0-7.4.

(6) Sterile filter into desired container using a 0.22 μm sterile filter.

(7) The prepared medium liquid should be stored in a dark environment at 2-8℃. It is recommended to be used in one month.

General culture recommendations

Subculture

- The recommended seeding density is 0.8-1.2×10^6 cells/ml for suspension culture. Subculture cells every 48 hours.
- Incubate the cells at 37℃ in a suitable incubator with 5% CO_2 in air atmosphere. Use shake flasks with vent cap. And the recommended shaker speed is 110-130 rpm.

Cryopreservation

Prepare the desired quantity of cells, harvesting in mid-log phase of growth with good condition. The recommended cryopreservation density is 2.5-3.5×10^7 cells/ml/vial. And the cryopreservation medium is prepared as 93% (v/v) fresh medium supplemented with 7% (v/v) DMSO. Centrifuge cells at 190× g for 5 min and discard the supernatant. Resuspend pellets with cryopreservation medium and dispense aliquots of this suspension into cryovials with 1 ml/vial.

Achieve cryopreservation in an automated controlled rate freezing container and keep it in -80℃ freezer overnight. Transfer frozen cells to liquid nitrogen for long-term storage.

Recovery

Thaw the vial by gentle agitation in a 37ºC water bath. This process should be rapid (approximately 2 minutes). Remove the vial from the water bath as soon as the contents are thawed. All the operations from this point on should be carried out under strict aseptic conditions. Transfer the vial contents to a centrifuge tube with 10 ml of growth medium and centrifuge cells at 190× g for 5 min to wash away DMSO. Discard the supernatant and resuspend the pellets with 20-30 ml of growth medium into a 125 ml shake flask. The recommended seeding density is 0.8-1.2×10^6 cells/ml.