Molecular dynamics simulation of heme-containing membrane protein prostaglandin H synthase

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Background

The Prostaglandin H_2 Synthase (or PGHS) is homodimeric membrane-bound glycoprotein, localized to the lumen of the EPR and to the inside of the nuclear envelope (Fig.1). The PGHS is presented in nearly all tissues as two isozymes with some structural differences. Biological role of PGHS is arachidonic acid two-step conversion into prostaglandin H_2, a crucial element of pain and inflammatory response mechanisms[1].

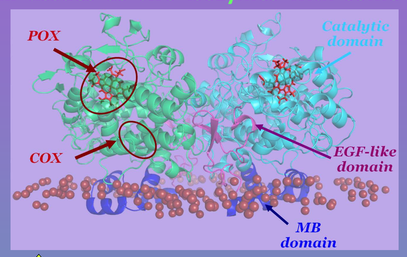


Figure 1. Structure of PGHS-1 (1EQH) integrated into membrane. Coloured by domains, red spheres correspond to the lipid carbonyl groups localization.

PGHS is a pharmacological target for a broad class of compounds belonging to non-steroidal anti-inflammatory drugs (NSAIDs), widely used in medical practice[2]. Therefore, PGHS was studied during last 30 years and was shown to be ineffective without heme or as isolated monomer. Despite this, nearly all PGHS models described in literature exclude heme or membrane because of computational complexity.

Authors	Verli H et al, 2011	Coveney PV et al, 2009	Smith WL et al, 2007	Coveney PV et al, 2006	Desiraju GR et al, 2006	Momani E et al, 2005	Ruiz J et al, 2002	Nina et al, 2000	Gago F et la, 2000	Filizol M et al, 1997
2 Subunits	-	+	+	-	-	+	+	-	+	-
Heme	?	+	+	+	+	+	?	-	?	+
Membrane	-	+	-	+	-	-	-	+	-	-
Explicit solvent	+	+	+	+	+	+	+	+	-	-

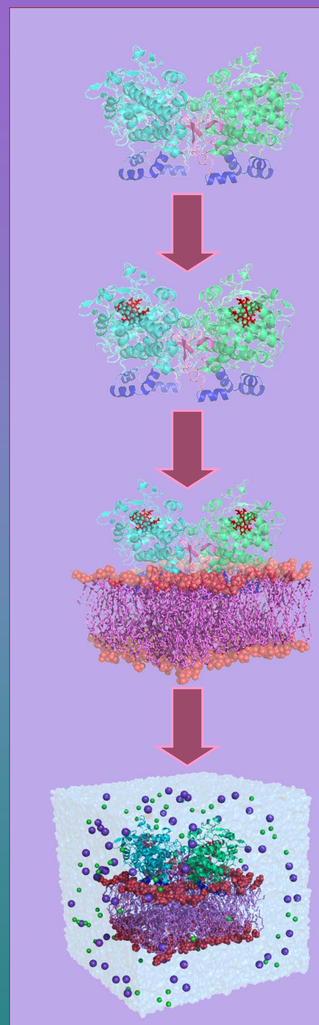
Table 1. PGHS molecular modeling studies described in literature

Summary

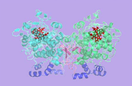
Heme binding, integration into membrane and dimerization are likely to significantly influence on the PGHS native structure and dynamic behavior. 3D models of the PGHS dimers containing heme and integrated into membrane were constructed for both isozymes. Molecular dynamics simulations of obtained PGHS-1 and PGHS-2 models were performed on "Lomonosov" supercomputer[3] in NAMD using CHARMM topology and parameters.

Also, we paid extra attention to the default CHARMM topology for heme residue and made some corrections in bond types and Internal coordinates of lateral substituents.

Methods

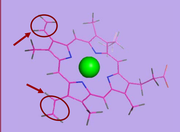


1. Initial structure selection



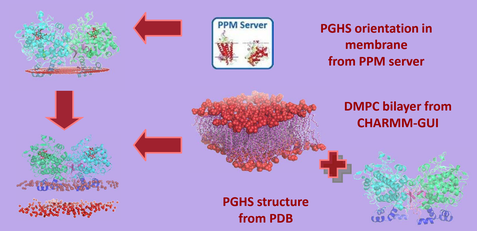
- X-ray structures
- Non-mutant
- Containing heme
- With identical inhibitors

2. Heme topology correction



- ✓ *Missing double bonds addition*
- ✓ *Some corrections in internal coordinates*
- ✓ *Non-planar Heme conformations*

3. Integration into membrane



4. Ions addition

PGHS-1	PGHS-2
• Water: 42809 molecules	• Water: 42856 molecules
• Box size : 144 Å × 150 Å × 144 Å	• Box size : 144 Å × 150 Å × 144 Å
• Ions : 115 K^+ 113 Cl^-	• Ions : 123 K^+ 113 Cl^-
• Lipids : 240 DMPC	• Lipids : 238 DMPC

5. Molecular dynamics run

- *NAMD 2.5*
- *CHARMM 27 topology*
- *T = 310 K, P = 1.01325 bar*
- *Periodic boundary conditions*

Aims of work

To construct a 3D PGHS model containing:

- *two monomers*
- *one heme group per monomer*
- *membrane bilayer*
- *explicit water and ions*

To perform PGHS molecular dynamics runs

Trajectory analysis

Fig. 3. Residue number vs. time for PGHS-1 and PGHS-2, coloured by RMSD according to the scale ranging from blue (low values) to red (high values).

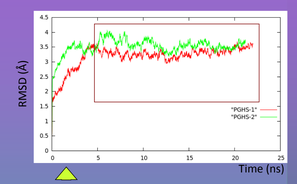
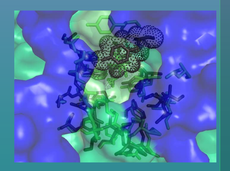


Fig. 2. The protein backbone RMSD vs. time for PGHS-1 (red) and PGHS-2 (green). Trajectory fragments from 5 to 20 ns were used for further analysis.

Fig. 4. Averaged structures superposition for PGHS-1 monomer (black) and dimer (green) in the COX site entrance region. COX site residues are shown in sticks model, other residues are presented as surface. MB and catalytic domains are coloured blue and green respectively. Tyr 355 displacement leads to substrate access blockage and hence monomer inactivation.



Results

1. *Default CHARMM topology for heme was corrected*
2. *Both for PGHS-1 and PGHS-2 20 ns dynamics runs were performed*
3. *70 ns dynamics was carried out for PGHS-1 monomer without membrane*

References

[1] Kulmacz R.J. et al. Comparison of the properties of prostaglandin H synthase-1 and -2 // *Progress in Lipid Research 42 (2003) 377–404.*
[2] Grosser T et al. Biological basis for the cardiovascular consequences of COX-2 inhibition: therapeutic challenges and opportunities // *J Clin Invest. 2006;116(1):4-15.*
[3] Information and Analytical Center on Parallel Computing (parallel.ru): http://parallel.ru