Biocompatible polymeric nanoparticles degrade and release cargo in response to biologically relevant levels of hydrogen peroxide

Caroline de Gracia Lux, Shivanjali Joshi-Barr, Trung Nguyen, Enas Mahmoud,
Eric Schopf, Nadezda Fomina, Adah Almutairi.
University of California, San Diego

Statement of Purpose: Methods of selective delivery of therapeutic and diagnostic reagents to sites undergoing oxidative stress would prove useful for the numerous diseases characterized by high concentrations of reactive oxygen species (ROS). Polymer-based nano- and microparticles are especially useful because they can be designed to degrade upon encountering ROS such as H_2O_2. To our knowledge, there are few if any polymeric systems able to undergo degradation and cargo release on encountering biologically relevant (50–100 μM) H_2O_2 concentrations. We sought to create such a nanoparticle by incorporating boronic esters into a polymer such that each H_2O_2-triggered deprotection would cause fragmentation. Such fragmentation in a polymeric nanoparticle would create pores, enabling cargo release.

Methods: The polymer (Fig. 1) was synthesized as described in our recent publication[1] and characterized by gel permeation chromatography (GPC). Nanoparticles were formulated by oil-in-water emulsification-evaporation and characterized by dynamic light scattering and scanning electron microscopy. Release kinetics were characterized by fluorescence measurements of nanoparticles encapsulating the solvatochromatic dye Nile red. Biological relevance was examined by measuring fluorescence upon exposure of nanoparticles encapsulating fluorescein diacetate (FDA) to PMA (phorbol 12-myristate 13-acetate)-activated neutrophils; FDA is cleaved by cellular esterases to fluorescein, so only released dye contributes to fluorescence. Cytotoxicity was assessed by ApoTox-glo assay (Promega).

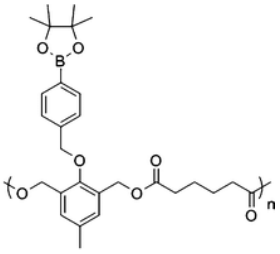

Figure 1. ROS-degradable polymer design.

Results: GPC of the polymer determined its molecular weight and polydispersity: 51.3 kDa, PDI = 1.4; DLS and SEM indicated that resulting nanoparticles were ~150 nm. Fluorescence measurements of Nile red-encapsulating particles revealed that cargo release in response to 50 μM H_2O_2 (Fig. 2). Exposure to activated neutrophils triggered ~2-fold greater release of FDA from the ROS-responsive polymer than from a control version lacking boronic ester groups (Fig. 3). Though activation induces only slight production of H_2O_2, local concentrations may be great enough to trigger polymer degradation. Apotoxglo results indicated that the polymer was equally as well tolerated by Raw264.7 macrophages as PLGA.

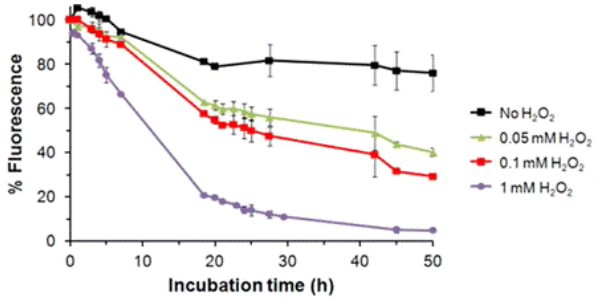

Figure 2. Release of Nile red from ROS-degradable polymer upon exposure to H_2O_2

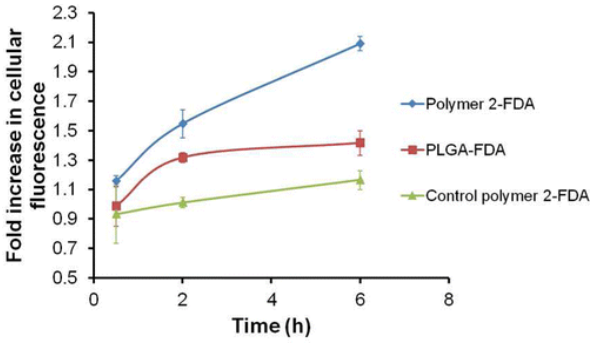

Figure 3. Release of FDA upon exposure to activated neutrophils.

Conclusions: This work represents a major advance in oxidation-responsive materials and paves the way for new imaging and drug delivery tools that can sense disease-relevant concentrations of this biochemical indicator.

References:

1. de Gracia Lux C et al., *J Amer Chem Soc* 2012; 134 (38), 15758–15764.