



Improving the Stability of Chitosan-Gelatin Based Injectable Hydrogel

for Cardiac Regeneration Therapy

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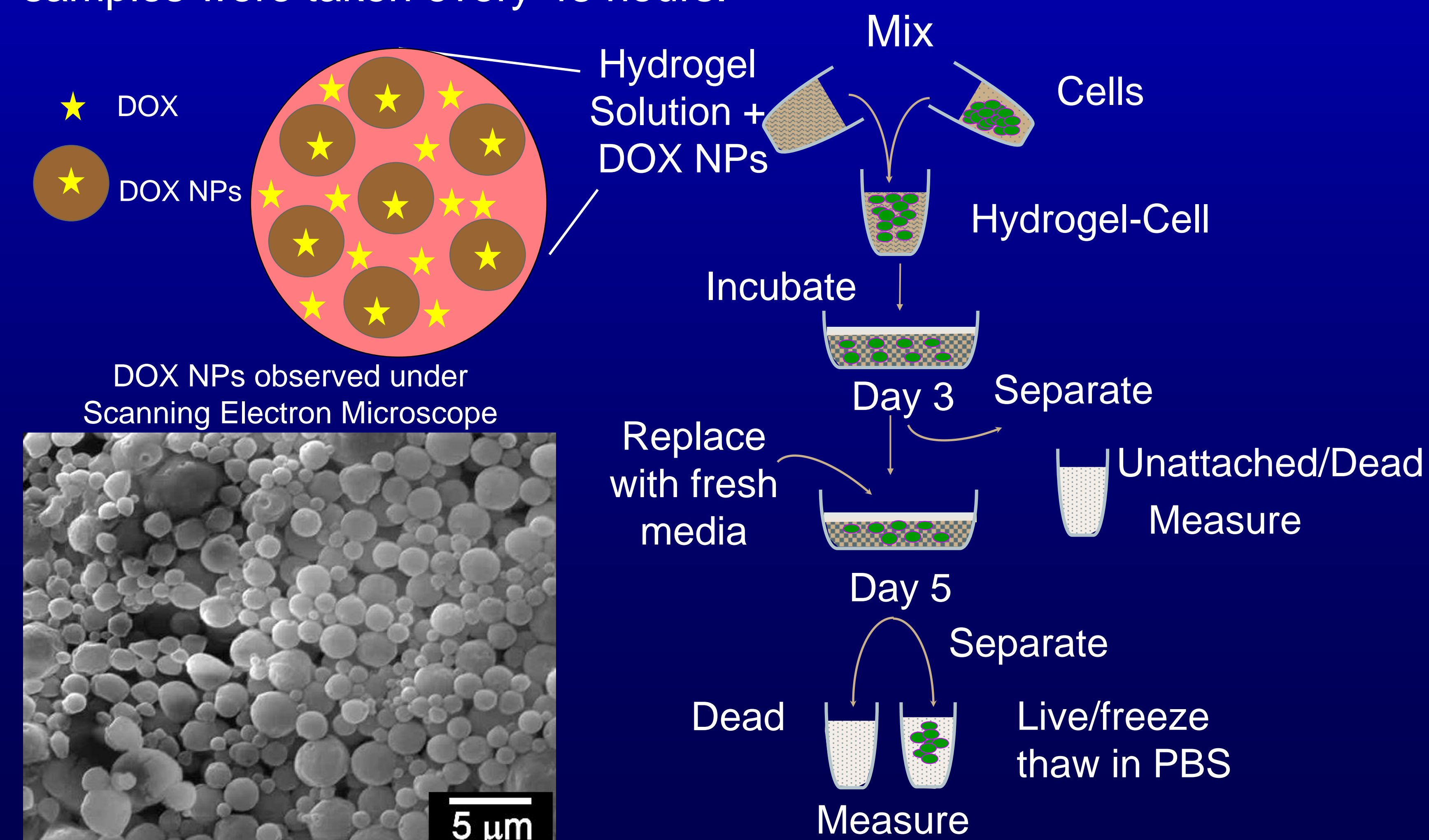
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INTRODUCTION

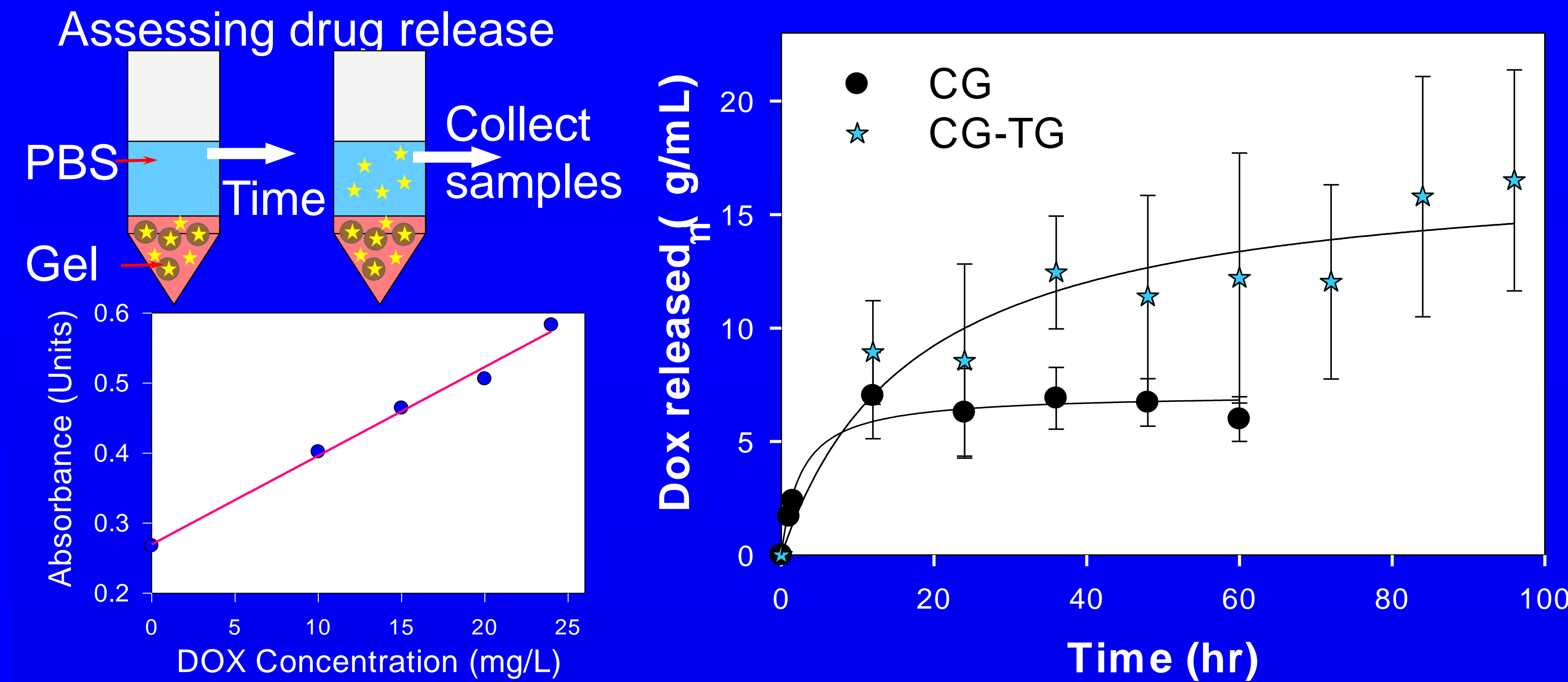
- Delivering cells using hydrogels to an injured area has been attractive option for in situ tissue regeneration.
- Clinical trials show significant attrition of injected human stem cells due to the poor stability of the hydrogel and lack of nutrients.
- To increase the stability of the resulting hydrogel and improve cell retention, Chitosan-gelatin (CG) with growth media was cross-linked with transglutaminase (TG).
- Cells secreted increased amounts of matrix metalloproteases (MMP-2/MMP-9) upon sensation of gelatin.
- Doxycycline hyclate (DOX), an antibiotic and inhibitor of MMP-2/MMP-9, was encapsulated in poly (lactic-co-glycolic acid) (PLGA) nanoparticles (NP) and embedded in the hydrogel.
- Release kinetics were determined and samples were evaluated for protein content.
- Cell viability was also determined by prestaining them with a fluorescent non-toxic dye.

Cell Culturing with the hydrogel

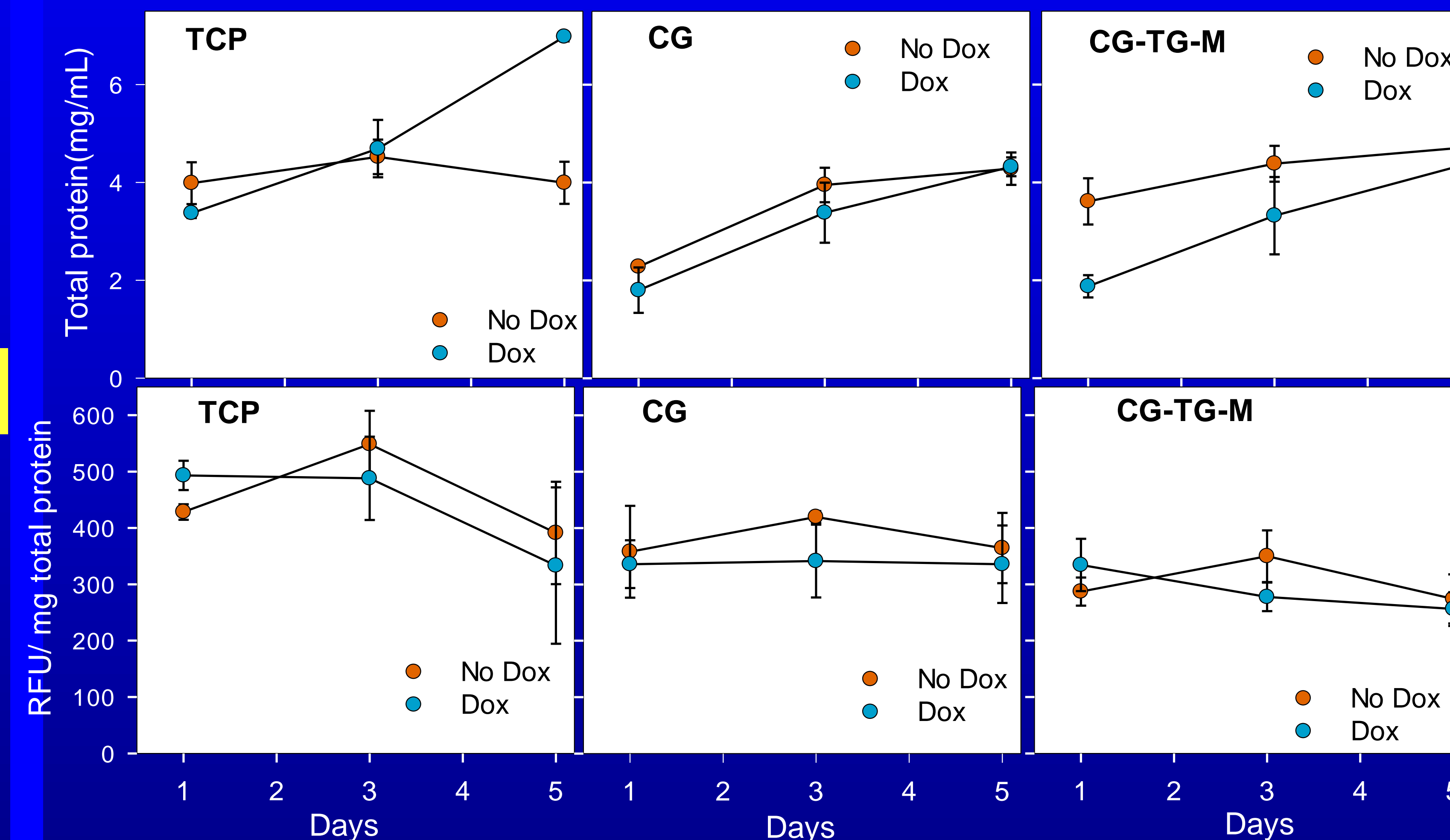
DOX NPs were prepared and added to 2% Chitosan Gelatin hydrogel along with cells. The resulting solution was incubated and samples were taken every 48 hours.



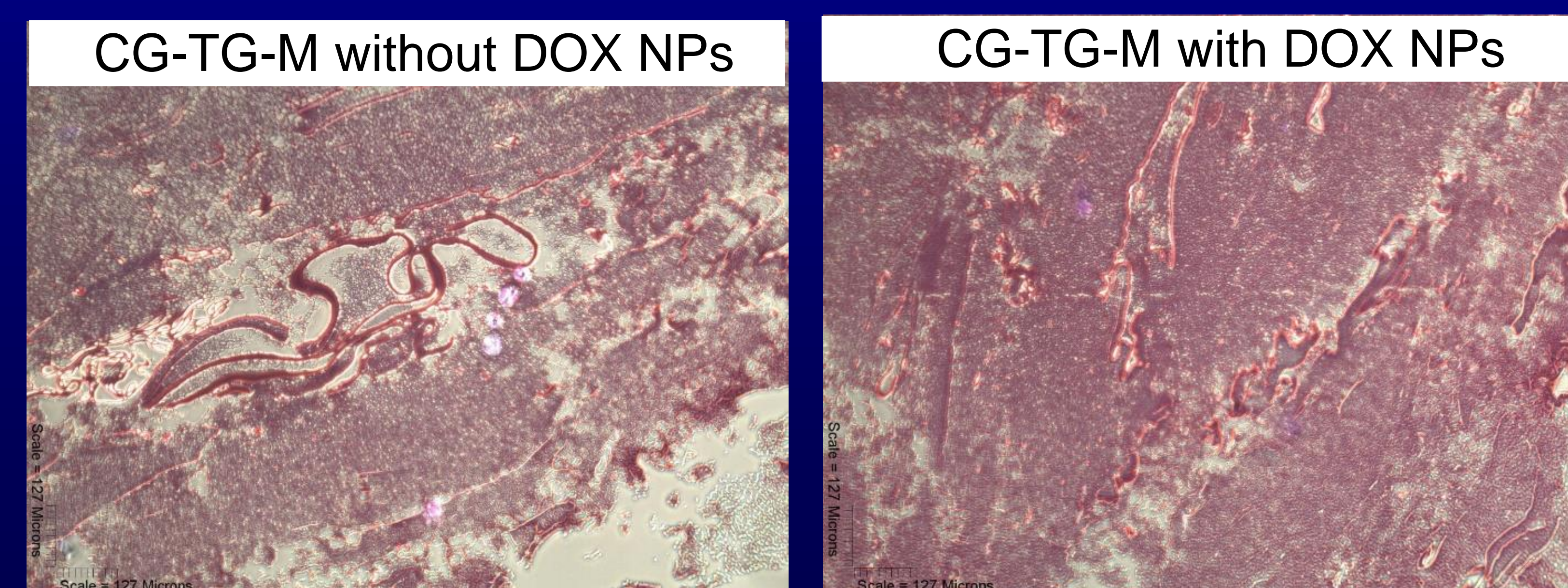
Improved DOX Release Time with TG



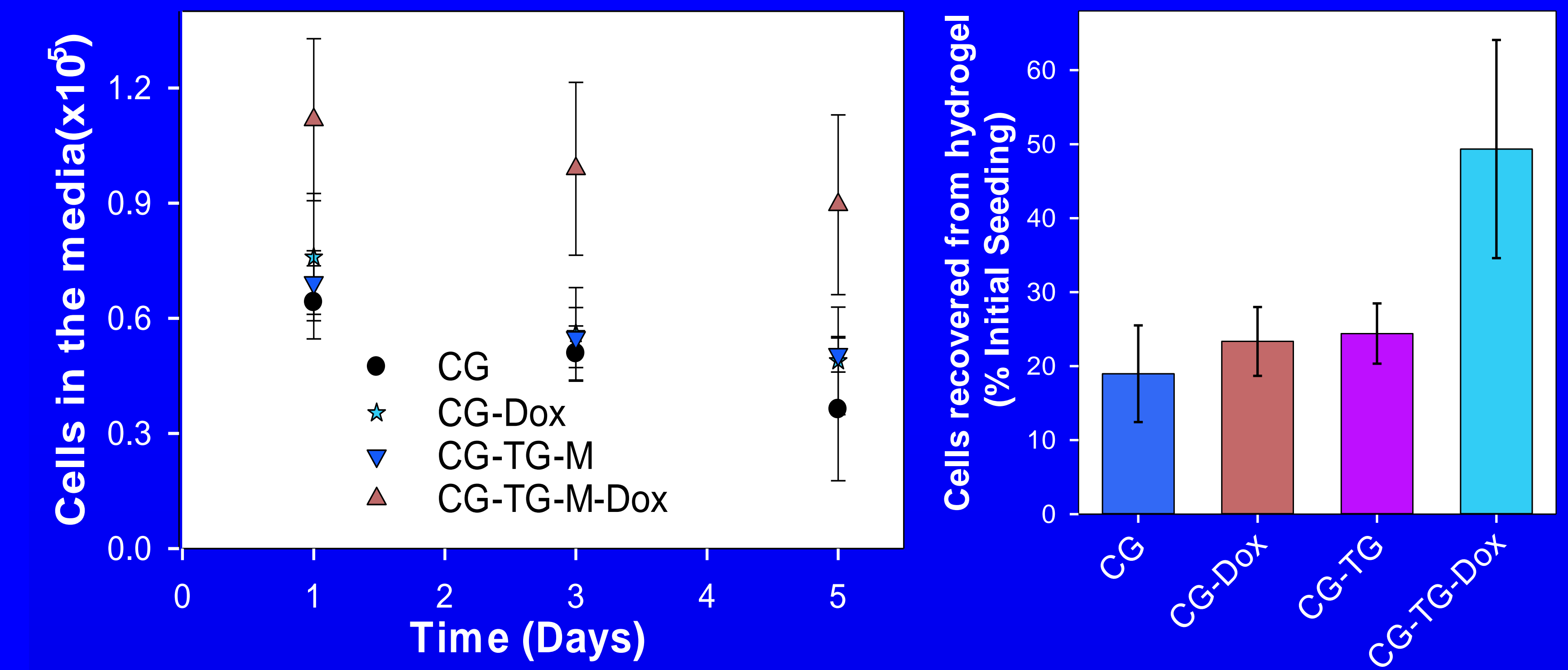
Prolonged Inhibition of Proteases by DOX



Histology of Hydrogels



Improved Cell Retention With DOX and TG



Societal Impact

With this research, this injectable hydrogel can be used to deliver human adult stem cells along with signaling molecules to stimulate stem cell differentiation in the cardiac region.

Summary

- DOX NPs were successfully formed and the particle size analysis showed 200-500 nm distribution, which was confirmed by SEM.
- Cross-linking of CG with TG showed an increased, sustained delivery of DOX as a function of time.
- Cell cultures showed no toxicity from DOX at the proposed concentration.
- Cell retention increased by over 250% with the combination of CG-TG-DOX. All cells were accounted for.
- Embedded DOX NPs decreased cell secretion of MMP-2/MMP-9.

Work in Progress

- Add signaling molecule involved in direct stem cell differentiation to injectable hydrogel.
- Testing long-term viability of human adipocyte stem cells.

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