Tracking isotopically labeled oxidants using boronate-based redox probes

Natalia Rios^{1,2}, Rafael Radi^{1,2}, Balaraman Kalyanaraman³, and Jacek Zielonka^{3*}

From the ¹Departamento de Bioquímica, ²Centro de Investigaciones Biomédicas (CEINBIO), Facultad de Medicina, Universidad de la República, Montevideo, Uruguay; ³Department of Biophysics, Medical College of Wisconsin, Milwaukee, WI 53226, USA

SUPPORTING INFORMATION



Supplementary Figure S-1. Generation of peroxynitrite and incorporation of oxygen atom into the phenolic and nitrated products during oxidation of *o*MitoPhB(OH)₂ by ONOO⁻. (*a*) Chemical structures of the products; (*b*) online mass spectra of the detected products; and (*c*) LC-MS/MS traces of the probe and products detected. LC-MS/MS analyses were performed after incubation (30 min) of *o*MitoPhB(OH)₂ (20 μ M) alone (1), or with *in situ*-generated fluxes of O₂⁻⁻ (2, 0.2 μ M O₂⁻⁻/min, formed during XO-catalyzed oxidation of HX in the solution saturated with oxygen), 'NO (3, 0.2 μ M/min, formed from thermal decomposition of spermine-NONOate), ON¹⁶O¹⁶O⁻ (4, 6), or ON¹⁸O¹⁸O⁻ (5). ON¹⁶O¹⁶O⁻ and ON¹⁸O¹⁸O⁻ were produced by co-generated fluxes of 'NO and ¹⁶O₂⁻⁻ or ¹⁸O₂⁻⁻. All samples contained phosphate buffer (25 mM, pH = 7.4), dtpa (0.1 mM), and catalase (5 kU/mL) in H₂¹⁶O (samples 1-5) or in H₂¹⁸O (sample 6), purged with ¹⁶O₂ (samples 1-4, 6) or ¹⁸O₂ (sample 5).