Technical seminar

ProteoTuner™ systems: The new revolutionary tool to analyze your Protein function

Date and Time	:	**Thursday, February 7, 2008** **16:30 – 17:30**
Venue	:	Seminar Room A7F, RIKEN CDB
Speaker	:	Suvarna Gandlur, Clontech Laboratories, Inc.
Host	:	Hiroshi Tarui Ph.D., RIKEN CDB

Abstract

Determining the function of a particular protein is crucial to understanding complex biological processes. There are multiple approaches to regulating the level of a protein of interest in order to study its functions. To date, regulation has been carried out either at the cDNA level via inducible promoter systems, e.g. tetracycline inducible systems, or at the mRNA level with RNA interference (RNAi) methods. Until recently, it has not been possible to regulate the abundance of a specific protein at the level of the protein itself. Direct control at the protein level allows for immediate analysis of cause and effect in a matter of minutes, not days. Clontech's new ProteoTuner™ systems allow rapid, ligand-dependent stabilization and destabilization of a specific protein of interest, directly at the level of the functional protein.

The ProteoTuner™ systems are based on a short tag derived from a mutant of the human FKBP12 protein, referred to as the destabilizing domain (DD). The DD can be expressed as a fusion tag to any protein of interest. The tag's presence causes very fast proteasomal degradation of the exogenous fusion protein, down to a negligible level. To accumulate the DD fusion protein in the cell, a membrane permeable ligand, Shield1, is used. Shield1 binds to the DD tag and causes rapid stabilization, and therefore accumulation, of the tagged protein. Tuning the concentration of Shield1 allows for various degrees of stabilization of the DD-tagged protein of interest. This process can be reversed in a matter of hours by removing Shield1, and can be repeated multiple times.

The ProteoTuner systems are especially powerful when used in conjunction with RNAi to "knock down" the endogenous levels of a protein of interest. This protein can now be expressed as a DD-tagged exogenous fusion protein, allowing very fast changes in its abundance for precise functional analysis simply by addition or removal of Shield1.

The rapid kinetics of stabilization and destabilization of your protein of interest make the ProteoTuner systems a quick and easy way to analyze the function(s) of the protein of interest. The system has been well tested in multiple cells types and with multiple proteins both cytosolic and membrane bound proteins.

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