Ratiometric Dual pH and Oxygen Sensors

Yanqing Tian, Liqiang Zhang, Fengyu Su, Hongguang Lu, Sean Buizer, Kevin Day, Jacob Messner, Deirdre R. Meldrum
Center for Biosignatures Discovery Automation, Biodesign Institute, Arizona State University, Tempe, AZ 85287

Statement of Purpose: pH and oxygen are two basic but important factors in cell biology, cell metabolism, and the formation of diseases and cancer. For example, it has been found that pH and oxygen play significant roles for wound-healing (1,2). We have been working on the development of optical sensors for pH, oxygen, potassium ions, and glucose as new biosensors and biomaterials for understanding cell metabolism (3-6). Herein, we will focus on our development of ratiometric dual pH and oxygen sensors for either extracellular sensing or intracellular sensing.

Methods: For extracellular sensing, the sensors are in the format of crosslinked polymeric hydrogels. For intracellular sensing, the sensors are in the forms of either amphiphilic block copolymer based nanostructured micelles or particles. Figure 1 shows a typical schematic drawing of the preparation of the hydrogel-based extracellular sensors with corresponding materials. The sensors were characterized using spectrophotometer and spectrofluorophotometer in buffer and/or cell culture medium.

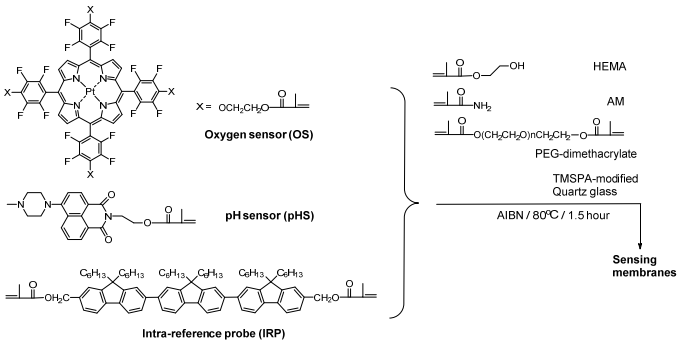

Figure 1. Typical chemical structures of the probes for tri-color ratiometric dual pH and oxygen sensor and the outline of the preparation of the sensing films. The sensor composes three probes - an oxygen probe (OS), a pH probe (pHS), and an internal built-in probe (IRP) which is inert to pH or oxygen. The probes were chemically immobilized in poly(2-hydroxyethyl methacrylate) (PHEMA)-containing polymer films (6).

Results: Figure 2A shows the emission spectra of a sensor film at different pH values. The blue emission at 421 nm from IRP doesn't change with pH value. The emission at 521 nm decreased with the increase of pH value, showing a good pH response of the sensing film due to the pHS. The red emission at 650 nm is from OS, which does not change with pH values. Figure 2B shows the emission spectral change of a sensor film under different dissolved O_2 concentration. The emission intensities at 421 nm and 521 nm have no change under the various oxygen conditions. A marked dependence of fluorescence intensities at 650 nm on O_2 concentrations was observed, showing the emission of the O_2 sensor was physically quenched by O_2. However, the IRP and pHS were not affected by O_2 concentration changes. These results showed that IRP is suitable as an internal reference probe for ratiometric analysis.

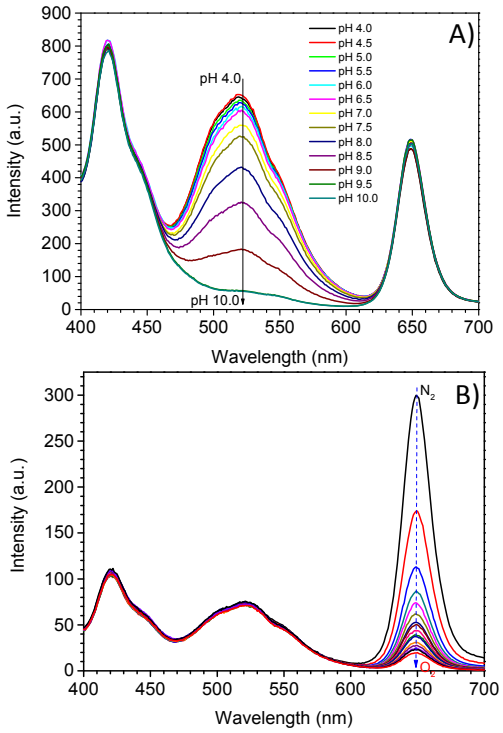

Figure 2. A) Emission spectral changes at different pH values in air saturated buffers (under oxygen partial pressure of 21 kPa corresponding to $[O_2]$ of 8.6 mg L^{-1}. B) Emission spectral changes at different dissolved oxygen concentrations (6).

Conclusions: We have synthesized and characterized ratiometric dual pH and oxygen sensors with triple emission colors. The use of ratiometric approach has been demonstrated to improve measurement accuracy in biological environments. Applications of these sensors for cell metabolism study and for cellular and molecular responses at the biomaterial-tissue interface are in progress.

References:

1. Schneider LA., et al. Arch Dermatol Res. 2007; 298: 413–420.
2. Schreml S., et al. Br J Dermatol. 2010; 163: 257-268.
3. Tian Y., et al. Chem Mater. 2010; 22:2069-2078.
4. Tian Y., et al. Biomaterials 2010; 31: 7411-7422.
5. Zhou X., et al. J Am Chem Soc. 2011; 133: 18530-3.
6. Lu HG., et al. J Mater Chem. 2011;21:19293-19301.

Acknowledgement: This work was supported by the NIH National Human Genome Research Institute, Centers of Excellence in Genomic Science, grant number 5 P50 HG002360, and NIH Common Fund LINCS program, grant number 5 U01 CA164250, Professor Deirdre Meldrum, PI.