

Attention: The patient samples are biological material and therefore safety precautions are necessary.

Purpose of Use

Prenaplus medium mainly serves the cultivation a) of cells from chorionic and abort material as well as b) of primary and amniotic cells. It is intended for human genetic *in vitro* diagnostics (IVD CE according to MPG).

Composition

It is a complete medium composed of basal-medium, pre-tested FBS, hormones and growth factors for the corresponding cell or tissue type, phenol red, NaHCO₃, plus gentamicin and L-glutamine.

Shelf life and storage

Prenaplus Medium is durable at a storage ≤ -18 °C for 18 months from the date of manufacture. For quality and sterility reasons, the use of Prenaplus medium is recommended after opening at a storage of + 2 °C to + 8 °C of a maximum of 7 days. Repeated thawing and freezing should be avoided. Prenaplus medium not exceed the expiry date indicated on the label.

Thawing

Thaw Prenaplus medium at + 2 °C to + 8 °C overnight. Thawing in a water bath at 37 °C is not recommended. Mix well before using Prenaplus medium. The normal pH value is 7,2 as indicated by the phenol red indicator. In the case of a pH deviation (yellow or pink), the pH value is obtained by incubating the slightly open bottle (approx. ¼ rotation of the lid) in a 5% CO₂ incubator equilibrated until the medium has reached the normal color red. Prenaplus medium contains no components whose quality is affected by pH fluctuations of +/-2. Heated medium at 37 °C and correct pH-value ensures an optimal start of the culture.

Sample preparation

Both the chorionic and the Abort material must be separated from maternal material (Dezidua) and other impurities (e.g. blood coagulate). Crush material with sterile scalpel. Now the samples can be coated in a culture bottle. Alternatively, use the enzyme dispase or collagenase to maceration for 10 minutes with adequate concentration. The cells are stored for an hour with the addition of 3 ml of Prenaplus in a "moist chamber", i.e. the culture bottle is placed upside down and then slowly and carefully turned so that the material does not detach!

Standard protocol

The method described below is a standard guide for the use of Prenaplus medium in the cultivation of corresponding tissue samples. Of course, this high-quality medium can be integrated into your own workflow. It is the responsibility of the user whether the protocol is taken over completely or only partially in the own protocols.

An optional protocol can be provided.

Bottle Method

- Concentration of cells by centrifugation at low speed
- Take off 90 – 95% of the Supernatant and resuspend cells in the remaining residue.
- Dilute the pellet with preheated Prenaplus to at least 2 ml to obtain 2 ml per culture bottle
- Incubate at + 37 °C and 5% CO₂ in incubator
- Check growth on the 5th day and replace the medium with fresh Prenaplus medium
- Replace the used medium to the harvest on a regular basis

- For optimal results medium the day before harvest change

in situ method

- Concentration of cells by centrifugation at low speed
- Remove 95% of the Supernatant and resuspend cells in the remaining excess
- Dilute the cell suspension with preheated Prenaplus medium to at least 2 ml to obtain 0.5 ml suspension per deck of glass (total number 4)
- Incubate at + 37 °C and 5% CO₂ in incubator
- 2 ml Prenaplus medium on the second day
- Control cell growth after 4 – 5 days
- Immediately afterwards, gently vacuum the entire medium and replace with 2 ml preheated fresh Prenaplus medium
- Recommendation: Media change every 2 days
- For optimal results replace the medium the day before harvest

Important observation

- Calciumoxalat crystals may occasionally form, but have not yet shown any negative effects on cell growth.
- Thawing in a water bath at 37 °C should be avoided since precipitates can form.

Important Notes

- Always handle biological materials with proper safety precautions.
- Exclusively for *In vitro* Diagnostics (IVD).
- Warning: Not suitable for therapeutic use in humans or animals. Prenaplus medium is for diagnostic purposes only.
- Each laboratory is required to carry out representative tests according to regulations before the media can be used in routine diagnostics.
- Acceptance of Prenaplus medium as a diagnostic reagent is at the sole discretion and responsibility of the testing laboratory and responsible management.
- Cytogen GmbH does not guarantee success of diagnostic testing solely through the use of Cytogen products.

CE marking

With Prenaplus medium provides Cytogen a CE marked medium for IVD, which meets the requirements of Directive 98/79 / EC, established by the European Commission.

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