Supplementary material

1. Characterization of synthesized products

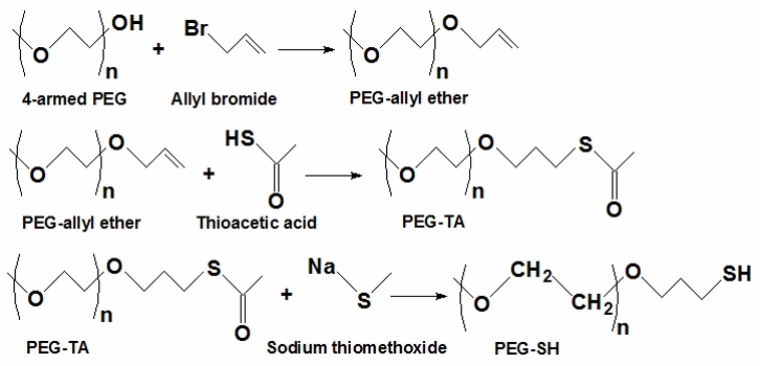

Figure 1. Schematic representation of three-step synthesis of PEG-SH.

As for the reaction of PEG alcoholate with allyl bromide，the product was confirmed by ^{1}H NMR (Figure 2a). Besides the signals from the PEG main-chain protons, new peaks appeared at δ 5.15-5.33 ppm and δ 5.87-5.98 *ppm* due to the double bond protons of allyl groups which confirmed the success of reactions. The second-step product PEG-TA was aynthesized by a radical addition reaction between thiol group and double bond whoes structure was proved by a strong signal at δ 2.33 ppm attributed to the thioacetate methyl protons (Figure 2b).

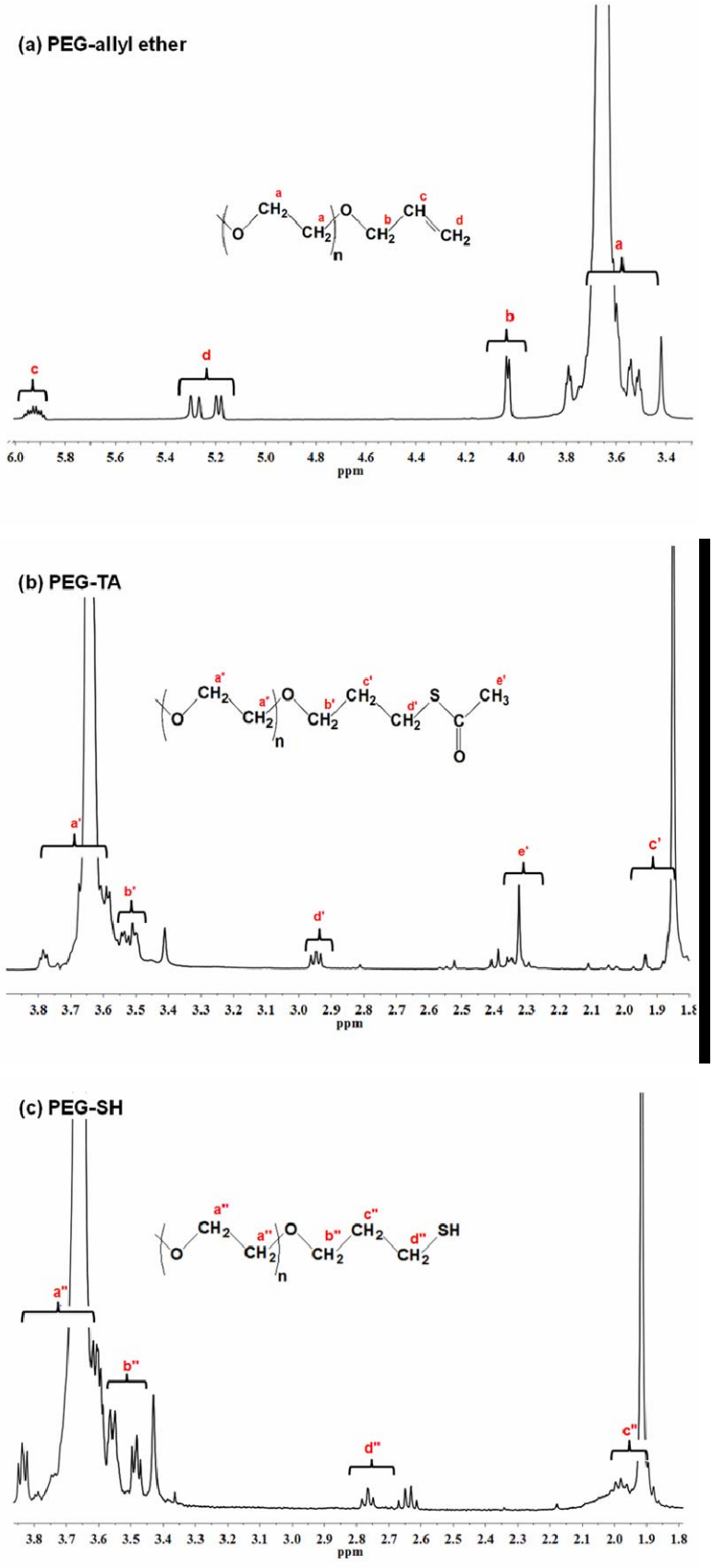

Figure 2. ^{1}H NMR spectra of synthesized products:
(a) PEG-allyl ether, (b) PEG-TA, (c) PEG-SH.

Thiol content was measured using Ellman's reagent method. 5,5'-dithio bis-(2-nitrobenzoic acid) (DTNB) can react with thiols and show yellow colour thus colorimetric methods can be ultilized for quantitative determination of thiols. Figure 3 showed the thiol conten standard curve with OD value as a function of thiol concentration. Extra pure cysteine was used as standard substance. Standard function of y=2.614x+0.01 (R=0.9998, x: thiol concentration, mM; y: OD value) was obtained and thiol content of PEG-SH was calculated under y=0.39. So we have the thiol concentration of 0.15 mM. As the concentration of PEG-SH solution was 1.0 mg/mL, we had the final thiol content of 0.15 mmol/g (-SH: mmol, PEG-SH: g).

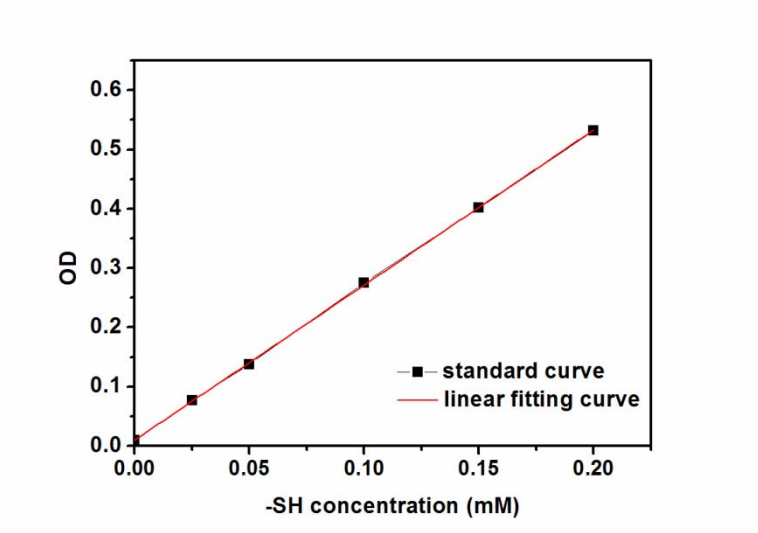

Figure 3. Standard curve of thiol concentration

2. Cell cytotoxity

The potential cytotoxicity of gels and degradation products was investigated with regards to their effects on in vitro viability of C2C12 cell by MTT on the sample of 10% concentration. Mouse myogenic cell C2C12 was cultured in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum (FBS) at 37 ºC and 5% CO_2. Cells were seeded onto a 96-well tissue culture plate at a

seeding density of 5000/well. Then, the medium was replaced with a fresh one containing serial extractives of gels and gel degradation products which were prepared under aseptic conditions as follows. 150 μL of precursors at 10% concentration were transferred in two parallel glass plates clamped together with a 0.8 mm gap and gelation was allowed to proceed at 37 ºC for 24h. A circular mold was used to cut the hydrogels into round disks with a diameter of 14 mm. Some of disks were incubated in DMEM for 24 h and the medium was used as extractive of gels. The others were incubated in DMEM with GSH (0.01 mM) till complete degradation and the degradation solution was used as extractive of degradation products. Cells were respectively incubated in the two extractives for 24 h before the cell number was determined by 3-(4, 5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay using a microplate spectrophotometer to measure optical density (OD) at 570 nm. Pure DMEM incubated cells were used for control. Cell viability was calculated by the percentage ratio of ODsample / ODcontrol ×100%. At least four replicates were tested for each variable.

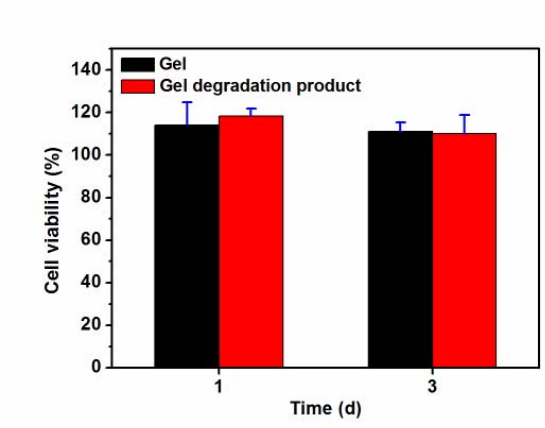

Figure 4. Cytotoxicity study by MTT method on 10 wt% hydrogel and hydrogel degradation extractives co-cultrued with C2C12 cells, 1 d and 3 d.

Both of the gel and its degradation products were found non-toxic in figure 4 with viability above 100% based on the formulation. In addition, PEG has already been extensively used as drug carrier or drug release modifier and proved to be nontoxic and safe. In terms of degradation products, the end groups were reduced to thoilates without significantly affecting the bulk PEG properties, which has been reported concerning with protein modifying PEG. Moreover, PEG with high molecular weight can be oxidized by cytochrome P450 to small molecules and then excreted through glomerular filtration.