FertiCult IVF medium™

CELL CULTURE MEDIUM FOR IN VITRO CULTURE OF HUMAN EMBRYOS, DURING THE FIRST 48 HOURS IN CULTURE

FertiCult IVF medium is sterilized by aseptic filling STERILE A

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GENERAL INFORMATION

FertiCult IVF medium is a ready to use formulation for the in vitro culture of mammalian embryos. It is designed for short term culture only (up to 48 hours in culture).
The medium is complete and needs no further additives.
If preferred 10 % patient serum v/v can be added.
FertiCult IVF medium is suitable for microdroplets under oil or equally suited to culture in Falcon 3037 organ culture dishes (also 4-well Nunc culture dishes).
As with all IVF media, FertiCult IVF medium ought to be **pre-incubated in the incubator for 24 hours before use** (with lid opened).

MATERIAL INCLUDED WITH THE KIT

Productcode: FECU-20

9 x 20mL FertiCult IVF medium

Productcode: FECU-50

5 x 50mL FertiCult IVF medium

Productcode: FECU-100

3 x 100mL FertiCult IVF medium

MATERIAL NOT INCLUDED WITH THE KIT

- Incubator
- Petri dishes
- Mineral oil
- Laminar flow
- Microscope
- Test tubes

PRODUCT SPECIFICATIONS AND QUALITY CONTROL

FertiCult IVF medium is manufactured according to these specifications:

- pH: 7.20 - 7.40
- Osmolality: 275 - 285 mOsm/kg
- Endotoxin: < 0.25 EU/mL
- Sterility: Sterile
- Mouse-embryo test: > 70% blastocysts after 96 hours in culture
- Use of Ph Eur or USP grade products if applicable

INSTRUCTIONS FOR USE

PREPARATIONS FOR USE OF MICRODROPLETS

For microdroplets, between 100-250µL of FertiCult IVF medium may be dispensed around the culture dish, up to 8 per 60mm dish.

The dish is then filled with 5 mL of pre-washed and pre-equilibrated light mineral oil (nontoxic and preferably embryo tested, e.g. FertiCult Mineral Oil).
One egg is usually placed with about 10.000 sperm per microdroplet, so if each patient during the next day is expecting 16 eggs, at least two dishes per patient should be prepared. The dishes are then placed in the incubator (usually non-humidified) to equilibrate overnight at 37°C and under an atmosphere of 5% CO_2 in air.

This overnight equilibration is highly recommended.

PREPARATIONS FOR USE IN OPEN SYSTEMS

In open systems such as with the Falcon or Nunc dishes, about 1 mL of medium is placed in each well.

A further 3 mL is placed in the reservoir surrounding the wells. This helps to maintain humidity as well as providing medium with which to wash the eggs and embryos.

Up to five eggs per dish (or well if using Nunc dishes) may be cultured together, usually with about 100.000 sperm per well. So if each patient during the next day is expecting 15 eggs or so at egg collection, at least six dishes per patient should be prepared (three for initial holding and cleaning of eggs during collection and then three dishes for overnight culture).
When using Nunc dishes, two would be enough (since there are 4 wells per dish, the resulting 8 wells would be more than enough).

The dishes are then placed in the incubator to equilibrate overnight at 37°C and under an atmosphere of 5% CO_2 in air.
As with the microdroplets method described above, **equilibration overnight is highly recommended**.

In the open system, paraffin oil is not necessary, but sometimes a 1mL layer of mineral oil may be added over the medium in the inner well. When using such an open system the incubator must be humidified.

CULTURE

After equilibration overnight, the prepared dishes are ready for use. During egg collection, eggs are identified in the follicular aspirates and then washed before placing into the wells or droplets in the dish (as appropriate). Once egg collection is over, eggs will be checked and may be rewashed, before then being placed into fresh droplets/wells for overnight culture.

Generally, within 6 hours of egg collection, eggs will be inseminated with prepared sperm and then left overnight in the incubator.

The procedures described above are for the first 24 hours of culture. Once embryos have been identified by the presence of two pronuclei, they are usually placed into fresh dishes containing fresh medium.

In the past, where people have used cord serum or patient serum, the initial medium contains 10% and the changeover (after fertilisation has been confirmed) 15%.

When using FertiCult IVF medium no extra protein is required at changeover. Dishes should be prepared as described above, equilibrated overnight, after which they will be ready to receive the newly fertilised embryos.

NB. Test tube culture is now rare, but for those still using this method, place 1mL of FertiCult IVF medium in each tube and then follow the same procedure as for Falcon or Nunc dishes.

STORAGE AND CONSERVATION

Store FertiCult IVF medium between 2-25°C.

Keep from light.

WARNING AND PRECAUTION

All human, organic material should be considered potentially infectious.

Handle all specimens as if capable of transmitting HIV or hepatitis. Always wear protective clothing when handling specimens.

FertiCult IVF medium does not contain antibiotics, always work under strict hygienic conditions (laminar flow) to avoid contamination, or add your own antibiotics (penicillin) at about 100 units per ml.

Don't use FertiCult IVF medium if cloudy.

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