Biomimetic Recyclable Microgels for On-demand Generation of Hydrogen Peroxide and Antipathogenic Application

Rattapol Pinnaratip[a], Hao Meng[a], Pegah Kord Forooshani[a], Pratik U. Joshi[b], Julie Osborne[a], Xue Mi[b], Christa Meingast[c], Jonathan Kelley[a], Ameya Narkar[a], Weilue He[a], Megan C. Frost[a], Caryn L. Heldt[b], Bruce P. Lee[a,*]

[a] Department of Biomedical Engineering, Michigan Technological University, Houghton, MI 49931, USA

[b] Department of Chemical Engineering, Michigan Technological University, Houghton, MI 49931, USA

[c] Department of Civil and Environmental Engineering, Michigan Technological University, Houghton, MI 49931, USA

Statement of Purpose: Hydrogen peroxide (H_2O_2) is commonly used as an oxidant in biomedical (i.e., disinfectant) applications due to its non-toxic degradation products (water and oxygen). However, diluted H_2O_2 is highly unstable while concentrated H_2O_2 is explosive. We prepared recyclable microgels using a biomimetic (reduction-oxidation) redox chemistry approach, which could be activated on-demand to generate H_2O_2. The marine mussels produced catechol adhesive containing a large amount of 3,4-dihydroxyphenylalanine (DOPA). The catechol side chain of DOPA is a redox-active adhesive moiety, capable of generating H_2O_2 while oxidizing to quinone. Catechol-incorporated microgels were synthesized through photo-initiated polymerization of dopamine methacrylamide (DMA) with N-hydroxyethyl acrylamide (HEAA) monomer. Our results indicate that millimolar concentration of H_2O_2 was continuously generated from these microgels for up to 4 days, which was dependent on catechol concentration, temperature, and pH of the solution. The microgels could be recovered for repeated use after a simple episode of washing in acidic water. The H_2O_2 generated from microgels was sufficient to kill both gram-positive (Staphylococcus epidermidis, S.epi) and gram-negative (Escherichia coli, E.coli) bacteria as well as inactivate both enveloped (Sindbis virus) and non-enveloped (Porcine parvovirus, PPV) virus. The tunable and repeatable production of H_2O_2 from this microgel makes it a potential candidate for different applications including surgical tool sterilizer, drinking water sanitizer or wound disinfectant in situations where refrigerated storage or modern conveniences (i.e., electricity, sterilization equipment) are not available.

Methods: For microgel preparation , catechol-containing microgels were prepared by photo-initiated polymerization of up to 10 mol% of dopamine methacrylamide (DMA), a hydrophilic monomer N-hydroxyethyl acrylamide (HEAA) and a bifunctional crosslinker methylene bis-acrylamide in an Emulsion (DMA microgel). For H_2O_2 release profile, the H_2O_2 concentration was quantified by using the Quantitative Peroxide Assay kit of the ferrous ion oxidation xylenol orange (FOX) assay. Foe antipathogenic activity, microgels were sterilized using ethylene oxide. The antipathogenic activity was tested against both gram-positive (Staphylococcus epidermidis, S. epi) and gram-negative (Escherichia coli, I) bacteria.

Results: DMA incorporated HEAA particle can be synthesized and store in a dry powder form. The particle readily hydrated and generated H_2O_2 in a neutral to mildly basic PBS solution (pH 6.7 or higher). There is no H_2O_2 generation observed at pH 3.5. The microgels can be repeatedly activated (pH 7.4) and deactivated (pH 3.5) for H_2O_2 generation (Figure 1.). The H_2O_2 generated was greater than 740 µM with a persistent release over 96 hour.

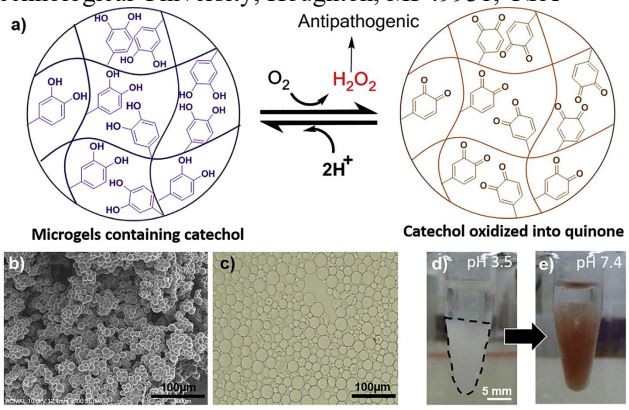

Figure 1. Catechol-containing microgels generate H_2O_2 during the autoxidation (a). SEM (b) and bright-field microscopy (c) images of the dried and hydrated microgels, respectively. Photographs of the microgels suspended in pH 3.5 DI water (d) and PBS (pH 7.4) (e) indicated that catechol only oxidizes (red color in panel e) in a basic solution. The dashed line in (d) encloses the microgels.

The amount of H_2O_2 Generated from the microgel completely killed both gram-negative and gram-positive bacteria strains within 24 h. Additionally, the microgel also reduced the infectivity of PPV by 3 log10 reduction value (LRV) and BVDV by 4-5 LRV after 12h.

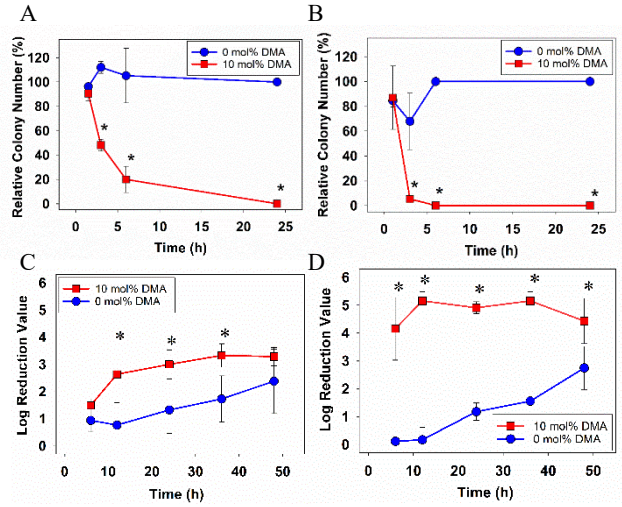

Figure 2. Relative colony number of S. epi (A) and E. coli (B) and LRV of PPV (C) and BVDV (D) with and without H_2O_2 generating microgel

Conclusions: The microgel can be repeatedly activated for an on-demand H_2O_2 generation by hydration in neutral pH.. The microgel in dried form and hydrated form under low pH does not contain H_2O_2, which minimize hazard associated with storage. The amount of H_2O_2 generated from the microgel was both bactericidal and veridical.