

AMMS[®]HEK293 serum-free medium

Size / Cat.No.: 1000mL /AS-11

Product Name

Generic Name AMMS[®]HEK293 serum-free medium

Product Information

Storage	2~8°C, keep it away from light
Validity	12 months
Status	Liquids
Scope of application	HEK293 cell suspension culture and transfection

Product description

AMMS[®]HEK293 serum-free medium is a serum-free, protein-free and animal-free chemical component qualified medium used in the HEK293 cell suspension culture and transfection process. The product contains HEPES, sodium bicarbonate, amino acids, vitamins, phenol red, trace elements, F-68 and other components conducive to the growth and expression of HEK293 cells. L-glutamide should be added before use.

Product features

1. No serum, no protein, no animal origin and clear chemical composition;
2. High transfection efficiency, suitable for HEK293 cell suspension culture and transfection;
3. Strict quality control and stable performance;

Instructions for use

1. Usage suggestions:

- 1.1 During conventional liquid replacement or transmission, the medium removed from the refrigerator of 4°C can be used immediately without preheating to room temperature or 37°C;
- 1.2 This medium contains 2mM L-glutamine, which can be appropriately supplemented according to the use scenario. It is recommended to add L-glutamine during protein expression and virus production to achieve a final concentration of 4-6 mM;
- 1.3 Without centrifugation and liquid exchange, the cell suspension can be directly mixed into the culture medium according to the required ratio;
- 1.4 It is recommended that the cell density after transmission be controlled at $5\sim 10 \times 10^5/\text{mL}$.

Note: Suggestions for cultivating HEK293 cells can be adjusted according to the actual situation.

2. HEK293 Suspension cell resuscitation and culture:

- 2.1 Material preparation: HEK293 serum-free medium, 15mL sterile centrifugal tube, sterile pipette tube, sterile gun head, sterile shaker bottle, L-glutamine, etc.;
- 2.2 Add 8mL HEK293 serum-free medium to the 15mL sterile centrifugal tube standby;
- 2.3 Quickly remove HEK293 cells to be resuscitated from the liquid nitrogen tank and slowly shake them in a 37°C water bath pot for 2 minutes to melt;
- 2.4 Disinfect the alcohol on the surface of the frozen storage tube and put it into the ultra-clean table, and transfer the cell fluid into the previously spare centrifugal tube with a pipette gun;
- 2.5 150×g centrifugation for 5 minutes;
- 2.6 Discard supernatant, gently flick the bottom of the centrifugal tube with your fingers to disperse the cells. After adding a 2mL medium, gently blow the cells with a pipette gun to disperse the cells;
- 2.7 Transfer cells to 125mL shakes that have been added to 18mL HEK293 serum-free medium, L-glutamine can be appropriately supplemented according to the actual situation;
- 2.8 Place the shake bottle in a carbon dioxide incubator (37°C, 5% carbon dioxide concentration, shaker 120rpm or adjusted according to the actual situation) for 3 to 4 days to determine cell density and cell activity.

Note: Cell viability may slightly decrease within 24 hours of recovery, but should be maintained at 70% or higher; Recovery takes 4 to 7 days to reach over 90%.

3. HEK293 Suspension Cell passage:

- 3.1 Material preparation: HEK293 serum-free medium, sterile centrifugal tube, sterile pipette tube, sterile gun head, sterile shaker bottle, L-glutamine, etc.;
- 3.2 Preparation before passage:
 - 3.2.1 Prepare the sterile shaker to be used;
 - 3.2.2 Remove the cells from the incubator, pay attention to tighten the cap before removing them, wipe the countertop of the microscope with an alcohol cotton ball, and then observe the cells under the mirror;
 - 3.2.3 Observe whether the cell is contaminated, and the normal cell medium is clear. If the color of the medium is observed to turn yellow and cloudy, it means that the cell has been contaminated and should be treated in time. Then the growth state of the cell should be observed. The cell suspension with good growth state is relatively uniform, and there are no flocculation or block objects, otherwise the cells may be contaminated.
 - 3.2.4 Count, if the cell density reaches $4\sim6\times10^6/\text{mL}$, it should be passaged.
- 3.3 Cell passage:
 - 3.3.1 Without centrifugation and liquid exchange, cell suspension can be directly exchanged into fresh HEK293 serum-free medium according to the required proportion;
 - 3.3.2 Appropriate amounts of L-glutamine can be added proportionally according to specific application situation;
 - 3.3.3 The cell density after passage should be controlled at $5\sim10\times10^5/\text{mL}$;
 - 3.3.4 Generally, it needs to be passaged 2 to 3 times a week (7 days);
 - 3.3.5 Place the shaker in a carbon dioxide incubator (37°C, 5% carbon dioxide concentration, shaker 120rpm or adjusted according to the actual situation).

4. HEK293 suspended cell freeze storage:

- 4.1 Preparation of dimethyl sulfoxide (DMSO) cell freeze storage solution, preparation as needed;
- 4.2 Cultivate cells to the logarithmic growth period (density about $4\sim6\times10^6/\text{mL}$), count and collect cells centrifugally;
- 4.3 Use cell freezing fluid to suspend centrifugal cells at a density of $10\times10^6/\text{mL}$;
- 4.4 Pack an appropriate amount of cell suspension into a frozen storage tube marked with information such as name and date, and tighten the tube cover;

- 4.5 Place the freezer tube in the freezer container and slowly cool down in the -80°C refrigerator;
- 4.6 Transfer the frozen storage tube to liquid nitrogen for long-term preservation the next day. This process needs to be completed as quickly as possible (recommended within 2 minutes).

Product Use

For research and manufacturing use