Review of Doctoral Dissertation
"Algorithms for the analysis of molecular protein structures and drug-like ligands for modeling and simulations of residence time drug-molecular target"
Candidate: mgr inz. Magdalena Lugowska
Reviewer: Prof. James Briggs, Professor of Biochemistry, Associate Provost for Faculty Development and Faculty Affairs, University of Houston, Houston, TX 77204, USA
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Summary:
In summary, the candidate has presented a dissertation describing the development and application of methods for the calculation and application of drug-target residence time as a way to predict drug efficacy. While a ligand may bind to a target strongly, it can only exert its effect while it remains bound to the target. If the residence time is short, that drug is unlikely to be very efficacious. The candidate has created PDBrt – a database that provides curated binding kinetics for structures available in the PDB. A set of specific systems were chosen for more detailed investigation (e.g., MTb enoyl ACP reductase (InhA), HSP90, ENR, EGFR, and HIV-1 PR). The τRAMD method, developed by R. Wade was employed to predict kinetic parameters, as validated against a set of the proteins above.

The thesis is well-written and describes an approach to an important problem in drug discovery and development. The selected set of proteins is reasonable and will hopefully continue to expand in future/ongoing studies in order that the machine learning step has more data. One of the key findings is that the τRAMD method works well for relative residence times if the ligands are only slightly different but is much less accurate when the ligands differ substantially. The final Chapter (5) is particularly valuable as it offers insights into factors that impact ligand residence time. The primary analysis for determinants of residence time was for InhA. As more and more systems are characterized, it would be good to see if more general trends are revealed, such as, hydrophobic interactions together with VDW contribute primarily to longer residence times, etc.

Overall, this is excellent work and is well-presented. If appropriate, I recommend in favor of granting of the doctoral degree.

Specific comments
- Pg. 27: Isoniazid is misspelled in the table.
- Pg. 39: The following sentence is repeated "The only parameter that needs careful definition is the magnitude of the applied force, which must not interfere with the calculated relative residence times".
- Pg 39: Input systems were protonated with PyMOL. How were Histidine residues treated? Were they all set to neutral or all protonated or was each His evaluated to determine if it should be neutral or protonated by looking at its environment and/or by conducting a protonation state prediction via APBS or some other tool?
- Energy minimization was conducted but no mention was made about when the min was stopped. In principle, it should have been when a threshold in max force or change in system

energy was reached (i.e., the system converged). Sometimes min is done for a fixed number of steps (which is not ideal), but not even this is mentioned.

- Pg. 82: “A similar situation was observed for interacting with amino acid Asp41, especially hydrophobic.” I do not understand this. Asp41 is hydroPHILIC.