TheWell BIOSCIENCE
Growing Cells in New Dimensions

Product Data Sheet

Revision v1.03

VitroGel® RGD High Concentration

Catalog Number: TWG003

Usage restrictions: For Research Use Only. Not For Use In Diagnostic Procedures.

Product Description

VitroGel® RGD High Concentration is a tunable, xeno-free hydrogel system modified with cell adhesive peptide RGD to promote cell attachment and cell-matrix interactions during the 3D cell culture. VitroGel RGD High Concentration comes with VitroGel Dilution Solution to adjust the final hydrogel strength from 10 to 4000 Pa.

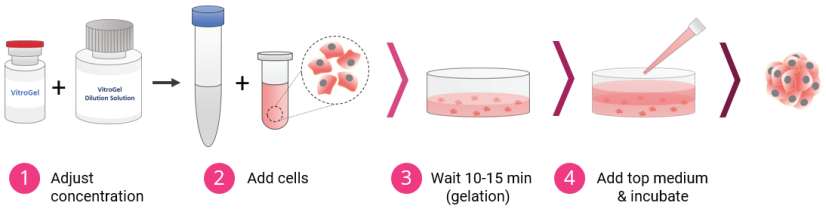
VitroGel® High Concentration hydrogels are our xeno-free, tunable hydrogels for researchers wanting full control to manipulate the biophysical and biological properties of the cell culture environment. The tunability of the hydrogel gives the ability to create an optimized environment for cell growth. The hydrogel system has a neutral pH, transparent, permeable and compatible with different imaging systems. The solution transforms into a hydrogel matrix by simply mixing with the cell culture medium. No cross-linking agent is required. Cells cultured in this system can be easily harvested for futher studies. The hydrogel is also injectable for in vivo studies. From 3D cell culture, 2D coating to animal injection, VitroGel makes it possible to bridge the *in vitro* and *in vivo* studies with the same platform system.

"Mix & Match" Unique to the VitroGel High Concentration hydrogels are its ability to be blended with other different types of VitroGel to create a customized multi-functional hydrogel.

SPECIFICATIONS	
Contents	VitroGel® RGD High Concentration, 3 mL VitroGel® Dilution Solution, 50 mL
Use	Good for adhesion cells or cells requiring stronger cell-matrix interactions.
Formulation	Xeno-free. Polysaccharide based hydrogel modified with RGD peptide.
Hydrogel strength	10 - 4,000 Pa of G' depending on dilution ratio. Use VitroGel Dilution Solution.
Physical State	Liquid
pH	Neutral
Cell Recovery	Use VitroGel® Cell Recovery Solution (Cat# MS03-100)
Storage	Store hydrogel at 2-8°C. Ships at ambient temperature.
Stability	24 months from date of manufacture
Uses	200 uses at 1:3 dilution for 96 well plate

VitroGel High Concentration Workflow

VitroGel High Concentration hydrogels are easy-to-use. There is no cross-linking agent required. Work confidently at room temperature.



Protocol

Visit www.thewellbio.com/faq-hydrogel for frequently asked questions on cell culture preparation and operation

Full protocol and video demonstrations can be found at > **www.thewellbio.com/protocols**

1. Bring VitroGel to room temperature and warm cell culture medium to 37°C if needed.

2. Adjust the concentration of VitroGel for different cell types by diluting the VitroGel with VitroGel Dilution Solution. After dilution, gently mix the diluted VitroGel with a cell suspension (in the desired media) without introducing bubbles.
   (Recommend cell concentration of 0.5-2 x 10^6 cells/mL)
   See Table 1 below for suggested solution/medium volume of different dilutions.

   Table 1. Volumes of solution/medium for different hydrogel dilutions for 3D cell culture (each well of a 24-well plate)

Dilution Ratio	VitroGel	Dilution Solution	Cell Medium with Cells
1:0	240 µL	0 µL	60 µL
1:1	120 µL	120 µL	60 µL
1:2	80 µL	160 µL	60 µL
1:3	60 µL	180 µL	60 µL
1:5	40 µL	200 µL	60 µL

If cells are to be cultured in complete cell culture medium with 10% FBS or other critical growth factors/supplement, prepare the cell suspension by following the step below:

a. Prepare 100% FBS with 10X of critical growth factors.
b. Prepare cells in regular 1X cell culture medium. (Do not make the medium at a high concentration as the ionic molecules would affect the hydrogel formation.)
c. Mix the solution from step a) and b) to get cell suspension in 50% FBS with 5X critical growth factors
d. Mix the diluted VitroGel with cell suspension at 4:1 v/v ratio (eg.400 µL diluted VitroGel with 100 µL cell suspension).

Note: If the cells need to culture at a higher FBS concentration (eg. 20%), prepare cells suspension directly in 100% FBS. Prepare the diluted VitroGel by mixing VitroGel with VitroGel Dilution Solution and wait 30-60 min before mixing it with cell suspension. Wait 20-30 min at room temperature (or 37°C) before adding the cover medium on top.

3 Transfer the hydrogel mixture to a well plate. Gently tilt/swirl the well plate to ensure there is an even coating on the bottom of each well.

Table 2. Recommended hydrogel volume for WELL PLATES

WELL PLATE	Volume of hydrogel (µL)	Volume of Cover Medium (µL)
6 well plate	1200	1200
12 well plate	600	600
24 well plate	300	300
48 well plate	150	150
96 well plate	75	75

Table 3. Recommended hydrogel volume for PLATE INSERTS

PLATE INSERTS	Volume of hydrogel (µL)	Volume of Cover Medium (µL)
6 well plate	800	800
12 well plate	400	400
24 well plate	200	200
48 well plate	100	100
96 well plate	50	50

4. Wait 10-20 min at room temperature for a soft gel formation. Note: During the hydrogel forming process, do not disrupt the hydrogel by tilting or shaking the well plate.
5. After soft gel formation, GENTLY tilt the well plate to check if hydrogel has formed and attached firmly to the bottom of the well plate.
6. Carefully cover hydrogel with additional medium to further stabilize the hydrogel. See Table 2 or Table 3 for recommended volume of cover medium.
7. Place the well plate in an incubator and change the cover medium every 48 hours.
   Note: We recommend to only change 60-80% of the top medium without disturbing the hydrogel.

Related Products

- VitroGel Cell Recovery Solution (MS03-100)
- Other versions of VitroGel High Concentration - www.thewellbio.com/hc-hydrogels

References

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2. Wang, F., Nan, L., Zhou, S., Liu, Y., Wang, Z., Wang, J., Feng, X., & Zhang, L. (2019). Injectable Hydrogel Combined with Nucleus Pulposus-Derived Mesenchymal Stem Cells for the Treatment of Degenerative Intervertebral Disc in Rats. Stem Cells International, 2019, 1–17. https://doi.org/10.1155/2019/8496025

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4. Huang J. 3D Cell Culture on VitroGel System. HSOA Journal of Cytology and Tissue Biology. https://doi.org/10.24966/CTB-9107/S1001

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