Selection of the most significant ligand-receptor interactions in GPCRs crystal complexes

Dawid Warszycki[1], Krzysztof Rataj[1], David E. Gloriam[2], Andrzej J. Bojarski[1], Stefan Mordalski[1]

[1]Institute of Pharmacology, Polish Academy of Sciences, 31-343 Kraków, 12 Smętna street, Poland
[2]Department of Drug Design and Pharmacology, University of Copenhagen, Jagtvej 162, DK-2100 Copenhagen, Denmark

e-mail: warszyc@if-pan.krakow.pl

Introduction

G Protein-Coupled Receptors (GPCRs) comprise a large superfamily of signalling proteins (~800 receptors in humans), which are involved in a number of physiological processes, like mood and behaviour regulation, perception of pain, autonomic nervous system transmission, inflammation, regulation of immune system, etc. [1]. Despite the fact that up to 59 receptors have been drugged, only 159 crystal structures are available in PDB in total (for 35 targets).

These structures were utilized in the new pharmacophore method [2] based on the library of interacting ligand moiety-residue pairs extracted from the crystal structures. Here we automate the methodology using interaction fingerprints for the systematic analysis of the growing number of crystal structures and the extraction of fragments. In addition we extend it to the homology models, investigating the effect of conformational flexibility in the quality of the pharmacophore models.

Materials and methods

- β_2-AR models were constructed on ten different crystal templates (Table 1, Fig. 1)
- Modeller 9v8 was used to prepare the models (manual alignment)
- A library of interacting ligand moiety – residue fragments was used (Fig. 1)[2]
- Fragments were mapped on each of the models
- Pharmacophore models were constructed using Phase [3] (Fig. 3)
- Retrospective screening was conducted with 319 active compounds (ChEMBL, K_i or equivalent less than 100 nM) and 20248 DUD decoys.
- Yourden's statistics and Matthiew's Correlation Coefficient (MCC) were used as the measures of model's efficiency (Fig. 4).

Table 1. Crystal structures used as templates for homology modeling of β_2-AR

Template name	PDB ID	Resolution [Å]
5-HT_{1B}R	4IAR	2.70
5-HT_{2B}R	4IB4	2.70
A_{2A}R	3QAK	2.71
β_1-AR	2Y00	2.50
CXCR4R	3OE0	2.90
D_3R	3PBL	2.89
H_1R	3RZE	3.10
M_2R	3UON	3.00
M_3R	4DAJ	3.40

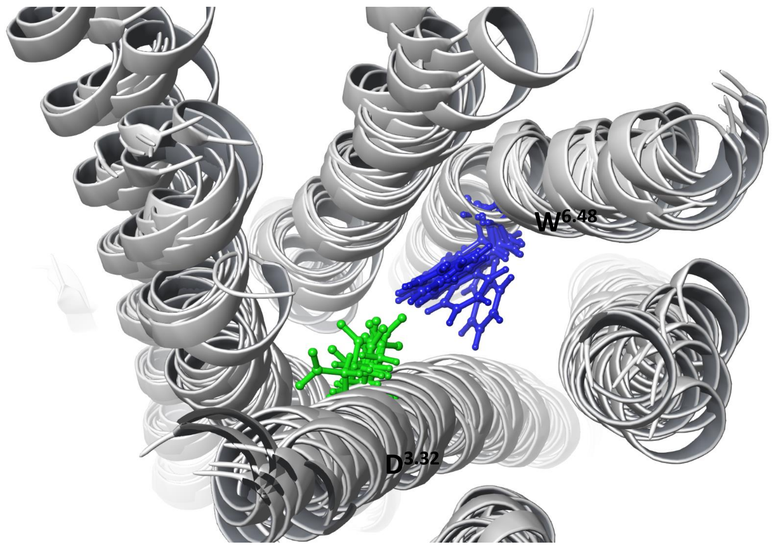

Figure 1. Conformational flexibility of the homology models comming from different crystal templates, as pictured by $D^{3.32}$ (green) and $W^{6.48}$ (blue).

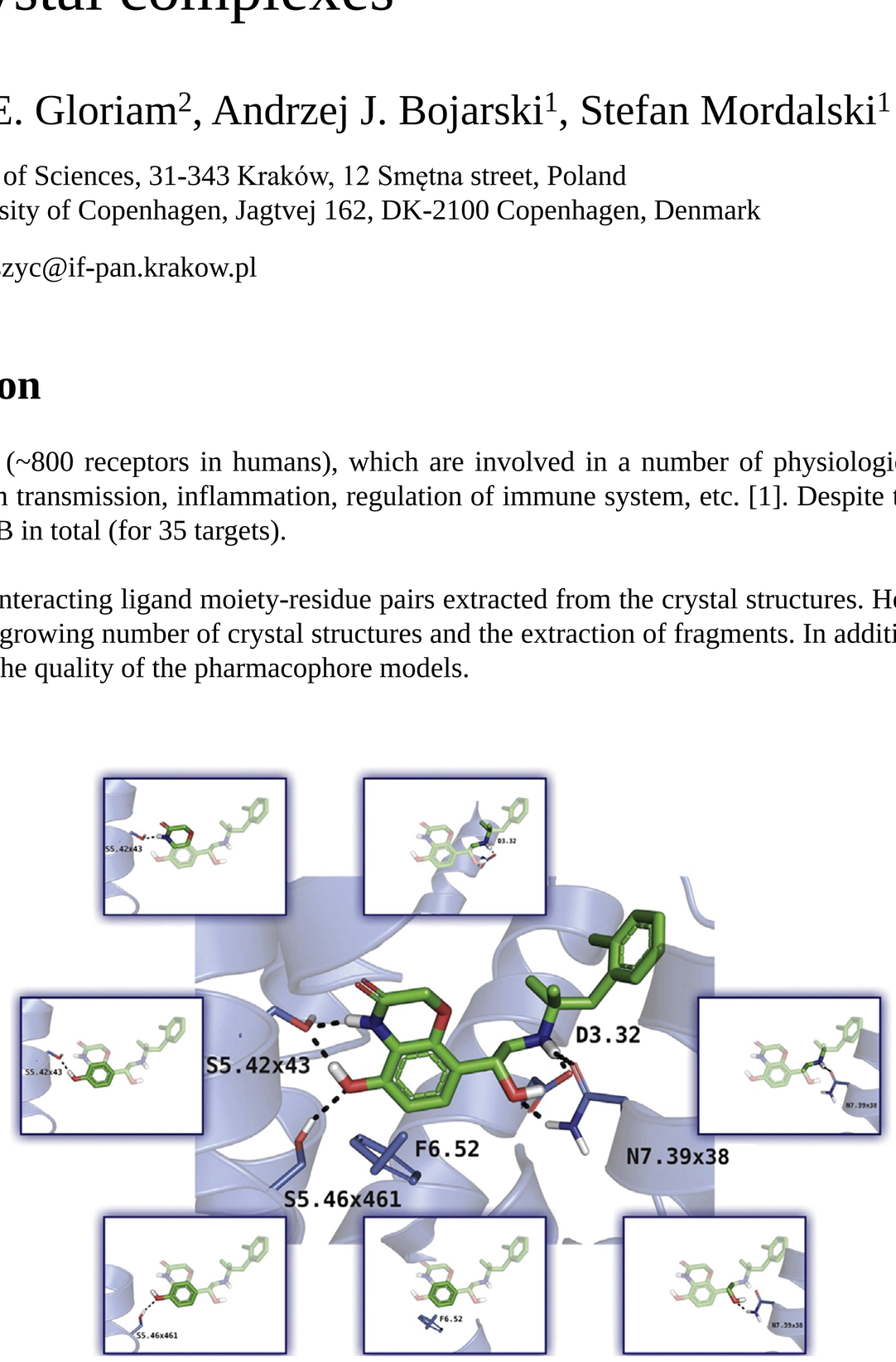

Figure 2. Extraction of fragments (in small boxes) from the ligand (green) – b2 adrenergic receptor (blue) complex crystal structure (PDB: 3SN6). The generic residue numbers are denoted in the GPCRDB scheme [4] (source: Fidom *et al.* [2]).

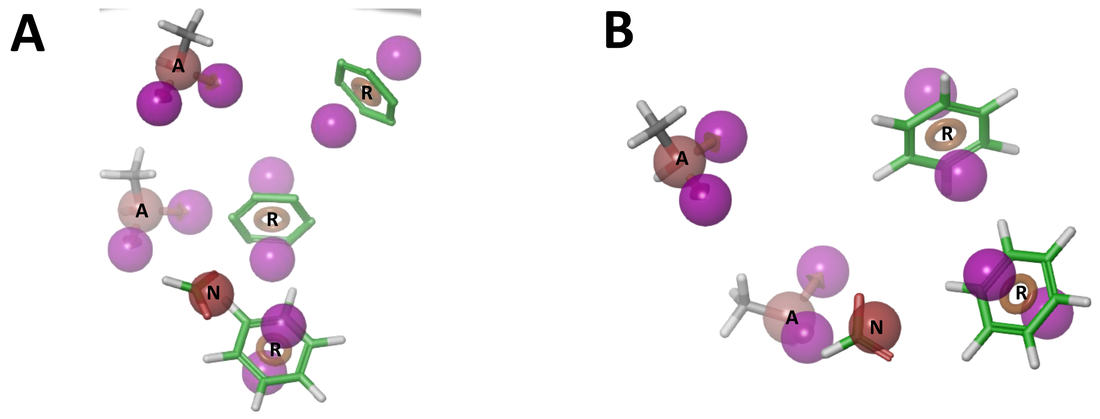

Figure 3. Exemplary pharmacophore hypotheses for β_2-AR homology models built on A_{2A} receptor (**A**) and D_3 (**B**). The feature abbreviations used are: hydrogen bond acceptor – A; negatively ionized group – N, aromatic ring – R. Ligand attachment points are marked in magenta.

Results and conclusions

Results (Figure 4.) clearly indicate that models generated on serotonin receptors templates outperformed the remaining ones. Nevertheless, relatively low values of calculated parameters suggests that for virtual screening purposes the additive model of different hypotheses (instead of single one) can be necessary. Moreover, this study did not include fact that multiple interactions can be caused by single ligand's fragments contacing with different residues which will be considered within further studies.

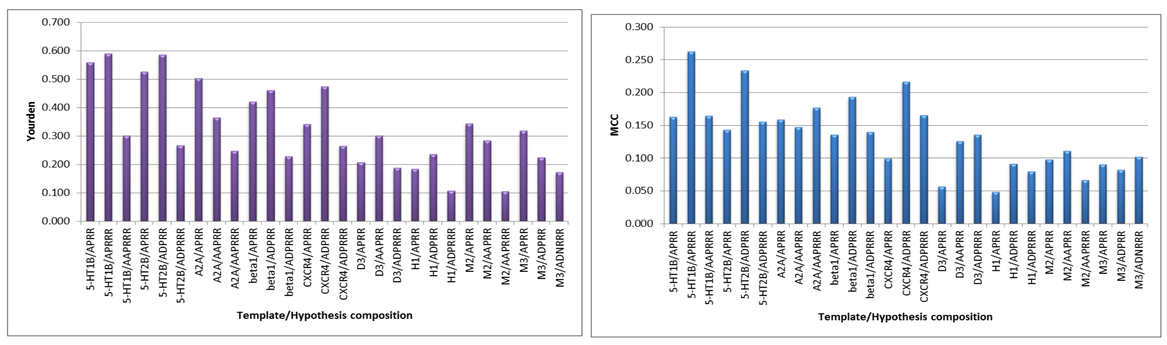

Figure 4. Screening performance of pharmacophore hypotheses with varying number of features for receptor models developed on 9 different templates.

References

[1] Sharman J. L. et al.; *Nucleic Acids Res.* 2013, 41, D1083–8.
[2] Fidom K. et al.; *Methods* 2015, 71, 104–12.
[3] Isberg V. et al.; *Trends Pharmacol. Sci.* 2015, 36(1): 22–31.
[4] Phase, Schrödinger, LLC, New York, NY, 2016

Acknowledgements

The work was supported by the National Science Centre (Poland) grant no. 2015/18/M/NZ2/00423.

NATIONAL SCIENCE CENTRE POLAND