Oral presentation

Inhibition of the human mitochondrial ClpP protease as a novel strategy against multiple myeloma

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Multiple myeloma (MM) is an incurable blood cancer affecting antibody-producing plasma cells, whose clonal expansion and dysregulated secretion of immunoglobulins leads to widespread organ failure. The available therapies, including hematopoietic stem cell transplantation and inhibition of the 20S proteasome, have limited efficacy, and relapses occur with high frequency. Thus, the identification of novel vulnerabilities of MM cells is a cogent requirement to develop effective strategies to benefit persons with MM. A particularly interesting aspect is the role of mitochondria in MM, involved not only in energetic metabolism but also acting as a central hub of cytotoxic processes such as the mitochondrial unfolded protein response (UPR).

Transcriptomic analysis of MM cells showed an upregulation of the human mitochondrial ClpP (hClpP) serine protease expression compared to both other tumours and cells from healthy donors [1]. hClpP has been long considered an attractive target against tumours through its hyperactivation [2], leading to disruption of oxidative phosphorylation. Yet, only limited information is available on the fine details of the hClpP active site that are required for the design of highly selective inhibitors. Downregulation of ClpP leads to rapid death of leukemic and MM cell lines in an electron chain transport-independent process.

Here, we report the structural characterization of hClpP in a truly unliganded state using X-ray crystallography, demonstrating a highly flexible active site with an "unloaded" catalytic triad. Then, we used high-resolution cryoEM to determine the structure of hClpP in complex with a boronic acid peptide inhibitor [3]. The inhibitor binds to the enzyme with a covalent two-step mechanism, modifying the active site serine and interacting with one hydrophobic specificity pocket. Binding of the inhibitor induces a major restructuring of the hClpP quaternary structure and active site that can be exploited for improving specificity and affinity. The boronic acid inhibitor is effective in selectively killing MM cell lines, through a synergic mechanism that involves both proteasome and hClpP inhibition. Our results pave the way for identification of novel compounds with favourable safety profiles and pharmacokinetics that can be tested in both model and patient-derived cells to achieve new tools for the treatment of this incurable disease.

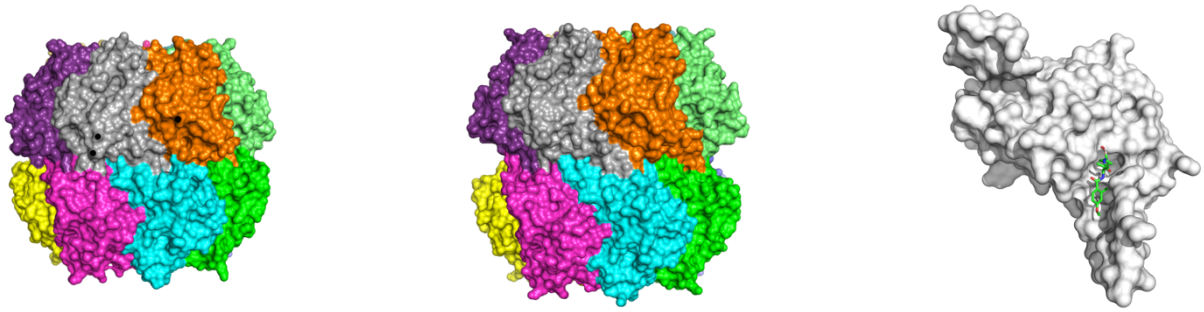

Figure 1. Side views of human ClpP in absence of ligands (left) and bound to a covalent inhibitor (middle), and the boronic acid peptide inhibitor bound to the active site of one ClpP monomer.

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