Title: Thiol–ene click chemistry enables low-temperature gelation of norbornene-functionalized gelatin and PEG-thiol hydrogel

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Abstract
Radical crosslinking of a hydrogel typically requires complex setup or lifted temperatures to produce high concentrations of radicals needed for crosslinking. [1] However, thiol-ene click reaction is a highly efficient reaction requiring lower radical concentration and being highly specific crosslinking which is not affected by, for example oxygen molecules. [2] Herein we discovered a hybrid hydrogel that crosslinks in room-temperature via very low concentration of ammoniumpersulfate.
In this study gelatin molecule was functionalized with norbornene using EDC coupling reaction in water solution. Modification was quantified with 1H-NMR and TNBS assay with UV-spectroscopy. Low-temperature gelation of a hybrid hydrogel was initiated using 0.5 w% of ammoniumpersulfate initiator to utilize thiol-ene click reaction in hydrogel formation. Hydrogel polymer concentration of 5 w% was used in 1:1 conversion of gelatin-norbornene and polyethylene-glycol (PEG) -thiol which were mixed together and pipetted into a mold. After pipetting the hydrogel its gelation began spontaneously at room temperature in 25 to 30 minutes forming a clear and stable hydrogel. Gelation time was determined using rotational rheometry and tube tilt test. With higher temperature or initiator concentration the gelation was accelerated. Viscoelastic properties of the hydrogel were characterized with rotational rheometry showing linear behaviour with storage modulus of 1000 Pa at 1 Hz frequency for 0.1 to 16 % strain.
This thiol-ene click chemistry offers safe and easy hydrogel gelation mechanism at room temperature with a gelation time window suitable for applications such as cell encapsulation.

[1] Lin et al., 2024
[2] Muñoz et al., 2014