

KREAvital Prenatal Medium

Medium for Human Amniotic Fluid and Chorionic Villi Samples

KREAvital Prenatal Medium (Basal)

Cat. No: KBI-90010 (90 ml)

Store at: 2°C - 8°C

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 cells in division. Amniocentesis and chorionic villi sampling are the major invasive diagnostic procedures used for the detection of fetal chromosomal abnormalities. KREAvital Prenatal Medium is specifically optimized for the primary culture of human amniotic fluid cells and chorionic villi samples used in prenatal diagnostic testing. The supplement is supplied frozen and contains serum.

Precaution and Disclaimer

1. For in vitro diagnostic use. The medium is not intended for therapeutic use.
2. Do not use if a visible precipitate is observed in the medium.
3. Use of KREAvital Prenatal Medium does not guarantee the successful outcome of any prenatal diagnostic testing.
4. Do not use KREAvital Prenatal Medium beyond the expiration date indicated on the product label.
5. Due to the residual risks of sample contamination, and the severity in such case, we recommend to establish parallel test systems as part of the procedure.
6. In case of questions please contact your local sales representative or send an email to customerservice.amsterdam@leicabiosystems.com

Storage and Stability

The supplemented medium is stable for 14 days when stored at 2 - 8°C.

Do not freeze the supplemented medium. Protect both the basal medium and the complete medium from light.

Instructions for Use

For the preparation of 100 ml complete medium, use KBI-90010 with KBI-90011. Thaw the KREAvital Prenatal Medium (Supplement) by swirling in a 37°C water bath, and

KREAvital Prenatal Medium (Supplement)

Cat. No: KBI-90011 (10 ml)

Store at: -20°C

aseptically transfer the contents to the bottle of KREAvital Prenatal Medium (Basal). Mix the complete medium by swirling the bottle, and add 2mM L-Glutamine. Antibiotics may be added if desired.

Procedure

KREAvital Prenatal 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 cells from 2.5 ml of amniotic fluid per one 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 from the cell pellet into a sterile test tube.
3. Re-suspend the cell pellet with 2 ml of amniotic fluid.
4. Add 2 ml of KREAvital Prenatal Medium and swirl gently.
5. Culture 0.5 ml of the cell suspension on each coverslip in a tissue culture dish.
6. Incubate cultures at 37°C in 5% CO₂ atmosphere.
7. Flood cultures on day 2 with 1.5 ml of KREAvital Prenatal 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 KREAvital Prenatal Medium.
10. When the cultures have colonies of sufficient size, proceed with harvesting.

Note: It is recommended to replace the medium with fresh KREAvital Prenatal Medium the day before harvesting.

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

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

1. Re-suspend the cell pellet with 4 ml of amniotic fluid. Add 16 ml of KREAvital Prenatal Medium and swirl gently
2. Culture 5 ml per each T25 flask. Place the cap loosely on the flask and incubate undisturbed at 37°C in 5% CO₂ atmosphere. For Closed Systems: Flush each culture flask with 5% CO₂ – 95% air through 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

KREAvital Prenatal Medium (Basal) and KREAvital Prenatal Medium (Supplement) are tested for sterility, pH and osmolality. In addition, each batch is tested for cell growth using primary human amniotic fluid cells.

