

PROTOCOL

XENO-FREE INJECTABLE HYDROGEL

XENOGRAFT | IN VIVO STUDIES | CELL THERAPY

VitroGel® xeno-free hydrogels are excellent for injection and a superior alternative to the animal-based extracellular matrix or plant-based hydrogel for xenograft, targeted delivery, control release, and cell therapy for tissue engineering.



Room temp stable

Ready-to-use. All protocols at room temperature. (5-15 min prep time)



Xeno-free

100% animal origin-free system with batch-to-batch consistency.



Long Injectable Status

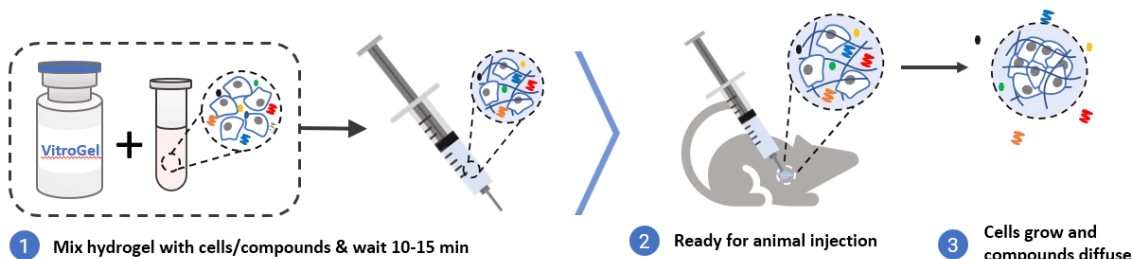
Maintain an injectable status for hours at room temp or at 37°C.



Full ECM Control

Full control of the ECM supplements to boost cell growth. Neutral pH. Bio-degradable and supports cell activities.

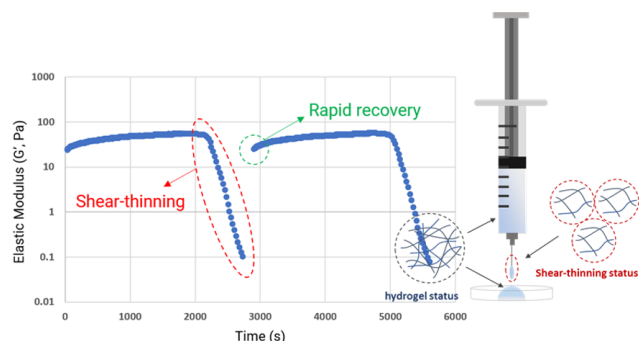
Simply mix the hydrogel solution with cells/compounds at room temperature and the hydrogel is ready for injection in 5-15 minutes. The system is biocompatible without showing a toxic or inflammatory response during the animal safety study.



Maintain an Injectable Status for Hours at Room Temperature or at 37°C

Researchers do not need to worry about the fast gel crosslinking and the rush for injection or putting the solution on ice. VitroGel has a unique rheological property that can maintain an excellent injectable status for hours after mixing with cells.

VitroGel hydrogel system has a unique shear-thinning and rapid recovery rheological property. After mixing the hydrogel solution and the cell medium, an injectable soft hydrogel will form. Under the mechanical shearing force such as injection through a syringe, the soft hydrogel performs a gel-sol transition and becomes free-flowing status. After injection, the mechanical strength can rapidly recover with a sol-gel transition and become a hydrogel status again. Such unique properties can enhance cell retention after injection.



TO LEARN MORE: <https://www.thewellbio.com/applications/in-vivo-studies/>

VIDEO EXAMPLE: <https://www.thewellbio.com/vitrogel-injection-xenograft-video-protocol/>



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For research use only

Growing Cells in New Dimensions

For Ready-To-Use Hydrogels

Animal Injection—EXAMPLE PROTOCOL

Applies to Cat. No.
VHM01, VHM01S, VHM02, VHM02S
VHM03, VHM03S, VHM04, VHM04S
VHM05, VHM05S, VHM06

All VitroGel hydrogels are injectable and excellent for xenografts. Researchers have full control of the supplement/growth factors in the hydrogel-cell mixture. **Simply adding 3-5X of FBS (or your key supplement) or 3-5% BSA/HSA to the cell suspension before mixing with hydrogel can boost the cell growth after injection.**

VitroGel Hydrogel Matrix (Cat# VHM01) is used as an example below. Any Ready-To-Use hydrogels can be substituted.

MATERIALS AND REAGENTS (AS AN EXAMPLE)

- VitroGel® Hydrogel Matrix
 - Cells or molecular compounds
 - Cell culture medium or PBS
 - Additional supplement (optional)
 - Conical tubes (15 mL or 50 mL)
 - Micropipette; low retention pipette tips
 - Syringe
1. Bring VitroGel hydrogel to room temperature or warm at 37°C.
 2. Prepare cell suspension in the cell culture medium.
 - Adjust the cell seeding concentration according to your experiment.
 - Adding 3-5X of FBS (or your key supplement) or 3-5% BSA/HSA to the cell suspension before mixing with hydrogel can boost the cell growth after injection.
 - If using VitroGel for molecular compounds injection, prepare the compounds in the buffer at 3X of the desired concentration. The compounds solution can then be mixed with the VitroGel hydrogel solution for a 1X final concentration in step 3.
 3. Add 1 mL VitroGel hydrogel solution to 500 µL cell suspension from step 2 and gently pipette up and down 5-10 times to mix thoroughly. (The recommended mixing ratios with other solutions are listed in the table below.)

Medium used to prepare cell suspension/drug solution	VitroGel	Cell suspension/drug solution
Cell culture medium	2 mL	1 mL
1X PBS	1 mL	0.5 - 1 mL

4. Transfer the hydrogel mixture to a syringe. Let mixture stabilize at room temperature for 5-15 min. The hydrogel is ready for animal injection. VitroGel has a unique rheological property that can maintain an injectable status for hours after mixing with cells. *Optional: After transferring the hydrogel mixture to the syringe, the hydrogel can be injected right away.*



For High Concentration Hydrogels

Animal Injection—EXAMPLE PROTOCOL

Applies to Cat. No.
TWG001, TWG003, TWG007, TWG008
TWG009, TWG010, TWG011

All VitroGel hydrogels are injectable and excellent for xenografts. Researchers have full control of the supplement/growth factors in the hydrogel-cell mixture. **Simply adding 3-5X of FBS (or your key supplement) or 3-5% BSA/HSA to the cell suspension before mixing with hydrogel can boost the cell growth after injection.**

VitroGel® MMP (Cat# TWG010) is used as an example below. Any High Concentration hydrogels can be substituted.

MATERIALS AND REAGENTS (AS AN EXAMPLE)

- VitroGel® MMP + VitroGel Dilution Solution
 - Cells or molecular compounds
 - Cell culture medium or PBS
 - Additional supplement (optional)
 - Conical tubes (15 mL or 50 mL)
 - Micropipette; low retention pipette tips
 - Syringe
1. Bring VitroGel hydrogel to room temperature or warm at 37°C.
 2. Prepare cell suspension or drug solution in cell culture medium (can also use 1X PBS or VitroGel Dilution Solution to substitute the cell culture medium).
 - Adjust the cell seeding concentration according to your experiment.
 - Adding 3-5X of FBS (or your key supplement) or 3-5% BSA/HSA to the cell suspension before mixing with hydrogel can boost the cell growth after injection.
 - If you use VitroGel for molecular compounds injection, prepare the compounds in buffer at 3-5X of the desired concentration. The compounds solution then can mix with VitroGel hydrogel solution to get 1X final concentration in step 3.
 3. Adjust the hydrogel concentration accordingly and then gently mix the adjusted VitroGel solution with cell suspension (drug solution) without introducing bubbles. The recommended ratios of different hydrogel dilutions and mixing are listed in the table below.

Type of medium to prepare cell suspension/ drug solution	Dilution Ratio	VitroGel	Dilution Solution	Cell Suspension/ Drug Solution	Waiting time for hydrogel stabilization
Cell culture medium	1:0	2 mL	0 mL	500 µL	5-15 min
	1:1	2 mL	2 mL	1 mL	
	1:2	2 mL	4 mL	1.5 mL	
	1:3	1 mL	3 mL	1 mL	
1X PBS	1:0	2 mL	0 mL	1-2 mL	5-15 min
	1:1	2 mL	2 mL	2-4 mL	
	1:2	2 mL	4 mL	2-4 mL	
	1:3	1 mL	3 mL	2-4 mL	



For High Concentration Hydrogels

Animal Injection—EXAMPLE PROTOCOL

CONTINUED

4. Transfer the hydrogel mixture to a syringe. Let mixture stabilize at room temperature for 5-15 min. The hydrogel is ready for animal injection. VitroGel has a unique rheological property that can maintain an injectable status for hours after mixing with cells. *Optional: After transferring the hydrogel mixture to the syringe, the hydrogel can be injected right away.*



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Page: 4

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