IJCRT.ORG ISSN : 2320-2882

INTERNATIONAL JOURNAL OF CREATIVE RESEARCH THOUGHTS (IJCRT)

An International Open Access, Peer-reviewed, Refereed Journal

IN-SILICO DOCKING STUDIES ON LEONURINE AND CANTHINE-6-ONE FOR NEPHROPROTECTIVE ACTIVITY

Mamatha. Atmakoor[1], V. Jyoshna[1], V. V. Rajesham[1], P. Roshan Ali[1,] T. Rama Rao[2]

[1]Department of Pharmacology, CMR College of Pharmacy, Kandlakoya (V), Medchal (M & D), Hyderabad–501401, Telangana, India.

[2]Professor and Principal, CMR College of Pharmacy, Kandlakoya (V), Medchal (M & D), Hyderabad–501401, Telangana, India.

ABSRACT

Kidney disease encompasses a diverse array of disorders impacting kidneys, often culminating in Chronic Kidney Disease (CKD), which progresses slowly and irreversibly. Cardiovascular events and fatality are more likely in people with chronic kidney disease (CKD). There is a complex association between Acute Kidney Injury (AKI) and Chronic Kidney Disease (CKD), which can range from modest functional abnormalities to severe renal failure. According to studies, certain alkaloids have been found to exhibit distinct pharmacological features, such as antioxidant, anti-inflammatory, and cytoprotective activities, which contribute to their nephroprotective effectiveness. Additionally, there are now greater possibilities for treatments against kidney illnesses owing to the potential of alkaloids, especially leonurine and canthine-6-one, as nephroprotective drugs.

***Key Words*:** Chronic Kidney Disease (CKD), cytoprotective, nephroprotective, leonurine and canthine-6-one

INTRODUCTION

Kidney disease is a heterogeneous group of disorders referring to a diverse range of conditions that affect the structure and operation of the kidneys, and a progressive increase in reduced function leads to Chronic Kidney Failure or complete kidney failure. Chronic Kidney Disease (CKD) characterized by its irreversibility and slow progressive evolution, is demonstrated by aberrant albumin excretion, evaluated by measured or estimated glomerular filtration rate (GFR), that lasts for longer than three months [1,2]. CKD being highly prevalent is associated with a higher risk of cardiovascular disease, severity, and death [2]. Identification of an individual's predisposing factors to CKD is essential in terms of personal and community health, as some

risk factors can be modified and can prevent or slow down the progression to End-stage renal disease (ESRD) [3]. In this context, any medical intervention that may prevent the progression of CKD towards end-stage renal disease (ESRD) is significant [4].

Acute Kidney Injury (AKI) encompasses the entire spectrum from small abnormalities in renal function to a requirement for renal replacement therapy (RRT). If not promptly detected or treated it might progress to CKD [5]. There is a complex relationship between AKI and CKD; where AKI can lead to CKD, while CKD raises the risk of AKI [6]. Although the pathogenesis of AKI is diverse and complex, ischemia [7], hypoxia [8], sepsis, and nephrotoxicity [9] are the most frequently acknowledged causes.

Various factors, such as exposure to nephrotoxic agents such as toxins, heavy metals (lead, mercury, arsenic, and cadmium), medications (aminoglycosides, antibiotics, chemotherapeutic agents, and NSAIDs); oxidative stress, inflammation, and ischemia-reperfusion injury can lead to acute renal failure, interstitial nephritis, and nephritic syndrome or contribute to the progression of chronic kidney disease (CKD). However, the majority of renal dysfunctions induced by nephrotoxic agents are reversible [10]. Diabetes and hypertension, the ultimate cause of chronic kidney disease, is now the major cause of End-stage renal failure worldwide [11]. Furthermore, once hypertension or diabetes are manifest, their renal complications can be mitigated by secondary prevention efforts aimed at blood pressure and glycaemic control. At the end-stage renal disease level, this is a severe condition that requires kidney transplantation or dialysis.

In the quest to combat kidney disorders, alkaloids such as Leonurine and Canthine-6-one have emerged as a promising class of compounds showing nephroprotective potential.

Leonurine (SCM-198), a chemically synthesized compound based on a bioactive alkaloid, is mainly used in Chinese traditional medicine. Leonurine has a major effect on strengthening the uterus [16] and diuresis and a positive effect on a wide range of biological activities, such as anti-inflammation, antioxidation, anti-apoptosis, excited uterine activity as well as, cardiovascular protective effects and neuroprotection [12]. Animal investigations suggest that Leonurine ameliorates the progression of kidney diseases such as AKI and CKD via the suppression of inflammatory factors and oxidative stress and protects against kidney damage [13,14]. Canthin-6-one is a subclass of β-carboline alkaloids found in a variety of medicinal plants and fungal sources has an anti-inflammatory effect on the intestines by downregulating immune molecular mediators and lowering oxidative stress [15]. The low toxicity and good biological activities of Canthin-6-one alkaloids mean that there is potential for them to be developed into new drugs.

MATERIALS AND METHODS

Docking Methodology

Molecular modeling studies were performed for the identified bioactive alkaloid compounds in order to assess the ability of bioactive molecules for nephroprotective activity. The ligand was docked to the active site of the target using the AutoDock program, version 4.2.

Preparation of Ligand

Ligand preparation is the first step in investigations involving molecular docking. The three-dimensional structures of alkaloid phytochemicals (Leonurine; CID-161464 and Canthine-6-one; CID-97176) were obtained from the NCBI –PubChem repository (“PubChem”) in “sdf” format required for the analysis.

Preparation of protein

X-ray crystal structure (resolution 2.30) of human soluble epoxide hydrolase associated with synthetic inhibitor (PDB ID: 1.98) and NF-B (PDB ID: 1NFK) p50 homodimer were retrieved from the Protein Data Bank (PDB) of the Research Collaboratory (RCSB). The co-crystallized DNA macromolecule of 1NFK and the synthetic inhibitor of 3ANS were removed from the structure and all the hydrogen atoms were added to the protein to stabilize them using the CHARMm force field. Kollmann charges were added to the molecules which is used to estimate the electrostatic properties between ligand and their target protein or receptor. These charges help in predicting the orientation of the ligands during their docking. The energy of the target was minimized using the traditional dynamics cascade technique in Discovery Studio 3.5. The residue of the binding site has been selected to carry out molecular docking experiments.

Receptor Grid Generation

On the specified receptor, the co-crystallized ligand was separated from the receptor chain's active site. The atoms were the same size as Van der Waals radii of 1.0> and had partial atomic charges less than 0.25 defaults. The active sites' depiction of an enclosing box is the centroid of the workspace ligand. This technique was followed, and the default Glide parameters were used to create a grid centered on the ligand. All the compounds were docked with the grid structure.

Molecular Docking Analysis

The high precision (XP) function of AUTODOCK was employed to perform flexible docking on the specified receptor grid. The described ligand-receptor interactions were not constrained in any way. The structure output format was converted to a pose viewer file so that the results of subsequent docking processes may be observed via the pose viewer.

Generation of E-Pharmacophore:

Phytochemicals leonurine and Canthin-6-one docked with 3ANS macromolecule protein, an e-pharmacophore was created. PHASE from Schrodinger, LLC in New York was utilized to automatically generate pharmacophoric areas with enhanced ligands. PHASE renders utilization of characteristics such as the positive ionizable group (P), aromatic ring (R), hydrogen bond acceptor (A), hydrogen bond donor (D), negative ionizable group (N), and hydrophobe (H). Each pharmacophoric site was assigned an energy value based on the Glide XP descriptions.

Selection of the Best-Scored Pose:

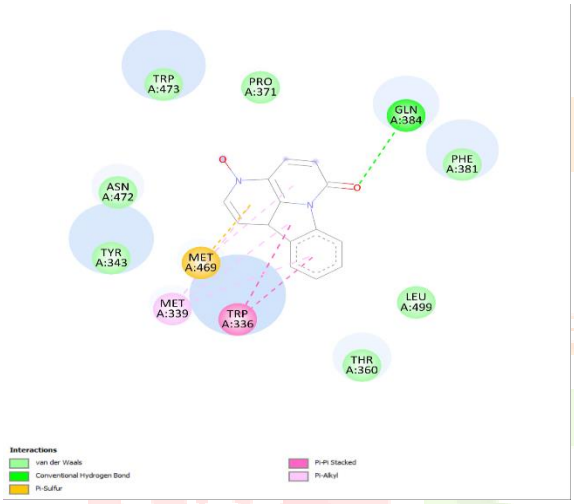
The docking scores were the most crucial factor to take into consideration when choosing the optimal docking poses for the PCNPs, but there were other elements to take into account as well, such as the values of various energies, the number of H bonds, and a visual analysis of all docking positions in Maestro (Schrodinger, USA). The energy of the interaction between the protein and the ligand is responsible for binding affinities. Many criteria were devised in order to find the best-docked structure for each ligand. Then the rank was determined directly using the Glide GScore.

RESULTS AND DISCUSSION

This study evaluated the docking simulation of Leonurine and Canthine-6-one with the protein 3 ANS [human soluble epoxide hydrolase (PDB ID:3ANS)]. The standard drug taken was Vitamin E (alpha-tocopherol). These alkaloid compounds showed promising results when compared to the standard.

Table: Interaction of alkaloid phytochemicals with target protein

S.NO	Phytochemical	Binding Affinity (K.cal/mol)
1	**Vit E (Standard)**	-9.2
2	Leonurine	-7.5
3	Canthin-6-one	-7.8



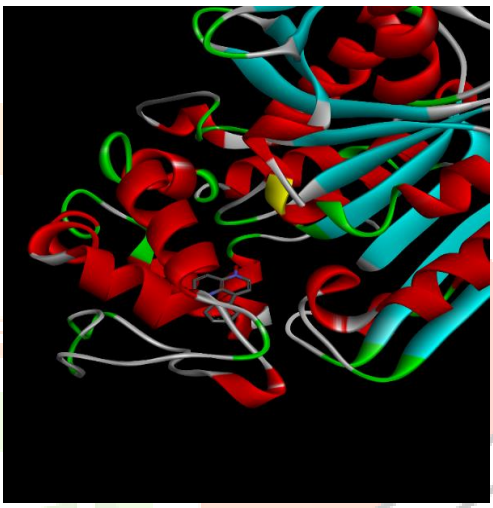
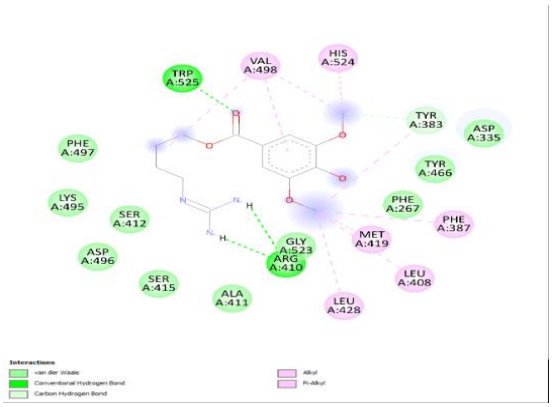


Figure 1: (A) **Figure 2: (B)**

(A) 3D interaction of phytocompound Canthin-6-one with the 3 ANS [human soluble epoxide hydrolase] protein.

(B) 2D interaction of phytocompound Canthin-6-one with the 3 ANS [human soluble epoxide hydrolase] protein



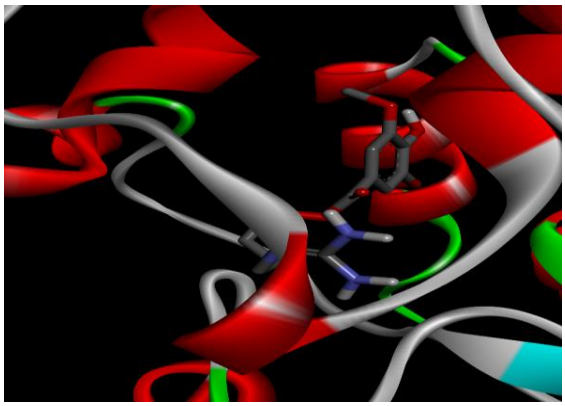


Figure 3: (A) **Figure 4: (B)**

(A) 3D interaction of phytocompound Leonurine with the 3 ANS [human soluble epoxide hydrolase] protein.

(B) 2D interaction of phytocompound Leonurine with the 3 ANS [human soluble epoxide hydrolase] protein.

DISCUSSION

NF-B is a crucial signal transduction mediator that is activated by a variety of pro-inflammatory cytokines, therefore participating in the effector phase of inflammation. Many pathophysiological events in renal cells also activate NF-B, which is responsible for inflammation and is associated with renal diseases. Chronic inflammatory disease and oxidative stress, both of which have been linked with NF-B activation, play a significant role in the development and progression of chronic kidney disease. Cisplatin nephrotoxicity has been associated with NF-B as well [17].

There have been beneficial benefits in experimental renal damage. Agents that inhibit or antagonize NF-B activating stimuli have been identified. Many herbal remedies have been shown during research to protect against CP-induced renal damage by inhibiting NF-B activation [19]. As a result, it is clear that treating inflammation (NF-B) is an intriguing therapeutic approach in the treatment of many renal illnesses. initially, it was believed that soluble epoxide hydrolase (sEH) was only involved in the metabolism of xenobiotic metabolism. Fatty acid epoxides are now well established as suitable substrates for this enzyme as a component of the arachidonic acid cascade, sEH plays a key role in eicosanoid epoxide metabolism. sEH is produced in a variety of cells and organs, including the liver, kidney, vascular endothelium, leukocytes, and adipocytes, which catalyzes the hydrolysis of epoxyeicosatrienoic acids (EETs) into the corresponding dihydroxyeicosatrienoic acids.

Several studies have revealed that EETs have nephroprotective properties due to their antioxidative, anti-inflammatory, and anti-apoptotic properties [18]. sEH inhibitors are a novel therapy option for nephrotoxicity. The aforementioned findings compelled us to investigate the molecular docking and dynamic behavior of Leonurine and Canthin-6-one bioactive compounds isolated from the title against NF-B and sEH.

CONCLUSION

Alkaloids are vital chemical substances that may be exploited to find new drugs. This present study concludes that the phytocompounds selected suggest the potential ability to exert nephroprotective activity. Leonurine and Canthin-6-one phytocompounds when docked with 3ANS protein and with Vit E as a standard drug. The binding affinity of the compound Leonurine and Canthin-6-one with 3ANS protein are observed to be -7.5 and -7.8 respectively. These results are the report that related alkaloids have played a nephroprotective role by regulating multiple mechanisms. Thus, our study indicated that these alkaloids could offer an efficient and novel strategy to explore novel drugs for nephroprotective activity.

REFERENCES

1. Ammirati AL. Chronic Kidney Disease. Rev Assoc Med Bras [Internet]. 2020; 66:s03–9. Available from: https://doi.org/10.1590/1806-9282.66.S1.3
2. Kazancioğlu R. Risk factors for chronic kidney disease: an update. Kidney international supplements. 2013 Dec 1; 3(4):368-71.
3. Atkins RC. The epidemiology of chronic kidney disease. Kidney international. 2005 Apr 1; 67:S14-8.
4. Sujana D, Saptarini NM, Sumiwi SA, Levita J. Nephroprotective activity of medicinal plants: A review on in silico-, in vitro-, and in vivo-based studies. Journal of Applied Pharmaceutical Science. 2021 Oct 3; 11(10):113-27.
5. Levey AS, Levin A, Kellum JA. Definition and classification of kidney diseases. American Journal of Kidney Diseases. 2013 May 1; 61(5):686-8.
6. Rifkin DE, Coca SG, Kalantar-Zadeh K. Does AKI truly lead to CKD?. Journal of the American Society of Nephrology: JASN. 2012 Jun; 23(6):979.
7. Sharfuddin, A., Molitoris, B. Pathophysiology of ischemic acute kidney injury. *Nat Rev Nephrol* **7**, 189–200 (2011).
8. Ow CP, Ngo JP, Ullah MM, Hilliard LM, Evans RG. Renal hypoxia in kidney disease: cause or consequence?. Acta Physiologica. 2018 Apr; 222(4):e12999.
9. Kwiatkowska E, Domański L, Dziedziejko V, Kajdy A, Stefańska K, Kwiatkowski S. The mechanism of drug nephrotoxicity and the methods for preventing kidney damage. International Journal of Molecular Sciences. 2021 Jun 6;22(11):6109.
10. Sebastian M. Renal toxicity. InHandbook of Toxicology of Chemical Warfare Agents 2009 Jan 1 (pp. 561-574). Academic Press.
11. Locatelli F, Vecchio LD, Pozzoni P. The importance of early detection of chronic kidney disease. Nephrology Dialysis Transplantation. 2002 Oct 2;17.
12. Huang L, Xu DQ, Chen YY, Yue SJ, Tang YP. Leonurine, a potential drug for the treatment of cardiovascular system and central nervous system diseases. Brain and behavior. 2021 Feb; 11(2):e01995.
13. Li J, Zhang S, Liu X, Han D, Xu J, Ma Y. Neuroprotective effects of leonurine against oxygen–glucose deprivation by targeting Cx36/CaMKII in PC12 cells. PLoS One. 2018 Jul 17; 13(7):e0200705.
14. Deng Z, Li J, Tang X, Li D, Wang Y, Wu S, Fan K, Ma Y. Leonurine reduces oxidative stress and provides neuroprotection against ischemic injury via modulating oxidative and NO/NOS pathway. International Journal of Molecular Sciences. 2022 Sep 5; 23(17):10188.
15. Al-Snafi, A.E. The pharmacological importance of Antirrhinum majus—A review. Asian J. Pharm. Sci. Technol. 2015, 5, 313–320.

16. LI X, CHEN FH, YUAN FL, LU WG, WU FR, ZHANG YN, LI XX. Effects of leonurine on uterus induced by drug-abortion in rats. Chinese Journal of Clinical Pharmacology and Therapeutics. 2009; 14(5):481.
17. Kpemissi M, Eklu-Gadegbeku K, Veerapur VP, Negru M, Taulescu M, Chandramohan V, Hiriyan J, Banakar SM, Thimmaiah NV, Suhas DS, Puneeth TA. Nephroprotective activity of Combretum micranthum G. Don in cisplatin induced nephrotoxicity in rats: In-vitro, in-vivo and in-silico experiments. Biomedicine & Pharmacotherapy. 2019 Aug 1; 116:108961.
18. Liu Y, Lu X, Nguyen S, Olson JL, Webb HK, Kroetz DL. Epoxyeicosatrienoic acids prevent cisplatin-induced renal apoptosis through a p38 mitogen-activated protein kinase-regulated mitochondrial pathway. Mol Pharmacol. 2013 Dec; 84(6):925-34. doi: 10.1124/mol.113.088302. Epub 2013 Oct 3. PMID: 24092818; PMCID: PMC3834146.
19. Gao H, Wang X, Qu X, Zhai J, Tao L, Zhang Y, Song Y, Zhang W. Omeprazole attenuates cisplatin-induced kidney injury through suppression of the TLR4/NF-κB/NLRP3 signaling pathway. Toxicology. 2020 Jul 1; 440:152487.