Peripheral Blood Medium product information

Product Name

Name: PBType-1 Peripheral Blood Karyotyping Medium

Cat. No.: C3601-0005, C3601-0100, C3601-0150

Size: 5 mL, 100 mL, 5 mL*30

Product Description

PBType-1 Peripheral Blood Karyotyping Medium is intended for short-term cultivation of peripheral blood lymphocytes for cytogenetic studies and in vitro diagnostic procedures. The medium is based on an optimized RPMI 1640 medium supplemented with L-Glutamine, fetal bovine serum (FBS), antibiotic, and phytohemagglutinin-M (PHA-M).

PBType-1 Peripheral Blood Karyotyping Medium is supplied as a frozen and complete medium, and it is ready to use without any further supplementation after thawing.

Note

- It will not affect cell growth performance if a flocculent precipitate is observed in the medium.
- Use of PBType-1 Peripheral Blood Karyotyping Medium does not guarantee the successful outcome of any chromosome/karyotyping analysis testing because the large variations in the subsequent steps can also affect the results of karyotyping.
- Do not use PBType-1 Peripheral Blood Karyotyping Medium beyond the expiration date indicated on the product label.

Storage and Stability

The product should be kept at **-20°C**.

After thawing, the medium should be stored at 2-8℃. The medium should be used within 10 days after thawing and should be used within 4 days for optimal performance.

The product is **light-sensitive** and therefore should be protected from light.

Shelf life: 24 months from the date of manufacture.

Procedure

Thaw PBType-1 Peripheral Blood Karyotyping Medium at refrigerator temperatures (2 - 8°C) or by swirling the bottle in a 37°C water bath. Mix gently during thawing.

Note that the medium already contains L-Glutamine, antibiotic, and PHA-M.

Culture of Peripheral Blood Lymphocytes for Chromosome Analysis

The peripheral blood cell karyotyping method has been developed to provide information about chromosomal abnormalities. Peripheral blood lymphocytes do not normally undergo cell divisions in normal and healthy adults. In the presence of a mitogen such as phytohemagglutinin, lymphocytes are stimulated to enter into mitosis. After 48 - 72 hours, a mitotic inhibitor such as Colcemid or colchicine Solution is added to the culture to stop mitosis in the metaphase stage. After treatment with a hypotonic solution, fixation, and staining, mitotic chromosomes can be microscopically observed and evaluated for abnormalities.

A simple procedure for blood culture is provided below:

1. Inoculate approximately 0.5 mL of heparinized whole blood into a glass or plastic tube with 10 mL of PBType-1 Peripheral Blood Karyotyping Medium.
2. Incubate the culture for 72 hours.
3. Add 0.1 - 0.2 mL of Colcemid Solution to each culture tube. Incubate the culture for an additional 15 - 30 minutes (the volume or concentration of Colcemid can be changed according to the experimental requirements).

4. Transfer the culture to a centrifuge tube and spin at 500 x *g* for 5 minutes.
5. Remove the supernatant and re-suspend the cells in 5-10 mL of hypotonic 0.075 M KCl solution to lyse the red blood cells. Incubate at 37ºC for 10 – 12 minutes.
6. Spin at 500 x *g* for 5 minutes.
7. Remove the supernatant, shake the remaining sediment, and add dropwise 5 - 10 mL of freshly prepared ice-cold fixative (1 part acetic acid to 3 parts methanol). Stay on ice for 10 minutes.
8. Repeat steps 6 and 7 to remove the red hemoglobin solution.
9. Spin at 500 x *g* for 5 minutes.
10. Resuspend the cell pellet in 0.5 - 1 mL of fresh fixative, drop the solution with the cells onto a clean slide, and allow to air-dry.
11. At this stage, the preparation can be stained with Giemsa. Giemsa staining is the most common method to stain the chromosomes. If G banding is desired, the slides need to be treated with trypsin-EDTA 10X solution for a few seconds. The timing of the trypsin treatment needs to be adjusted after the first slide.

Quality Control

PBType-1 Peripheral Blood Karyotyping Medium is tested for appearance, capacity limit, sterility, pH, osmolality, cell culture, and endotoxin concentration. In addition, each batch is tested for karyotyping in a leading clinical cytogenetics laboratory for its performance.

Quality Assurance

- Manufactured under ISO 13485 QMS.
- Manufactured under controlled environments and processes per:

1. ISO 13408 – Aseptic Processing of Health Care Products
2. ISO 14644 – Cleanrooms and associated controlled environments

Manufacturer

Shanghai Dr. Cell Co., Ltd.

Issue Date

January 2025

Precaution and Disclaimer

For in vitro diagnostic use, not for treatment.