

CO₂ Independent Medium, including Phenol red, L-glutamine and Sodium Pyruvate, and does not including HEPES

Cat: C3190

Storage: 2~8°C, away from light.

Validity: 12 months

Product Parameters:

Appearance(character): red clarified liquid

Source: Non-animal source

Endotoxin: ≤1 EU/mL

Osmotic pressure: 270-340mOsm/kg

pH: 7.0-7.4

Serum addition: 10% fetal bovine serum

Culture conditions: 37°C, 5-10% CO₂

Cell type: a variety of suspended and attached mammalian cells

Introduction:

The carbon dioxide independent medium was buffered by phosphate. The formula contains a small amount of sodium bicarbonate to satisfy the necessary bicarbonate-dependent function. Since synthetic buffers such as Hepes are not used, the cytotoxic effects associated with such buffer systems are eliminated. In addition, the optimized formulation of the medium improves cell production and CO₂ utilization, so that CO₂-dependent cell function can be maintained without exogenous CO₂. CO₂ independent media are used to support the growth of a variety of suspended and attached mammalian cell lines, such as epithelial cells, fibroblasts and lymphoid cell lines, without the need for CO₂ incubators. The medium is ideal for transporting cells or tissues under atmospheric conditions and for handling mouse embryos. For long-term culture, traditional standard CO₂-dependent medium is recommended. Carbon dioxide independent medium contains no proteins, lipids or growth factors. Therefore, more nutrients need to be added in culture, ITS needs to be added in low serum culture, and 10% fetal bovine serum needs to be added in conventional culture. This medium does not require a CO₂ environment to maintain physiological pH. To achieve the highest possible growth performance, some cell lines may need to be adapted directly or sequentially to the medium.

Prepare completely carbon dioxide independent medium

1. L-glutamine or L-alanyl glutamine was added to the final concentration of 200mM;
2. Adding fetal bovine serum to the final concentration of 10%, such as adding ITS, can reduce the serum concentration to 2-5%;
3. Antibiotics can be added if needed.

Cell adaptation to carbon dioxide independent medium

For maximum growth performance, some cell lines may need to be adapted directly or sequentially to CO₂-independent media. In both cases, the protocell line should be in a logarithmic growth phase with a high survival rate(>90%), and the success of the adaptation process will depend on the cell line used and the culture conditions adopted. It is recommended that users first evaluate this medium with unadapted cells, as not all cell lines need to be adapted.

Direct adaptation

1. The culture was inoculated at normal inoculation density, using a closed culture bottle with a tight cap, and cultured in an incubator connected to a humid atmosphere(37°C).
2. The cell growth was monitored daily, and when the cells reached 80-90% confluent, the passage was carried out. If the cell culture cannot maintain acceptable growth and survival during direct adaptation, the sequential adaptation method is used.

Sequential adaptation

1. Cells with normal seeding density were inoculated into carbon dioxide independent medium and the medium configured with a 50:50 ratio(v/v) of the current medium, cell growth was monitored daily, and then passed when the cells reached 80-90% confluence.
2. Pass to carbon dioxide independent medium and the current medium in a 75:25 ratio(v/v) configuration. The cell growth was monitored daily, and when the cells reached 80-90% confluent, the passage was carried out.
3. Subculture to completely carbon dioxide independent medium. The cell growth was monitored daily, and when the cells reached 80-90% confluent, the passage was carried out.
4. Complete carbon dioxide independent culture medium was used for subsequent generations.

Notes:

1. If the biochemical reagents produced by our company are not specially marked, they are basically non-sterile packaging. If they are used in cell experiments, please pre-treat them in advance.
2. Once the solution is prepared, please store it separately to avoid product failure caused by repeated freezing and thawing.
3. Product information is for reference only, if you have any questions, please call 400-968-6088 for consultation.
4. This product is for scientific use only. Do not use for medicine, clinical diagnosis or treatment, food and cosmetics. Do not store in ordinary residential areas.
5. For your safety and health, please wear a lab coat, disposable gloves and a mask.