In-Silico Docking Studies of Angiotensin Converting Enzyme Using Natural Inhibitor

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Abstract

Lowering blood pressure (BP) using antihypertensive medicines lowers the risk of target organ failure as well as the occurrence of cardiovascular disease. The most common modifiable risk factor for death and disability is hypertension, which is associated with strokes, increased coronary and systemic atherosclerosis, heart problems, and chronic kidney diseases (CKD). The majority of deaths and disabilities globally now result from cardiovascular diseases (CVDs), mainly in low- and middle-income nations. Numerous genetic, behavioral, and environmental risk factors all contribute to hypertension, a significant component in the advancement of CVD. Given the significance of protein-ligand interactions in structure-based therapy development, we molecularly docked nifedipine to the cardiovascular target protein to determine the drug’s binding affinity. The Angiotensin Converting Enzyme (Target hypertension Protein) three-dimensional (3D) structure was docked using the Autodock tool, which was retrieved from the Protein Data Bank (PDB). Our analysis indicates that nifedipine is an effective choice for treating hypertension and reducing the symptoms of angina (chest pain).

Keywords

AutoDock Tool, Angiotensin Converting Enzyme (ACE), Cardiovascular Disease, Hypertension, Nifedipine, Systolic

1. INTRODUCTION

Hypertension, defined as a systolic or diastolic blood pressure measurement that is at least 10 points greater than the upper limit for a specific individual’s age group, is a global pandemic that worsens with age. One-fourth of the world’s adult population has hypertension now, but that percentage is expected to grow to one-third by 2025 [1]. However, isolated systolic hypertension becomes more common as people become older. Systolic hypertension alone, as contrasted to systolic hypertension + diastolic hypertension, increases from twenty (20) percent in those under forty to eighty (40-80) percent in those sixty to sixty-nine (60-69) and ninety five (95) percent in those older than eighty [2]. Since systolic blood pressure is an excellent predictor of coronary and cerebrovascular risk, particularly in the elderly, it is receiving more attention than ever before. Particularly in elderly individuals with a high-risk profile, treating systolic hypertension, characterized by a broad pulse pressure, is successful in terms of blood pressure management and decreased morbidity [3].

Around 16.5% of all fatalities globally are attributable to hypertension; it causes more deaths and disabilities than any other kind of cardiovascular disorder. It is predicted that by 2030, 23.5 million people would die every year. Hypertension contributes significantly to the development of peripheral artery disease, heart failure, coronary artery disease, and atherosclerosis, stroke also damages the kidneys and may cause dementia or blindness [4]. Primary (essential) hypertension is the more common kind of raised blood pressure, whereas secondary hypertension occurs for other reasons [5]. Secondary hypertension, although affects 5-10% of hypertensive persons, has a better chance of being treated since its causes are known and treatable, like diabetes and renal impairment. However, essential hypertension develops due to a combination of variables. Essential hypertension is more challenging to treat than other types of hypertension because its etiology could be harder to determine or establish. It’s interesting to note that essential hypertension accounts for the vast majority of individuals (90%-95% vs. 5-7%) [6].
When an angiotensin-converting enzyme (ACE) inhibitor is administered, both hypertensive and normotensive patients have considerable reductions in mean arterial blood pressure, systolic blood pressure, diastolic blood pressure, and pulse rate. Several randomized controlled studies have examined the effectiveness of angiotensin-converting enzyme inhibitors as hypertension treatments. One of four pharmacological classes suggested for the first treatment for people with increased blood pressure, ACE inhibitors were included in the 2014 evidence-based recommendations released by the Eighth Joint National Commission (JNC8) [7]. Calcium channel blockers (CCBs), thiazide diuretics, and angiotensin receptor blockers are often used for initiation therapy in the non-black population. Only thiazides and calcium channel blockers should be utilized to treat high blood pressure initially in the black population [8]. The “American Heart Association”/“American College of Cardiology” (AHA/ACC) and the “European Society of Cardiology (ESC)” have both released guidance recommending “angiotensin-converting enzyme (ACE)” inhibitors as first-line antihypertensive therapy, especially for patients with diabetes mellitus (DM) and cardiovascular diseases. One of the most effective hypertension drugs, angiotensin-converting enzyme (ACE) inhibitors, has been demonstrated to have a smaller effect on hypertensive Black persons than on White people [9].

When the membrane potential of smooth muscle cells drops, calcium enters the cells via voltage-gated channels. Nifedipine lowers calcium ion entry in vascular smooth muscle and cardiac cells by blocking voltage-dependent L-type calcium channels. Systemic blood pressure drops and oxygen supply to the heart muscle increases as a result of decreased intracellular calcium levels. This means that nifedipine may lower blood pressure and reduce angina. The dihydropyridine subclass of calcium channel blockers includes nifedipine. It is mainly used as an antianginal and antihypertensive drug. NIFEDIPINE, a calcium channel blocker, and its analogs are employed here. In cases when the desired first-line medications are either ineffective or contraindicated, calcium-channel blockers are often used as a second line of defense. They work well for controlling blood pressure in those who also have angina or diabetes. Dihydropyridines and non-dihydropyridines are the two main chemical families of calcium-channel blockers, and they each have their unique pharmacokinetic features and therapeutic indications [10].

2. LITERATURE REVIEW

The research by Syed Awais Attique et al. found that high blood pressure (hypertension) is mostly caused by a combination of hereditary, lifestyle, and environmental variables. In this work, putative inhibitors of ACE were identified and evaluated from botanicals, other natural sources, and synthetic sources using a molecular docking-based method. In addition, Lipinski’s rule was used to foretell whether or not these inhibitors would be useful as medications in biological systems by taking into account their adsorption, distribution, metabolism, and excretion (ADME). In conclusion, our research offers a fresh and more precise understanding of the interactive features of established potential ACE inhibitors [11].

For this objective, Dr. Cüneyt Turkos et al. isolated glutathione S-transferase (GST) using the inhibitory effects of many calcium channel blockers (CCBs) were investigated using GSH-agarose affinity chromatography. These CCBs included amlodipine, cinnarizine, isradipine, nifedipine, and nilvadipine. Micromolar levels of inhibitory action against GST were shown by the CCBs (KIs in the range of 98.84 ±0.53, M-502.7±02.5 M). Isradipine was the most effective inhibitor of GST. Moreover, Docking studies were carried out to further define the interactions between nilvadipine and the active site of GST, confirming the inhibitory effect of this competitive inhibitor [12].

The researchers Ruidan Wang et al. stated in their research isolated and identified sesame protein into “angiotensin I-converting enzyme (ACE)” inhibitory peptides using in vitro stomach digestion modeling and then investigated the underlying processes through docking studies”. Digestion time increases breakdown and peptide production. Digestion increased ACE inhibitory action. SPDS-VII (3 kDa) demonstrated the highest ACE inhibition after ultrafiltration using various “molecular weight cut-off (MWCO)” membranes. With the use of the NGC Quest™10 plus Chromatography System, SPDS-VII was purified. From peak 4, 11 peptides were found utilizing Nano UHPLC-ESI-MS/MS (nano ultra-high performance liquid chromatography-electrospray ionization mass spectrometry/ mass spectrometry). According to molecular docking, GHIITVAR inhibits ACE by forging strong hydrogen bonds with its binding sites. These results demonstrate...
the potential of GHIITVAR for the development of new functional foods by identifying sesame protein as a rich source of ACE inhibitory peptides [13].

3. METHODOLOGY

3.1. Design:
We found the most probable and efficient ligand structure for controlling blood pressure with complementary and alternative therapies (CAM) by combining data mining with a thorough review of the existing research. In this study, a computational approach and docking mechanism are used to examine the targeting capability of certain ligands and target proteins. The human angiotensin-converting enzyme was shown to be a protein of interest in the therapy of hypertension, and also the chemical benazepril was found to have this effect using data mining and comprehensive analysis of the reviewed studies. Autodock4 was then used to prepare the protein and chemicals for docking, and BIOVIA Drug Discovery Studio was used to view the resulting complex shown in Figure 1.

3.2. Instrumentation
The Angiotensin Converting Enzyme (ACE) protein's three-dimensional structure is available for download in PDB format from the RCSB. By making available the PDB data on the three-dimensional proteins, nucleic acids, and other macromolecules' 3D structures and the interactions between smaller molecules (cofactors, medicines), RCSB aids in scientific study and teaching across the world. Tools for exploring, viewing, and interacting with the PDB databank may be found on the RCSB website; they include, for example, an easy way to look for the chemical interactions that stabilize macromolecules and play a crucial part in the interactions and functionalities of proteins and chemicals. All users, including structural biologists, computational biologists, and many others, have direct access to the PDB data conserved by the "Research Collaboratory for Structural Bioinformatics Protein Data Bank (RCSB PDB)" in the United States. Proteins and nucleic acids, two of life's most fundamental building components, have three-dimensional forms that are recorded and maintained by this system. "X-ray crystallography", "cryo-electron microscopy", and "nuclear magnetic resonance" (NMR) spectroscopy are frequently used techniques, and data is contributed by biotechnologists and scientists from all over the globe.

A chemical’s biological test activity may be looked up in PubChem, an online database. The "National Center for Biotechnology Information (NCBI)" is the organization most people think of when they hear “National Library of Medicine.” Using a web interface, PubChem may be accessed for free. Millions of compounds’ chemical structures and corresponding descriptions may be downloaded for free through FTP. One of the numerous categories of chemicals covered by PubChem is those with less than 100 atoms and fewer than 1,000 bonds. More than 80 separate database sources have contributed to PubChem’s fast expansion.

For the docking studies, we used Autodock4, one of the most popular programs in the research literature. The program simulates the binding of a drug or ligand to a protein or receptors in a biological system that used a docked log file. To assess the significance of this data, we use a graphical application called BIOVIA drug discovery studio.

3.3. Sample
“Angiotensin-converting enzyme (ACE)” inhibitors are commonly used to treat hypertension. An-
giotensin II synthesis may be inhibited by angiotensin-converting enzyme (ACE) inhibitors, a peptide that constricts blood arteries. Hypertension is a major contributor to heart failure (HF), cardiovascular disorders, stroke, and some other CVD, and ACE inhibitors are used to manage and control it. In the vast majority of instances, the cause cannot be pinpointed. The essential knowledge for members of an interprofessional team guiding the treatment of hypertensive patients and their associated illnesses and sequelae are examined, including clear signs, contraindications, activities, adverse events, and more. Thus, utilizing crystals produced under various circumstances, we report the 1.85-resolution crystal structure of ACE (in a novel crystal form). With the potential to create a new generation of superior ACE inhibitors, this new structure could be more suited for researching the binding of novel chemicals as shown in Figure 2.

Figure 2: Displays the displays of homology model of the crystalline human angiotensin-converting enzyme based on the PDB.

Nifedipine is a dihydropyridine-type calcium channel blocker. It is most often used for the treatment of hypertension and angina. Chronic stable angina and hypertension are two of the FDA-approved conditions. Other, unapproved uses exist for it as well. This exercise will help healthcare practitioner’s effectively lead patient treatment when nifedipine is indicated by outlining its uses, effects, dosages, risks, precautions, and adverse events of a drug, and the chemical structure of Nifedipine as shown in Figure 3.

Nifedipine lowers blood pressure and boosts the heart’s ability to get oxygen by inhibiting L-type voltage-gated calcium channels. Due to the short half-life of immediate-release nifedipine, it must be taken three times daily. The typical daily dosage of nifedipine is 10–120 mg. Patients need to be warned about the potential for life-threatening low blood pressure, chest pain, and heart attacks.

3.4. Data Collection

Docking generates several confirmations and locations for effective protein-ligand interaction. To begin the docking process, the (. gpf) file was used to initiate the auto grid, and the (. dpf) file had been used to initiate autodock4. A rigid protein or macromolecule as well as a flexible ligand were used to complete the docking. Autodock was used to get the binding energy for each docked confirmation in the DLG file. Figure 4 shows how the confirmations with the biggest negative binding energy were used to display the interaction, and this was proceeded by the generation of a complex.pdbqt file for additional analysis and visualization.

3.5. Data Analysis:

Using BIOVIA Drug Discovery Studio, we determined the binding energy of each resulting complex and focused on the one with the lowest value. In Figure 5 below, we can see the distinct features of the interactions that occur between the protein’s building blocks and the ligand molecules. Following the guidelines in Table 1, the 2-dimensional structure was constructed
to better understand the many types of bonds necessary to form a stable complex.

4. RESULTS AND DISCUSSION

Although ACE inhibitors are effective, it is not yet clear how they work. Although their action is not proportional to blood levels of renin, they indeed affect the renin-angiotensin-aldosterone system. A common class of hypertension drugs called ACE inhibitors works by blocking an angiotensin-converting enzyme, therefore preventing the production of angiotensin II. Natriuresis, blood pressure, and the reorganization of smooth muscle and cardiac myocytes are all positively impacted by a reduction in angiotensin II production. Having lower venous and arterial pressure reduces preload and volume overload. Bradykinin is a peptide that promotes vasodilation, and ACE inhibitors are thought to prevent its breakdown [14]. An angiotensin-converting enzyme regulates the balance between bradykinin’s vasodilatory and natriuretic actions and Angiotensin II’s salt-retentive and vascular-sclerosing properties. ACE inhibitors alter this balance by reducing Angiotensin II and decreasing the breakdown of Bradykinin. It is unknown whether ACE inhibitors’ effects on the production and degradation of many other vasoactive molecules, such as substance P, contribute to the drug’s favorable or detrimental outcomes [15].

An overdose is treated depending on several factors, including the quantity ingested, the time since ingestion, the patient’s age, and any existing medical conditions. Securing the airway, breathing, and circulation, as well as doing the necessary blood work and testing for contestants, are all part of the first examination. A quick call to poison control or toxicology should be the priority. Because high-dose insulin has been found to reduce mortality and improve hemodynamics, it is a viable treatment choice. Monitoring of electrocardiogram readings, vital signs, renal function, urine output, and electrolytes is essential. Consultation with a psychiatrist is also required in cases of purposeful consumption. Overdosing on immediate-release medications requires close monitoring for 4-7 hours after the patient presents. There is no better time than 24 hours of telemetry monitoring for extended-release formulations. Currently, there is no known effective treatment for this [16].

The sum of the energies of polar, non-polar, and unbonded interactions may be used to calculate the binding free energy. The binding energy is calculated by

![Figure 4: Displays the Protein-Ligand Interactions in the Docked State.](image)

![Figure 5: Displays the Molecular Interaction Between Amino Acids in a Protein and a Ligand.](image)

<table>
<thead>
<tr>
<th>Amino Acids</th>
<th>Bond Length</th>
<th>Types of bond</th>
</tr>
</thead>
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<tr>
<td>UNL1-ARG238</td>
<td>2.00413</td>
<td>Hydrogen Bond</td>
</tr>
<tr>
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<td>2.55948</td>
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<td>2.20082</td>
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</tr>
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adding the internal energy, intermolecular energy, and torsional free energy. This energy is subtracted from the total energy of an unrestrained system to arrive at the total energy. To get docking outcomes and a binding energy database, the DLG file format was employed. Decreases in binding energy result in a more stable protein-ligand complex. In Figure 6, we can see that the binding energy study between the ligand “Nifedipine” and the receptor/protein “Angiotensin Converting Enzyme” revealed that the complex produced was stable. This molecule has a binding energy of -10.87 kcal/mol.

![Figure 6: Different Clusters of Conformations’ Binding Energies are Shown.](image)

5. CONCLUSION
High blood pressure (hypertension) affects blood vessels to become rigid and not operate as well. Long-term, this damages essential body parts including the lungs, kidneys, heart, and brain raising the probability of experiencing a cardiac arrest or a cerebrovascular accident. Keeping your blood pressure under control is easier if you live a healthy lifestyle. You may get nifedipine in both fast-acting and slow-acting forms. It was first sold in a formulation that had a limited half-life and needed numerous daily doses. Rapid vasodilation and subsequent reflex sympathetic activation generated by these preparations led to undesirable symptoms such as headaches, palpitations, and flushing. Because of these issues, a new category of drugs known as extended-release formulations was produced. These drugs have been proven to have an anti-hypertensive effect that lasts for 24 hours with fewer side effects.

References
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