BESTCHROM
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Bestdex Cell 1
Microcarriers for Cell Culture

Instruction for use

Contents

1. Introduction 1

2.Technical characteristics 1

3. Method of chromatography 2

4. Sterilization 3

5. Storage 4

6. Disposal and recycling 4

7. Order information 4

1. Introduction

Microcarrier culture is a technique which enables the high yield culture of anchorage-dependent cells. Bestdex Cell 1 has been developed specifically for growing a wide range of animal cells in volumes ranging from a few milliliters to several thousand liters. The use of Bestdex Cell 1 in a simple suspension culture system provides a yield of millions of cells per milliliter. It changes the available surface area simply by changing the microcarrier concentration. Applications include large-scale production of cells, viruses, and recombinant cell products (eg., interferon, enzymes, nucleic acids, hormones), cell adhesion, differentiation, and cell function. This product is intended for research purposes only and is not intended for clinical or in vitro diagnostic use.

Bestdex Cell 1 is formed by coupling diethylaminoethyl groups to Bestdex G-50.In the process of Cell culture media, the microcarriers provide bioinert matrix, and the cells are highly adhered to the surface of the microcarriers. In addition, the density of Bestdex Cell 1 is similar to that of water, which is conducive to the uniform suspension of cells.

Bestdex Cell 1 is suitable for most established Cell lines. It can also be used for the culture of primary cells and normal diploid cell lines.

The main characteristics of this resin include:

- The optimization of particle size and density provides high yield growth conditions for various cells.
- After swelling, the microcarrier is transparent, which is convenient for the microscopic detection of the attached cells.
- Microcarrier scaffolds are biologically inert and can provide a firm but non-rigid substrate for stirring culture of cells.

2.Technical characteristics

Matrix	Dextran
Functional group	Diethylaminoethyl
Particle size +	150~250μm
Ionic capacity	1.40-1.60mmol Cl^-/g resin
Deposition velocity	~ 90cm/h
Density	~1.03g/mL
Swelling factor ++	20~25mL
Approx. area	~4400 cm^2/g dry powder
Approx. no. microcarriers /g dry resin	~4.3×10^6

+ Dry powder particles swelling with 0.15M NaCl wet glue particle size. The volume of particles in this range accounts for

more than 80% of the total volume.

++ Particle size may vary in different solutions, and this data is the result of swelling in 0.15M NaCl

3. Method of chromatography

3.1 Preparation of microcarriers

Note：Bestdex Cell 1is supplied in the form of dry power，which needs swelling before use.

- At room temperature, the microcarriers were swelted in a PBS solution without Ca^{2+} and Mg^{2+} (consumption: 50-100 mL PBS/g microcarriers) for at least 3h.
- Slowly pour out swelling PBS and wash the microcarriers with fresh PBS solution free of Ca^{2+} and Mg^{2+} (30-50 mL/g relative to microcarriers) for 2-3 min.
- Slowly pour out the supernatant and re-pour in fresh PBS solution free of Ca^{2+} and Mg^{2+} (consumption: 30-50 mL/g relative to the amount of microcarriers).
- Sterilized the microcarriers at 115℃ or 121℃, PH7.4, 0.1MPa for 15~30min.

  Note：The microcarrier can be repeatedly sterilized for more than 5 times without affecting the performance, and the pH of all the solutions during sterilization is 7.4. It can withstand high temperature sterilization at 130℃.
- Before use, make the microcarrier settled and remove the supernatant.
- The microcarriers were quickly rinsed with a resin restored to room temperature (20-50 mL/g relative to the amount of microcarriers).
- After the microcarriers settled, the supernatant was poured out and transferred to the culture vessel.

3.2 Culture vessel

- Microcarriers culture can be performed in a general purpose cell culture media. The cell culture media with slow stirring function, ensuring uniform suspension of microcarriers, and not producing high shear force is the best. The agitator must not collide with the inner surface of the container during the agitation process, and the container in which the agitator bearing is immersed in the resin is not suitable because the microcarriers may be ground in the agitator bearing.

  Note：Glass culture containers should be silicified before use.

3.3 Culture procedure

- The specific culture procedure of microcarriers depends on the cell type and culture container. In the process of culture, the content of microcarriers was 1-5 g/L, the inoculation amount was 5×10^4~2×10^5 cells /mL, and the rotation speed was generally 20-60 rpm depending on the structure of the tank, so the microcarriers needed to be suspended.
- Microcarriers culture can be carried out by referring to the following steps (the specific volume and inoculation amount can be adjusted proportionally according to the actual situation, and the following steps should be modified for different types of cell culture media).

➢ The 30 mL culture media was supplemented with 0.3g microcarriers (the final culture volume

was 100 mL).

- Inoculate the culture with 10^7 cells, mixed slowly and evenly, and incubate at 37℃.
- After the cells were firmly attached to the surface of the microcarrier, continuous stirring began.

  Note：The time required for cell attachment depends on the attachment efficiency of different cell types. If the time required is too long, the culture can be stirred intermittently (e.g., 2 minutes every 30 min) to ensure an even distribution of cells and microcarrier .
- After the microcarriers were evenly distributed, increase the culture volume to 50 mL.
- After 1 to 2 days, increase the culture volume to 100 mL, and after 3 to 5 days, partial replacement of the resin may be required.

3.4 Detection of cell growth

- Representative microcarrier samples were taken from the culture and analyzed directly or stained with hematoxylin for microscopic examination. The most appropriate method to determine the number of cells is to use standard nuclear counting.

3.5 Harvesting cells

- Cells can be removed from the surface of the microcarrier in a various mrthods. The most common method is to treat with proteolytic enzymes such as trypsin.
- The microcarrier were settle and remove the culture media.
- The microcarriers were washed in a PBS solution free of Ca^{2+} and Mg^{2+} containing 0.02% (w/v) EDTA (50~100 mL/g for microcarriers) for 5 min at pH 7.6.

  Note: At higher serum concentrations, more washing processes are required.
- Remove the EDTA-PBS, EDTA solution containing trypsin or collagenase was added (about 30-50 mL/g relative to the microcarriers). Max well and incubated at 37℃ for 15 min with occasional agitation. the reaction was stopped by adding serum-containing resin (about 20-30 mL/g relative to the microcarrier).

  Note: At this stage, the remaining cells in the microcarriers can be removed by gentle agitation.
- The detached cells can be separated from the microcarriers under gravity by sedimentation or by a 100μm cell filter and can be used for subsequent microcarriers culture inoculation during scale-up production.

  Note: When the above cells are used as inoculants for subsequent microcarrier culture, cell viability, membrane integrity and high adhesion efficiency need to be ensured. Therefore, the speed of washing and acquisition is very important.

4. Sterilization

Bestdex Cell 1 is supplied in the form of dry powder and must be swollen and sterilized before use. The Bestdex Cell 1 is very stable and can be autoclaved at 115℃ or 121℃ for 15~30min. When sterilizing, the pH of all solutions should be 7.4.It can withstand high temperature sterilization at 130℃.

5. Storage

After use, the Bestdex Cell 1 should be stored aseptically in PBS solution at 2~8℃.

6. Disposal and recycling

Bestdex Cell 1 is very difficult to degrade in nature, incineration is recommended to protect the environment.

7. Order information

Product	Cat. No.	Pack size
Bestdex Cell 1	AI132305	25g
	AI132307	100g
	AI132311	500g
	AI132312	1kg
	AI132313	5kg
	AI132316	10kg
	AI132314	25kg