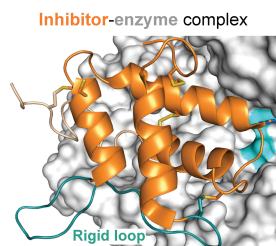


IN THIS ISSUE

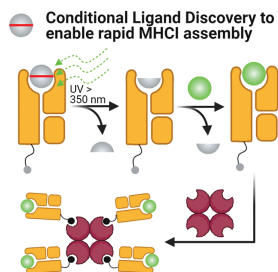


1144

An ultra-high affinity protein–protein interface displaying sequence-robustness

Marie Sofie Møller, Sita Vaag Olesen, Ingemar André

Structures and energetics behind stability of ultra-high affinity protein–protein interactions are not well understood. Here computational guided engineering was used to show that the picomolar affinity complex between a starch debranching enzyme and its proteinaceous inhibitor is not relying on a classical hot-spot arrangement interaction, but on several interactions spread over the interaction interface including residues of a long rigid loop and that many interface mutations can be introduced without substantially reducing the affinity. This property could make the complex more robust to mutational drift in evolution. Furthermore, energetic coupling of distant residues in the interface was demonstrated to play a role as well. <https://onlinelibrary.wiley.com/doi/10.1002/pro.4080>

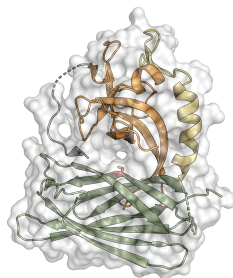


1169

High-throughput identification of conditional MHC I ligands and scaled-up production of conditional MHC I complexes

Martine Darwish, Sara Wichner, Jenny Li, Jiun Chiun Chang, Christine Tam, Yvonne Franke, Hong Li, Pamela Chan, Craig Blanchette

Cancer immunotherapy (CI)/Immuno-Oncology (IO) is one of the most rapidly growing therapeutic areas and relies on using drugs to harness the immune system in controlling tumor growth. One primary goal of CI/IO treatments is to drive tumor specific CD8 T-cell responses; however the number of tools available to track these responses is highly limited. This limitation is rooted in the fact that the T-cell responses are unique to each patient and would require the development of personalized reagents. Here we describe a method to identify conditional ligands to enable high throughput generation of reagents to track patient specific T-cell responses and enable development of patient specific immune monitoring program to support CI/IO clinical programs. <https://onlinelibrary.wiley.com/doi/10.1002/pro.4082>

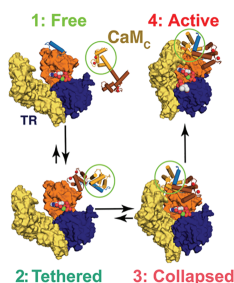


1196

Crystal structure and molecular dynamics of human POLDIP2, a multifaceted adaptor protein in metabolism and genome stability

Anastasija A. Kulik, Klaudia K. Maruszczak, Dana C. Thomas, Naomi L. A. Nabi-Aldridge, Martin Carr, Richard J. Bingham, Christopher D. O. Cooper

Cellular pathways of redox metabolism and genome stability are intimately linked, as the reactive oxygen by-products of the former are a major influence on the latter. We determined the crystal structure of one protein that bridges these pathways, POLDIP2 (Polymerase δ -interacting protein 2). This demonstrated that the two tightly associated YccV and DUF525 domains are centred over a central channel that comprises a modified cysteine residue, suggesting a potential link to redox regulation for POLDIP2. Furthermore, molecular dynamics simulations indicate the flexible N-terminus and loop regions are key to determining how POLDIP2 functions as a 'moonlighting' protein, interacting with many other proteins such as PCNA and PrimPol in genome stability. <https://onlinelibrary.wiley.com/doi/10.1002/pro.4085>



1221

Structural dynamics of the complex of calmodulin with a minimal functional construct of eukaryotic elongation factor 2 kinase and the role of Thr348 autophosphorylation

Andrea Piserchio, Kimberly Long, Kwangwoon Lee, Eric A. Kumar, Rinat Abzalimov, Kevin N. Dalby, Ranajeet Ghose

The α -kinase, eEF-2K regulates protein synthesis through a unique mechanism mediated by calmodulin and reliant on autophosphorylation at a primary activating site, Thr348. Using TR, a minimal functional construct of eEF-2K, we demonstrate the presence of a long-range network that integrates both components of the TR/calmodulin complex. The C-terminal lobe of calmodulin plays a key role within this network, in tethering TR to its vicinity, in facilitating the formation of an intimate collapsed complex to enable efficient chemistry, and in sensing primary regulatory signals like Thr348 phosphorylation, and secondary ones such as pH changes. <https://onlinelibrary.wiley.com/doi/10.1002/pro.4087>