第10回細胞生物学セミナー

日時：平成25年11月 27日（水）16:30～（１時間程度）
場所：手形キャンパス総合研究棟２階講義室

Eukaryotic cytosolic chaperonin CCT: the actin folding mechanism and cell biology in yeast

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The chaperonin of the eukaryotic cytosol CCT performs an essential role in cells for the folding of newly translated actin and tubulin polypeptides. CCT is a double-ring ATPase machine constructed from 8 independent, but homologous, 60kDa protein subunits (Dekker et al; 2011). In the case of actin newly translated and experimentally unfolded actin polypeptides adopt similar, stable conformational ensembles, Ac_I and I_3 respectively, which are kinetically and thermodynamically trapped under physiological conditions. The CCT machine binds these states of unfolded actin through a highly specific, mutual recognition mechanism and folds actin to a less kinetically stable but now productive folding intermediate I_2 by coupling its own nucleotide hydrolysis cycle to the phases of actin substrate maturation (Altschuler and Willison; 2008). We have assembled *in vitro* the yeast CCT-actin-PLP2 folding machine (McCormack et al; 2009) and have used a spectroscopic assay to monitor actin as it is folded by CCT (Stuart et al; 2011). In addition to actins and tubulins only a relatively small group of other proteins depend absolutely on CCT for their biogenesis. Several WD40-motif proteins are members of this group; the regulators of the anaphase promoting complex, Cdh1 and Cdc20; the Cdc55 phosphatase regulatory subunit and the TAF5 regulator of mediator complex (Dekker et al; 2008). We will describe a combination of yeast genetic approaches mass spectrometric approaches to the behaviour and quantification of substrate and co-factor proteins bound to yeast CCT and a set of ATP-site mutants (Amit et al; 2010). Co-variance analysis of the raw spectral signals shows strong correlations between expected binding partners such as Act1p and Plp2p (McCormack et al; 2009) and also reveals new couplings between CCT-binding proteins. CCT is intimately involved in co-ordinating cell cycle and cell growth control because it directly couples the rate of actin and tubulin biogenesis to the activities of critical regulators of these processes and a model will be discussed.

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