Hundreds of starting points to develop protein-protein interaction modulators

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Protein-protein interactions (PPIs) are vital for cellular processes and have been found to be involved in several diseases [1] und thus are promising targets in drug discovery and tool compound development. Targeting PPIs also broadens the available target space by including e.g. scaffold or hub proteins. The discovery of starting points for PPI modulators is possible by crystallographic fragment screening (CFS). It has evolved into an attractive application due to enormous efforts of the community, by increasing the throughput and reducing manual labor. Fragments are small organic compounds (< 300 Da) that can efficiently probe a protein surface. The application of CFS has the advantage of readily available 3D information of the fragment's binding position and mode, which can be utilized for structure-guided hit optimization.

Here, we targeted a yeast spliceosomal PPI that is necessary for the formation of an active spliceosome, Prp8RNaseH and Aar2 (AR) [2]. Prp8 is one of the key scaffold proteins in the spliceosome, engaging in various PPIs throughout the process. Aar2 is a shuttling protein, transporting Prp8 into the nucleus. Thus, several target sites are investigated for possible development of PPI modulators.

AR was screened against the novel F2X-Universal Library developed at HZB [3]. The library contains 1103 compounds representing the chemical space of all available fragments. In total over 1000 crystals were soaked at HZB [4], utilizing an evaporation reduction device termed the EasyAccess Frame [5]. Data were collected at the BioMAX beamline at MAX IV [6] and analyzed via FragMAXapp [7] with additional clustering by cluster4x [8]. Overall, 269 fragment hits (377 binding events) could be observed that are scattered across both protein surfaces as well as at the AR interface. The found hit sites include additional known PPI interfaces of Prp8RNaseH - Prp18, Prp8RNaseH - Slu7/Prp3 and Aar2 – Prp8Endonuclease. In summary, the experiment yielded several different starting points for further optimization into potential spliceosomal PPI modulators.

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