Molecular docking and Identification of *Anisomeles* derived inhibitors against Plasmodium falciparum glutathione s-transferase (PF GST) causing Malaria

[1]Mahendran Radha*, [2] Jeyabasker Suganya, [2] SharanyaManoharan [2]Amrutha.P.Murugan, [2]Dhananya.S

[1]Professor & Head, [2]Assistant Professor,
[1&2] Department of Bioinformatics, School of Life Sciences,
Vels Institute of Science, Technology and Advanced Studies (VISTAS), Chennai, India.

ABSTRACT:

Malaria is a life-threatening disease caused by Plasmodium parasite that are transmitted to people through the bites of infected female Anopheles mosquitoes. In 2017 there were estimated 219 million cases of malaria in 90 countries and WHO reported that malarial death reached 435000 during the same year. Thus the complexity of the malarial parasite made development of malarial vaccine a difficult task. Hence to combat the situation there is need for several research that can identify more efficient anti-malarial drugs. Medicinal plants has the ability to cure many types of infectious disease and hence the present researchers are pointing towards various plant kingdoms. *Anisomeles malabarica* is a traditional medicinal plant and has been used as a folk medicine for the treatment of various diseases. The pharmacological studies of *Anisomeles malabarica* were reported for the use of Antibacterial diseases. The 34 compounds from *Anisomeles malabarica* were retrieved through several journal surveys. The protein PfGST (plasmodium falciparum glutathione S-transferase) of PDB ID 1OKT is responsible for causing malaria in human. Molecular docking is performed using Argus Lab protocol for these 34 compounds against the protein PfGST. The best docking pose were identified and compared with the docking interactions of the PfGST with synthetic compounds S-Hexylglutathione and Protoporphyrin. Thus the docking results and comparative studies shows that the compounds of *Anisomeles malabarica* exhibit the best binding interactions than the synthetic compounds interactions.

KEYWORDS: Malaria, *Anisomeles malabarica*, Antibacterial, Synthetic compounds, Docking, Phytochemical.

INTRODUCTION:

Anisomeles malabarica (L) is perhaps the most useful traditional medicinal plant. It is a highly aromatic plant belonging to the family Lamiaceae (Labiatae). *Anisomeles malabarica* is a species of herbaceous plant native to tropical and subtropical regions. Mosquitoes act as a vector for most of the life threatening disease like malaria, yellow fever, dengue fever. Different aspects of *Anisomeles malabarica* medicinal values are briefly demonstrated such as anti-bacterial, anti-allergic, anaphylactic, anticancer, anti-carcinogenic, and anti-inflammatory. *Anisomeles malabarica* has the pharmacological activity which can cure the malaria. (Ramesh Ramaraj and Yuwalee Unpaprom, 2013)

Malaria is a life-threatening mosquito-borne blood disease. The Anopheles mosquito transmits it to humans. The parasite in mosquitoes that spread malaria belongs to the Plasmodium genus. Over 100 types of plasmodium parasite can infect a variety of species. Different types replicate at different rates, changing how quickly the symptoms escalate, and the severity of the disease. Five types of Plasmodium Parasite can infect human. They are *P.falciparum, P.vivax, P.ovale, P.malaria, P.knowlesi*. These single-celled organisms that cannot survive outside of their host.

The 34 compounds of the medicinal plant *Anisomeles malabarica* were identified through several literature survey by using PubMed. PubMed is a freely accessible online tool which is developed and maintained by National Center for Biotechnology Information and is part of the United States National Library of Medicine.

The protein PF GST (*plasmodium falciparum* glutathione s-transferase) is retrieved from the PubMed database. Glutathione S-transferase of the malarial parasite Plasmodium falciparum (PfGST) represents a novel class of GST isoenzymes. Since the architecture of the PfGST substrate binding site differs significantly from its human counterparts and there is only one isoenzyme present in the parasite, PfGST is considered a highly attractive target for antimalarial drug development. The structure of the protein is retrieved from PDB by using the PDB id 1OKT. The Protein Data Bank archive contains information about experimentally determined structure of proteins, nucleic acids and complex assemblies. Molecular Docking is performed for the 34 compounds with the protein PfGST. Molecular docking involves the interaction of two or more molecules to give the stable adduct depending upon binding properties of ligand and target and also it predicts the 3D structure of any complex.

MATERIALS AND METHODS:

FILTERING OF SMALL MOLECULES:

The small molecules for 34 compounds were identified through literature search for the medicinal plant *Anisomeles Malabarica* using PubMed. PubMed comprises more than 29 million citations for biomedical literature from MEDLINE, life science journals, and online books. It is developed and maintained by the National Center for Biotechnology Information (NCBI) at the National Library of Medicine (NLM). For further studies and computational analysis, the compounds should have at least 2D structure. Using PubChem database the structures are retrieved. PubChem Compound Database to be queried by chemical structure or chemical structure pattern. The PubChem Sketcher allows a query to be drawn manually. Users may also specify the structural query input by PubChem Compound Identifier (CID), SMILES,SMARTS, In ChI, Molecular Formula, or by upload of a format. PubChem is maintained by the National Center for Biotechnology Information (NCBI), a component of the National Library of Medicine, which is part of the United States National Institutes of Health (NIH). PubChem can be accessed for free through a web user interface. It is a database of chemical structures of validated small molecules. The database contains entries for PubChem entities which are grouped by structural identity and similarity. Users can submit compounds and download via FTP.

FILTERING OF PROITEINS:

Glutathione *S*-transferase of the malarial parasite *Plasmodium falciparum* (PfGST) represents a novel class of the GST isoenzymes. This GST is highly abundant in the malaria parasite, its activity was found to be increased in chloroquine-resistant cells, and it has been shown to act as a ligandin for parasitotoxichemin. Thus, the enzyme represents a promising target for antimalarial drug development for malaria. The three dimensional structure of the protein was retrieved using Protein Data Bank (PDB) (https://www.rcsb.org/) which was determined by experimental studies by X-Ray Diffraction. The Protein Data Bank archive (PDB) has served as the single repository of information about the 3D structure of protein, nucleic acids, and complex assemblies. The Worldwide PDB (wwPDB) organization manages the PDB and ensure that the PDB is freely and publicly available to the global community.

MOLECULAR DOCKING USING ARGUS LAB:

After the preparation of the Protein and Ligand, molecular docking studies were performed to evaluate the interaction using Argus lab software. Molecular docking is a study of how two or more molecular structures, for instance, drug and catalyst or macromolecule receptor, match along to be a perfect fit of protein. Argus Lab is a molecular modelling, graphics, and drug design program. It is the new drug docking code. Contains both GADock and ArgusDock docking engines and the A score scoring function with a preliminary set of parameters. Argus lab is a free molecular package that runs under windows. It is installed on all public computers in Shoker Science Center (an icon should be on your desktop), and you may also download it for personal use from www.arguslab.com/downloads.htm. The program allows you to draw very complex protein configurations, obtain helical chains of amino acids and folded leaves etc. Argus Lab uses a tree system to organize all the elements to add to any structure before representing this data as a drawing, allowing you to analyses it a visual manner. The docking was carried out for the protein PF GST and the 34 compounds of *Anisomeles malabarica* using the software Argus Lab.

Comparative Studies:

The best interaction of the phytochemical compounds were compared with the interactions of synthetic compound S-Hexylglutathione and Protoporphyrin for the parasitic disease Malaria. These synthetic compounds were docked with the same protein PF GST (PDB ID 10KT). The comparative Docking studies were performed and it proves that the phytochemical compounds has the best interactions than the Synthetic compounds. So it proves that the phytochemicals of *Anisomeles malabarica* could lead to the discovery and development of potential drugs against parasitic diseases such as Malaria.

RESULTS AND DISCUSSIONS:

Preparation of small molecules:

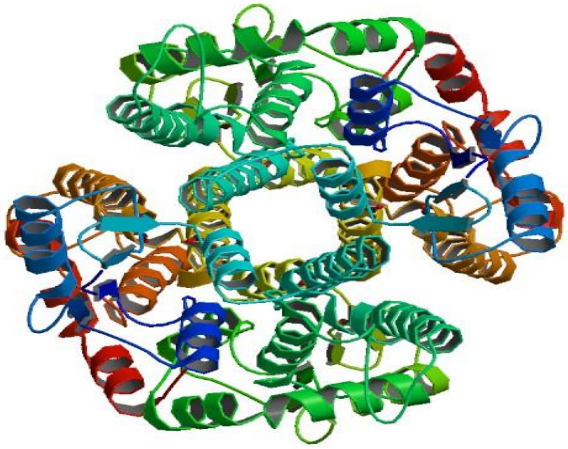
The 34 phytochemical compounds through literature were identified from the plant *Anisomeles malabarica* with the help of PubMed database.

Table 1: Pytochemical Compounds from *Anisomeles malabarica*

S.No	Pytochemical Compounds
1.	Alpha pinene
2.	Camphene
3.	Beta pinene
4.	3-Octanol
5.	1,8-cineole
6.	Cis-sabinene hydrate
7.	Linalool
8.	Camphor
9.	Borneol
10.	Myrtenol
11.	Alpha-thujone
12.	Linalyl acetate
13.	Nerol
14.	Geraniol
15.	Geranial
16.	Thymol
17.	Bornyl acetate
18.	Terpenyl acetate
19.	Anisole
20.	2-Isopropylbenzaldehyde
21.	Eugenol
22.	N-nonanyl acetate
23.	Delta-cadinene
24.	Isocaryophyllene
25.	Caryophyllene oxide
26.	Epiglobulol
27.	Globulol
28.	Nerolidyl acetate
29.	Farnesyl acetate
30.	Alpha bisabolol
31.	Trans-phytol
32.	Citronellol
33.	Isomenthol
34.	Azulene

Preparation of proteins:

The structure of protein PF GST (plasmodium falciparum glutathione s-transferase) was predicted and retrieved from the Protein Data Bank. The PDB ID is 10KT. The Protein Data Bank (PDB) is a database for the three-dimensional structural data of large biological molecules, such as proteins and nucleic acids.



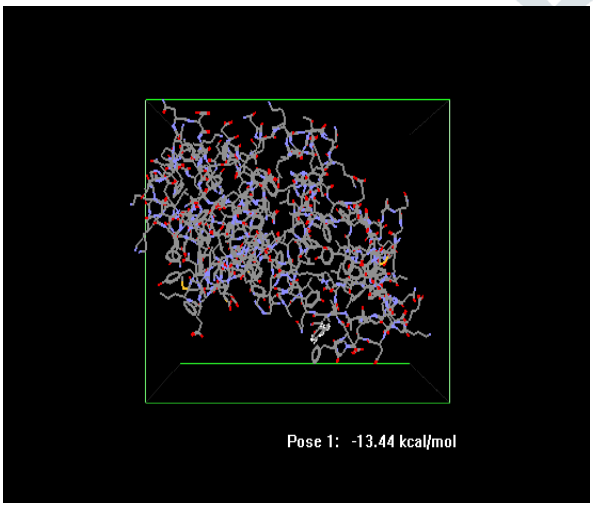
3D Structure of plasmodium falciparum glutathione s-transferase 1OKT

Docking Interactions:

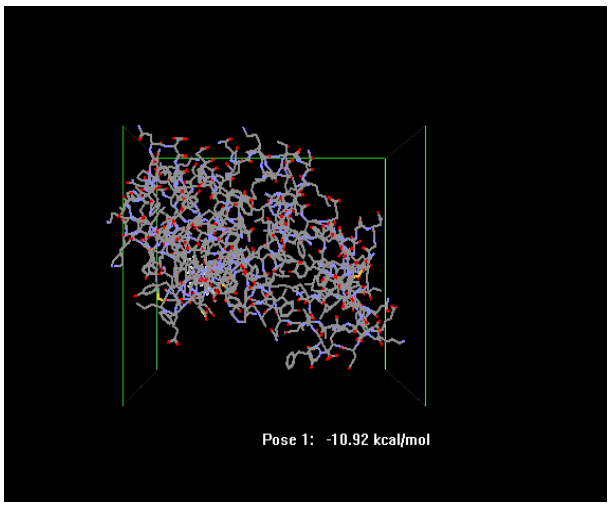
Table 2: Docking Interactions of the ligands and proteins.

S.No	pytochemical Compounds	1OKT Kcal/mol
1.	Alpha pinene	-12.66
2.	Camphene	-12.17
3.	Beta pinene	-11.48
4.	3-Octanol	-8.93
5.	1,8-cineole	-8.33
6.	Cis-sabinene hydrate	-9.55
7.	Linalool	-10.32
8.	Camphor	-9.48
9.	Borneol	-11.02
10.	Myrtenol	-10.15
11.	Alpha-thujone	-7.75
12.	Linalyl acetate	-7.98
13.	Nerol	-11.92
14.	Geraniol	-8.95
15.	Geranial	-10.72
16.	Thymol	-10.27
17.	Bornyl acetate	-11.13
18.	Terpenyl acetate	-9.47
19.	Anisole	-8.35
20.	2-Isopropylbenzaldehyde	-10.52
21.	Eugenol	-8.58
22.	N-nonanyl acetate	-10.23
23.	Delta-cadinene	-12.04
24.	Isocaryophyllene	-9.97
25.	Caryophyllene oxide	**-13.44**
26.	Epiglobulol	-12.08
27.	Globulol	-12.24
28.	Nerolidyl acetate	-9.64
29.	Farnesyl acetate	-8.95
30.	Alpha bisabolol	-12.81
31.	Trans-phytol	-10.72
32.	Citronellol	-10.07
33.	Isomenthol	-11.08
34.	Azulene	-12.74

Best binding energies of Phytochemical and Synthetic compounds:



1OKT for Caryophyllene oxide
(Phytochemical compound)



1OKT for Protoporphyrin
(Synthetic compound)

Comparative Studies:

The synthetic compounds were identified for the disease Malaria through literature and journal survey using PubMed. The synthetic compounds are S-Hexylglutathione and Protoporphyrin. These proteins were retrieved for performing Molecular docking studies to predict the interactions. This was performed to compare the interactions between Synthetic compounds and phytochemical compounds. The Synthetic Compounds S-Hexylglutathione and Protoporphyrin were docked with the same protein *Plasmodium falciparum* glutathione s-transferase (PF GST) of the PDB id 1OKT. And we identified that the interactions of synthetic compounds were lower than the Phytochemical compounds.

Table 3: Docking interactions of Synthetic and Phytochemical compounds.

S.No	Protein	Synthetic Compounds	Ligand Pose Kcal/mol
1	1OKT	S-Hexylglutathione	-6.27
2	1OKT	Protoporphyrin	**-10.92**

S.No	Protein	Phytochemical compounds	Ligand Pose Kcal/mol
1.	1OKT	Caryophyllene oxide	**-13.44**
2.	1OKT	Alpha bisabolol	-12.81

CONCLUSION:

Molecular Docking studies proved that the phytochemical compounds from the medicinal plants has the best interactions than the Synthetic compounds which are used for treatment nowadays. While comparing both the docking interactions of phytochemical compounds and the synthetic compounds, it proves that the phytochemical compounds Caryophyllene oxide has more effect to cure the pathogenic disease Malaria. In future the phytochemical compounds of *Anisomeles malabarica* can lead to the discovery and development of potential drugs against various pathogenic diseases. Although the synthetic compounds are very effective against malaria, it has reported to possess several side effects. Hence it is suggested to use natural compounds from the medicinal plants as a good alternative. Further clinical and in vitro studies can be performed on these natural compounds *which* could lead to the discovery of novel potential effective drugs against malarial disease.

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