

## Hybridoma Serum-Free Medium

Cat.No.: SF3001

Size: 500mL

### Applicable cell lines

Hybridoma serum-free medium is a serum-free product developed by Pricella's R&D team. It is suitable for the high-density growth and stable protein expression of hybridoma and myeloma cells.

### Medium characteristics

1. This product does not contain serum and contains small amounts of hydrolysates, proteins, and growth factor components.
2. The medium contains L-glutamine and HT.

### Culture conditions

1. The culture temperature is  $37 \pm 0.5$  °C, and the shaker speed is 100-120 rpm (50 mm).
2. Under the condition of non-automatic pH control, it is recommended to control the CO<sub>2</sub> concentration at 5%.
3. Process conditions such as pH, DO and temperature can be set according to process development results or platform process parameters.

### Culture medium adaptation

1. Direct adaptation: In the initial culture stage, the cells are recommended to be inoculated at the density of  $0.75 \times 10^6$  cells/mL, and the original medium is directly replaced with Hybridoma serum-free medium for cell culture. After the cells have been cultured for 2-3 generations and the cell growth is stable, subsequent experiments could be carried out. When hybridoma cells grow stably, they are seeded at a density of  $0.75 \times 10^6$  cells/mL and the density can reach  $3-5 \times 10^6$  cells/mL after 48 hours of culture.
2. Indirect adaptation: hybridoma serum-free culture medium and cell original culture medium are mixed at a ratio of 1:1 to culture cells, and continuously passage for 2-3 generations. After the cells grow stably, the medium can be replaced with hybridoma serum-free medium and the cells continuously passage for 2-3 generations. Follow-up experiments can be carried out after the cells grow stably. When hybridoma cells grow stably, they are seeded at a density of  $0.75 \times 10^6$  cells/mL and the density can reach  $3-5 \times 10^6$

cells/mL after 48 hours of culture.

## Cell cryopreservation

1. Use the freezing medium made from this product for cell freezing. Prepare cell cryopreservation solution:  
The recommended ratio is 70% culture medium + 20% FBS (it is recommended to use high quality fetal bovine serum) +10% DMSO. After the cryopreservation solution is prepared, place it in a 4°C refrigerator to pre-cool (When preparing the cryopreservation solution, add the culture medium first, followed by the serum or DMSO. This helps prevent the DMSO concentration from becoming too high, which could otherwise lead to denaturation of serum components.)
3. Cell freezing density is recommended to be  $1.5\text{-}2.0 \times 10^7$  cells/mL, with  $2.0 \times 10^7$  cells/mL being preferred.

## Storage

Store in 2-8 °C; protect from light.

Shelf life: 12 months

## Precautions

1. Depending on the specific conditions of the cells, choose whether to adapt the culture medium directly or indirectly.
2. To ensure stable cell growth, dilute passage is performed when the cell expansion ratio is  $\geq 1:7$ , and centrifugal passage is performed when the cell expansion ratio is  $< 1:7$ .
3. Dilute passage: Calculate the required cell suspension during passage according to the cell density, remove excess cell suspension, and replenish with fresh culture medium to reach the desired culture volume.
4. Centrifugal passage: Calculate the cell suspension required for passage according to the cell density, centrifuge at  $100\times g$  -  $300\times g$  for 5 minutes, remove the supernatant, and resuspend the cells in fresh culture medium.