International Journal of ChemTech Research
CODEN (USA): IJCRGG, ISSN: 0974-4290, ISSN(Online):2455-9555
Vol.13 No.03, pp 149-165, 2020

In Silico Anti- HIV Analysis of FTIR identified Bioactive compounds present in *Vitex altissima* L and *Vitex leucoxylon* L

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Abstract : The knowledge of the traditional plants in India is a collection over millennia by our ancient people. The Siddha System of Medicine (Traditional Tamil System of medicine) is the foremost of all other medical systems in the world which provide service to the humanity for more than 5000 years in combating diseases and also in maintaining its physical, mental and moral health. *Vitex* species were used in siddha for its anti- viral activity for several years. However, the present study deals with the Human Immunodeficiency Virus because of its complexity and killing effects. FTIR analysis of *Vitex altissima* L and *Vitex leucoxylon* L revealed the presence of 21 and 17 bioactive compounds respectively. These compounds were analysed further for its binding affinity mechanism against one of the virulence causing protein, reverse transcriptase (target protein) of Human Immunodeficiency Virus (HIV) by using molecular docking and bioinformatics tools. Interaction rate was determined between bioactive compounds against the protein target based on binding free energy requirements. Molecular docking was also made to the commercially available drugs (Zidovudine, Stavudine, and Nevirapine) against the target protein. By comparing the results between bioactive compounds in the *Vitex* species and the commercially available drugs, it was clear that the bioactive compounds were much more effective than the commercially available drugs, thereby suitable for the treatment of AIDS. Hence, this study will form the basis for promoting therapeutic lead molecules from the traditional plants which restore the tradition and also eliminates the harmful side effects.

Keywords : FTIR analysis, *Vitex altissima* L, *Vitex leucoxylon* L, Zidovudine, Stavudine, Nevirapine, Binding free energy, Molecular docking.

DOI= http://dx.doi.org/10.20902/IJCTR.2019.130312

1. Introduction

Traditional systems of medicine (TM) are the most important medicinal system in the world that has been practiced in many countries such as China, Japan and India since immemorial time [1]. These are usually known as "non- conventional" medicine by the world [2]. Siddha system of medicine (SSM) is one of the oldest and the most important traditional systems of medicine, which has been originated in India (southern parts of the country) for treating various disorders and it can even cure chronic illness [3][4]. Most of the therapeutic approaches aim at symptomatic aid rather than providing the clear and permanent cure to the sickness. Nowadays, there is a growing interest in traditional systems of medicine because of the fact that the therapeutic approaches do not cure the sickness permanently which is posing a greater health issue for a wider population across the globe, especially in the developing countries. Even the World Health Organization (WHO) recommends the practice of the traditional system of medicine as it is affordable, safe and culturally acceptable [5]. TM practices generally uses plants as a basic component for the development of drugs and this practice have been followed for thousands of years by people in China, India, and rest of the other countries [6]. Various evidences prove that plants were used for treating medical illness by various people. The earliest known medical document is a 4000-year-old Sumerian clay tablet that recorded plant remedies for various illnesses [7]. Its origin dates back to BC 10,000 to BC 4, 000 [8]. These plants can also cure various disease caused by microorganisms such as bacteria, viruses, fungi etc.

Acquired immune deficiency syndrome (AIDS) is a spectrum of events caused by the infection of Human Immunodeficiency Virus (HIV). HIV is a highly variable virus which mutates very rapidly. HIV/AIDS has had a great impact on society, both as an illness and as a cause of discrimination. Generally, there are two major types of the human immunodeficiency virus. HIV-1, which was discovered first, is the most widely spread all over the world. HIV-2 is more than 55% genetically different from HIV-1 [9]. Due to this genetic difference, HIV-1 and HIV-2 antigens are distinct enough that if a test is developed only to detect HIV-1; it will not reliably detect HIV-2. In India, HIV-1 is very common. According to Traditional medicinal system, there are various species of plants which shows anti-HIV activity.

1.1 Siddha system of medicine (SSM) and Vitex species

India is a land of various important plant species. It is the land which shows unique characteristic of having six renowned systems of traditional medicinal practices: Siddha, Ayurveda, Unani, Yoga, Naturopathy and Homoeopathy [7]. Siddha medicine is the most ancient indigenous system of medicine of Indian origin that has been in practice in Tamil Nadu and in some parts of southern India. The SSM is the oldest traditional treatment system generated from Dravidian culture and it has flourished during the period of Indus valley civilization [10]. According to the siddha concepts, matter and energy are the two dominant elements, which have great influence in shaping the nature of the universe. Matter cannot exist without energy and vice-versa and so they are inseparable [11]. These two entities are, therefore, termed as Siva and Sakthi in the siddha system.

The important feature of SSM is to study the relationship between the mind and body and aims at maintaining the physical, mental and moral health of an individual. Application of Siddha medicine includes a wide range of activities, from physical cures using herbal medicines and other remedies, to the promotion of psychological and spiritual well-being of humans.

Various plants are found to possess medicinal values and are used for treating various chronic and acute diseases. The genus *Vitex* consists of over 270 species, predominantly trees and shrubs and is restricted to tropical and subtropical regions, while a few species are also found in temperate zones [12]. *Vitexaltissima* L. is a large tree with a grey, scaly, fibrous bark. The leaves are 3-foliolate; petiole angular or winged; the leaflets are sub sessile, elliptic-lanceolate while the flowers are bluish-white, terminal paniculate cymes [13]. It is commonly known as 'Mayilainotchi' [14] (in Tamil) and is widely distributed in South East Asia. It is used for the treatment of stomatitis, cardiac diseases, anorexia, blindness, leprosy, worm infestation, rheumatic swellings and chest pains [15].It has also anti-inflammatory [16] and antioxidant [17] activities. Stem bark is for the treatment of ephemeral fever [18], snake bite [19]. Leaves are used to treat wounds [20][21], skin allergies [22], snake and scorpion bites [23] and rheumatism [24].

Vitexleucoxylon L., commonly known as "nir-notchi" (in Tamil), is the most important medicinal plant widely distributed in Eastern Ghats and Deccan plateau regions located in India. The leaves of *V.leucoxylon* L are used in traditional medicine (mainly in SSM) for relieving headache, fever [25]. General pharmacological

studies revealed various properties of aqueous and ethanolic extracts of leaves of *V. Leucoxylon* L such as anti-psychotic, anti-depressant, analgesic, anti- inflammatory, anti-parkinsonian and anti-microbial activities [26]. Sarma et al. have studied the anti-inflammatory and wound healing properties of the crude alcoholic extract of the leaves in acute inflammation model [27]. The roots and bark are astringent and the roots are reported to be used as a febrifuge.

1.2 HIV and its mechanism

A virus is a unique pathogen which is incapable of replicating without a host cell. It utilizes the host cell environment and cellular factors for its propagation. This unique feature makes it difficult to develop a process or drugs to attack the virus or its replication directly without any adverse effects on the infected cells. However, viruses share a common stage in their replication cycle [28], which includes:

Attachment and entry to the host cell,

Transcription of viral mRNA,

Replication of viral genome,

Assembly and budding as progeny virus particles.

RNA viruses, such as HIV, HCV, and influenza, are genetically highly variable, because of the presence of viral reverse transcriptase or RNA-dependent RNA polymerase that cannot perform proofreading procedure. Thus, subsequent mutations in viral RNA genome have been proven to be associated with the surfacing of drug-resistant viruses [29][30][31]. The emergence of such drug-resistant viruses poses a challenge for the design of new drugs. These problems emphasize the need to develop new antiviral drugs targeting different steps in the viral replication cycle in comparison with the old traditional way of action.

Human immunodeficiency Virus (HIV) is a very dangerous virus comes under a group of retroviruses. They possess RNA which can be transferred to DNA during infection with the help of Reverse transcriptase enzyme. Generally, there are two types of HIV, HIV-1 and HIV-2 (http://www.avert.org/hiv-types.htm). However, both are transmitted by sexual intercourse, blood transfusion, and from mother to child and so, they materialize to cause clinically indistinguishable AIDS infection. On the other hand, it is observed that the HIV-2 is less easily transmitted. This is because their period of infection and the illness caused by both these types differs from one another. In case of HIV-2, the period between initial infection and illness is longer compared to that of HIV-1.

HIV-1 is the most common pathogenic strain of HIV. It is further classified into 4 groups: the "major" group M, the "outlier" group O and two new groups, N and P (http://www.avert.org/hiv-types.htm). These groups are classified based on the independent transmission of HIV into humans [32]. Among all, the Group M is the most common and highly dangerous type of HIV.

1.2.1 Mechanism of action

HIV-1 is unique in terms of its transmission and replication [33]. HIV-1 is transmitted both by sexual contact and hematogenously through contaminated needles or blood products, so the virus can initiate infection by crossing a mucosal barrier and by direct entry into a T cellor monocyte/ macrophage lineage cell in the peripheral blood. HIV-1 can spread after a long latent period of infection. The reverse transcriptase (RT) of the human immunodeficiency virus (HIV) is a heterodimer. HIV-1 RT consists of a 66-kda (p66) and a 51-kda (p51) protein with identical amino-terminal sites. The p51 subunit arises from the p66 subunit by viral proteolysis [34] between the amino acid residues Phe440 and Tyr441 [35]. The heterodimer catalyses the reverse transcription of the viral RNA genome, an essential step in the viral replicative cycle. During this process, the single-stranded viral RNA is copied to a double-stranded DNA genome by the multifunctional reverse transcriptase, containing RNA-dependent DNA polymerase, RNase H, and DNA-dependent DNA polymerase activity. Subsequently, the viral DNA can be inserted into the host cell genome by the action of the viral integrase. Recent advances in the understanding of the cellular and molecular mechanisms of HIV-1 entry and replication have provided the basis for novel therapeutic strategies to prevent viral penetration of the target cell membrane and inhibit virus multiplication. The inhibitors affect the essential proteins that are involved in replication cycle, reverse transcriptase and protease, to obtain optimum therapeutic effects. Currently, the number of anti- HIV/AIDS therapeutic drugs approved by the FDA has increased to 26 drugs, the first approved

drug being AZT, in 1987 [36]. These anti-HIV/AIDS drugs can be categorized into several types, nucleoside/ nucleotide reverse transcriptase inhibitors (NRTIS), non-nucleoside reverse transcriptase inhibitors (NNRTIS), and protease inhibitors [37][38][39][40]. However, HIV-1 has developed an astonishing degree of genetic mutations in protease, reverse transcriptase, and gp41 that have been associated with decreased vulnerability to the presently available antiretroviral drugs. In addition to these three types of inhibitors, there are further developments of drugs based on mutations called as integrase inhibitors and maturation inhibitors that target Gag and it showed promising effects in preclinical and clinical trials [40][41][42][43].

1.3 Commercial drugs

The drugs that are used against Reverse transcriptase enzyme are listed in the World Health Organization's List of Essential Medicines [44]. The following are the three top ranked medicines that are in use for the treatment of HIV infections.

1.3.1 Zidovudine: Zidovudine (INN) or azidothymidine (AZT) (also called as ZDV) is a nucleoside analog reverse-transcriptase inhibitor (NRTI), a type of antiretroviral drug used for the treatment of HIV/AIDS infection. AZT is a thymidine analog that inhibits the enzyme (reverse transcriptase) by terminating the synthesis of viral DNA, thus preventing the spread of HIV virus [45]. It is the first commercially available drug against HIV that was approved by FDA.

1.3.2 Stavudine: Stavudine (2′, 3′-didehydro-2′, 3′-dideoxythymidine, d4t, brand name Zerit) is a nucleoside analog reverse-transcriptase inhibitor (NARTI) active against HIV [46]. It is an analog of thymidine, thus during replication it gets incorporated into the viral DNA in place of natural thymidine triphosphate thus terminating the replication of viral DNA strand.

1.3.3 Nevirapine: Nevirapine (NVP), also marketed under the trade name Viramune, is a non-nucleoside reverse transcriptase inhibitor (NNRTI) used to treat HIV-1 infection and AIDS. It binds allosterically to the enzyme awy from its actual active site, thus the effect is low. As with other antiretroviral drugs, HIV rapidly develops resistance if nevirapine is used alone, so recommended therapy consists of combinations of three or more antiretrovirals [47].

1.4 Insilico analysis

In silico is a latin word which was coined in 1989 and it literally means, "performed on computer". It involves the series of experiments on living organisms that are carried outside their body (computer simulation) using various important software tools called as bioinformatics tools. In drug discovery and development, in silico studies reduces the need for expensive lab work and clinical trials. This can be made possible by using Protein- ligand docking method to identify the potential inhibitors to an enzyme associated with any disease. This approach differs from the use of expensive high-throughput screening (HTS) labs to physically test all the samples over a long period of time and also their hit rate will be very low.

2. Materials and Methods

2.1 FTIR analysis of Vitex species

Identification of the chemical nature of phytochemical compounds present in the medicinal plants will provide various information regarding the different functional groups that are responsible for their medicinal properties. *Vitexaltissima* L. And *Vitexleucoxylon* L were collected from various regions of Thanjavur district, Tamil Nadu. The leaves were shade dried, powdered and the leaf extracts of *Vitexaltissima*L. And *Vitexleucoxylon* L were subjected to FTIR analysis [48]. The results revealed that 21 and 17 important bioactive compounds respectively are found in both these species. These compounds are used in further Insilico analysis.

2.2 Retrieval of 3D structure of reverse transcriptase (target protein)

The first step of insilico analysis involves the retrieval of the structure of reverse transcriptase (a protein present in HIV type- 1) from databases. A non-liganded high resolution 3D structure was retrieved from protein data bank in PDB format [49]. PDB ID: 1DLO (http://www.rcsb.org/pdb/explore/explore.do?Structureid=1dlo)

2.3 Construction of 3d structures of ligands

The compounds present in *Vitexaltissima* L and *Vitexleucoxylon* L was reported using Fourier transform infrared spectroscopy (FTIR) analysis. This analysis forms the basic concept for the present study. Chemsketch, a chemically intelligent drawing interface freeware, developed by Advanced Chemistry Development, Inc., (http://www.acdlabs.com) was used to construct the structure of the ligands (phytochemicals). Using the DRAW mode of Chemsketch, the ligands were generated and the three dimensional optimizations were done and the ligand files were saved in .mol format [Original Arguslab docking tutorial available on-line (www.arguslab.com) [50].

2.4 Preparation of target protein

By using Argus lab, the ligands and the crystallographic water molecules were removed from the protein. Crystallographic disorders and unfilled valence atoms were corrected. Then the protein was subjected to energy minimization and on the final stage, the hydrogen atoms were added to the target protein molecule before docking [51].

2.5 Preparation of ligands

Geometry optimization of the ligands was performed according to the Hartree-Fock (HF) calculation method using arguslab 4.0.1 software [52].

2.6 Binding site prediction

Various analyses showed that the binding of reverse transcriptase occurs allosterically in a hydrophobic pocket located approximately 10 Å from the catalytic site in the palm domain of the p66 subunit site of the protein [53]. The NNRTI binding pocket (NNIBP) contains five aromatics (Tyr-181, Tyr-188, Phe-227 and Trp-229), six hydrophobic (Pro-59, Leu-100, Val-106, Val-179, Leu-234 and Pro-236) and five hydrophilic (Lys-101, Lys-103, Ser-105, Asp-132 and Glu-224) amino acids that belong to the p66 subunit and additional two amino acids (Ile-135 and Glu-138) belonging to the p51 subunit [54]. Each NNRTI interacts with different amino acid residues in the NNIBP [55]. The sites are verified and confirmed using online bioinformatics tools like Metapocket (http://projects.biotec.tu-dresden.de/metapocket/index.php/).

2.7 Creation of local database for ligands

In order to accelerate the process, the local database of prepared ligands (phytochemicals) was created using Open Babel GUI software [56]. Ligands which were stored in .mol format were integrated into .sdf (acceptable database format for ARGUS LAB software) file.

2.8 Molecular docking using argus lab

Insilicomodeling is an efficient way for the traditional drug testing compounds that reduces the time consuming multi step process against screening and clinical trials. Molecular docking is amethod which helps to confirm the binding mode andinteraction energy for the ligands with the target protein.

Molecular docking was carried out in Argus lab. Argus Lab is the electronic structure program which operates on the basis of quantum mechanics and helps to predict the potential energies, molecular structures, geometry optimization of structure, vibration frequencies of coordinates of atoms, bond length, bond angle and reactions pathway. Based on predicted binding residues, grid box was constructed and molecular docking was performed. All computational docking studies were carried out using Argus lab 4.0.1 installed in a single machine running on a 2.5 GHz core i7 processor with 6 GB RAM and 320 GB hard disk with windows 8.1 as an operating system.

Target protein (Reverse trascriptase) was docked against the obtained ligands using arguslab 4.0.1 (Mark A. Thompson, Planaria Software LLC, Seattle, WA, USA, www.arguslab.com) to find out the reasonable binding geometries and explore the protein ligand interactions. Docking of the protein - ligand complex was mainly performed only on to the predicted active site. Docking simulations were performed by selecting "argusdock" as the docking engine [57].

The selected residues of the receptor were defined to be a part of the binding site. A spacing of 0.4 Å between the grid points was used and an exhaustive search was performed by enabling "High precision" option in Docking precision menu, "Dock" was chosen as thecalculation type, "flexible" for the ligand and the Ascore wasused as the scoring function.

The A Score function was used to calculate the binding energies of the resulting docked structures [58]. All the compounds in the dataset were docked into the active site of reverse transcriptase, using the same protocol. After the completion of docking, the docked protein (protein-ligand complex) was analysed to investigate the quality of interactions.

2.9 Retrival and optimization of 3d structures of commercially available drugs

3D structures of the following commercially available drugs were collected and confirmed by studying several literatures.

- **Zidovudine**
- **Stavudine**
- **Nevirapine**

The retrieved 3D structures were optimized to avoid Crystallographic errors. The Geometry optimization of the drugs was performed according to the Hartree-Fock (HF) calculation method using arguslab 4.0.1 software [52].

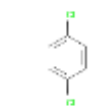
2.10 Molecular docking using argus lab

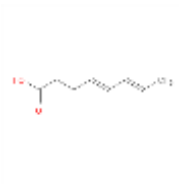
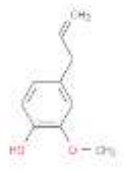
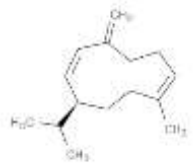
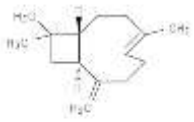
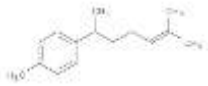
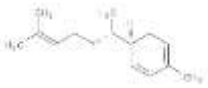
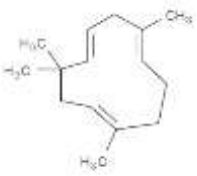
The obtained 3D structures of commercially available drugs were considered as the ligand molecules and were docked against the reverse transcriptase to find out the reasonable binding geometries and also to investigate the protein - ligand interactions. Also, the docking was performed only on to the predicted active sites.

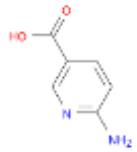
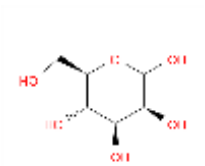
3. Results

The samples (leaf extracts of *Vitexaltissima* L. And *Vitexleucoxylon* L) first undergo FTIR analysis and all the bioactive compounds present in it were identified. The necessary information such as Molecular formula, molecular structure and molecular weight about the phytochemicals was retrieved from Pubchem database. The 3D structures of these bioactive compounds are docked against the protein target (reverse transcriptase) and the following results were obtained.

Table 1: FTIR Analysed Result of Leaf extraction of *Vitex altissima* L. and the Results of Docking with the Target Protein (Reverse Transcriptase)

Serial number	Compound name	Molecular formula	Molecular weight	Structure	Binding free energy (kcal/mol)
1	Benzene, 1, 4-dichloro	$C_6H_4Cl_2$	146		-9.6699

2	4, 6-Octadienoic Acid	$C_8H_{12}O_2$	140		-10.656
3	Eugenol	$C_{10}H_{12}O_2$	164		-10.4401
4	Germacrene D	$C_{15}H_{24}$	204		-12.3021
5	**Caryophyllene**	**$C_{15}H_{24}$**	**204**		**-15.4645**
6	Benzene,1-(1,5-dimethyl-4-hexenyl)-4-methyl,[S-(R*, S*)]	$C_{15}H_{24}$	202		-14.0318
7	1, 3-Cyclohexadiene, 5-(1, 5-dimethyl-4-hexenyl)-2-methyl	$C_{15}H_{24}$	204		-11.7252
8	À-Caryophyllene	$C_{15}H_{24}$	204		-14.1437

9	1,6,10-Dodecatriene, 7, 11-dimethyl-3-methylene-[Z]	$C_{15}H_{24}$	204		-12.6587
10	Dodecanoic acid	$C_{12}H_{24}O_2$	200		-4.60943
11	3-Pyridine carboxylic acid,6-amino	$C_6H_6N_2O_2$	138		-7.71027
12	D-Mannose	$C_6H_{12}O_6$	180		-10.8761
13	Tetradecanoic acid	$C_{14}H_{28}O_2$	228		-14.671
14	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	$C_{20}H_{40}O$	296		-10.9167
15	1, 2-Benzene Dicarboxylic acid, butyl octyl ester	$C_{20}H_{30}O_2$	334		-11.2288

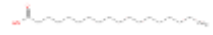
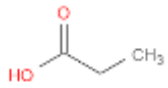
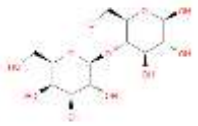
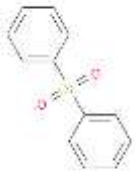
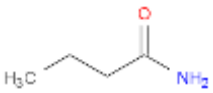
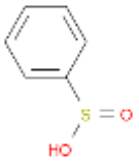
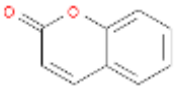
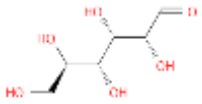
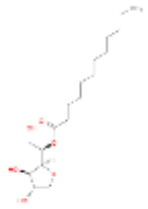
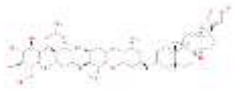
16	N-Hexadecanoic Acid	$C_{16}H_{36}O_2$	256		-10.7807
17	Hexadecanoic acid ethyl ester	$C_{18}H_{36}O_2$	284		-12.8909
18	Phytol	$C_{20}H_{40}O$	296		-10.1599
19	9, 12-Octadecadienoic acid [Z, Z]	$C_{18}H_{36}O_2$	280		-12.0161
20	Octadecanoic acid	$C_{18}H_{36}O_2$	284		-10.3974
21	Squalene	$C_{30}H_{50}$	410		-12.3085

Table 2: FTIR Analysed Result of Leaf extracts of Vitex leucoxylon L and the Results of Docking with the Target Protein (Reverse Transcriptase)

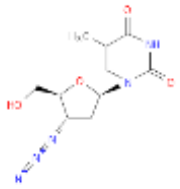
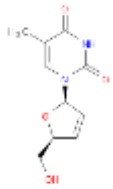
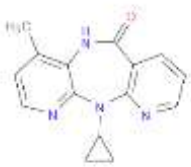
Serial number	Compound name	Molecular formula	Molecular weight	Structure	Binding free energy (kcal/mol)
1	Nonyl Aldehyde	$C_9H_{18}O$	142.239 Da		-7.7597
2	Triacontane	$C_{30}H_{62}$	422.813 Da		-9.70885
3	Propionic Acid	$C_3H_6O_2$	74.078 Da		-10.5
4	(+)-Beta-D-Lactose	$C_{12}H_{22}O_{11}$	342.297 Da		-6.9179
5	**Phenyl Sulfone**	**$C_{12}H_{10}O_2S$**	**218.272 Da**		**-11.6156**
6	Butyramide	C_4H_9NO	87.120 Da		-4.97783

7	Benzenesulfinic acid	$C_6H_6O_2S$	142.176 Da		-9.48683
8	3-Nonanone	$C_9H_{18}O$	142.239 Da		-10.7358
9	Coumarin	$C_9H_6O_2$	146.143 Da		-9.44295
10	Maltotriose hydrate	$C_{18}H_{32}O_{16}$	504.437 Da		-5.86672
11	Dextrin	$C_6H_{12}O_6$	180.156 Da		-6.84262
12	Maltopentaose hydrate	$C_{30}H_{54}O_{27}$	846.734 Da		-5.8316

13	Tridecanal	$C_{13}H_{26}O$	198.345 Da		-9.012
14	Sorbitanmonolaurate	$C_{18}H_{34}O_6$	346.459 Da		-10.2215
15	Gamma-Cyclodextrin hydrate	$C_{48}H_{80}O_{40}$	1297.125 Da		-7.16
16	Lanatoside a	$C_{49}H_{76}O_{19}$	969.116 Da		-10.2237
17	Dodecyl aldehyde	$C_{12}H_{24}O$	184.318 Da		-10.106

The commercial drugs were also docked against the target protein (Reverse transcriptase) and the following results of its binding efficiency was obtained.

Table 3: Structure and the Docking Results of the Commercially available drugs

Serial no	Compound name	Molecular formula	Molecular weight	Structure	Binding free energy (kcal/mol)
1	Zidovudine	$C_{10}H_{13}N_5O_4$	267.241 Da		-8.8776
2	Stavudine	$C_{10}H_{12}N_2O_4$	224.213 Da		-8.6193
3	Nevirapine	$C_{15}H_{14}N_4O$	266.298 Da		-9.68587

4. Discussion

The phytochemicals which were used in this study showed the binding energies in the range of -4.6 kcal/mol to -15.6 kcal/mol which is in very good agreement with the standard and ideal binding energy. **Caryophyllene** present in *Vitexaltissima* L. And **Phenyl Sulfone** present in *Vitexleucoxylon* L showed the maximum effectiveness against Reverse transcriptase than the available commercial drugs. Docked poses of commercial drugs and efficient bioactive compounds were shown below.

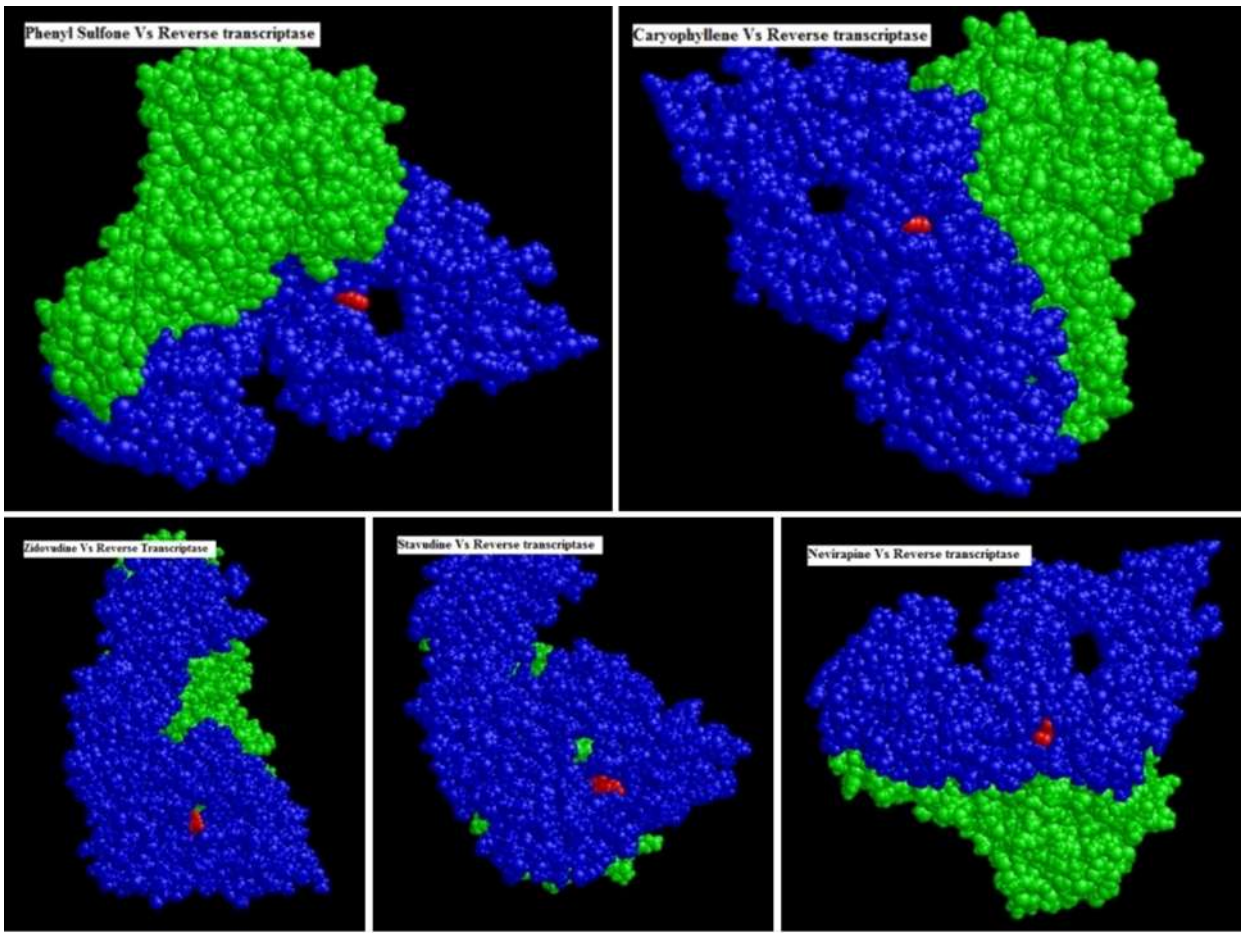


Fig 1: The docked poses of commercially available drugs and efficient bioactive compound against reverse transcriptase. (from clockwise) a. Phenyl sulfone VS Reverse transcriptase, b.Caryophyllene VS Reverse transcriptase, c. Nevirapine VS Rreverse transcriptase, d. Stavudine VS Rreverse transcriptase, e. Zidovudine VS Reverse transcriptase

5. Conclusion

The protein-ligand interaction plays a significant role in structural based drug designing. In this present work, receptors for HIV targets has been taken and the potential drugs have been identified that can be used against AIDS. By applying computational approaches, it has been tried to understand the mechanism of interactions and the binding affinity between phytochemicals and HIV targets. Hence these natural compounds could be used as a template for designing therapeutic lead molecules which could results into massive reductions in therapeutic development time. This study may be the subject of experimental validation and clinical trials to establish these phytochemicals as more potent drug for the treatment of AIDS. In future, the ADME/T (Absorption, Distribution, Metabolism, Excretion / Toxicity) properties of these compounds can be calculated using the commercial ADME/T tools available thereby reducing the time and cost in drug discovery process. These results will be an influential factor for determining a lead phytochemical for further drug discovery process.

6. Acknowledgement

We would whole heartedly thank the Tamil Nadu Veterinary and Animal Sciences University, Chennai for their support in providing the authenticated results for FTIR of the leaf samples.

7. References

[1] A.B.D. Vaidya and T.P.A. Devasagayam, Journal of Clinical Biochemistry and Nutrition, vol. 41, pp. 1-11, 2007.

[2] WHO, "Traditional Medicine Strategy 2002-2005", World Health Organization, Geneva, Switzerland, 2002. WHO/EDM/TRM/2002.1. Available at: http://whqlibdoc.who.int/hq/2002/who_edm_trm_2002.1.pdf (accessed on 25th May 2012).
[3] S.V. Subramanian, V.R. Madhaven (Eds), "Heritage of Tamil Siddha medicine". (Madras, India: International Institute of Tamil Studies), pp. 171-184, 1983.
[4] J.J Thas, Clinics in Dermatology, vol. 26, pp. 62-78, 2008.
[5] X. Zhang,"Regulatory situation of herbal medicines – a worldwide review", World Health Organization, Geneva, Switzerland. 1998. Available at: http://apps.who.int/medicinedocs/pdf/whozip57e/whozip57e.pdf (accessed on 25thMay 2012).
[6] W. Sneader, "Drug Discovery: A History", Chichester, UK: Wiley, 2005.
[7] K. Karunamoorthi, K.Jegajeevanram, J. Xavier, J. Vijayalakshmi, L. Melita,"Tamil traditional medicinal system- Siddha: An indigenous health practice in the international perspectives", TANG Humanitas Medicine, vol. 2(2), pp. 1-4, 2012.
[8] T.V. Sambasivapillai,"Dictionary based on Indian medical science, Chennai, India: National Institute of Siddha", 1931.
[9] J.O. Kahn, B.D. Walker,"Acute human immunodeficiency virus type 1 infection", N Engl J Med,vol. 339, pp. 33–39, 1998.
[10] P.K. Mukherjee, A. Wahile, "Integrated approaches towards drug development from Ayurveda and other Indian system of medicines", J Ethnopharmacol,vol. 103, pp. 25-35,2006.
[11] V. Narayanaswamy, "Introduction to the Siddha System of Medicine", Madras, India: Research Institute of Siddha Medicine, 1975.
[12] K. Padmalatha, K. Jayaram, N.L. Raju, M.N.V. Prasad and R. Arora,"Ethnopharmacological and Biotechnological Significance of *Vitex*", Bioremediation, Biodiversity and Bioavailability, vol.3(1), pp. 6-14,2009.
[13] A.K. Meena, U.S. Niranjan,M.M. Rao, M.M. Padhi, R. Babu, "A review of the important chemical constituents and medicinal uses of *Vitex* genus", Asian Journal of Traditional Medicines, vol. 6(2), pp.54-60, 2011.
[14] S. S. Sathish, N. Janakiraman, M. Johnson, "Phyto chemical analysis of *Vitex alitssima* L. Using UV-Vis, FTIR and GC-MS", International journal of Pharmaceutical Science and Drug Research, vol.4 (1), pp. 56-62, 2012.
[15] J.A. Parrotta, "Healing plants of Peninsular India", CABI Publishers, USA, pp. 770.
[16] C. Sridhar, G.V. Subburaju, Y. Venkateshwaralu, R.T. Venugopal. "New acylatediridoid glycosides from *Vitexaltissima*". Journal of Natural Products, Vol. 67, pp. 2012-2016, 2004.
[17] C. Sridhar, K.V. Rao, G.V. Subburaju, "Flavonoids, triterpenoids and a lignin from*Vitex altissima*". Phytochemistry, Vol. 66, pp. 1707-1712, 2005.
[18] P.M. Pragada, G.M.N. Rao, "Ethnoveterinary medicinal practices in tribal regions of Andhra Pradesh, India". Bangl J Plant Taxon Vol. 19(1), pp. 7-16, 2012.
[19] K.N. Reddy, C.S. Reddy, V.S Raju, "Ethnomedicinal observations among the Kondareddis of Khammam district, Andhra Pradesh, India", Ethnobotanical Leaflets Vol. 12, pp. 916-926, 2008.
[20] M. Ayyanar, S. Ignacimuthu,"Herbal medicines for wound healing among tribal people in Southern India: Ethnobotanical and scientific evidences". International Journal of Applied Research in Natural Products, Vol. 2(3),pp. 29-42, 2009.
[21] M.K.R. Narayanan, S. Mithunlal, P. Sujanapal, N. Anil Kumar, M. Sivadasan, A.H. Alfarhan, A.A. Alatar,"Ethnobotanically important trees and their uses by Kattunaikka tribe in Wayanad Wildlife Sancturary, Kerala, India". Journal of Medicinal Plants Research, Vol. 5(4), pp. 604-612, 2011.
[22] N. Rajakumar, M.B. Shivanna,"Traditional herbal medicinal knowledge in Sagar taluk of Shimoga district, Karnataka, India". Indian Journal of Natural Products and Resources, Vol. 1(1), pp. 102-108, 2010.
[23] M. Ayyanar, S. Ignacimuthu,"Medicinal plants used by the tribals of Tirunelveli hills, Tamil Nadu to treat poisonous bites and skin diseases". Indian J Trad Knowledge, vol. 4(3), pp. 229-236, 2005.
[24] K. Narayana Rao, T. Thammanna, "Medicinal Plants of Tirumala. T.T.D, Tirupati, India", 1990.
[25] Y.R. Chanda, "The wealth of India:A dictionary of Indian Raw materials and Industrial products", Publication and Information Directorate, CSIR, New Delhi, pp: 520-521, 1982. http://www.niscair.res.in/activitiesandservices/products/woi1.htm

[26] H.G. Makwana, B. Ravishankar, V.J. Shukla, N.P. Vijayan, C.K. Sasikala, V.N. Saraswathy and S.V. Bhatt, "General pharmacology of *Vitexleucoxylon*linn leaves". Indian J. Physiol. Pharmacol, Vol. 38, pp. 95-100, 1994. http://indianmedicine.eldoc.ub.rug.nl/root/M/65647/

[27] S.P. Sarma, K.S. Aithal, K.K. Srinivasan, A.L. Udupa, V. Kumar, D.R. Kulkarni and P.K. Rajagopal, "Anti-inflammatory and wound healing activities of the crude alcoholic extract and flavonoids of Vitexleucoxylon". Fitoterapia, vol. 61, pp. 263-265, 1990.

[28] Goudsmit, Jaap, "Viral Sex; the Nature of AIDS". Oxford University Press. New York, New York, pp. 51-58. Retrieved May 25, 2008.

[29] D.D. Richman, "Antiviral drug resistance". Antiviral Res, vol. 71, pp. 117-121, 2006.

[30] P.D. Yin, D. Das, H. Mitsuya,"Overcoming HIV drug resistance through rational drug design based on molecular, biochemical, and structural profiles of HIV resistance". Cell Mol Life Sci., Vol. 63, pp. 1706-1724, 2006.

[31] R.W. Shafer, S.Y. Rhee, D. Pillay, V. Miller, P. Sandstrom, J.M. Schapiro, D.R. Kuritzkes, D. Bennett, "HIV-1 protease and reverse transcriptase mutations for drug resistance surveillance". AIDS, vol. 21, pp. 215-223, 2007.

[32] "Origins of HIV and the AIDS Pandemic". Cold Spring Harbor Perspectives in Medicine 1 (1): a006841a006835. Doi:10.1101/cshperspect. a006841. PMC 3234451.PMID 22229120.

[33] KaioKitazato, Yifei Wang, Nobuyuki Kobayashi,"Viral infectious disease and natural products with antiviral activity". Drug DiscovTher, Vol. 1(1), pp. 14-22, 2007.

[34] F. DimarzoVeronese, D.T. Copeland, A.L. Devico, R. Rahman, S.Oroszlan, R.C. Gallo, M.G. Sarngadharan, "Characterization of highly immunogenic p66/p51 as the reverse transcriptase of HTLV-III/LAV". Science, vol. 231(4743), pp. 1289–1291, 1986.

[35] I.C. Bathurst, L.K. Moen, M.A. Lujan, H.L. Gibson, P.H. Feucht, S. Pichuantes, C.S. Craik, D.V. Santi, P.J. Barr, "Characterization of the human immunodeficiency virus type-1 reverse transcriptase enzyme produced in yeast". Biochem. Biophys. Res. Commun., vol. 171, pp. 589-595, 1990.

[36] E. De Clercq,"Current lead natural products for thechemotherapy of human immunodeficiency virus (HIV)infection". Med Res Rev, vol. 20, pp. 323-349, 2000.

[37] Y. Mel Safadi,V. Vivet-Boudou, R. Marquet R, "HIV-1 reverse transcriptase inhibitors". ApplMicrobiol Biotechnol, vol. 75, pp. 723-737, 2007.

[38] M.J. Camarasa, S. Velázquez, A. San-Félix, M.J. Pérez-Pérez, F. Gago,"Dimerization inhibitors of HIV-1 reversetranscriptase, protease and integrase: a single mode ofinhibition for the three HIV enzymes", Antiviral Res, vol. 71, pp. 260-267, 2006.

[39] V. Nair, G. Chi,"HIV integrase inhibitors as therapeuticagents in AIDS". Rev Med Viol (in press).

[40] Y. Pommier, A.A. Johnson, C. Marchand,"Integraseinhibitors to treat HIV/AIDS". Nat Rev Drug Discov, vol. 4, pp. 236-248, 2004.

[41] B. Grinsztejn, B.Y. Nguyen, C. Katlama, J.M. Gatell, A. Lazzarin, D. Vittecoq, C.J. Gonzalez, J. Chen, C.M. Harvey, R.D. Isaacs,"Safety and efficacy of theHIV-1 integrase inhibitor raltegravir (MK-0518) intreatment-experienced patients with multidrug-resistantvirus: a phase II randomized controlled trial". Lancet, vol. 369, pp. 1261-1269, 2007.

[42] B.S. Min, M. Hattori, H.K. Lee, Y.H. Kim,"Inhibitory constituents against HIV-1 protease from Agastacherugosa". Arch Pharm Res, vol. 22, pp. 22:75-77, 1999.

[43] B.S. Min, H.J. Jung, J.S. Lee, Y.H. Kim, S.H. Bok, Ma C. M,Nakamura N, M. Hattori, K. Bae, "Inhibitory effect oftriterpenes from Crataegus pinatifida on HIV-I protease". Planta Med, vol. 65, pp.374-375, 1999.

[44] "WHO Model List of essentialmedicines". World Health Organization. October 2013. Retrieved 22 April 2014.

[45] "Zidovudine". Pubchem Public Chemical Database. NCBI. Retrieved 2011-04-10.

[46] "New HIV recommendations to improve health, reduce infections and save lives". World Health Organization. November 30, 2009.

[47] "New drug profile: nevirapine". Clinical Immunotherapeutics, vol. 6 (4), pp. 307–317.

[48] R.Ashokkumar and M. Ramaswamy, "Phytochemical screening by FTIR spectroscopic analysis of leaf extractsof selected Indian Medicinal plants". Int.J.Curr.Microbiol.App.Sci., vol. 3(1), pp. 395-406, 2014.

[49] Y. Hsiou, J. Ding, K. Das, A.D. Clark Jr., S.H. Hughes, Arnold. "Structure of unliganded HIV1 reverse transcriptase at 2.7 A resolution: implications of conformational changes forpolymerization and inhibition mechanisms", E.Journal: (1996) Structure 4: 853-860.

[50] Thangaraj, Sindhu, Sundaraj, Rajamanikandan, Dhanapal, Durgapriya ,Jebamalai, Raj, Anitha, Selvaraj, Akila and Velliyur Kanniyapan,Gopalakrishnan. "Molecular Docking and Qsar Studies On Plant Derived Bioactive Compounds as Potent Inhibitors of DekOncoprotein". Asian Journal of Pharmaceutical and Clinical Research, vol. 4(2), 2011; ISSN-0974-2441.

[51] K. Rohini, P.S. Srikumar, "In Silico Approach of Anticancer Activity of Phytochemical Coumarins against Cancer Target Jnks". International Journalof Pharmacy and Pharmaceutical Sciences, ISSN-0975-1491.Vol 5, Suppl 3,2013.

[52] K.V. Lisina, EashwaranMurugesh, Shanmughavel Piramanayagam. "In Silico Analysis of Compounds Characterized from Nymphaea stellata Flower with MUC1". Int. J. Pharm. Sci. Rev. Res., vol. 23(2), Nov – Dec 2013; pp. 180-184. ISSN 0976 – 044X.

[53] A. Ivetac, J.A. McCammon, "Elucidating the Inhibition Mechanism of HIV-1 Non-Nucleoside Reverse Mechanism of Inhibitors through Multicopy Molecular Dynamics Simulations", Journal of Molecular Biology, vol. 388 (3), pp. 644- 658, 2008. doi: 10.1016/j.jmb.2009.03.037, PMC 2744402, PMID 19324058.

[54] M.P. De Béthune, "Non-nucleoside reverse transcriptase inhibitors (NNRTIs), their discovery, development, and use in the treatment of HIV-1 infection: A review of the last 20 years (1989-2009)", Antiviral Research, vol. 85 (1), pp. 75–90, 2009. doi: 10.1016/j.antiviral.2009.09.008, PMID 19781578.

[55] K. Das, P.J. Lewi, S.H. Hughes, E. Arnold, "Crystallography and the design of anti-AIDS drugs: Conformational flexibility and positional adaptability are important in the design of non-nucleoside HIV-1 reverse transcriptase inhibitors", Progress in Biophysics & Molecular Biology,vol. 88 (2), pp. 209–231, 2005. doi: 10.1016/j.pbiomolbio.2004.07.001.

[56] Noel M O'Boyle1, Michael Banck, Craig A James, Chris Morley, Tim Vandermeersch and Geoffrey R Hutchison,"Open Babel: An open chemical toolbox", Journal of Cheminformatics,vol. 3:33, 2011.

[57] SabaFerdous, Muhammad Usman Mirza, Usman Saeed. "Docking Studies reveal Phytochemicals as the long searched anticancer drugs for Breast Cancer". International Journal of Computer Applications (0975 - 8887) Vol. 67 - No. 25, April 2013.

[58] S. Rajeswari, S. Kriushnapriya, "Anticancer Activities of Phytochemicalsfrom a Wood Rot Mushroom Phellinus Adamantinus - in Silico and in Vitro Approach". International Conference on Bioscience, Biochemistry and Bioinformatics IPCBEE vol.5, 2011, IACSIT Press, Singapore.

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