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3D Cell Culture Substrate MatriMix (511)
Culture methods -On gel culture, Sandwich culture-

MatriMix (511) comprises of three components: Solution A (DMEM, Laminin 511E8 fragment and hyaluronan), Solution B (sodium bicarbonate), and Solution C (mixture of pepsin-solubilized type I/III collagen and acid-extracted type I collagen). The three solutions are mixed to form a gel at 37°C prior to cell culture.

Instructions for use

I. Preparation of MatriMix solution

The mixed solution must be prepared before each use.

To change the collagen concentration of Solution C, please read "IV. How to dilute Solution C, collagen solution".

(1) Calculate the volume ratio to be 5.4:0.6:4.0 (Solution A: Solution B: Solution C = 5.4:0.6:4.0) according to the amount used.

(2) Add Solution B to the aliquoted Solution A[*1]. Then add solution C[*2]. The operation should be performed at low temperature.

*1 When Solution B is added to Solution A, the color changes from grayish blue-green to pink.

*2 Since the collagen solution in liquid C is highly viscous, do not remove the pipette tip from the liquid surface too early when pipetting. If the tip is pulled up too quickly, air will enter the tip end. After adding collagen solution, mix slowly and thoroughly.

(3) Store at low temperature until ready for use.

II. "On Gel culture"

The following is an example of culturing cells in a 24-well plate. If you change the culture plate/dish, adjust the volume of MatriMix solution.

(1) Preheat the culture vessel in a CO_2 incubator set at 37°C.

(2) Add 300 µL of MatriMix solution to cover the entire bottom of the culture plate.

(3) Allow to gel in a CO_2 incubator set at 37°C for at least 30 minutes.

*Before gelation, move the culture plate gently and carefully.

*When gelation occurs, the color of the gel becomes slightly cloudy. If gelation does not occur, extend the incubation time.

(4) Prepare the required number of cells.

(5) Gently seed the cells onto the gel.

(6) Incubate in a CO_2 incubator set at 37°C until cells adhere to the gel.

(7) Gently add medium along the well wall to prevent the gel detachment. Move the plate slowly into the incubator.

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III. "Sandwich culture"

The following is an example of culturing cells in a 24-well plate. If you change the culture plate/dish, adjust the volume of MatriMix solution.

(1) Preheat the culture vessel in a CO_2 incubator set at 37°C.
(2) Add 300 µL of MatriMix solution to cover the entire bottom of the culture plate.
(3) Allow to gel in a CO_2 incubator set at 37°C for at least 30 minutes.
*Before gelation, move the culture plate gently and carefully.
*When gelation occurs, the color of the gel becomes slightly cloudy.
If gelation does not occur, extend the incubation time.
(4) Prepare the required number of cells.
(5) Gently seed the cells onto the bottom gel.
(6) Incubate in a CO_2 incubator set at 37°C until cells adhere to the bottom gel.
(7) Gently remove the medium on the bottom gel.
(8) Add 300 µL of MatriMix solution over the cells adhered to the bottom gel.
(9) Allow to gel in a CO_2 incubator set at 37°C for at least 30 minutes.
*Before gelation, move the culture plate gently and carefully.
*When gelation occurs, the color of the gel becomes slightly cloudy.
If gelation does not occur, extend the incubation time.
(10) Gently add medium along the well wall to prevent the gel detachment. Move the plate slowly into the incubator.

IV. How to dilute solution C, collagen solution

To adjust gel stiffness, Solution C, collagen solution, can be diluted up to x1/2.

(1) Prepare cold sterile water.
(2) Dispense sterile water into a new tube to achieve the volume ratio set in the maximum 1/2-fold dilution range.
(3) Dispense the required volume of Solution C and mix thoroughly with cold sterile water on ice.
(4) Prepare MatriMix solution according to **I. Preparation of MatriMix solution** above.

If you need a higher concentration of collagen, or want to mix other molecules (such as growth factors or other extracellular matrix components), please contact us at MatriMix@nippi-inc.co.jp.

V. Other Information

If you have any questions, please refer to the MatriMix website (https://matrimix.nippi.bio/) for FAQs or contact us at MatriMix@nippi-inc.co.jp.