For life science research only.
Not for use in diagnostic procedures.

Polyethylene Glycol 1500 (PEG 1500)

Solution, filtered through 0.2 μm pore size membrane

Cat. No. 10 783 641 001 10 × 4 ml

Version 16
Content version: November 2014
Store at at +2 to +8°C

1. What this Product Does

Number of Tests

50% PEG 1500 (w/v) in 75 mM Hepes, pH 8.0, bottled under nitrogen; filtered through 0.2 μm pore size membrane, ready for use.

Storage and Stability

Stable at +2 to +8°C until the expiration date printed on the label.

⚠ Store protected from light

Application

Cell fusion induced by polyethylene glycol (PEG) has become a standard method in somatic cell genetics (1–4). PEG promoted cell fusion is also the standard procedure for the production of hybridoma cells (5 –12).

2. How to Use this Product

Working instruction for fusion of myeloma cells and mouse spleen cells

🔍 The medium used for the following protocol must be serum-free in every step of the procedure until Step 12, as the cell surface must be free of serum during the cloning steps.

Step	Action
❶	Mix 10^8 spleen cells (in 25 ml) and 2×10^7 myeloma cells (in 25 ml) in serum-free medium in a conical tube.
❷	Spin the cells down (5-10 min, 200 - 400 × *g*).
❸	Remove the supernatant with a pasteur pipette. 🔍 Complete removal of the supernatant is essential to avoid dilution of PEG.
❹	• Break the pellet by gently tapping the bottom of the tube. • Place the tube in a 37°C water bath and kept there during the fusion.
❺	Add 1 ml 50% PEG 1500 pre-warmed to 37°C to the pellet using a 1 ml pipette, over a period of 1 min, continually stirring the cells with the pipette tip.
❻	Continue stirring the cells in 50% PEG 1500 for further 1 to 2 min.
❼	Add 1 ml medium pre-warmed to 37°C to the fusion mixture, continuously stirring as before, over a period of 1 min.
❽	Add 3 ml medium pre-warmed to 37°C over a period of 3 min, continuously stirring the cells.
❾	Add slowly 10 ml medium pre-warmed to 37°C.
❿	Incubate for 5 min at 37°C.
⓫	Spin the cells down.
⓬	Discard the supernatant and resuspend the pellet in selection medium, *e.g.,* RPM1 1640, 10% FCS (fetal calf serum) (v/v), non essential amino acids (1×), 2 mM glutamine, 1 mM sodium pyruvate, HAT-media supplement* (1 ×), 10% BM Condimed1) H1 * (v/v) or 50-100 U/ml IL-6*.
⓭	Seed cells as usual.

3. Additional Information on this Product

3.1 How this Product Works

Typical analysis

Peroxides and aldehydes not detectable; free from Ca^{2+}.

Formula

$HO\ (CH_2CH_2O)_nH$

Molecular weight

1,500 Da.

Biological activity

PEG 1500 is biologically evaluated for high fusion efficiency.

4. Supplementary Information

Ordering Information

For a complete overview of related products, please visit http://www.lifescience.roche.com

Changes to Previous Version

- Addition of information note to the section "How to Use this Product".
- Links and contact information updated.
- Editorial changes.

Text Conventions

To make information consistent and understandable, the following text conventions are used in this Instruction Manual:

Text Convention	Use
Numbered instructions labeled ❶, ❷, etc.	Steps in a procedure that must be performed in the order listed.

Symbols

Symbols are used in this Instruction Manual to highlight important information:

Symbol	Description
🔍	Information Note: Additional information about the current topic or procedure.
⚠	Important Note: Information critical to the success of the procedure or use of the product.

Trademarks

BM CONDIMED is a trademark of Roche.

References

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To call, write, fax, or email us, visit **www.lifescience.roche.com**, and select your home country. Country-specific contact information will be displayed.

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