FLUORESCENCE RESONANCE ENERGY TRANSFER (FRET)-BASED SENSOR OF RANGTP-IMPORTIN B INTERACTION

Tech ID: 17268 / UC Case 2003-067-0

ABSTRACT

Scientists at the University of California Berkeley have designed FRET sensor of a complex of Ran-GTP-importin b that is a fluorescent protein construct consisting of Importin b-domain (IBB) of importin a flanked by fluorescent proteins (donor and acceptor) capable of FRET. The sensor functions as an indirect sensor of Ran-GTP through its Ran-GTP sensitive specific interaction with importin b.

See:

P. Kalab, Weis, K., and Heald, R. (2002), Visualization of a Ran-GTP Gradient in Interphase and Mitotic Xenopus Egg Extracts. Science, March 29, 2002, Vol. 295, 2452-2456.

The small guanosine triphosphate Ran is loaded with guanosine tripohosphate (GTP) by the chromatin bound guanine nucleotide exchange factor RCC1 and releases import cargoes in the nucleus during interphase. In mitosis, Ran-GTP promotes spindle assembly around the chromosomes by locally discharging cargoes that regulate microtubule dynamics and organization. We used fluorescence energy transfer-based biosensors to visualize gradients of Ran-GTP and liberated cargoes around chromosomes in mitotic Xenopus egg extacts. Both gradients were required to assemble and maintain spindle structure. During interphase, Ran-GTP was highly enriched in the nucleoplasm, and a steep concentration difference between nuclear and cytoplasmic Ran-GTP was established, providing evidence for a Ran-GTP gradient surrounding chromosomes throughout the cell cycle.

APPLICATIONS

spatially resolved and quantitative analysis of Ran-GTP-induced release of importin b cargoes.

readout system in screens for the modulators and inhibitors of Ran, RCC1, Ran GAP and other components of Ran system.

ADVANTAGES

The sensor functions as an indirect sensor of Ran-GTP through its Ran-GTP sensitive specific interaction with importin b.

CONTACT

Terri Sale
terri.sale@berkeley.edu
tel: 510-643-4219.

OTHER INFORMATION

KEYWORDS

research tool, vector

CATEGORIZED AS

» Research Tools

» Vectors

RELATED CASES

2003-067-0