HEK293 Serum-Free Feed

Catalog Number	Name	Size
SF1002	HEK293 Serum-free Feed A	50mL
SF1003	HEK293 Serum-free Feed B	5mL

Applicable cell lines

HEK293 serum-free feed medium (A&B) is independently developed and produced by Procell. It is suitable for the fed-batch culture process of different subtypes of Human Embryonic Kidney 293 Cells (HEK293) to achieve large-scale amplification of adenovirus and stable expression of products.

Medium characteristics

1. HEK293 serum-free feed A and feed B are both feed media and do not contain any animal-derived components (Animal-Derived Component-Free, ADCF).
2. Feed A contains amino acids, vitamins, glucose, hydrolysates, inorganic salts and trace elements; Feed B is a mixture of amino acids. Both feed media are free of growth factors, peptides, and phenol red.
3. Feed A contains P188 and HT, but does not contain L-glutamine.
4. Feed B does not contain P188, HT and L-glutamine.

Feeding strategy for transient transfection cells(Recommended)

Feeding addition amount (V/V): Feed A 10%、Feed B 1%

On the day before transfection, inoculate cells at a density of 1.5×10^6 cells/mL, and transfection will be carried out the next day if the density reaches about $3\text{-}4\times10^6$ cells/mL and the viability is above 97%:

1. Dilute the plasmid and PEI separately, and adjust the dosage of plasmid and PEI according to the actual process;
   (1) Calculate the required amount of plasmid, dilute it to 1.5 mL with Transbuffer, mix well and let it stand at room temperature for 5 minutes;
   (2) Take the corresponding amount of PEI, dilute it to 1.5 mL with Transbuffer, mix well and let it stand at room temperature for 5 minutes;
   (3) Slowly add the diluted PEI to the diluted plasmid, mix gently and slowly, and incubate at room temperature for 10 minutes (or adjust the incubation time according to the actual process).
2. Add the 3 mL incubated mixture to the transfection shaker flask and continue culturing. At this time, the working volume is 30 mL and the time is recorded as D0;
3. When 18-24 h after transfection, feed once with feed A 10% and feed B 1%.
4. During the subsequent culturing process, only sufficient glucose needs to be added (if the concentration is below 4 g/L, replenish it to 6-8 g/L). No additional feeding is required.
5. Take samples every day to measure cell density, viability, residual glucose, lactate, and other culture parameters. Harvest and measure the yield on Day 7.

(Note: The specific cultivation process can be adjusted according to actual project needs)

Feeding strategy for stable transfection cells (Recommended)

1. Cooling strategy: HEK293 stably transfected cells generally do not need to be cooled.
2. Feeding strategy:

   Feeding addition amount (V/V): Feed A 3%、Feed B 0.3%

   (1) The feeding amount of D7-D13 is determined based on osmotic pressure. The osmotic pressure at the harvest point is controlled within an appropriate range based on historical tolerance data. The highest osmotic pressure at harvest is generally 400-600 mOsmol/kg;

   (2) During the initial test experiment, the appropriate feeding amount can be explored according to the osmotic pressure. It is recommended to measure the osmotic pressure every day starting from D3, and at least detect the osmotic pressure on D2, D5, D7, D9, and D11;

   (3) Before D7, it is recommended that the osmotic pressure before feeding on the day of feeding is 300-320 mOsmol/kg, and for D7-D10, it is recommended that the osmotic pressure before feeding on the day of feeding be 330-350 smol/kg;

   (4) If the osmotic pressure is too high after D10, the amount of feed needs to be reduced, and the feed needs to be adjusted according to the actual tolerance of the osmotic pressure and metabolism of the cells.
3. Adding Glucose strategy: When the concentration is lower than 4 g/L, supplement it to 6-8 g/L.
4. Glucose concentration: The Glucose content of feed A is 77 g/L.
5. Lactic acid control strategy: If the lactic acid concentration increases in the later period, it is recommended to adjust by reducing the feed amount.

Storage

Store in 2-8°C; protect from light.

Shelf life: 12 months

Notes

1. This product is only used for scientific research or further research, not for diagnosis and treatment.
2. This product is sterilized by 0.1 μm filtration.
3. It is necessary to pay attention to the aseptic operation and avoid the contamination.