

## Noviogel-RGD

### Introduction

Noviocyte's core technology is a polyisocyanopeptide (PIC) hydrogel, which displays several unique properties. PIC hydrogels are an improved platform for cell studies, because they combine the unique benefits of both natural and synthetic biomaterials. PIC hydrogels mechanically mimic collagen, while possessing the reproducibility and tunability of synthetic hydrogels. The porous fibrous polymer network has a stiffness that increases under applied force similar to collagen or fibrin.

PIC hydrogel is fully reversible thermo-sensitive which allows for ease of cell/tissue recovery and - downstream processing after culturing. By adding peptide sequences or growth factors, the hydrogels can be readily functionalized for different cell types. The unique combination of tunable bio functionality and biomechanics of the PIC hydrogels makes them excellent matrices for 3D stem cell culture or regenerative medicine.

### Formulation

15 mg of Noviogel-RGD powder is shipped at ambient temperature. Store at -20 °C upon arrival. *We recommend making a stock solution of 5mg/ml, this stock solution has a shelf life of 4 months at -20°C.*

- Before opening the vial with polymer, let it slowly reach room temperature to avoid moisture condensation.
- Add the desired volume of cold solvent (e.g. cell culture medium or PBS at 0-4 °C ) to the polymer, shake manually and incubate 3 hours on ice or at 4 °C. Manual shaking/swirling once every hour could speed up dissolving.
- After 3 hours, place the vial on ice and pipet gently but thoroughly up and down to homogenize the solution.
- If air bubbles are present, these can be removed by centrifuging, preferably in a cooled centrifuge at 4 °C
- Prepare aliquots and store at -20 °C until use. This stock solution has a shelf life of 4 months at -20°C

This protocol is suitable for concentrations up to 5 mg/ml.

Warning: Do not vortex the Noviogel solution.

Storing the solution for longer periods of time at 4 °C causes a lower stiffness after gelation (circa 5% loss after 16 hours)

### Application Cell Culture

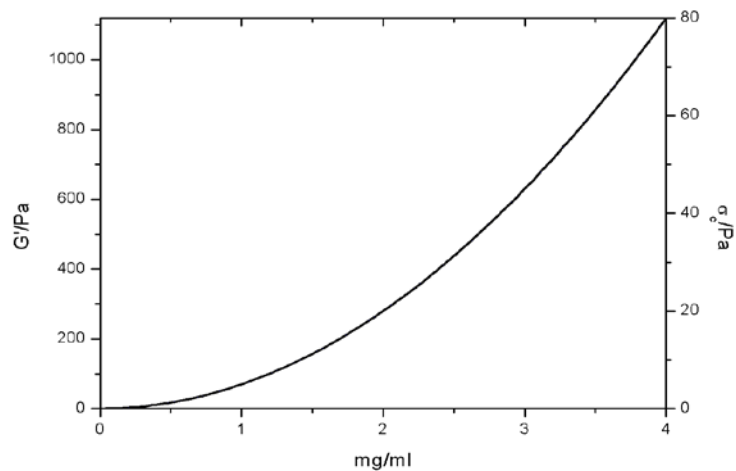
1. Advice: put your well plates at least a few hours before use at 37°C or even the day before.
2. Put your cell culture medium at 37°C.
3. Put your harvested cells and dissolved polymer on ice.
4. Add your cells to the polymer and mix it gently by pipetting up and down.
5. If you use a 24 wells plate then add 50 µl of cell / polymer suspension as a droplet on the surface of the well.
6. Start gelation by placing the plate for 10 minutes at 37°C.
7. The hydrogel with encapsulated cells have now been formed. Add gently 500 µl of cell culture medium to the well.
8. Culture the cells at 37°C and change cell culture medium when necessary but always in a gently fashion.

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9. After cell culture, cells or cell structures can be recovered by placing the plate at 4°C for approximately 10 min.
10. Centrifuge the solution and remove the hydrogel solution by pipetting.

In the table below, the concentration can be found that corresponds with the approximate value of  $\sigma_c$  and  $G'$ . The figure shows the relation between concentration,  $G'$  and  $\sigma_c$ .

mg/ml	$\sigma_c$ (Pa)	$G'$ (Pa)
1.0	5	70
1.2	7	100
1.4	10	140
1.6	13	180
1.8	16	225
2.0	20	280
2.2	24	340
2.4	29	405
2.6	34	470
2.8	39	550
3.0	45	630
3.2	51	710
3.4	58	810
3.6	65	910
3.8	72	1010
4.0	80	1120



### Dilutions from the stock solution of 5mg/ml to get to above concentrations:

Sigma c	$\sigma_c=9$	$\sigma_c=15$	$\sigma_c=25$	$\sigma_c=100$
Stock Solution (5mg/ml)	ml stock solution to dilute to a final 1 ml			
Noviogel-RGD	0.262	0.338	0.436	0.872

### Storage and stability

Store at -20°C upon arrival. Prepare aliquots and store at -20°C until use. This stock solution has a shelf life of 4 months at -20°C.

### Precautions

For research use only. Not for use in or on humans or animals or for diagnostics. It is the responsibility of the user to comply with all local/state and federal rules in the use of this product. Noviocyte is not responsible for any patent infringements that might result from the use or derivation of this product.

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### Warnings

1. The PIC hydrogel is reversible thermosensitive, this means that the gel is a liquid at 4°C and a gel at 37°C. Also after gelation the hydrogel will return to solution at 4°C.
2. Always warm up your cell culture medium at 37°C before use, otherwise hydrogel becomes liquid.
3. Please handle the hydrogel with care and caution.
4. Do not use the vortex machine!

### References

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