Journal of Molecular Graphics and Modelling 51 (2014) 193–202

Contents lists available at ScienceDirect

Journal of Molecular Graphics and Modelling

journal homepage: www.elsevier.com/locate/JMGM

Investigating the binding preferences of small molecule inhibitors of human protein arginine methyltransferase 1 using molecular modelling

Wei Hong[a,b], Jingyang Li[c], Charles A. Laughton[d], Lee Fah Yap[b], Ian C. Paterson[b], Hao Wang[c,*]

[a] School of Chemistry and Chemical Engineering, Beifang University of Nationalities, Yinchuan, PR China
[b] Department of Oral Biology and Biomedical Sciences, and Oral Cancer Research and Coordinating Centre, Faculty of Dentistry, University of Malaya, Kuala Lumpur, Malaysia
[c] School of Pharmacy, Ningxia Medical University, Yinchuan, PR China
[d] School of Pharmacy and Centre for Biomolecular Sciences, University of Nottingham, Nottingham, UK

ARTICLE INFO

Article history:
Accepted 28 May 2014
Available online 4 June 2014

Keywords:
PRMT1 inhibitors
Homology modelling
Active Site Pressurisation
Molecular dynamic simulation
Binding mode

ABSTRACT

Protein arginine methyltransferases (PRMTs) catalyse the methylation of arginine residues of target proteins. PRMTs utilise S-adenosyl methionine (SAM) as the methyl group donor, leading to S-adenosyl homocysteine (SAH) and monomethylarginine (mMA). A combination of homology modelling, molecular docking, Active Site Pressurisation, molecular dynamic simulations and MM-PBSA free energy calculations is used to investigate the binding poses of three PRMT1 inhibitors (ligands 1–3), which target both SAM and substrate arginine binding sites by containing a guanidine group joined by short linkers with the SAM derivative. It was assumed initially that the adenine moieties of the inhibitors would bind in sub-site 1 (PHE44, GLU137, VAL136 and GLU108), the guanidine side chain would occupy sub-site 2 (GLU 161, TYR160, TYR156 and TRP302), with the amino acid side chain occupying sub-site 3 (GLU152, ARG62, GLY86 and ASP84; pose 1). However, the SAH homocysteine moiety does not fully occupy sub-site 3, suggesting another binding pose may exist (pose 2), whereby the adenine moiety binds in sub-site 1, the guanidine side chain occupies sub-site 3, and the amino acid side chain occupies sub-site 2. Our results indicate that ligand 1 (pose 1 or 2), ligand 2 (pose 2) and ligand 3 (pose 1) are the predominant binding poses and we demonstrate for the first time that sub-site 3 contains a large space that could be exploited in the future to develop novel inhibitors with higher binding affinities.

1. Introduction

Protein arginine methyltransferases (PRMTs) can catalyse the methylation of arginine residues of a variety of proteins involved in cell signalling, RNA splicing and gene regulation, such as histones H2A, H3 and H4 [1]. All PRMTs utilise S-adenosyl methionine (SAM) as the methyl group donor, leading to S-adenosyl homocysteine (SAH, Fig. 1) and monomethylarginine (mMA) [2]. Further methylation often follows, with Type I PRMTs delivering asymmetrical dimethylarginine (aDMA) and Type II PRMTs symmetrical dimethylarginine (sDMA), respectively [2,3]. PRMT1 is the predominant Type I PRMT in mammals and its activity is deregulated in some human pathologies, such as cardiovascular disease and cancer [4,5]. Therefore, specific PRMT1 inhibitors could prove useful as therapeutic agents.

The crystal structure of human PRMT1 has not been resolved, but the crystal structures of rat PRMT1 (rPRMT1, PDB 1OR8) [6] and rat PRMT3 (rPRMT3, PDB 1F3L) [7] homologues are available. Both these crystal structures show that the active site of PRMT can be divided into two parts, namely the SAH and substrate arginine binding sites. These two sites are close to each other, thus enabling the methyl group to be transferred from SAM to the substrate arginine (Fig. 1A). Recently, three bisubstrate inhibitors of hPRMT1 (ligands 1–3, Fig. 1B; IC_{50} ~5 μM for each inhibitor) have been described [8]. These ligands target both SAM (blue and yellow in Fig. 1C and D) and substrate arginine (red in Fig. 1C and D) binding sites by containing a guanidine group joined by short linkers with the SAM derivative. When these three ligands were designed, it was assumed that the adenine moieties of the ligands would bind in sub-site 1 (composed of PHE44, GLU137, VAL136 and GLU108, blue), the guanidine side chain would occupy sub-site 2 (composed of GLU 161, TYR160, TYR156 and TRP302, red), with the amino acid side chain occupying sub-site 3 (composed of GLU152, ARG62, GLY86 and ASP84, yellow), as shown in Fig. 1C; in

* Corresponding author. Tel.: +86 9516880582; fax: +86 9516980193.
E-mail address: paxhw@yahoo.co.uk (H. Wang).

http://dx.doi.org/10.1016/j.jmgm.2014.05.010
1093-3263/

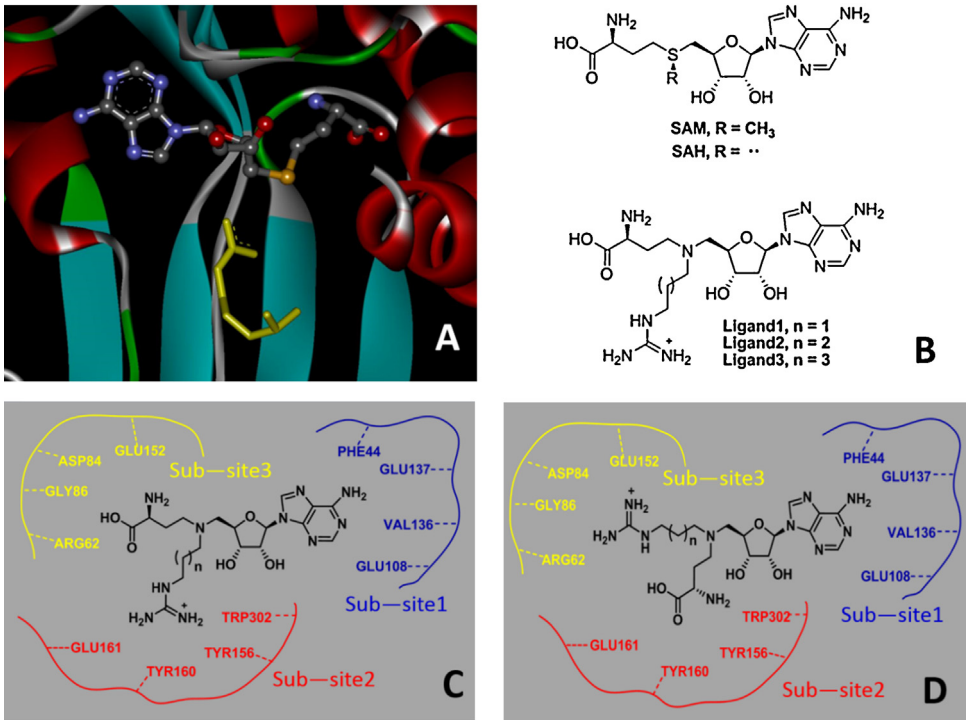

Fig. 1. (A) Active site of PRMT1 in complex with SAH (ball and stick) and substrate arginine (yellow tube); (B) the chemical structures of SAM and related compounds; (C) binding pose 1; (D) binding pose 2. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of the article.)

the present study, this is referred to as binding mode pose 1. However, the cavity of sub-site 3 in Fig. 2 is in fact quite large and the SAH homocysteine moiety only occupies part of the space. Therefore, another binding pose is possible (pose 2), whereby the adenine moiety binds in sub-site 1, the guanidine side chain occupies subsite 3, and the amino acid side chain occupies sub-site 2. A clear understanding of how these inhibitors occupy the PRMT1 binding site is required to fully understand the mechanism of inhibition and to inform medicinal chemistry efforts to synthesise even more effective inhibitors.

Molecular modelling and computational aided drug design have been used extensively to study the reaction mechanism, discovery of potential inhibitors, and interactions between ligands with arginine methyltransferases [9–13]. Consideration of receptor flexibility is essential in computational aided drug design, and it is particularly important when the crystal structure of the receptor is not available [14–16]. Various docking software can take receptor flexibility into account during the docking process. However, they cannot fully explore the binding site conformations and are not suitable for large chemical database screening. An alternative approach is to consider the flexibility of receptor prior to the docking progress using molecular dynamic simulations [17,18]. However, with limited computational time, the accessible trajectories (typically in the nanosecond to microsecond time scale) suffer from not being able to overcome high conformational barriers in the potential energy surface of the receptors and complexes. This limits the technique to exploring only conformations of the ensemble that are around the starting structures. An additional problem is that when there is no ligand binding to maintain the conformation of the active site, the conformation of the receptor will tend to convert to that of the apo form during the simulation, which was an issue in the current study. Active Site Pressurisation (ASP) [19] was created recently to examine the flexibility of the active sites of receptors. The basic idea of ASP is to 'pump' Lennard–Jones (LJ) particles into a protein cavity and see how this expands by exploiting natural directions of the weakest resistance in the protein structure, which usually cannot be achieved by unperturbed MD with limited simulation time.

In the present study, we used a combination of homology modelling, molecular docking, ASP, molecular dynamic simulations and MM-PBSA (Molecular Mechanics – Poisson–Boltzmann Surface Area) free energy calculations to investigate the binding poses of

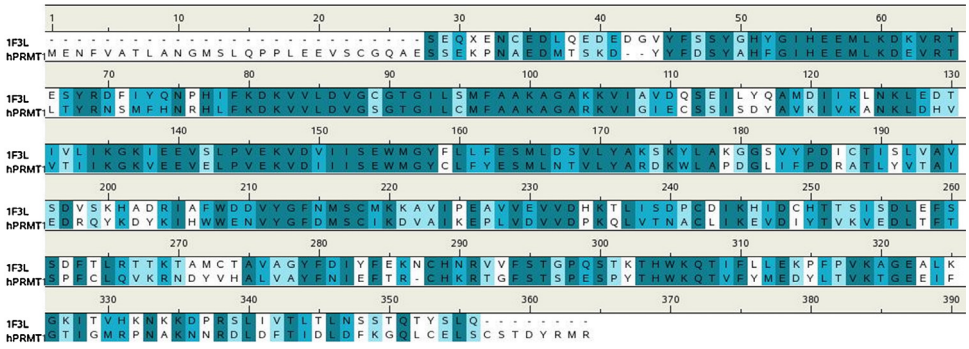

Fig. 2. Sequence alignment of hPRMT1 and rPRMT3 (PDB 1F3L).