Instruction Manual

SERVA Electrophoresis

Collagen R Solution 0.2 % Cat. No. 47254

Product description:

General	Collagen is the major structural component of extracellular matrices found in connective tissues and internal organs, but is most prevalent in the dermis, tendons and bones. Type I collagen is a heterodimer composed of two α_1(I) chains and one α_2(I) chain that spontaneously forms a triple helix scaffold at neutral pH and 37 °C:
Application	• Excellent substrate for the cultivation of epithelial cells and a number of other cell lines • Propagation of cells which are not able to grow on glass or plastic surfaces[1-2] • Cell adhesion in culture media without serum or fibronectin[3-4] • Experiments in cell migration[5] • Changes in cell morphology in three dimensional collagen gels[6-7] • Morphological studies[8] • Preservation of differentiation status of higher cells *in vitro*[9-10] • Influence of substrate and cell morphology on DNA-synthesis and cell proliferation[11] • Development of tissue-like structures *in vitro* and the use in wound healing processes[12]
Composition	2 mg/ml acid soluble collagen (Type I) from rat trail in 0.1 % acetic acid
Storage	Store solution at +2 °C - +8 °C

Preparation of collagen gels:

Additional required material	• 10x medium, sterile (e.g. BME with Earle's BSS or MEM Eagle with Earle's BSS) • 0.34 M NaOH, sterile • Petri dishes (polystyrene or glass) of ca. 60 cm diameter
Pouring of collagen gels	1. Mix 20 ml 10x medium and 10 ml 0.34 M NaOH directly before use. 2. Dispense 1.7 ml Collagen R solution. Evenly on the bottom of the culture dish (you may have to dilute it with 0.1 % acetic acid). 3. Add NaOH/medium mixture until the color of the indicator changes from yellow to slightly pink (pH 7.0 -7.5) and turn the dish in circles for 15 seconds. 4. Let the gel set at room temperature or at 37 °C. Duration 15 – 60 minutes.

Coating of cell culture dishes with Collagen R:

Optimal conditions for attachment and growth must be determined for each cell line and application by the user.

Described is a 2 ml formulation.

Additional required material	• 9.0 % NaCl solution • 0.17 M NaOH, sterile • Petri dshes (Polystyrene or glass) of ca. 10 cm diameter
Coating	1. 0.2 ml 9.0 % NaCl solution 0.2 ml 0.17 M NaOH 1.6 ml Collagen R solution Mix 2. Coat Petri dish with the mixture evenly. 3. Place it in the incubator for at least 1 hour at 37 °C. 4. Aspirate excess fluid and wash 2x with e.g. PBS, pH 7.0. Cells can now be seeded.

Floating collagen membranes:

After sowing of the cells in the collagen layer, the collagen membrane can be removed from the bottom with a sterile spatula under circle movement of the culture dish and will then float in the medium as membrane.

Literature:

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Ver. 08/11

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