

## Scalable One-Step Synthesis of Gelatin-Dithiolane for Neural Tissue Engineering

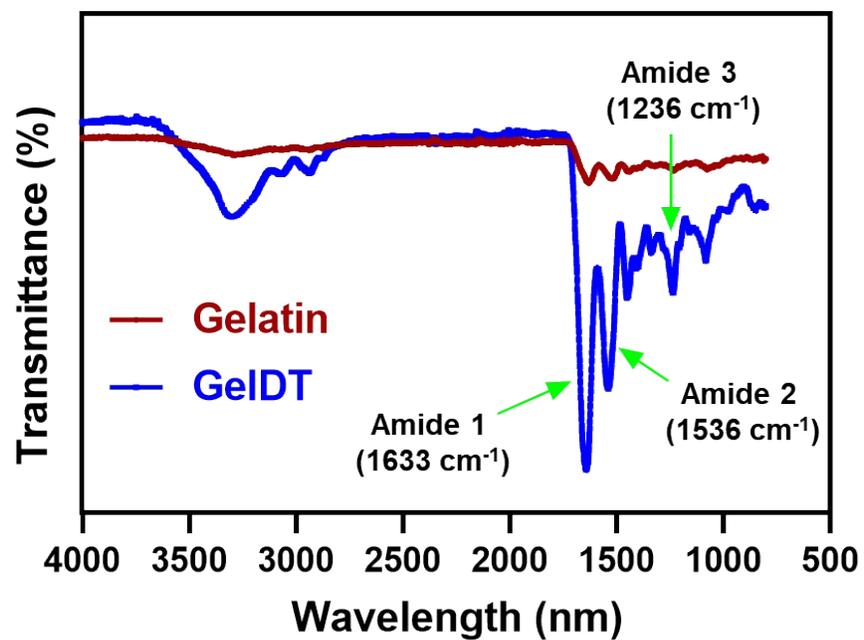
Muhammad Waqas Ishaq<sup>1</sup>, Asma Talib Qureshi<sup>1</sup>, Saad Asim<sup>1</sup>, Akanksha Subbarao<sup>1</sup>, Muhammad Rizwan<sup>1,2,3\*</sup>

<sup>1</sup> Department of Biomedical Engineering, University of Texas Southwestern Medical Center, Dallas, Texas 75390, USA

<sup>2</sup> Department of Ophthalmology, University of Texas Southwestern Medical Center, Dallas, TX, 75390 USA

<sup>3</sup> Harold C. Simmons Comprehensive Cancer Center, University of Texas Southwestern Medical Center, Dallas, TX, 75390 USA

\* Corresponding author. Dr. Muhammad Rizwan. Contact: [muhammad.rizwan@UTSouthwestern.edu](mailto:muhammad.rizwan@UTSouthwestern.edu)



**Fig. S1. FTIR spectrum of GeIDT.** GeIDT retained characteristic amide bands: amide 1 (1633  $\text{cm}^{-1}$ ), amide 2 (1536  $\text{cm}^{-1}$ ) and amide 3 (1236  $\text{cm}^{-1}$ ) found in gelatin's backbone.

**Table S1:** Dialysis pattern for synthesis of GelDT in DMSO/MES (2:1) as solvent.

Dialysis pattern for reaction in DMSO/MES (2:1) solvent	Start	First change	Second change	Third change
Day 1	MilliQ water, pH 9			
Day 2	MilliQ water, pH 7.4			
Day 3	MilliQ water, pH 7.4			
Day 4	MilliQ water, pH 7.4	MilliQ water, pH 7.4	MilliQ water, pH 7.4	<b>Freeze dry</b>

**Table S2:** Dialysis pattern for synthesis of GelDT in carbonate-bicarbonate (CB) buffer as solvent.

Dialysis pattern for reaction in CB buffer as solvent	Start	First change	Second change	Third change
Day 1	MilliQ water, pH 9			
Day 2	MilliQ water, pH 7.4			
Day 3	MilliQ water, pH 7.4			
Day 4	<b>Freeze dry</b>			

### 10% GeIDT-3

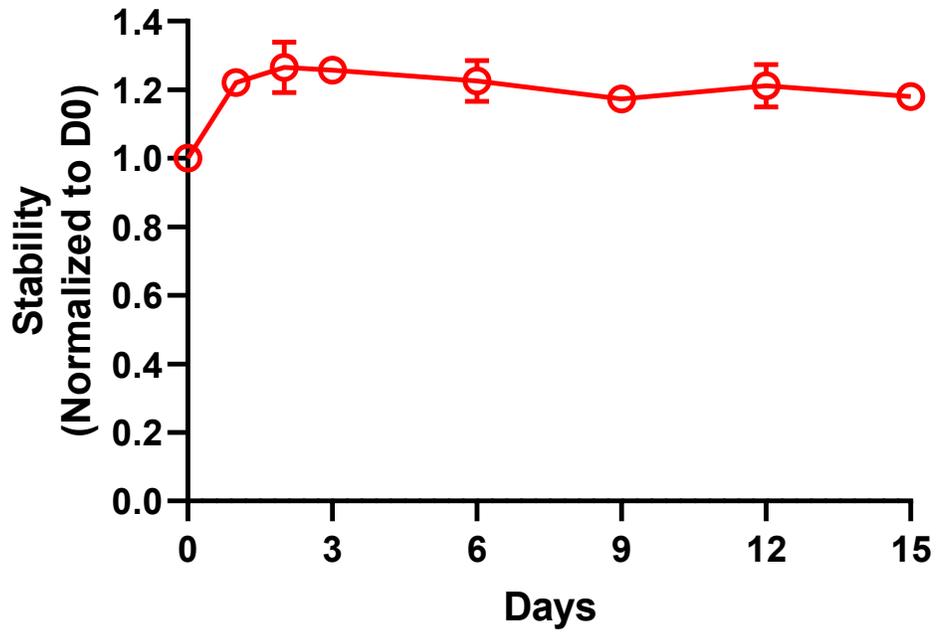


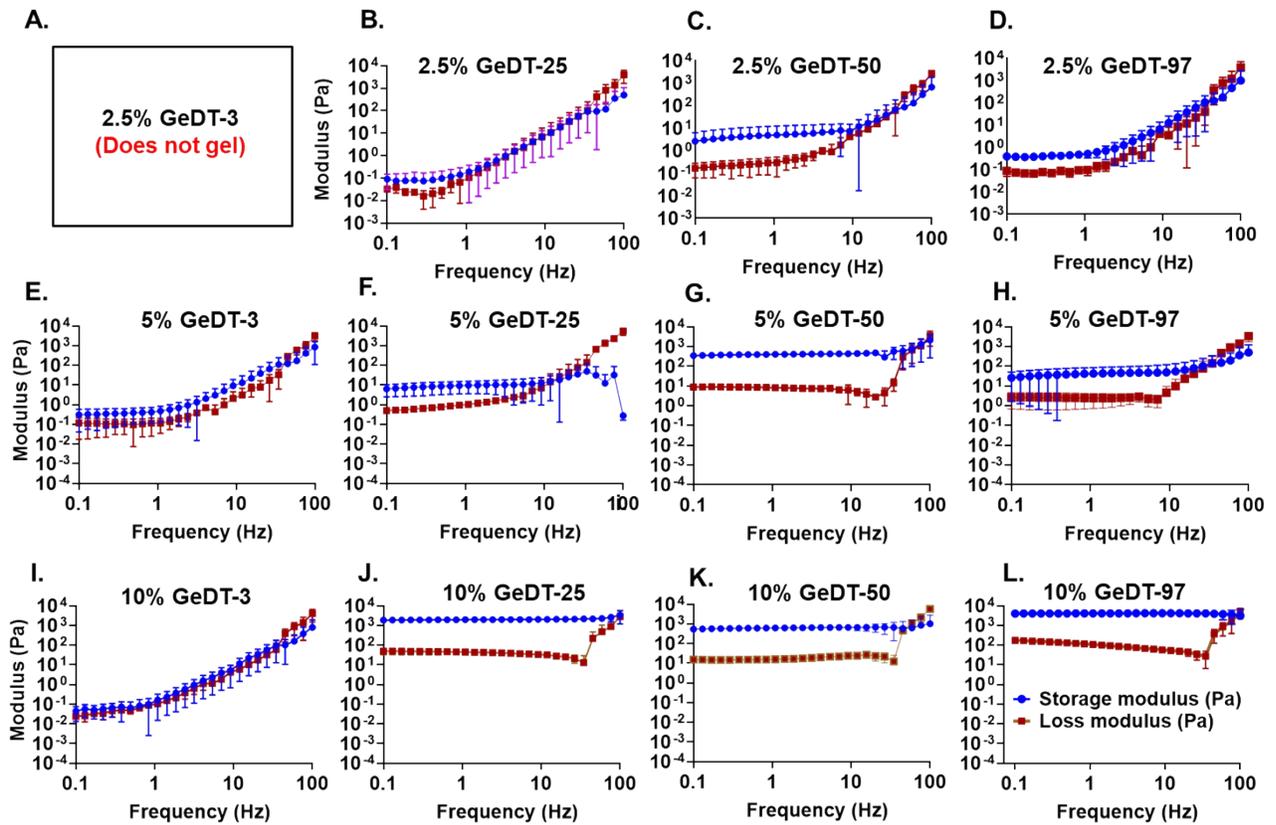
Fig. S2. Stability test of GeIDT-3 at 10% w/v in PBS + N<sub>3</sub>.

**Transparent**



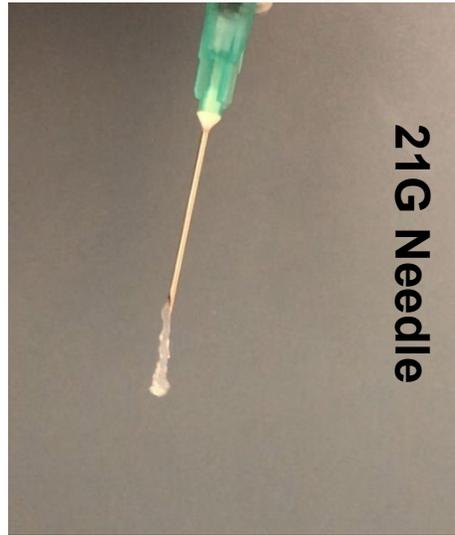
**5%GeIDT-97**

*Fig. S3. Transparency of GeIDT-97 at 5% w/v.*



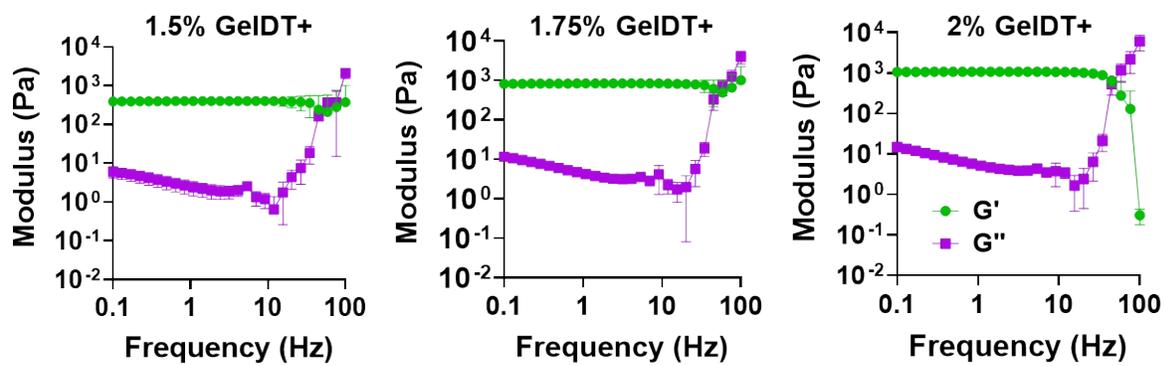
**Fig. S4.** Frequency sweep analysis of different DoF of GeDT ((DoF) 3, 25, 50, and 97) at 2.5-10% w/v.

**Injectable**

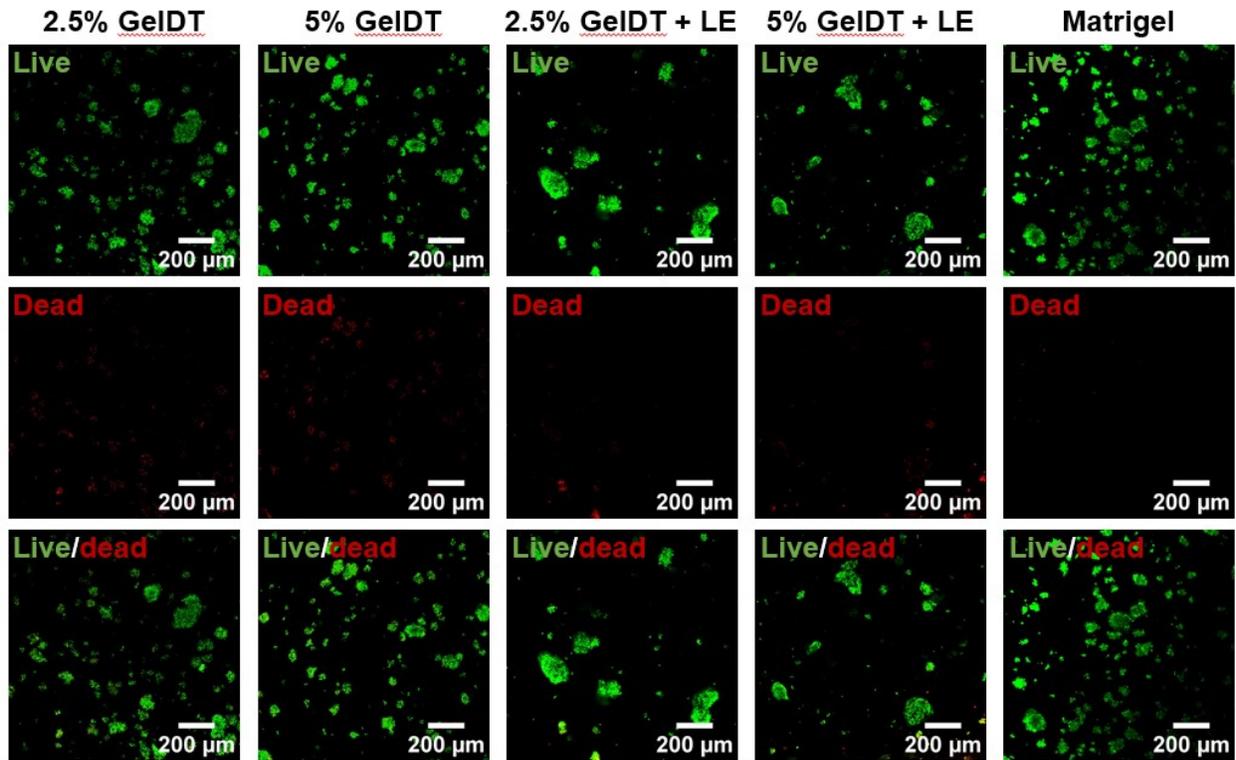


**5%GelDT-97**

*Fig. S5. Injectability behavior of GelDT-97 at 5% w/v.*



**Fig. S6.** Frequency sweep of physiochemically crosslinked GelDT+ hydrogels at ultra-low gelatin content (1.5 – 2%w/v).



**Fig. S7.** Live/dead assay on day 1 with PC-12 cells encapsulated in 2.5% GeIDT, 5% GeIDT, 2.5% GeIDT+LE and 5% GeIDT+LE hydrogels.