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REVIEW ARTICLE

In-silico screening: Piperine and Mangiferin bioactive compound as potential anticancer bioactive molecule

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ABSTRACT
Lung cancer has been identified as the leading cause of cancer deaths worldwide. The mortality rate from lung cancer has been estimated to be 18.4%. Until now, conventional treatments have not yielded optimal results, thus necessitating an investigation into the use of traditional herbal plants as potential candidates for its treatment. This study aimed to determine the inhibitory and apoptotic activity of the Piperine and Mangiferin bioactive molecule by in silico molecular docking studies. In this research work we was select the piperine bioactive compound and evaluated In-silico for their ability anti-cancerdrug. Mangiferin and Piperine bioactive compound identified and were found to obey all the criteria of Lipinski's rule of five for drug-likeness with two Lipinski violations. In addition, Mangiferin and Piperine bioactive compound interacted with Human Epidermal Growth Factor Receptor protein (PDB: 1MOX) validated as anti-lung cancer drug at very degrees. Moreover, analysis of the binding interaction suggests that structural bulkiness and present of polar groups are essential for favorable binding contact across the entire considered anti-lung cancer protein target. Given the physicochemical features of the Mangiferin and Piperine bioactive compound and their interesting binding interactions with all the anticancer protein targets, further attention on them is advised to exploit their potentials as revealed in this study for the advancement of anti-lung cancer drug development and the communal knowledge of anti-psoriatic science as a whole.
Keywords: *In-silico screening, Piperine, Mangiferin, bioactive compound, Human Epidermal Growth Factor Receptor protein.*

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INTRODUCTION

According to projections, there will be 15.7 lakh cancer cases in the nation by 2025, up from 14.6 lakh in 2022. Despite the fact that there are many different types of cancer, men are more prone than women to develop cancer of the breast, lungs, and thyroid, as well as cancers of the prostate, stomach, and liver (1). On the continent, cancer is becoming more common and is killing people at an alarming rate. Finding the precise causes of this growth is difficult. However, the ageing population, demographic growth, and changes in the epidemiology are all important cancer risk factors (2).

Another issue is lung cancer, which is the most common type of cancer worldwide in terms of incidence and the leading cause of death for male patients, despite being the second most common disease in terms of mortality for female patients (3). The forecasts show that lung cancer kills 2.5 times as many people as colorectal cancer and more than breast, prostate, and pancreatic cancer combined (4). The 130,180 lung cancer deaths anticipated in 2022 are predicted to be 81% related to tobacco use, with an estimated 3,650 deaths coming from secondhand smoking (5-7). The creation of novel medications for the early and metastatic stages of lung cancer treatment has not advanced significantly during the past 40 years. Even if there are many therapeutic alternatives available to treat lung cancer when it is still in its early stages, their reduced efficiency harms healthy cells and lowers patients' chances of survival. Surgery, radiation therapy, and chemotherapy are currently the most common therapies for lung cancer patients, albeit they

have only partially been successful in curing the disease. In order to decrease cancer-related fatalities, effective and promising treatments for lung and breast cancer are urgently needed. Cancer patients still struggle because standard treatments are ineffective and have side effects, despite the fact that cancer treatment has advanced significantly over the past two millennia (8-9).

The amide alkaloid piperine has pleiotropic qualities, including antioxidant, anticancer, anti-inflammatory, antihypertensive, hepatoprotective, and neuroprotective effects. It also increases bioavailability and has effects on fertility. Drug-metabolizing enzymes, gastrointestinal problems, and the bioavailability of a number of medications can all be affected by piperine. Piperine may be a promising drug candidate for the treatment of lung cancer, according to the available research. Therefore, we chose the piperine molecule for the in silico approach of piperine screening as a potential anticancer bioactive molecule.

Mangiferin is a bioactive mango component that protects against illnesses caused by way of life. By preventing the expression of tumour necrosis factor, it defends against a variety of human malignancies, including lung, colon, breast, and brain tumours.

A receptor for extracellular protein ligands that are a part of the epidermal growth factor family (EGF family), the transmembrane protein known as the epidermal growth factor receptor (EGFR; ErbB-1; HER1 in humans) has this role [11]. The EGFR is a member of the ErbB family of receptors, a subfamily of four closely related receptor tyrosine kinases. Other names for the EGFR include ErbB-1, HER2/neu, ErbB-2, ErbB-3, and ErbB-4. Numerous different cancer types may be affected by mutations that change EGFR expression or function[12].There is currently no information on the effectiveness of piperine and nagiferin, two active chemicals, in treating lung cancer. To test their efficacy against lung cancer using an *In-silico* technique, we chose the bioactive compounds piperine and manniferin (10). The study's validity was furthered by the use of in silico ADMET (Absorption, Distribution, Metabolism, Excretion, and Toxicity) assessments of certain natural chemicals to determine their pharmacokinetics and pharmacodynamic features to forecast potent medications for the treatment of lung cancer.

MATERIALS AND METHODS

1. Piperine and Mangiferin sdf structures were downloaded from the PubChem and converted to pdb format for the docking analysis and Human Epidermal Growth Factor Receptor (1MOX) was downloaded from the PDB website.
2. Chain A was selected for the prediction of active site in the target protein (1MOX). Active site prediction server (http://www.scfbio-iitd.res.in/dock/ActiveSite.jsp)
3. Docking was performed using autodock4. Docking results were analysed using the AutoDockTools v1.5.7, PLIP server and Chimera packages.

Proteins

Human Epidermal Growth Factor Receptor (1MOX) proteins linked to lung cancer were the main focus of the recent studies. Using the URL http://www.rcsb.org/PDB, the 3D structures for these proteins were retrieved from the Protein Data Bank (PDB). Human Epidermal Growth Factor Receptor (1MOX), the target protein's name, was retrieved from the PDB database. Human Epidermal Growth Factor Receptor (1MOX) proteins' PDB structures were created by eliminating all water molecules and then including hydrogen atoms and Kollman charges. The protein files were saved in PDBQT format for additional research, and then they were optimised for docking with AutoDock Tools. 4.

Ligands

These proteins were chosen as targets for piperine and retrieved from PubChem (https://pubchem.ncbi.nlm.nih.gov). Chem sketch programme was used to draw the structures of all compounds using SMILES and file them in MOL format. Using the Open Babel programme, all chemicals were translated from MOL format to PDB format and stored. The ligand files were stored in PDBQT format for additional research, and Auto Dock Tools 4 was then used to optimise them for docking. For the prediction of the target protein's active site (1MOX), Chain A was used. The top binding site was located at the coordinates X=7.943 Y=49.874 Z=34.936 by the active site prediction server (http://www.scfbio-iitd.res.in/dock/ActiveSite.jsp).

Molecular Docking of Protein and Ligand

The "key and lock" theory, which is utilised to determine the orientation of ligand and protein that fits them best, is known as molecular docking. Target proteins were docked with a few natural chemicals using AutoDock Tools 4, and binding energy was determined. AutoDock Tool received the produced files for the protein and ligands. Different conformations for the ligand were created during the docking process, but the optimal conformations with the least amount of energy were chosen as the output.

Autodock4 was used for docking. Using the AutoDockTools v1.5.7, PLIP server, and Chimaera packages, docking results were examined.

Docking simulation

From the protein data bank, the crystal structures of the anti-lung cancer protein targets under consideration and their co-crystallized inhibitors were downloaded. For use in docking calculations, the enzyme-inhibitor complexes were produced in accordance with standards. The co-crystallized ligands and protein targets were then separated and stored separately. Using the Auto Dock Tools 4, piperine, an amide alkaloid, was docked towards its binding site on the Human Epidermal Growth Factor Receptor protein (PDB: 1MOX) targets. The docking procedure included the conformational analysis of the ligands, placement, and scoring steps. By superimposing ligand atom triplets and triplet locations in the receptor binding region, several poses were produced. The alpha sphere centers at the receptor site points, which stand for areas of dense packing.

The Auto Dock Tools 4 was used to score the poses created during the placement step utilising the scoring stage. The scoring function in Auto Dock Tools 4 determines the ligand's free energy of binding from the given rotational and translational entropy terms, energy lost due to the ligand's flexibility, hydrogen bonds, metal contacts, and a desolvation term resulting from the volumes of the protein and ligand's atoms in contact with the solvent. The goal of docking validation was to find the docking parameters with the lowest root mean square deviation (RMSD) from the experimental binding mode (X-ray crystal structure), i.e., those that best mimic the ligand conformation (docking poses) within the binding pocket. The native ligands found in each protein complex's binding pocket were docked towards their corresponding reception sites during the docking validation method utilising various grid parameters. The data set was then docked towards the binding sites of the 15 anti-lung cancer medication targets using the parameters retained with the lowest RMSD values.

ADME and Toxicity analysis

For the estimation of physicochemical parameters like logP and logS, a trustworthy ADMET prediction model may be helpful. The ADME analysis of the chosen natural substances was carried out with the help of Swiss software. To investigate the various pharmacokinetic and pharmacodynamic aspects of the compounds, such as blood brain barrier permeability, carcinogenicity, subcellular localization, LD50, and category of acute oral toxicity, the molecular structures of the compounds were uploaded to the ADMET-SWISS server (http://www.swissadme.ch).

RESULTS AND DISCUSSION

The piperin and mangiferin SDF structures were purchased from PubChem and converted to pdb format for the docking experiment. The 3D structure of the target protein Human Epidermal Growth Factor Receptor (1MOX) was downloaded from the PDB database. For the prediction of the target protein's active site (1MOX), Chain A was used. The top binding site was determined using the active site prediction server (http://www.scfbio-iitd.res.in/dock/ActiveSite.jsp), and it is located at the coordinates X=7.943, Y=49.874, and Z=34.936. Autodock4 was used for docking. Using the AutoDockTools v1.5.7, PLIP server, and Chimaera packages, docking results were examined.

Piperine interaction with 1MOX residues

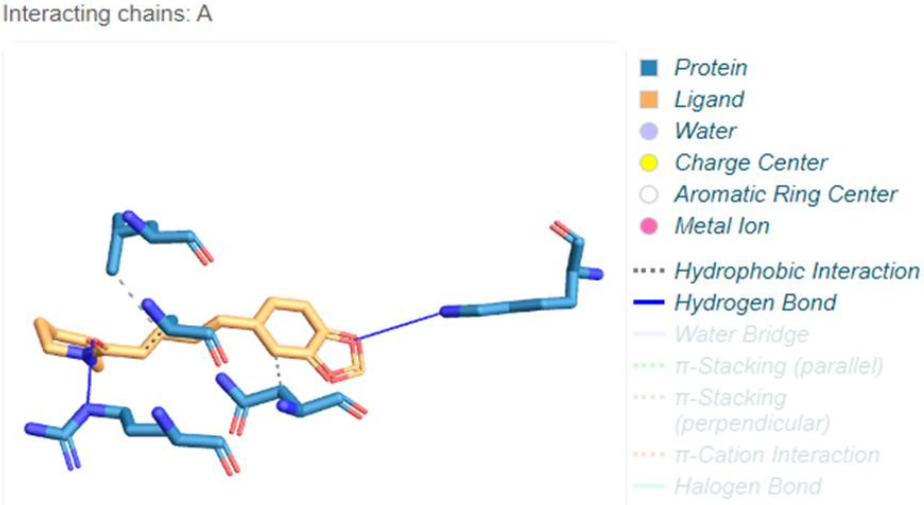

Figure 1: Piperine interaction with 1MOX residues visualised using PLIP server.

Identified interactions were (hydrogen bond and hydrophobic interaction)

Table 1: Piperine interaction with 1MOX residues

Identified interactions (hydrophobic and hydrogen bonds) and their properties are summarised.

Hydrophobic Interactions

Index	Residue	AA	Distance	Ligand Atom	Protein Atom
1	38A	LEU	3.25	4724	373
2	62A	ALA	3.15	4723	616
3	86A	ASN	3.16	4719	844

Hydrogen Bonds

Index	Residue	AA	Distance H-A	Distance D-A	Donor Angle	Protein donor?	Side chain	Donor Atom	Acceptor Atom
1	84A	ARG	2.42	3.24	137.00	✓	✓	825 [Ng+]	4725 [O2]
2	322A	LYS	3.04	3.86	138.18	✓	✓	3011 [N3+]	4715 [O3]

Mangiferin interaction with 1MOX

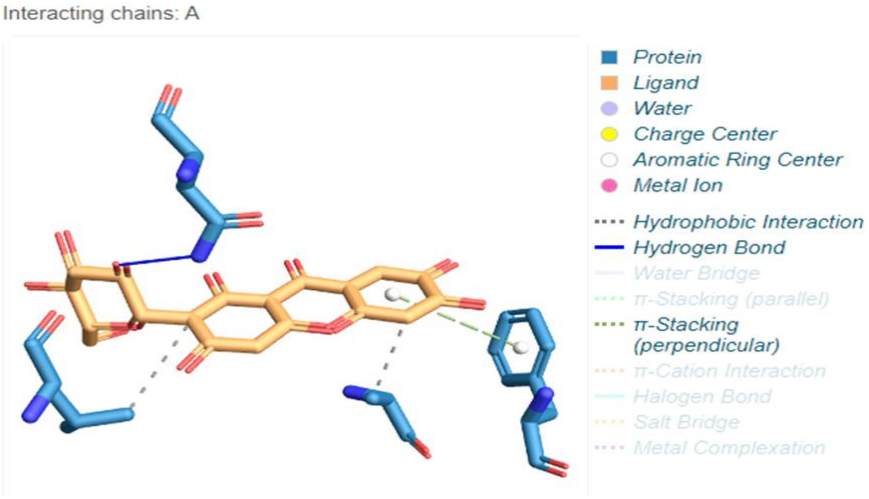

Figure 2: Mangiferin interaction with 1MOX residues visualized using PLIP server. Identified interactions were (hydrogen bond, hydrophobic interaction and **π stacking**)

Table 2: Mangiferin interaction with 1MOX residues

Identified interactions (hydrophobic, hydrogen bonds and **π stacking**) and their properties are summarised.

Hydrophobic Interactions

Index	Residue	AA	Distance	Ligand Atom	Protein Atom
1	38A	LEU	3.99	4714	373
2	265A	ALA	3.19	4724	2537

Hydrogen Bonds

Index	Residue	AA	Distance H-A	Distance D-A	Donor Angle	Protein donor?	Side chain	Donor Atom	Acceptor Atom
1	86A	ASN	1.86	2.81	153.96	✓	✓	847 [Nam]	4733 [O3]

π-Stacking

Index	Residue	AA	Distance	Angle	Offset	Stacking Type	Ligand Atoms
1	230A	PHE	5.12	71.55	1.87	T	4721, 4722, 4723, 4724, 4725, 4726

Docking of mangiferin and piperine to the Human Epidermal Growth Factor Receptor protein (PDB: 1MOX) reveled efficient interaction to the selected site. The lowest binding energy for the mangiferin was identified as -5.33 kcal/mol with inhibition constant of 124.08 uM while piperine showed binding energy -5.15 kcal/mol with inhibition constant of 167.58 uM. This suggests both molecules can be lead molecule for the treatment of lung cancer.

ADME and Toxicity analysis

The estimation of physicochemical parameters like logP and logS can benefit from the usage of a trustworthy ADMET prediction model. The results of Swiss software ADME analysis of mangiferin and piperine selected natural compounds. To investigate the various pharmacokinetic and pharmacodynamic aspects of the compounds, such as blood brain barrier permeability, carcinogenicity, subcellular localization, LD50, and category of acute oral toxicity, the molecular structures of the compounds were uploaded to the ADMET-SWISS server (http://www.swissadme.ch).In comparison to mangiferin bioactive polyphenol, the report suggested that the piperine alkaloid bioactive molecule had good properties. Mangiferin has a low water content and a limited bioavailability. Mangiferin's lower bioavailability feature can be eliminated by utilizing piperine. The piperine alkaloidal molecule has reportedly been utilised as a bioavailability-improving drug, according to numerous researchers. The outcomes are displayed in Table 3.

Table 3: ADMET-SWISS results data of Piperine and Mangiferin

Molecule	**Mangiferin**	**Piperine**
Formula	C19H18O11	C17H21NO3
MW	422.34	287.35
#Heavy atoms	30	21
#Aromatic heavy atoms	14	6
Fraction Csp3	0.32	0.35
Rotatable bonds	2	7
H-bond acceptors	11	3
H-bond donors	8	0
ESOL Solubility (mg/ml)	1.54E+00	6.50E-02
ESOL Solubility (mol/l)	3.64E-03	2.26E-04
ESOL Class	Soluble	Soluble
Silicos-IT class	Soluble	Soluble
GI absorption	Low	High
BBB permeant	No	Yes
Pgp substrate	No	No
CYP1A2 inhibitor	No	Yes
CYP2C19 inhibitor	No	Yes
CYP2C9 inhibitor	No	Yes
CYP2D6 inhibitor	No	No
CYP3A4 inhibitor	No	No
log Kp (cm/s)	-9.14	-5.49
Lipinski #violations	2	0
Ghose #violations	1	0
Veber #violations	1	0
Egan #violations	1	0
Muegge #violations	3	0
Bioavailability Score	0.17	0.55
PAINS #alerts	1	0
Brenk #alerts	2	2
Leadlikeness #violations	1	1
Synthetic Accessibility	4.76	3.09

CONCLUSION

Mangiferin and piperine, two bioactive compounds, were found to meet all five requirements of Lipinski's rule of five for druglikeness, with only two Lipinski infractions. Additionally, Mangiferin and Piperine bioactive compounds had very low-level interactions with Human Epidermal Growth Factor Receptor

protein (PDB: 1MOX), which was verified as an anti-lung cancer medication. Furthermore, a study of the binding interaction reveals that the presence of polar groups and structural bulkiness are necessary for a favourable binding contact across the full target protein for the anti-lung cancer protein under consideration. Further research on the Mangiferin and Piperine bioactive compounds is advised to take advantage of their potentials as revealed in this study for the advancement of anti-lung cancer drug development and the general knowledge of anti-psoriatic science, given their interesting binding interactions with all the anticancer protein targets and their unique physicochemical characteristics.

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CONFLICT OF INTEREST: The authors declared no conflict of interest.

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REFERENCES

1. Rahib L., Smith B. D., Aizenberg R., Rosenzweig A. B., Fleshman J. M., Matrisian L. M. (2014). Projecting cancer incidence and deaths to 2030: the unexpected burden of thyroid, liver, and pancreas cancers in the United States. *Cancer research* 74, 2913–2921. doi: 10.1158/0008-5472.CAN-14-0155
2. Sung H., Ferlay J., Siegel R. L., Laversanne M., Soerjomataram I., Jemal A., et al.. (2021). Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *Cancer Journal for Clinicians* 71, 209–249. doi: 10.3322/caac.21660
3. Jemal A., Siegel R., Xu J., Ward E. (2010). Cancer statistics, 2010. *Cancer Journal for Clinicians* 60, 277–300. doi: 10.3322/caac.21660.
4. Siegel R. L., Miller K. D., Fuchs H. E., Jemal A. (2022). *Cancer statistics*, 2022. 69, 7–34. doi: 10.3322/caac.21551.
5. Islami F., Goding Sauer A., Miller K. D., Siegel R. L., Fedewa S. A., Jacobs E. J., et al.. (2018). Proportion and number of cancer cases and deaths attributable to potentially modifiable risk factors in the United States. *Cancer Journal for Clinicians* 68, 31–54. doi: 10.3322/caac.21440
6. Rahman M. M., Islam M. R., Akash S., Harun-Or-Rashid M., Ray T. K., Rahaman M. S., et al.. (2022). Recent advancements of nanoparticles application in cancer and neurodegenerative disorders: At a glance. Biomedicine & Pharmacotherapy 153, 113305. doi: 10.1016/j.biopha.2022.113305.
7. Siegel R. L., Miller K. D., Fuchs H. E., Jemal A. (2022). Cancer statistics, 2022. 69, 7–34. doi: 10.3322/caac.21551.
8. Hirsch F. R., Scagliotti G. V., Mulshine J. L., Kwon R., Curran W. J., Jr., Wu Y.-L., et al.. (2017). Lung cancer: current therapies and new targeted treatments. The Lancet 389, 299–311. doi: 10.1016/S0140-6736(16)30958-8.
9. Huang C.-Y., Ju D.-T., Chang C.-F., Reddy P. M., Velmurugan B. K. (2017). A review on the effects of current chemotherapy drugs and natural agents in treating non–small cell lung cancer. Biomedicine 7, 4–7. doi: 10.1051/bmdcn/2017070423.
10. Wang Y., Wang M., Wu H. X., Xu R. H. (2021). Advancing to the era of cancer immunotherapy. *Cancer Communications* 41, 803–829. doi: 10.1002/cac2.12178.
11. Muhammad Imran, Muhammad Sajid Arshad, Masood Sadiq Butt, Joong-Ho Kwon, Muhammad Umair Arshad, Muhammad Tauseef Sultan.(2017). Mangiferin: a natural miracle bioactive compound against lifestyle related disorders.*Lipids Health Dis*. 2017; 16: 84.
12. Herbst RS (2004). "Review of epidermal growth factor receptor biology". *International Journal of Radiation Oncology, Biology, Physics*. 59 (2 Suppl): 21–6. doi:10.1016/j.ijrobp.2003.11.041. PMID 15142631.
13. Zhang H, Berezov A, Wang Q, Zhang G, Drebin J, Murali R, Greene MI (2007). "ErbB receptors: from oncogenes to targeted cancer treatment". *The Journal of Clinical Investigation*. 117 (8): 2051–8. doi:10.1172/JCI32278. PMC 1934579. PMID 17671639.