<u>BRC Science Highlight</u> December 2019

A Photo-Regulated Aptamer Sensor for Spatiotemporally Controlled Monitoring of ATP in the Mitochondria of Living Cells

Background/objective

Monitoring cellular metabolite production is important for testing the efficacy of engineered microbes. In contrast to biological macromolecules such as nucleic acids and proteins, *in-situ* monitoring of metabolites such as ATP in living cells is much less advanced. Here, researchers describe a method for the design of a photo-regulated aptamer sensor for spatiotemporally controlled monitoring of ATP in the mitochondria of living cells.

Approach

- A photo-regulated ATP aptamer probe (PC-Apt) was designed to undergo photolysis upon irradiation with 365 nm light, exposing the ATP binding region.
- The stability and sensitivity of PC-Apt was tested in cell lysate.
- PC-Apt was packaged with DQAsomes, liposome-like vesicles that target the mitochondria in living cells.

Results

- The detection limit for PC-Apt was calculated to be 3.7 μM, while minimal fluorescence was detected in the presence of CTP, GTP, and UTP, establishing specificity for ATP.
- PC-Apt was stable in cell lysate for up to 12 hours and maintained its ability to bind ATP when photo-activated.
- Confocal laser scanning microscopy of cells incubated with the PC-Apt/DQAsome complex showed variation of red fluorescence upon irradiation at 365 nm reflecting fluctuation of ATP concentration.

Significance

This work represents the first successful delivery and implementation of a DNA aptamer sensor in mitochondria and, via the PC-Apt DQAsome system, provides a new platform for targeted aptamer delivery to organelles. These tools will help monitor energy production processes relevant to bioengineering applications for production of bioproducts.



Schematic of DNA probe/ DQAsome complex formulation (A) and depiction of targeted delivery of DNA probe/DQAsome to mitochondria (B).