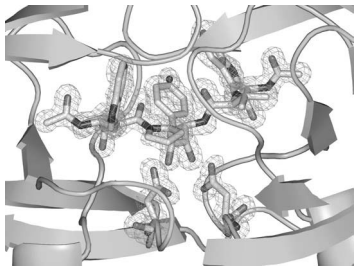
synchrotron radiation, clearly show that the catalytic site is fully occupied by a single ordered molecule (see Figure). This permitted unambiguously the identification of nature and stereochemistry of the bound inhibitor. Furthermore, the clear electron density map, without residuals, suggests that the inhibition constant of this compound should be at least one order of magnitude lower than the constants of the other compounds. The full occupancy of the site indicates that its value is less than 1μM. This biocrystallographic study has allowed a first assessment of inhibition properties without the purification of the mixture and the classic activity assays that are normally conducted on each compound. The co-crystallization strategy could be applied in conjunction with combinatorial chemistry synthesis to discover, by self selection, new potent inhibitors.



Keywords: single-crystal structure analysis, inhibitor binding, isomers

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Crystal Structure of a Disintegrin Heterodimer from *Echis carinatus* at 1.9 Å Resolution

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Disintegrins are a family of small proteins that bind to integrins specifically. Their binding site is characterized by the presence of Arg-Gly-Asp motif which indicates an RGD-dependant mode of interaction with integrins. The disintegrins interfere with the functions of integrins as antagonists. Disintegrin was isolated from the venom of *Echis carinatus* and crystallized in the tetragonal space group $P4_32_12$ with a=b=90.7Å and c=55.5Å. It exits as a heterodimer unlike the low resolution structure which existed as a homodimer with its two subunits related by a two fold crystallographic symmetry. It is interlinked by two disulfide bonds at the N-terminal region and contains 64 amino acid residues in each chain. Each monomer contains three pairs of six antiparallel β-strands and is stabilized by four disulphide bridges. It has been refined to an R-factor of 0.212 and R_{free} of 0.251 for all the data. The two chains of the dimer are anchored at N-terminal but diverge away at their C-termini exposing the Arg-Gly-Asp motif onto opposite directions, thus enhancing their binding efficiency. This is one of its unique features. The structural studies of disintegrins can provide a useful framework for the design of potent antagonists of integrins.

Keywords: disintegrin, heterodimer, drug design

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Structure-assisted Design of Inhibitors Targeting Coronavirus Main Proteases

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Coronaviruses (CoVs) are important etiologic agents of upper respiratory and digestive tract diseases in humans and animals; especially, the severe acute respiratory syndrome (SARS). The viruses are characterized with a highly complex cascade of proteolytic processing the replicative polyproteins to control viral gene expression and replication, which was predominantly mediated by the viral main proteinase (M^{pro}, also called $3CL^{pro}$), therefore, an attractive target for drug development[1].

A series of novel compounds with Michael receptor was designed according to the crystal structures of 3 coronaviruses M^{pro}s. The solved structures of SARS-CoV and porcine transmissible gastroenteritis virus (TGEV) M^{pro}s individually complexed with these compounds revealed that inhibitors possessing *α,β*-unsaturated ester combined with peptidyl-binding elements specific for CoV M^{pro}s undergo a nucleophilic addition of the protease's catalytic Cys, resulting in covalent-bond formation and irreversible inactivation of the viral proteases. One compound in this series has exhibited potent and extensive inhibition effect on 6 CoV M^{pro}s covering all 4 groups within genus *Coronavirus*. Meanwhile, the novel small molecules showed low micromolar concentration of EC_{50} for inhibition of viral replication and very low cell toxicity. We suppose further modification of these compounds assisted with structural information might lead to discover drug candidates against all CoV-associated diseases, including SARS.

[1] Yang H., Yang M., Ding Y., Liu Y., Lou Z., Sun L., Zhou Z., Ye S., Pang H., Gao G., Anand K., Bartlam M., Hilgenfeld R., Rao Z., *Proc. Natl. Acad. Sci. USA*, 2003, **100(23)**, 13190-13195.

Keywords: SARS, main protease, drug design

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Structural Insights into the Substrate Binding Mechanism, Inhibition and Regulation of Pim-1

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Pim-1 is a highly conserved cytoplasmic serine/threonine kinase that was first discovered as a preferential proviral insertion site in Moloney Murine Leukemia Virus (MoMuLV) induced T-cell lymphomas. The expression pattern of Pim-1 is widespread and the protein is over-expressed in a series of tumors but highest expression levels are found in cells of the hematopoietic and lymphoid system. Pim-1 phosphorylates a number of signal transduction proteins involved in the regulation of cell cycle, apoptosis, differentiation and proliferation.

We determined the structure of human Pim-1 in complex with an inhibitor of the bisindolyl maleimide (BIM) class as well as in ternary complex with its consensus peptide (pimtide) and BIM-1/AMPPNP that provides interesting insight into the substrate binding and inhibition of Pim-1 and suggests further applications of BIM-like compounds for treatment of leukaemia and other Pim-1 dependent cancer types.

Structural analysis of the monophosphorylated Pim-1 and auto-phosphorylation studies show that the human Pim-1 kinase activity is not influenced by auto-phosphorylation of activation loop residues. The N-terminus of Pim-1 has been shown to be important for several Pim interacting proteins, it is therefore likely that phosphorylation at Ser8 indicated by phosphorylation mapping plays a role in modulating these interactions.

Keywords: kinase structure, phosphorylation, drug design

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Interdomain Communication in HCV Polymerase Abolished by Small-Molecule Inhibitors

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The hepatitis C virus (HCV) polymerase is required for replication of the viral genome and is a key target for therapeutic intervention against HCV. We have determined the crystal structures of the HCV polymerase complexed with two indole-based allosteric inhibitors at 2.3 Å and 2.4 Å resolution. The structures show that these inhibitors