PROT-ON: Structure-Based Detection of Critical Mutations in Redesigning Protein-Protein Interfaces

Mehdi Koşaca[1,2], İrem Yılmazbilek[1,3] Ezgi Karaca[1,2*]

[1] Izmir Biomedicine and Genome Center, Dokuz Eylül Health Campus, Izmir
[2] Izmir International Biomedicine and Genome Institute, Dokuz Eylül University, Izmir
[3]Middle East Technical University, Ankara

***Correspondence:** Ezgi Karaca +90 (533) 456 6096 ezgi.karaca@ibg.edu.tr

Deformation of protein-protein interactions (PPIs) causes several diseases. Many therapeutic agents have been developed by time-consuming experimental techniques. On the other hand, computational methods have been used to estimate the impact of interfacial mutations faster and more easily. The most used and accurate algorithms, EvoEF1 [1] and FoldX [2], are specialized to estimate the impact of mutations. However, their manual pipeline is insufficient to analyze all interfacial mutation possibilities. To address this issue, we have developed PROT-ON.

PROT-ON offers a novel automatic pipeline that scans the interfacial residues of a protein complex in the user interest cut-off distance. Then, they are mutated into other 19 amino acids and their binding affinities are predicted by EvoEF1 or FoldX. In the final step, estimated ΔΔG binding affinities are statistically analyzed with a box-and-whisker analysis. The most enriching and depleting mutations are proposed by filtering stability and evolutionary information (if accessible), and interactive heatmap and box-whiskers are generated (Figure 1).

We have tested the PROT-ON on the receptor binding domain (RBD) of Spike, and the catalytic domain of human ACE2 to detect dangerous mutations. Tian et al. [3] worked on experimental studies on RBD variants called Alpha, Beta, and Gamma. The most famous one is determined as N501Y mutation and led to an increased binding affinity. Moreover, K417N, G446S, Q493R, G496S, and Q498R are identified as interfacial RBD mutations that are observed in variants. PROT-ON detected mutations lead to enriching positions as Q493R, and G496S which were previously defined on the variants.

PROT-ON could be used by drug development and protein design researchers to redesign the interface. Interfacial mutations even not emerge in the evolutionary time can be predicted in a short time with our approach. We have presented a user-friendly web server and a standalone version. It can be accessed via http://proton.tools.ibg.edu.tr:8001.

References

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3. Tian, F., Tong, B., Sun, L., Shi, S., Zheng, B., Wang, Z., Dong, X., Zheng, P., 2021. Mutation N501Y in RBD of Spike Protein Strengthens the Interaction between COVID-19 and its Receptor ACE2. BioRxiv.

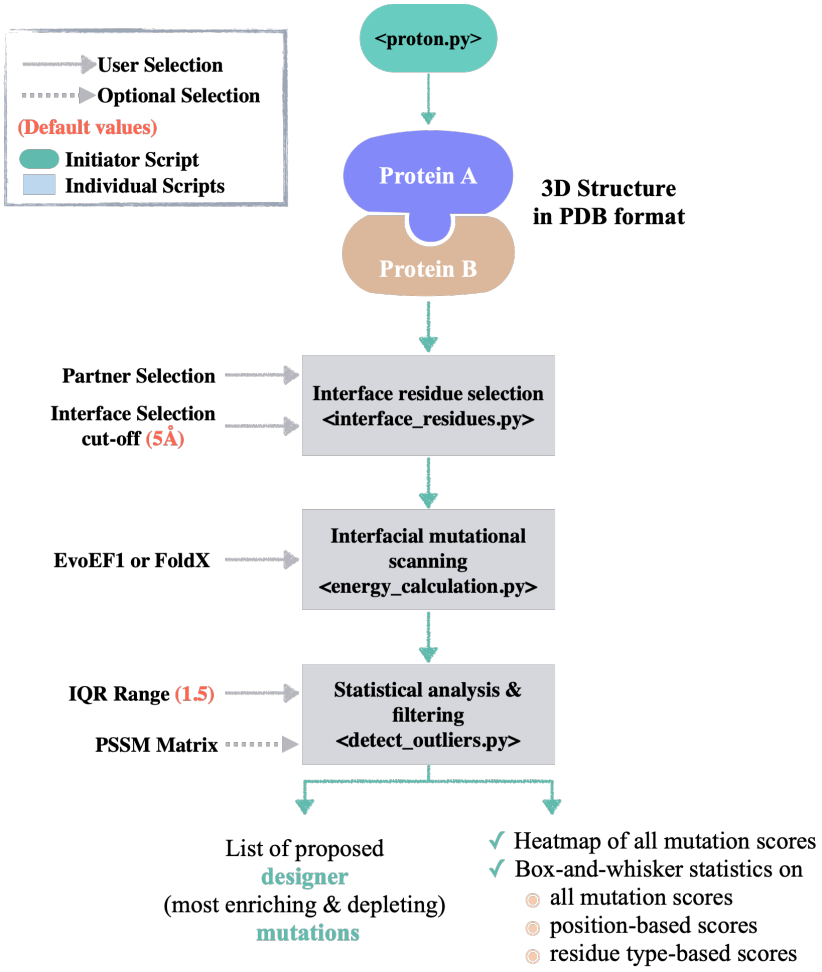

Figure 1. *The automatic pipeline of PROT-ON to propose designer interface mutations.*