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WATER-SOLUBLE FLUORESCENT POTASSIUM INDICATORS FOR CELL-BASED ASSAYS AND HIGH-THROUGHPUT SCREENING

Tech ID: 19062 / UC Case 2006-008-0

FULL DESCRIPTION

Potassium-sensing fluorescent indicators have applications in the measurement of cellular K+ content. For example, K+ sensors could be used to study K+ transport from K+ channels both in vivo and in vitro. K+ channels are important targets for drug discovery as they are involved in cardiac and neuronal excitability and epithelial fluid transport. Currently, patch clamp is the standard technique to assay K+ channel function. However, it is technically tedious, especially for high-throughput screening. There is thus a need for a robust assay for screening and cellular assays.

DESCRIPTION: UCSF investigators have synthesized a fluorescent K+ sensor, called TAC-red. The sensor is constructed so that the fluorescence of the compound is rendered sensitive to K+ binding. Thus, the fluorescence strongly increases in the presence of increasing K+ concentrations. Additionally, the compound is highly sensitive to K+, has a rapid response, and is water-soluble. The researchers also synthesized TAC-Crimson and TAC-Lime, both of which have similar properties to TAC-red. The investigators performed experiments demonstrating proof-of-concept that TAC-conjugated compounds can be used for in situ neurobiological assays to detect extracellular K+ levels (e.g. detecting differences in K+ concentrations in the extracellular space between communicating neurons) and simple, in vitro cell-based assays for high-throughput screening (e.g. for compounds that affect K+ efflux).

FEATURES/BENEFITS

- ▶ Water-soluble
- ► Multiple fluorophores
- ▶ High potassium sensitivity that facilitates detection of cellular K+ levels
- ► High K+ vs. Na+ selectivity
- ▶ Bright, long-wave fluorescence that minimizes background cellular fluorescence and photobleaching
- ► Insensitive to pH
- ▶ Rapid response especially useful for rapid neural signal transduction and ion channel gating
- ► Impermeable to cell membranes

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OTHER INFORMATION

KEYWORDS

potassium sensor, assay,

screening, high-throughput

CATEGORIZED AS

- **▶** Research Tools
 - ▶ Other
 - Reagents
 - Screening Assays

RELATED CASES

2006-008-0

- Low cellular toxicity
- Quantitative
- ► Can be used with commercially available plate-readers

APPLICATIONS

- ▶ Cell-based assays
- ► High-throughput screening
- ▶ Potassium sensing in the brain

STATE OF DEVELOPMENT

The investigators are currently working on a simpler, faster, cheaper, high-yield synthesis scheme for the TAC compounds.

PUBLICATIONS

- ▶ Padmawar et. al. (2005) Nature Methods 2, 825 827. K+ waves in brain cortex visualized using a long-wavelength K+-sensing fluorescent indicator.
- ▶ Magzoub et. al. (2006) J. Phys. Chem. B, 110 (42), 21216 -2122. Millisecond Association Kinetics of K+ with Triazacryptand-Based K+ Indicators Measured by Fluorescence Correlation Spectroscopy.
- Namkung et al. (2008) J. Am. Chem. Soc., 130 (25), 77947795. Cell-Based Fluorescence Screen for K+Channels and Transporters Using an Extracellular Triazacryptand-Based K+ Sensor.

PATENT STATUS

Country	Туре	Number	Dated	Case
United States Of America	Issued Patent	8,129,365	03/06/2012	2006-008

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