

P042 Mutational and *in vivo* analysis of the *Haloferax volcanii* CCT complex

Andrew Large and Peter Lund

School of Biosciences, University of Birmingham, UK

Chaperonins are essential protein folding machines found in virtually all organisms. There are two classes: Type I (found in bacteria, mitochondria and chloroplasts) and Type II (found in archaea and eukaryotic cytosol). We are interested in the roles of the archaeal type II chaperonins, and in using them as a simple model for eukaryotic chaperonins. To study them, we developed an expression system for the halophilic archaeon *Haloferax volcanii* based on the tryptophanase (*tna*) promoter, which is tightly off until induced by tryptophan. *H. volcanii* has three chaperonin (*cct*) genes, all of which are expressed. Either one of *cct1* or *cct2* alone is sufficient for growth. We have constructed a strain which lacks *cct2* and *cct3*, with the *cct1* gene under the control of the *tna* promoter. This strain, which cannot grow without tryptophan, is being used to test the ability of mutated and heterologous CCTs to complement for the loss of CCT1. Interestingly, only one of four homologues in *Haloarcula marismortui* could complement for loss of CCT1. Using a functional His-tagged *cct1* gene, we are now constructing a library of point mutations in parts of the protein believed to be important for its function.