Product Name

Name: AmnioType-1 Medium

Cat. No.: C3620-0100

Size: 100 mL

Product Description

The *in vitro* cultivation of amniotic fluid cells and chorionic villi is an essential part of every diagnostic cytogenetics laboratory since the preparation of metaphase chromosome spreads is dependent upon obtaining enough cells in the division. Amniocentesis and chorionic villi sampling are the major invasive diagnostic procedures used for the detection of fetal chromosomal abnormalities. AmnioType-1 Medium is specifically optimized for the primary culture of human amniotic fluid cells and chorionic villi samples used in prenatal diagnostic testing. The medium is supplied frozen and contains serum, glutamine, and an antibiotic.

Note

- For *in vitro* diagnostic use. The medium is not intended for therapeutic use.
- Use of AmnioType-1 Medium does not guarantee the successful outcome of any prenatal diagnostic testing.
- Do not use AmnioType-1 Medium beyond the expiration date indicated on the product label.
- The product is prepared by an aseptic, strict, validated process in order to minimize any possible risk of contamination and to ensure the safety and quality of the product. The membrane filtration and aseptic process are validated by media fill to ensure a safety level of 1 to 5000 bottles. Due to the residual risks of sample contamination, and the severity of such cases, we recommend establishing parallel test systems as a part of the procedure.

Storage and Stability

The product should be kept at **-20°C**.
The product is **light-sensitive** and therefore should not be left in the light.
Shelf life: 24 months from date of manufacture.

Procedure

Thaw AmnioType-1 Medium at refrigerator temperature (2 - 8°C). Swirl gently and intermittently during thawing.

Note that the medium already contains L-Glutamine and an antibiotic.

AmnioType-1 Medium may be used for:

- Primary culture of amniotic fluid cells
- Culture of passaged amniotic fluid cells
- Propagation of chorionic villus cells

The medium may be used in both open and closed culture systems.

It is recommended to use the cells from 2.5 mL of amniotic fluid per coverslip.

The following protocol and the volumes indicated are only general guidelines for use.

In Situ culture of amniotic fluid cells:

1. Centrifuge 20 mL of amniotic fluid at 750 rpm for 10 minutes.
2. Carefully decant the amniotic fluid (supernatant) from the cell pellet into a sterile test tube.
3. Resuspend the cell pellet with 2 mL of amniotic fluid.
4. Add 2 mL of AmnioType-1 Medium and swirl gently.
5. Culture 0.5 mL of the cell suspension on each coverslip put in a tissue culture dish.
6. Incubate the coverslips at 37°C in a 5% CO_2 atmosphere.
7. Flood cultures on day 2 with 1.5 mL of AmnioType-1 Medium.
8. After 5 days, check the cultures for the presence of colonies.
9. After the colonies first appear (5 - 7 days), replace the medium with fresh AmnioType-1 Medium.
10. When the size of colonies is sufficiently large, proceed with harvesting.

Note: It is recommended to replace the medium with fresh AmnioType-1 Medium the day before harvesting.

Flask Culture Method of Amniotic Fluid Cells – Open and Closed Systems

Use the same procedure as for the *in-situ* culture, with the following adaptations:

1. Resuspend the cell pellet with 4 mL of amniotic fluid. Add 16 mL of AmnioType-1 Medium and swirl gently.
2. Culture 5 mL per T25 flask. Place the cap loosely on the flask and incubate undisturbed at 37°C in a 5% CO_2 atmosphere. For Closed Systems: Flush each culture flask with 5% CO_2 - 95% air through a 0.2 µm sterile filter for 20 seconds. Tighten the caps and incubate the flasks at 37°C.
3. Check all flasks for growth after 5 days.

Quality Control

AmnioType-1 Medium is tested for sterility, pH, and osmolality. In addition, each batch is tested for cell growth and karyotyping using primary human amniotic fluid cells in a leading clinical cytogenetics laboratory. For full specifications please check the lot-specific Certificate of Analysis (CoA).

Quality Assurance

- Manufactured under ISO 13485 QMS and in compliance with applicable cGMP guidelines.
- Manufactured under controlled environments and processes in accordance with:
  1. ISO 13408 – Aseptic Processing of Health Care Products;
  2. ISO 14644 – Airborne Particulate Cleanliness Classes in Clean Rooms and Clean Zones.