DMEM/F12 (with glucose, glutamine, sodium pyruvate, HEPES, without phenol red) Product manual

Basic Information

Cat.NO	Size	Shelf	Form	Storage	Transportation
CMB0021	5L/10L	24 months	Powder	Store at 2-8°C away from light	Room temperature

Product Introduction

DMEM/F12 medium is a mixture of DMEM and Ham's F12 medium in a 1:1 ratio. It contains a variety of trace elements and is widely used in the culture of mammalian cells such as MDCK, glial cells, fibroblasts, human endothelial cells and rat fibroblasts.

HEPES is an excellent biological buffer with no toxic effect on cells. The culture medium with HEPES added can maintain a constant pH range for a long time, which can effectively prevent the adverse effects of large pH fluctuations in the culture medium on cell growth.

Phenol red is used as a pH indicator in culture media to continuously monitor the pH of the culture medium. At low pH values, phenol red makes the culture medium yellow, while at higher pH values, the culture medium turns purple. It turns red at pH 7.2-7.4, which is most suitable for cell culture. However, phenol red also has some disadvantages. Studies have shown that phenol red can simulate the effects of steroid hormones (especially estrogen). Therefore, when using estrogen-sensitive cells (such as breast tissue), it is best to use a culture medium that does not contain phenol red. Phenol red can interfere with detection during flow cytometry analysis. In addition, the presence of phenol red in some serum-free culture medium formulas can interfere with sodium-potassium balance.

Preparation of powder

1. Purified water, ultrapure water or water for injection should be used for preparation, and the water temperature should be controlled at 20-30℃ during the preparation process;
2. Add 90% of the preparation volume of preparation water to the preparation container (if 1L is required, add 900mL of preparation water here), turn on the mixing system of the culture medium preparation container (it is recommended that the input power per unit volume of the mixing system is greater than 10W/m³), stir well, and avoid the generation of bubbles during stirring;
3. Weigh the powder culture medium according to the required preparation volume. Add the accurately weighed culture medium powder to the preparation container of step 2 and stir thoroughly for more than 20 minutes until the powder is completely dissolved;
4. Add preparation water to accurately make the completely dissolved solution of step 3 to 100% of the preparation volume (if 1L is required, make it to 1L);
5. Measure the pH value and adjust the pH value to 7.20-7.30 with 1mol/L sodium hydroxide solution or 1mol/L hydrochloric acid solution if necessary; since filtration will slightly increase the pH value of

the culture medium, it should be lower than the target pH value (7.20-7.40);
6. Sterilize by positive pressure filtration with a filter membrane with a pore size of 0.2μm (pay attention to aseptic operation);
7. After filtration, take a small amount of liquid culture medium for bacterial inspection and use it after it passes the test;
8. The filtered culture medium liquid should be used immediately or stored in a glass bottle, culture medium bottle (PET) or disposable liquid storage bag with oxygen barrier coating, and stored at 2-8℃ away from light. At this time, the shelf life of the liquid culture medium is 1 year.

Instructions

1. Balance the culture medium and related solutions in a water bath or at room temperature, and prepare the culture medium required for the experimental cells;

2. Cell inoculation: Remove the cells to be cultured from the original culture container, wash with appropriate culture medium or PBS, and adherent cells need to be digested with trypsin;

3. Collect the cells by centrifugation, centrifuge at 1000rpm for 3 min at room temperature, and discard the supernatant;

4. Add fresh culture medium to resuspend the cells. Then inoculate the cell suspension into the culture bottle with the corresponding volume of culture medium, mix gently, and culture at 37°C and 5% CO_2 saturated humidity. Observe and replace fresh culture medium regularly according to cell growth and cell density.

Precautions

1. During the entire process, be sure to pay attention to aseptic operation to avoid contamination;
2. To maintain the best use effect of this product, do not perform freeze-thaw treatment;
3. This product is only used for research or further research, not for diagnosis and treatment.