Molecular Docking: A Comprehensive Screening of Eight Well-Known Softwares and Their Accuracy, Versatility, and Speed Regarding The Docking of a Functionally Diverse Library of Pharmaceuticals to their Receptors

Veda Kamaraju[1,*], Tanusree Banerjee[1,*], Shreya Sundar[1], Aditi Karthik[1], Ria Kolala[1], and Krithikaa Premnath[1]
Advisor: Harman Brah[1]

[1]Aspiring Scholars Directed Research Program, 43505 Mission Blvd, Fremont, CA 94539
[*]Denotes equal authorship

With molecular docking being a crucial tool for drug discovery, the number of docking softwares, both commercial and open-web, have significantly grown over the years. This raises the necessity of identifying the predicting software that is the most accurate, efficient, and multifaceted. After docking 10 ligands with their respective receptors through 8 different softwares, resulting binding affinities were compared to literature values in order to analyze accuracy. The speed and versatility of each software was recorded as well. Due to AutoDock Vina's major prevalence and reputation in the scientific community, we predicted that it would successfully rank the highest given all the criteria. Out of the 8 softwares evaluated, ParDock produced values closest to each molecule's real binding values, followed closely by DINC, AutoDock Vina, and CB-Dock. After taking into account both speed and versatility, AutoDock Vina ranked the highest followed by DINC and CB-Dock. The understanding of these accessible software's features and results can potentially improve the future of molecular docking.

molecular docking | protein-ligand complex | PDB files | structural optimization | binding affinity | percent error

Correspondence: ***harman.brah@fremontstem.com***

Introduction

Structure-based Drug Design

Molecular modeling for the purpose of structure based evaluations of molecular level interactions has been critical in modern pharmaceutical chemistry. Methods for biomolecular spectroscopy allow for critical examination of key macromolecular drug targets[1]. As such, structure based design methods which utilize three dimensional structural information regarding its targets is an emerging aspect of modern pharmaceutical chemistry. Among such methods, molecular docking allows for the analysis of binding energies, molecular interactions, as well as conformational shifts. Molecular docking seeks to predict the conformation of two biological binding partners, typically a peptide or protein with a small macromolecule, such as ligands or drugs[2].

Docking Softwares

Docking at the molecular level is a process which involves conforming the ligand to its target receptor in the right pose so as to minimize binding energies. Geometric and electrostatic interactions play a critical role in quantifying the accuracy of the orientation of the ligand to the active site of its targeted receptor. As such, Coulombic and Van der Waals interactions, which quantify the interactions between the electrical charges of the molecules, in addition to the formation of hydrogen bonds, are summed together to form a binding score which is indicative of the binding potential between the two molecules[3]. Docking softwares work by incorporating search algorithms which recursively search the orientation of the ligand until the binding energy of the ligand to the receptor is minimized.

Theoretical Basis of Docking

Over the past few decades, theories and methodologies developed in regard to molecular docking are used as the fundamental base of operation for the majority of docking softwares. The earliest records of docking methodology are based off of Fischer's lock and key model which proposes that the optimal conformation is when the substrate, such as the ligand of the drug fits into the active site of the macromolecule such as a protein or a peptide, similar to the conformation in which a lock fits into a key[4,5]. In essence, it ideated the concept of rigid docking in which both the ligand and the receptor are treated as rigid bodies, and in which the binding affinity score holds a proportional relationship to the geometric fit of the ligand to its targeted receptor. The induced-fit theory, introduced by Daniel Koshland in 1958, proposes a more flexible style of docking in which both the target and receptor make minor conformational and geometric changes to adapt to each other's core shapes in order to optimize their

best fit[2]. The movement affects several side chains in contrast to typically independent side chains. As a result of this flexibility and ability to adapt, flexible docking algorithms are able to implement higher accuracy and efficiency in predicting both binding affinities and modes in comparison to their rigid body docking counterparts[3]. In addition to the small induced fit methodology, the ensemble of conformational states model observes the more major conformational changes undergone by proteins and peptides and capitalizes on the plasticity of proteins which allows them to switch from one state to another[6]. These individual models and methodologies of molecular docking developed over the years each highlight a specific portion of the molecular recognition process. While Fischer's model focuses on the 3D complementarity of the protein and the ligand, the induced fit model works on the process of molecular modification to achieve the best complementarity, and the ensemble model highlights the conformational complexities of the molecules[5].

In recent years of study in structure based drug design, more than 60 softwares have been developed for the purposes of both academic and commercial use, such as AutoDock, Autodock Vina, Blaster, HADDOCK, ParDock, PatchDock, DINC, and SwissDock[3]. The approaches utilized by these softwares range from shape-based algorithms to incremental recursive approaches to genetic construction approaches. Albeit some exceptions amongst these softwares, a vast majority of flexible docking programs capitalize on the flexibility of the ligand while treating the receptor as rigid[7]. The programs are evaluated based on their ability to predict the optimal binding pose while minimizing the binding energy it takes to achieve the most optimal conformation.

Algorithms and Scoring Functions of Docking

In order to distinguish among numerous softwares, two components of molecular docking are assessed: sampling algorithms and scoring functions. Due to the several degrees of freedom of both the ligand and protein, as well as the six degrees of translational and rotational freedom, the number of possible binding poses between the two molecules is too large for even modern computers to generate in a short amount of time. Thus, sampling algorithms are incorporated to identify the best possible conformations[8]. Matching algorithms (MA) match a ligand to an active site based on molecular shapes and chemical properties. Ligand conformations are generated by recording distances between the pharmacophore, within the protein and ligand, and the respective ligand atoms. Hydrogen bonding is taken into consideration during the match as well. With the high speed feature, these algorithms are used in DOCK and SANDOCK softwares[9]. Another common method is the Monte Carlo. These algorithms utilize bond rotation and rigid-body translation or rotation to form possible modes of the ligand. The resulting conformation is weighed against an energy-based criterion[5]. This method, which is integrated in the AutoDock and Affinity program, allows the ligand to pass through energy barriers at a higher degree. Genetic algorithms (GA) indicate that the ligand's degrees of freedom are translated to genes, which make up chromosomes. These chromosomes represent the modes of the ligand. Two factors are highlighted in GA: mutation and crossover[5,8]. Mutations can change the genes in an unpredictable manner, while crossover switches genes on two chromosomes. When these factors impact the genes, new ligand structures are formed. AutoDock, GOLD, and DARWIN are popular softwares that use genetic algorithms[10]. Molecular dynamics (MD) is one of the most powerful simulation methods in molecular modeling. Compared to other algorithms, MD simulations depict the flexibility of the ligand and protein more accurately and effectively. However, MD simulations may lead to insufficient sampling, as they have a hard time passing through high energy barriers. To overcome this disadvantage, a random search, as well as mini MD simulations, can be utilized to recognize the ligand conformation[4].

Scoring functions make up the second component of molecular docking, and are crucial in ranking resulting poses. These functions assess the quality of docking conformations, while guiding the search algorithms towards accurate ligand modes. The first necessity of a scoring function is to discern the experimentally observed poses from the poses found by the search algorithms[11]. The second requirement is to distinguish active and inactive compounds, and the final goal is to predict binding affinities. This step of binding affinity prediction ranks the compounds in order, placing the most potent poses at the top. Scoring functions are generally classified into force-field based, knowledge-based, and empirical functions[11,12]. Force-field functions, utilized by DOCK and DockThor, involve calculating energy terms from a force field and considering interactions between the protein-ligand complex. Knowledge-based functions include a statistical analysis of atom pairs in the complex, and are used by DrugScore and PMF. Empirical scoring functions reproduce experimental data and employ binding affinity data of experimentally based structures. In other words, absolute free energy of binding can be predicted from 3D structures of the complex[4,11].

Types of Docking

Rigid body docking focuses on the existent surface complementarity of the ligand and the receptor, thus producing a large number of conformations and potential binding poses which undergo the process of ranking in accordance with the free energy of approximation associated with each of the conformations. The Fast Fourier transformation utilizes electrostatic interactions to explore the potential docking space, with the docking potential being limited to a correlation function form. The deployment of the transformation allows the search space to be expanded to a 3D space via spherical decomposition. Albeit having good surface complementarity, rigid docking algorithms often produce false-positives and do not adapt well to unbound crystal structures[3].

In recent studies, the flexibility of side chains has proven to have a critical role in the docking of protein-ligand complexes[3]. As a result, the binding site of the receptor can be adapted to the specific orientation of the ligand. Presently,

Monte Carlo methods, in site combinatorial searches, ligand buildup, and site mapping are the primary strategies used in the flexible docking of ligands[3,13]. As such, the large conformational modifications used in the mentioned methods allows the ligand to overcome energy barriers. Potential molecular affinities combined with conformational searches allows for an increased efficiency in the process of substrate docking for known structures[13].

This study aims to identify the most optimal software for molecular docking, while weighing in factors of accuracy, speed, and versatility. 10 molecules were docked using 8 different docking softwares and web servers, and resulting binding affinities were found. These values were compared against the literature binding affinities. Molecular docking has long been used in the scientific community for the purposes of drug discovery. The analysis of this study seeks to aid in the debate over best methods of docking both commercially and academically.

Methods

Optimization Via Molecular Mechanics

A total of 10 known ligands and their receptors were used to test the accuracy of 8 docking softwares respectively: Aspirin and Cyclooxygenase, Taxol and Beta-Tubulin, Doxorubicin and DNA, Acetaminophen and Naloxone, Monastrol and EG-5 Kinesin, Berberine and TLR4-MD-2, Anthramycin and DNAse 1, Donepezil and Acetylcholinesterase, Ibuprofen and Cyclooxygenase, and Atorvastatin and HMG-coA reductase, depicted with their respective structures and PDB codes in Figure 1. Prior to performing density functional theory (DFT) optimization, molecular mechanic optimization was performed on the 10 subject ligands. To execute the above described optimization, Avogadro (2016), which allows for molecular editing and visualization, was utilized[14]. The optimization was performed using the Universal Force Field (UFF) to 10,000 steps. The optimized structure of the molecules was then used as the input for DFT structural optimization.

DFT Optimization of Molecules

Density functional theory optimization, a method that performs ground-state calculations on rigid molecules, is used to increase the accuracy of the molecular shape and increase its versatility in terms of usage with regard to molecular docking. ORCA (2018-4.2.1), a quantum chemistry program, was used side by side with Avogadro to perform DFT optimizations on the ten ligands. A conductor-like polarizable continuum (CPCM) solvation model of water was used side by side with the RIJCOSX approximation approach while performing DFT with the B3LYP functional, a hybrid functional chosen for its speed and accuracy in comparison to other functionals that would have a more unfavorable trade-off on duration for higher precision. The def2-SVP basis set, containing the polarization of all atoms, was used alongside the approximation and functional systems. The completed optimized files were converted to the appropriate mediums to adapt to the input requirements of the eight following docking softwares.

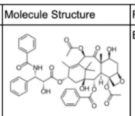
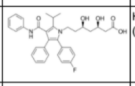
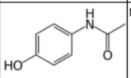
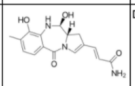
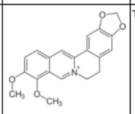
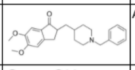
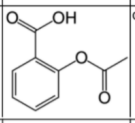
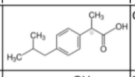
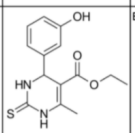
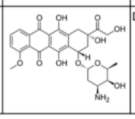
Ligand	Molecule Structure	Receptor (PDB Code)	Complex PDB Code
Taxol		Beta-Tubulin	5SYF
Atorvastatin		HMG-CoA Reductase (1DQ9)	1HWK
Acetaminophen		Naloxone	4YJ6
Anthramycin		DNase 1	274D
Berberine		TLR4-MD 2	3NP6
Donepezil		Acetylcholinesterase	5NAU
Aspirin		Cyclooxygenase 2	5F19
Ibuprofen		Cyclooxygenase 2	4PH9
Monastrol		EG-5 Kinesin (3HQD)	1X88
Doxorubicin		DNA	1P20

Fig. 1. Ligand, Receptor, and Complex Information

SwissDock

Formulated by Swiss Institute of Bioinformatics, the SwissDock software utilizes the EADock DSS algorithm methodology wherein several binding modes are generated within given grid coordinates and parameters or within the general area surrounding the target site, as in the case of blind docking[15,16]. In parallel to this, the CHARMM energies of the binding modes are approximated[17]. The binding energies formulated are evaluated using FACTS, an efficient generalized Born implicit solvent model, and clustered accordingly. The conformations of the most optimal energies are then outputted for visualization through the software[16]. In this study, we evaluated the efficiency of the docking potential of the software by establishing a comparison between literature binding affinities of the tested molecules and the free binding energies produced by the molecular docking software. We focused on docking the optimized ligand structure to the molecular complex of the ligand bound to the receptor. The software produced the best optimized conformation alongside a ranking of the different binding poses and their corresponding binding affinities. The free binding energies are presented in kcal/mol, being directly comparable to the

literature binding affinities, and is indicative of the binding energy of the ligand to its receptor.

PatchDock

PatchDock, created by the Swiss Bioinformatics Group alongside the team of the Tel Aviv Computer Science School, implements a molecular docking algorithm based on shape complementarity principles to dock proteins, peptides, and drugs through techniques typically utilized in Computer Vision, such as object recognition and image segmentation[18,19]. When implemented, the algorithm produces a wider interface area as well as a series of minor steric clashes. The algorithm transitions through the stages of molecular shape representation, surface patch matching, and filtering and scoring respectively. The program first computes the surface of the molecule so as to filter out geometric patches and maintain only the patches with "hot spot" residues, organizing the Connolly dot surface representations into concave, convex, and flat sectors in order to do so[18,20]. Geometric hashing and pose-clustering matching methods are then applied to match the patches which were previously filtered. Individual candidate transformations are further evaluated via a scoring function, taking into consideration both desolvation and geometric binding energies[18]. An RMSD, or root mean square deviation clustering, is then applied to remove redundancy in the solution set of conformations. The increased efficiency of the molecular docking program is instigated by the quickened pace of its transformational search which relies on the feature matching system rather than a search of the potential conformational space. This allows for average sized protein jobs to range less than 10 minutes in complete execution of the docking process[21]. For the purpose of this study, along the inputted receptor and ligand, the clustering RMSD condition was left at 4.0 and the complex type was set to protein-small ligand. The software produces a web results page with the top 20 binding conformations and poses with a solutions table which incorporates a geometric score, desolvation energy, size of the interfaced area, and the actual rigid transformation within itself.

ParDock

Developed by Professor B. Jayaram and team of the Indian Institute of Technology, New Delhi, PARDock, or Parallel Dock, utilizes a web based Monte Carlo docking system which works as an automated, parallelly processed function[6]. In the program, the Monte Carlo methodology is used in a six dimensional space in order to explore a larger quantity of conformations in search for the optimal location to bind to the targeted molecule. The docking system contains a broad dataset of a total of 226 protein-ligand complexes comprising 81 unique proteins taken from RCSB online portal. The software uses the inputted protein-ligand complex to automate a process for optimizing conditions for docking the inputted candidate ligand molecule. The docking program optimizes the ligand's geometry via the AM1, or Austin Model 1 method, followed by the process of determining the partial charge via the AM1-BCC methodology, which uses Mulliken-type partial charges taken from the AM1 quantum mechanical wave functiontripathi2017molecular,sousa2013protein. The AMBER force field is deployed to automatically assign the atom types, bond angles, dihedral and Van der Waals energies, and other parameters for the subjected ligand. Albeit the efficiency of the automated process, the accuracies of binding site predictions, and the capacity for flexibility of both the protein and ligand, remain as deficit qualities of the docking software program[22]. For this study, the ligand was specified to be docking to the active site of the protein as opposed to the entirety of the molecule, using 500 minimal cycles, and a ligand flexibility mode of 5. The software outputs the most optimal conformation as a visualization, while providing the top 4 binding poses with the optimal binding energies ΔG(kcal/mol) with downloadable conformation visualizations for each.

DockThor

DockThor is a web server flexible docking software that prepares ligands and receptors for all types of docking methodologies and utilizes the JSMOL java-based graphical user interface to visualize 3D versions of predicted complexes[23–25]. It was used to blindly dock the ligands to the receptors, which were uploaded in .pdb format. No protection states were altered, no cofactors were used, the rotatable bond editor was disabled, and the algorithm precision section was not specified.

AutoDock Vina

Unlike the other web softwares, AutoDock is an open-source program for molecular docking that can function on an executable file[26]. Compared to AutoDock 4, Vina achieves a significantly higher binding accuracy in less time. This study utilized Chimera, a visualization application, rather than through the more-commonly seen command-line approach, to run AutoDock Vina. Using a gradient optimization method, Vina can rank the poses effectively. The predicted binding energy is calculated through assessing intermolecular attractions of the lowest-scoring conformation[27]. The receptor was prepared using the DockPrep feature, adding charges where necessary. Grid coordinates were specified and adjusted accordingly to visually encompass the residue at hand, indicating a potential active site.

DINC

Docking Incrementally, otherwise identified as DINC, was created by the Kavraki Lab at Rice University to perform more seamless docking processes of large ligands[28,29]. It deploys an incremental algorithm in order to perform an efficient search for the space in which potential binding modes and conformations may be created between the ligand and the receptor[28,30]. The incrementing algorithm allows for easier processing of information by breaking down the search so as to optimize each one before proceeding to the next. Through the software, the ligand is treated as the superposition of a rigid body and rotatable component respectively. The search space is thus expanded for the ligand with numerous rotat-

able components. With its incremental approach, the "partial solutions" formulated at the end of each stage of docking correspond to contiguous sections of the ligand and are expanded through the process of adding atoms to the individual fragments till the ligand's original structure and composition is restored[30]. The software treats a certain set of rotatable bonds as active in each of the stages of incremental docking. Furthermore, DINC follows a meta-docking approach, relying on AutoDock Vina, a standard docking tool to perform sampling and score in each stage of the process. At the end of the process, the most optimal conformations are ranked and presented, based on binding energy ranking, with the highest ranked conformation visualized[31]. Along with binding energy, DINC also sorts out conformations based on RMSD clustering, or the root mean square deviation of atomic positions, which measure the average distance between atoms of the superimposed proteins, and provides the most optimal distance through a visualization[32].

Achilles

Achilles is a simple blind docking web server that specializes in small-molecule docking by incorporating AutoDock Vina's functionality. Utilizing pose clustering, cluster distance, and binding energy plots, the docking results can easily be imported to PyMOL, its supported visualization system, for further examination. The ten ligands and their receptors were uploaded in .pdb format to dock.

CB-Dock

CB-Dock (Cavity-detection guided Blind Docking) is a web server which automatically recognizes binding sites, calculates grid coordinates for the center and size, and completes molecular docking using AutoDock Vina[33,34]. Cavity-based docking has been proved to output a higher accuracy of conformations and hit-ratios. The software goes through 4 stages. Because this software was developed to perform blind docking at sites which have already been predicted, the process begins with detecting cavities and respective binding sites (cavity detection). The next step, cavity sorting, selects various top cavities according to the size. The final stage, Dock and Rerank, arranges and ranks the resulting poses according to a docking score. Input files remain in the .pdb format for ligands and .pdb or mol2 for receptors[35]. The best binding pose is given as the first conformation listed.

Results/Discussion

Ligands	DINC	DockThor	PatchDock	ParDock	AutoDock Vina	SwissDock	CB-Dock	Achilles	Literature Binding Affinity
Taxol	-9.2	-8.638	-19.44	-9.95	-8	-8.88	-8.6	error [ligand]	-11 ± 0.02 [40]
Atorvastatin	-9.2	-8.721	-19.95	-5.8	-8	-9.54	-8.3	error[ligand]	-10.8 [41]
Acetaminophen	-5.8	-6.703	-8.65	-6.26	-6.4	-6.38	-6.6	-6.2	-5.3 [42]
Anthramycin	-5.2	can't dock [nucleic acids]	-13.78	error	-6.3	error [receptor]	-5.1	Error [receptor]	-7 [43]
Berberine	cannot dock	can't dock [nucleic acids]	-17.82	error	-7.1	error [receptor]	-7	error [receptor]	-7.70 [44]
Donepezil	-10.4	-10.688	-17.3	-10.21	-10.6	-9.24	-11.1	-10.8	-11.9 [45]
Aspirin	-5.4	-6.238	-7.05	-4.34	-6.2	-6.92	-7	-7	-3.8 ± 0.5 [46]
Ibuprofen	-7.2	-7.88	-9.61	-9.21	-6.6	-7.9	-7.7	-7.1	-9.52 [47]
Monastrol	-6.2	-7.768	-16.03	-6.17	-7.9	-7.89	-7.2	-6.7	-6.67 [48]
Doxorubicin	-5.4	can't dock [nucleic acids]	-15.72	error	-5.8	error [receptor]	-5.9	error [receptor &ligand]	-6.88 [49]

Fig. 2. Predicted Binding Affinities (values shown in kcal/mol)

Given the wide range of computed binding affinities and number of softwares, multiple trends have been observed centering specific softwares or molecules. As seen in Figure 2, all 8 softwares overestimated the binding affinity of Aspirin to Cyclooxygenase and the binding affinities all softwares predicted with the lowest percent error were that of Donepezil's to Acetylcholinesterase and Monastrol's to EG-5 Kinesin. The percent errors of all molecules per software and the average percent error per software is shown in the heat map (Figure 3). As seen in Figure 4, PatchDock significantly overestimated the binding affinities of nine out of ten ligands to their receptors with an average percent error rate of 85%. The computed binding affinities for Acetaminophen to Naloxone were relatively similar (not the most accurate) across all softwares in comparison to the predictions for other molecules for which softwares had computed a much larger range of values for. Jobs for Anthramycin, Doxorubicin, and Berberine were the most frequent to fail, most likely because their receptors were nucleic acid macromolecules which many softwares like DockThor, SwissDock, Achilles, and ParDock did not support. The highest percent error of all the queued docking jobs was found in PatchDock's computation of Monastrol bound to EG-5 Kinesin at 140% and the lowest percent error of all the jobs was surprisingly Achilles's computation of Monastrol bound to EG-5 at 0.45%. The software with the lowest average percent error was found to be ParDock with an average error of 16%, followed by DINC at 19%, AutoDock Vina at 23%, CB-Dock at 24%, DockThor at 25%, Achilles and SwissDock at 27%, and then PatchDock at 85%. The majority of the softwares excluding PatchDock, DINC, and ParDock, remained in the 20-30% error range. Giving the least accurate affinities, PatchDock surprisingly took the longest amount of time, around 4-6 days on average, to compute the binding affinities of the 10 molecules, followed by DockThor which took 4-6 hours, SwissDock which took 30 minutes-1 hour, ParDock which took 15-30 minutes, Achilles which took 5-10 minutes, CB-Dock which took 3-5 minutes, DINC which took 1-2 minutes, and AutoDock Vina which took less than one minute. The most versatile softwares, judged based on their ability to dock various ligand shapes and sizes, support different types of atoms on ligands, and support different types of receptor macromolecules, were seen to be PatchDock, AutoDock Vina, and CB-Dock which computed results for all 10 molecules, followed by DINC which produced results for 9 molecules, DockThor, ParDock, and SwissDock which produced results for 7 molecules, and Achilles which produced results for 5 molecules. This disparity is due to some softwares not supporting certain atoms on ligands, larger ligands, and nucleic acid receptors, which will be explained below.

Docking Errors

Out of the eight softwares that our project revolved around, only three gave results for all ten molecules: Autodock Vina, Patchdock, and CB-Dock. DINC was unable to dock the berberine-TLR4-MD-2 complex, saying the docking job couldn't be processed. The remaining had the common error

Ligand	DINC	DockThor	PatchDock	ParDock	AutoDock Vina	SwissDock	CB-Dock	Achilles
Taxol	16 %	21 %	77 %	9.5 %	30 %	19 %	22 %	
Atorvastatin	15 %	19.3 %	84.7 %	46 %	30 %	11.7 %	23 %	
Acetaminophen	9.4 %	26 %	63 %	18 %	21 %	20. %	25 %	17 %
Anthramycin	30 %		100%		10 %		30 %	
Berberine			131 %		7.8 %		9 %	
Donepezil	12.6 %	10.2 %	45.4 %	14.2 %	10.9 %	22.4 %	6.72 %	9.24 %
Aspirin	42 %	64 %	86 %	14 %	63 %	82 %	80 %	80 %
Ibuprofen	24 %	17.2 %	0.945 %	3.26 %	31 %	17 %	19 %	25 %
Monastrol	7.0. %	16.5 %	140. %	7.50. %	18 %	18.3 %	7.9 %	0.45 %
Doxorubicin	22 %		126 %		16 %		14 %	
AVERAGE	19 %	25 %	85 %	16 %	23 %	27 %	23.782	27 %

0-10 % error	11-20 % error	21-30 % error	31-40 % error	41-60 % error	61-80 % error	81-100 % error	>100 % error	NA

Fig. 3. Heat Map Depicting Percent Errors of All Softwares By Molecule and Accuracy Trends. The ± shown in Figure 2 beside the literature affinities for aspirin and Taxol were not taken into consideration while calculating percent error.

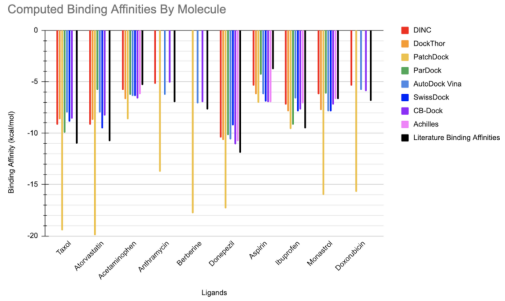


Fig. 4. Column Chart Depicting Further Trends Related To Accuracy and Versatility

of not being able to run 3 specific molecules: Anthramycin, Berberine, and Doxorubicin. Receptors for all three of these molecules are DNA or nucleic acids. Hence, the discrepancy with non-standard residues necessitates an alteration of fundamental settings within the software. This feature contributes to the versatility factor, as only 3 software settings were successfully able to accept nucleic acids. DockThor, Pardock and Swissdock produced results for all molecules except the aforementioned complexes, and in these softwares, the receptors produced errors while uploading. The three receptors would not load in Achilles, due to no alpha carbons being detected in the ligands or receptors. Along with those three complexes, Achilles could not run Taxol and Beta-Tubulin and Atorvastatin-HMG-CoA reductase, giving the error that the ligand had too many torsional degrees of freedom.

Scoring Rubric

As seen so far, softwares might perform well in one quota but very poorly in other categories. AutoDock Vina was the fastest and one of the most versatile softwares, but its predictions of binding affinities, even though fairly accurate, were outdone by other softwares like DINC and ParDock. PatchDock, even though it was one of the most versatile softwares, did not as accurately and quickly predict the binding affinities like other softwares did. Since the versatility, accuracy, and speed of the software are all crucial components of efficient and effective molecular docking, the softwares were all assessed on these three quotas which were weighted equally. Each quota, accuracy, versatility, and speed, was scored out of 100 points. Accuracy of each software was calculated as the difference of the average percent error and 100 (ex. ParDock which had an average percent error of 16% had an accuracy of 84%). For each job successfully completed, softwares received 10 points in the versatility category (ex. Since Vina successfully ran all 10 molecules so it received 100/100 points in this category and Achilles successfully ran 5 of the 10 molecules so it received 50 points in this category). Since there were only 8 softwares, points were designated as 12.5, 25. 37.5, 50, 62.5, 75, 87.5, and 100, from slowest to fastest software (ex. Since Vina was the fastest it received 100 points out of 100 and DINC being the second fastest received 87.5 points out of 100). Figure 5 displays the rubric and the final scores, showing that AutoDock Vina, DINC, and CB-Dock were the top three softwares respectively.

	DINC	DockThor	PatchDock	ParDock	AutoDock Vina	SwissDock	CB-Dock	Achilles
Versatility	90/100	70/100	100/100	70/100	100/100	70/100	100/100	50/100
Accuracy	81/100	75/100	15/100	84/100	77/100	73/100	76/100	73/100
Speed	87.5/100	25/100	12.5/100	50/100	100/100	37.5/100	75/100	62.5/100
Final Score	259/300	170/300	128/300	204/300	277/300	181/300	251/300	186/300

Fig. 5. Scoring Rubric for All Eight Docking Softwares

Conclusion

From this study, it can be concluded that AutoDock Vina, DINC, and CB-Dock are the top proficient softwares given their prompt speed, multifaceted capabilities, and their near-precise predictions of binding poses and affinities. Due to molecular docking's high prevalence and importance in structural drug design, the results from this study can be virtually applied to all other computational lab investigations and pharmaceutical research. Given the hundreds of docking servers available for commercial and leisure use in the industry, many users face a challenge when it comes to choosing the correct software given everyone's varying needs and all of the softwares' varying potential. More accurate softwares tend to require more time to predict binding poses while faster softwares are less accurate, but even these trends are not seen across all softwares given the various algorithms servers are built on and the types of molecules that they are capable of docking. All chosen softwares for this experiment are free and open to the public so in order to cater to everyone's varying needs, versatility, speed, and accuracy were weighted equally, resulting in the aforementioned softwares performing the best out of all eight. AutoDock Vina, even though the fastest and most versatile, requires a lot of receptor preparation before queuing a docking job. CB-Dock, a more user-friendly software is not as accurate as DINC and AutoDock and takes a little more time. DINC, even though it is not as fast as Vina or as versatile as it and CB-Dock, it produces results very comparable to Vina and requires no receptor preparation. For these reasons, AutoDock Vina is the best docking software for professional use and DINC, followed by CB-Dock, is the best docking software for leisure use and beginners.

AUTHOR INFORMATION

All authors contributed to the research and writing of this paper. Veda Kamaraju worked on the data collection through various docking softwares, troubleshooting of specific errors, and paper introduction, methods, and formatting. Shreya Sundar worked on data collection, conducted multiple molecular optimizations and analysis of the data, constructed the figures, and wrote a large portion of the paper. Aditi Karthik worked on data collection via many different docking softwares, data analysis, and contributed to the paper. Tanusree Banerjee performed several DFT optimizations, worked on collecting data from several docking softwares, worked on paper introduction, methods, and results.

ACKNOWLEDGEMENTS

We would like to thank the Olive Children Foundation, Western Digital Foundation, and BioLink Depot for research funding of the Aspiring Scholars Director Research Program. We would also like to thank Thermo Fisher Scientific and Nanalysis Corporation for scientific support, and DuPont pioneer and BioLink Depot for material support. We would also like to thank the faculty, especially Harman Brah, the advisor for this project, and the rest of the staff of Fremont STEM.

Bibliography

1. Ruben Abagyan and Maxim Totrov. High-throughput docking for lead generation. *Current opinion in chemical biology*, 5(4):375–382, 2001.
2. Tatu Pantsar and Antti Poso. Binding affinity via docking: fact and fiction. *Molecules*, 23(8): 1899, 2018.
3. Nataraj S Pagadala, Khajamohiddin Syed, and Jack Tuszynski. Software for molecular docking: a review. *Biophysical reviews*, 9(2):91–102, 2017.
4. Xuan-Yu Meng, Hong-Xing Zhang, Mihaly Mezei, and Meng Cui. Molecular docking: a powerful approach for structure-based drug discovery. *Current computer-aided drug design*, 7(2):146–157, 2011.
5. Veronica Salmaso and Stefano Moro. Bridging molecular docking to molecular dynamics in exploring ligand-protein recognition process: An overview. *Frontiers in pharmacology*, 9: 923, 2018.
6. Ashutosh Tripathi and Vytas A Bankaitis. Molecular docking: From lock and key to combination lock. *Journal of molecular medicine and clinical applications*, 2(1), 2017.
7. Zhe Wang, Huiyong Sun, Xiaojun Yao, Dan Li, Lei Xu, Youyong Li, Sheng Tian, and Tingjun Hou. Comprehensive evaluation of ten docking programs on a diverse set of protein–ligand complexes: the prediction accuracy of sampling power and scoring power. *Physical Chemistry Chemical Physics*, 18(18):12964–12975, 2016.
8. Aaftaab Sethi, Khusbhoo Joshi, K Sasikala, and Mallika Alvala. Molecular docking in modern drug discovery: Principles and recent applications. In *Drug Discovery and Development-New Advances*. IntechOpen, 2019.
9. N Moitessier, P Englebienne, D Lee, J Lawandi, and CR Corbeil. Towards the development of universal, fast and highly accurate docking/scoring methods: a long way to go. *British journal of pharmacology*, 153(S1):S7–S26, 2008.
10. Connie M Oshiro, Irwin D Kuntz, and J Scott Dixon. Flexible ligand docking using a genetic algorithm. *Journal of computer-aided molecular design*, 9(2):113–130, 1995.
11. Isabella A Guedes, Felipe SS Pereira, and Laurent E Dardenne. Empirical scoring functions for structure-based virtual screening: applications, critical aspects, and challenges. *Frontiers in Pharmacology*, 9:1089, 2018.
12. Jie Liu and Renxiao Wang. Classification of current scoring functions. *Journal of chemical information and modeling*, 55(3):475–482, 2015.
13. Nelly Andrusier, Efrat Mashiach, Ruth Nussinov, and Haim J Wolfson. Principles of flexible protein–protein docking. *Proteins: Structure, Function, and Bioinformatics*, 73(2):271–289, 2008.
14. Robert Kiss, Mark Sandor, and Ferenc A Szalai. http://mcule. com: a public web service for drug discovery. *Journal of cheminformatics*, 4(S1):P17, 2012.
15. David S Goodsell and Arthur J Olson. Automated docking of substrates to proteins by simulated annealing. *Proteins: Structure, Function, and Bioinformatics*, 8(3):195–202, 1990.
16. Aurélien Grosdidier, Vincent Zoete, and Olivier Michielin. Fast docking using the charmm force field with eadock dss. *Journal of computational chemistry*, 32(10):2149–2159, 2011.
17. Aurelien Grosdidier, Vincent Zoete, and Olivier Michielin. Swissdock, a protein-small molecule docking web service based on eadock dss. *Nucleic acids research*, 39(suppl_2): W270–W277, 2011.
18. Dina Schneidman-Duhovny, Yuval Inbar, Ruth Nussinov, and Haim J Wolfson. Patchdock and symmdock: servers for rigid and symmetric docking. *Nucleic acids research*, 33(suppl_2):W363–W367, 2005.
19. Dina Duhovny, Ruth Nussinov, and Haim J Wolfson. Efficient unbound docking of rigid molecules. In *International workshop on algorithms in bioinformatics*, pages 185–200. Springer, 2002.
20. Efrat Mashiach, Dina Schneidman-Duhovny, Nelly Andrusier, Ruth Nussinov, and Haim J Wolfson. Firedock: a web server for fast interaction refinement in molecular docking. *Nucleic acids research*, 36(suppl_2):W229–W232, 2008.
21. Irina Kufareva and Ruben Abagyan. Methods of protein structure comparison. In *Homology Modeling*, pages 231–257. Springer, 2011.
22. Sergio F Sousa, Antonio JM Ribeiro, JTS Coimbra, RPP Neves, SA Martins, NSHN Moorthy, PA Fernandes, and MJ Ramos. Protein-ligand docking in the new millennium–a retrospective of 10 years in the field. *Current medicinal chemistry*, 20(18):2296–2314, 2013.
23. Karina B. Santos, Isabella A. Guedes, Ana L. M. Karl, and Laurent E. Dardenne. Highly flexible ligand docking: Benchmarking of the dockthor program on the leads-pep protein–peptide data set. *Journal of Chemical Information and Modeling*, 60(2):667–683, 2020.
24. Karina B Santos, Isabella A Guedes, Ana LM Karl, and Laurent E Dardenne. Highly flexible ligand docking: Benchmarking of the dockthor program on the leads-pep protein–peptide data set. *Journal of Chemical Information and Modeling*, 60(2):667–683, 2020.
25. Camila Silva de Magalhães, Diogo Marinho Almeida, Helio José Correa Barbosa, and Laurent Emmanuel Dardenne. A dynamic niching genetic algorithm strategy for docking highly flexible ligands. *Information Sciences*, 289:206–224, 2014.
26. Oleg Trott and Arthur J Olson. Autodock vina: improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. *Journal of computational chemistry*, 31(2):455–461, 2010.
27. Xing Du, Yi Li, Yuan-Ling Xia, Shi-Meng Ai, Jing Liang, Peng Sang, Xing-Lai Ji, and Shu-Qun Liu. Insights into protein–ligand interactions: mechanisms, models, and methods. *International journal of molecular sciences*, 17(2):144, 2016.
28. Dinler A Antunes, Mark Moll, Didier Devaurs, Kyle R Jackson, Gregory Lizée, and Lydia E Kavraki. Dinc 2.0: A new protein–peptide docking webserver using an incremental approach. *Cancer research*, 77(21):e55–e57, 2017.
29. Rob L.M. van Montfort, Paul Workman, Rob L.M. van Montfort, and Paul Workman. Structure-based drug design: aiming for a perfect fit. *Essays in Biochemistry*, 61(5):431–437, 11 2017. ISSN 0071-1365. doi: 10.1042/EBC20170052.
30. Didier Devaurs, Dinler A Antunes, Sarah Hall-Swan, Nicole Mitchell, Mark Moll, Gregory Lizée, and Lydia E Kavraki. Using parallelized incremental meta-docking can solve the conformational sampling issue when docking large ligands to proteins. *BMC molecular and cell biology*, 20(1):42, 2019.
31. Ankur Dhanik, John S McMurray, and Lydia E Kavraki. Dinc: a new autodock-based protocol for docking large ligands. *BMC structural biology*, 13(S1):S11, 2013.
32. Hyoungrae Kim, Cheongyun Jang, Dharmendra K Yadav, and Mi-hyun Kim. The comparison of automated clustering algorithms for resampling representative conformer ensembles with rmsd matrix. *Journal of cheminformatics*, 9(1):21, 2017.
33. Yang Liu, Maximilian Grimm, Wen-tao Dai, Mu-chun Hou, Zhi-Xiong Xiao, and Yang Cao. Cb-dock: a web server for cavity detection-guided protein–ligand blind docking. *Acta Pharmacologica Sinica*, 41(1):138–144, 2020.
34. Yang Cao and Lei Li. Improved protein–ligand binding affinity prediction by using a curvature-dependent surface-area model. *Bioinformatics*, 30(12):1674–1680, 2014.
35. Sérgio Dias, Tiago Simões, Francisco Fernandes, Ana Mafalda Martins, Alfredo Ferreira, Joaquim Jorge, and Abel JP Gomes. Cavbench: A benchmark for protein cavity detection methods. *PloS one*, 14(10):e0223596, 2019.