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Ayur-informatics: Establishing an *in-silico-ayurvedic* remedy for AIDS.

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***Abstract*— Acquired Immune Deficiency Syndrome (AIDS) is an incurable and terminal disease of the human immune system caused by the Human Immunodeficiency Virus (HIV). 3D structures of CCR5 and CXCR4 proteins were generated using Homology Modeling. Active compounds from the medicinal herbs- *Azadirachta indica, Vitex negundo, Emblica officinalis, Aegle marmelos, Adhatoda vasica, Berberis aristata* and *Swertia chirajita* tested to have anti-viral property were used in this work. Chemical structures of the phyto-component of these herbs were retrieved from pubchem & converted to .pdb. Both the proteins were successfully docked with the *phyto*- components.**

Keywords- AIDS, CCR5, CXCR4, Bioinformatics, Homology Modelling, Drug designing.

I. INTRODUCTION

Acquired Immuno Deficiency Syndrome (AIDS) is a clinical syndrome that is the result of infection with Human Immuno Deficiency Virus (HIV), which causes profound immunosuppression. It has been a serious, life-threatening health problem since the first case was identified in 1981 and is the most quickly spreading disease of the century. Worldwide, it is the fourth biggest killer [1]. HIV emerged later in India than it did in many other countries [2]. Infection rates soared throughout the 1990s, and today the epidemic affects *all* sectors of Indian society, not just the groups – such as sex workers and truck drivers – with which it was originally associated. In a country where poverty, illiteracy and poor health are rife, the spread of HIV presents a daunting challenge. The spread of HIV in India has been diverse, with much of India having a low rate of infection and the epidemic being most extreme in the southern states [3, 4, 5].

The Envelope glycoprotein (Env) is the sole viral protein present on the surface of Human Immunodeficiency Virus-1 (HIV-1) virions. Env is synthesized as a 160 kDa precursor protein (gp160). It folds and trimerizes in the endoplasmic reticulum (ER) of the host cell, where it obtains ten disulfides and ~30 N-linked glycans depending on the viral isolate. In the Golgi complex, gp160 is cleaved by a cellular protease into a soluble subunit, gp120, and a transmembrane subunit, gp41. They remain non-covalently associated on the surface of infected cells and on virions. Together, the two Env subunits mediate viral entry: gp120 is responsible for binding to the receptor (CD4) and the coreceptor (CCR5 and CXCR4) on the host cell, and gp41 is needed for subsequent fusion of the viral and cellular membranes [6].

HIV uses CCR5 chemokine receptor as a coreceptor to gain entry into macrophages. The most commonly transmitted strains of HIV are those which bind to CCR5.The level of CCR5 expression is upregulated in chronic HIV infection. Measuring the expression of CCR5 is therefore one indicator of an individual's disease progression. Studies have found that CCR5 expression decreases after a person receives Highly Active Anti-Retroviral Therapy (HAART) treatment. CXCR4 is a chemokine receptor of GPCR gene family. It is expressed by cells in the immune system and the central nervous system. In addition to binding its to ligand SDF-1 (stromal cell-derived factor-1), CXCR4 triggers the migration and recruitment of immune cells mainly HIV receptor cells. CXCR4 functions as a co-receptor for entry of HIV into T cells and ligands of CXCR4, including SDF-1 may help in preventing HIV infection. CXCR4 induces downstream signaling. As a GPCR pathway candidate, CXCR4 binding with SDF-1 activates G-protein mediated signaling, including downstream pathways such as ras, and PI3 kinase [7]. Several reviews on the natural products for chemotherapy of HIV infection have been published earlier. Matthee *et al.*[8] reviewed naturally occurring HIV reverse transcriptase inhibitors. Jung *et al.*[9] discussed anti-HIV agents according to their chemical classes. Yang *et al.*[10] reviewed natural products-based anti-HIV drug discovery and development facilitated by NCI development

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programme. Recently, Cos *et al.*[11] reviewed different plant substances as anti-HIV agents according to their mechanism of action.

Table 1: Ayurvedic herb with corresponding active compound

Plant Name (Scientific name)	Common Name	Phyto compound used
Azadirachta indica	neem	Nimbin and nimbanene
Vitex negundo	nishinda	Isoorientin and casticin
Emblica officinalis	amlaki	Punicafolin and phyllanemblin
Aegle marmelos	bel	Limonene and germacrene B
Adhatoda vasica	vasak	Vasicine and vasicinone
Berberis aristata	daruharidra	Berberine and oxycanthine
Swertia chirata	chirata	Mangeferin and sawertiamarine

In this particular work authors wish to establish a remediation for AIDS using computational tools. The ayurvedic herbs used in this work are tested for their efficacy in treating diseases caused by viral infections.

II. METHODOLOGY

CCR5 protein and CXCR4 receptors were retrieved from Genbank [Table 1]. Homology modeling was carried out using Modeller 9.15 software, for predicting 3 D structures for the above mentioned proteins. Templates were downloaded from RCBS PDB database. The following templates were used [Table 1]:

Table 2: Receptor template information

Receptor	Genbank Accession Number	Template 1	Template 2	Template 3
CCR5	P51681.1	4MBSA	2LNLA	3ODUA
CXCR4	CAA12166.1	3OE6A	3OE0A	3ODUA

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Five models of each of the above proteins were generated. The models were analyzed by Rampage Ramchandran plot server and the best model of each was selected.

Chemical structures of active component of plants in Table 1 was retrieved from pubchem database converted to *.pdb file using Chimera software.

Each of the proteins used for this work, namely CCR5 & CXCR4 was docked with phyto-compounds in Table 1.

III. RESULTS and DISCUSSION

CCR5 & CXCR4 receptors were retrieved from Genbank and their homologous templates were downloaded from RCSB database. Using Modeller 9.15 software, the 3d structure of CCR5 & CXCR4 receptors were modelled. Modeller generated 5 models. These models were verified using Rampage Ramachandran Plot Server [Table 3, 4].

According to ramachandran plot CCR5 model 2 is selected as the best model for further docking studies [Fig. 1, 2].

Table 3: Ramachandran Plot analysis of CCR5

	Number of residues in favoured region	Number of residues in allowed region	Number of residues in outlier region	
Model 1	326 (93.1%)	18 (5.1%)	6 (1.7%)	
Model 2	338 (96.6%)	7 (2.0%)	5 (1.4%	(selected)
Model 3	332 (94.9%)	13 (3.7%)	5 (1.4%)	
Model 4	331 (94.6%)	15 (4.3%)	4 (1.1%)	
Model 5	330 (94.3%)	16 (4.6%)	4 (1.1%)	

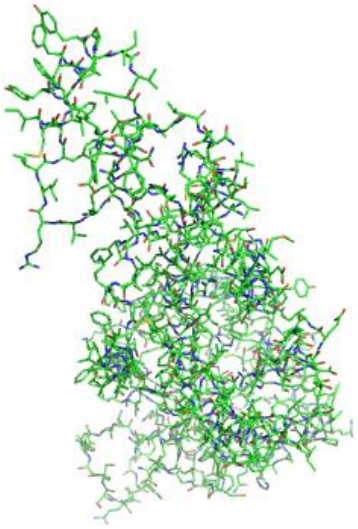

Fig. 1- Visualization of CCR5 receptor (model 2) in pymol.

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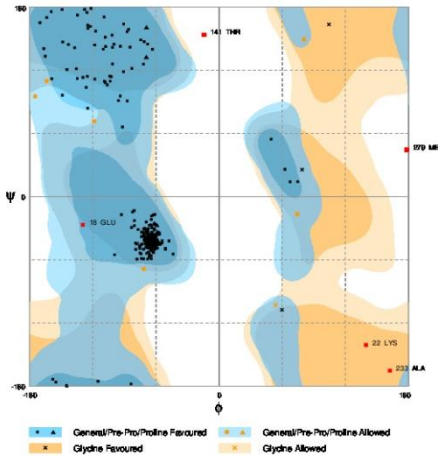

Fig. 2- Ramachandran plot analysis of CCR5 receptor (model 2).

CXCR4 receptor, model 2 is selected as the best model by ramachandran plot and selected for further studies [Fig. 3, 4].

Table 4: Ramachandran Plot analysis of CXCR4

	Number of residues in favoured region	Number of residues in allowed region	Number of residues in outlier region	
Model 1	335 (93.6%)	14 (3.9%)	9 (2.5%)	
Model 2	344 (96.1%)	6 (1.7%)	8 (2.2%)	Selected
Model 3	330 (92.2%)	19 (5.3%)	9 (2.5%)	
Model 4	338 (94.4%)	14 (3.9%)	6 (1.7%)	
Model 5	334 (93.3%)	15 (4.2%)	9 (2.5%)	

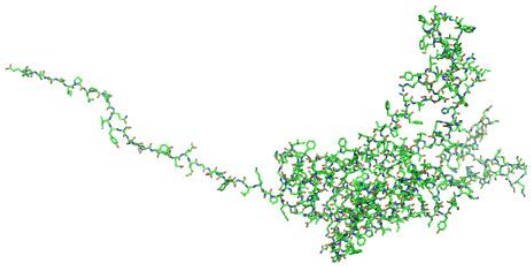

Fig. 3- Visualization of CXCR4 receptor (model 2) in pymol.

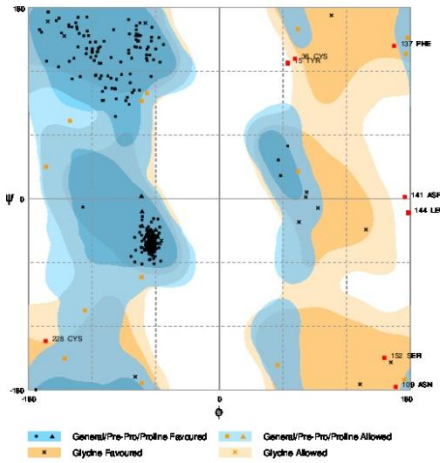

Fig. 4- Ramachandran plot analysis of CXCR4 receptor (model 2).

CCR5 is docked with the phytocompounds in Table 1 [Table 5, Fig. 5].

Table 5: Docking analysis of CCR5

Plant name	Active compound	Docking score (kcal/mol)	Interacting amino acids	Number of interactions	Docking (yes/no)
Azadirachta indica	nimbanene				No
	nimbin	6.626	SER 180 LYS 303 ARG 334	3 1 1	Yes
Vitex negundo	casticin	5.156	TYR 307 THR 167 SER 179 TYR 89	1 1 1 1	Yes
	isoorientin	5.282	GLU 333 SER 337 GLY 111	1 3 1	Yes
Emblica officinalis	punicafolin	7.674	SER 349 GLU 345 SER 63 ARG 140 GLN 277	2 1 1 3 1	Yes
	phyllanemblin	7.590	SER 6 SER 7 ASN 98 SER 169 LYS 171 GLN 4 TYR 3	1 2 1 1 2 1 2	Yes
Aegle marmelos	limonene				No
	germacrene				No

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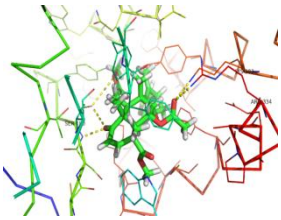
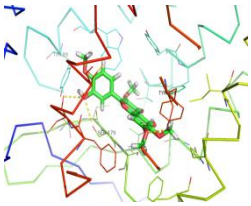
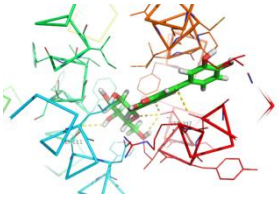
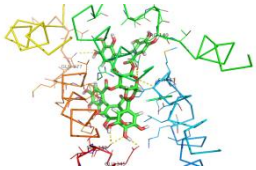
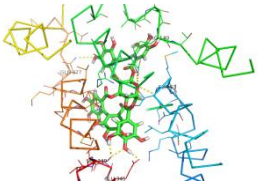
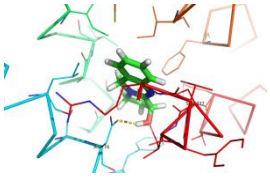
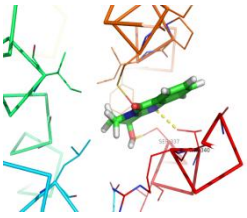
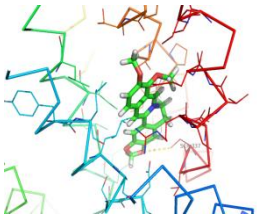
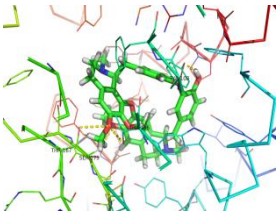
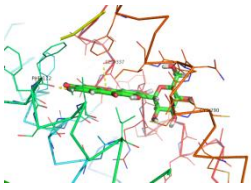
	B				
Adhatoda vasica	vasicine	3.586	SER 337 ASP 76	1 1	Yes
	vasicinone	3.432	THR 340 SER 337	1 1	Yes
Berberis aristata	berberine	5.362	SER 337	1	Yes
	oxycanthine	7.562	TYR 108 SER 179 THR 167 THR 105	1 1 1 1	Yes
Swertia chirata	sawertiamarine	4.570	LYS 303 ARG 334	1 1	Yes
	mangeferin	4.916	CYS 290 PHE 112 SER 337	1 1 1	Yes

	
Nimbin	Casticin
	
Isoorientin	Punicafolin
	
Phyllanemblin	Vasicine
	
Vasicinone	Berberine
	
Oxycanthine	Sawertiamarine

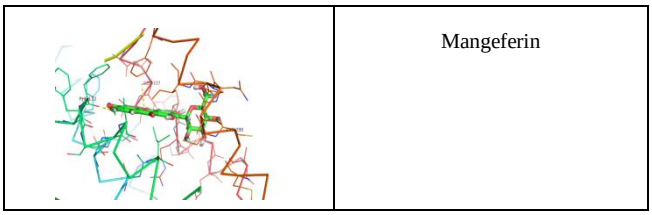


Fig. 5: Docking analysis of CCR5

CXCR4 is docked with the phytocompounds in Table 1 [Table 6, Fig. 6].

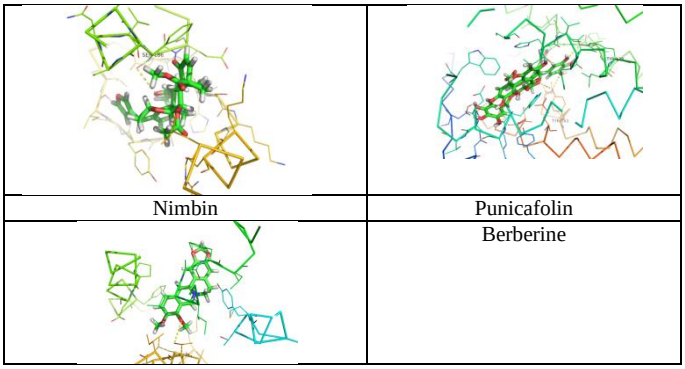


Fig. 6: Docking analysis of CXCR4

Table 6: Docking analysis of CXCR4

Plant name	Active compound	Docking score (kcal/mol)	Interacting amino acids	Number of interactions	Docking (yes/no)
Azadirachta indica	nimbanene				No
	nimbin	6.100	SER 186	1	Yes
Vitex negundo	casticin				No
	isoorientin				No
Emblica officinalis	punicafolin	7.592	THR 126 TYR 263	1 2	Yes
	phyllanemblin				No
Aegle marmelos	limonene				No
	germacrene B				No
Adhatoda vasica	vasicine				No
	vasicinone				No
Berberis aristata	berberine	4.766	THR 249	1	Yes
	oxycanthine				No
Swertia chirata	sawertiamarine				No
	mangeferin				No

IV. CONCLUSION

The CCR5 receptor docks best with punicafolin and phyllanemblin with docking scores of 7.674 kcal/mol and 7.590 kcal/mol respectively and interactions of 8 and 10 respectively.

Also, punicafolin docks best with CXCR4 receptor with docking score of 7.592 kcal/mol and with 3 interactions.

Hence it can be concluded that punicafolin from *Emblica officinalis* can be successfully used as ligand for CCR5 and CXCR4.

In.vitro receptor ligand binding studies can be done further to prove the efficacy of punicafolin as ligand for CCR5 and CXCR4.

REFERENCES

[1] Sepkowitz KA, 2001", AIDS--the first 20 years". *N. Engl. J. Med*. 344 (23): 1764–72

[2] Ghosh TK, 1986, "AIDS: a serious challenge to public health", Journal of the Indian Medical Association, 84(1):29-30

[3] Simoes EA, Babu PG, John TJ, Nirmala S, Solomon S, Lakshminarayana CS, Quinn TC, 1987, "Evidence for HTLV-III infection in prostitutes in Tamil Nadu (India)", Indian Journal of Medical Research, 85:335-8

[4] Panda S. (2002), 'The HIV/AIDS epidemic in India: an overview', in Panda S., Chatterjee A. and Abdul-Quader A.S. (Eds.), 'Living with the AIDS virus: The epidemic and the response in India', p.20

[5] Kumar R, Jha P, Arora P, Mony P, Bhatia P, Millson P, Dhingra N, Bhattacharya M, Remis RS, 2006, "Trends in HIV-1 in young adults in south India from 2000 to 2004: a prevalence study", The Lancet vol. 367:1164-1172.

[6] Alirezaei M, Watry DD, Flynn CF, Kiosses WB, Masliah E, Williams BRG, Kaul M, Lipton SA and Fox HS, 2007, "Human Immunodeficiency Virus-1/Surface Glycoprotein 120 Induces Apoptosis through RNA-Activated Protein

Kinase Signaling in Neurons", The Journal of Neurosciences, 27(41): 11047-11055.

[7] Poumbourios P, Maerz AL and Drummer HE, 2003, "Functional Evolution of the HIV-1 Envelope Glycoprotein 120 Association Site of Glycoprotein 41", 278(43):42149-42160.

[8] Matthee G, Wright AD. and Konig GM, "HIV reverse transcriptase inhibitors of natural origin". *Planta Med.*, 1999, 65: 493–506.

[9] Jung M, Lee S, Kim H and Kim H, "Recent studies on natural products as anti-HIV agents". *Curr. Med. Chem.*, 2000, 7: 649–661.

[10] Yang SS, Cragg GM, Newman DJ and Bader JP., "Natural product-based anti-HIV drug discovery and development facilitated by the NCI developmental therapeutics program". *J. Nat. Prod.*,2001, 64: 265–277.

[11] Cos P, Mees L, Berghe DV, Hermans N, Pieters L and Vlietinck A, "Plant substances as anti-HIV agents selected according to their putative mechanism of action". *J. Nat. Prod.*, 2004, **67**: 284–293.

[12] Boyd MR, Hallock YF, Cardellina JH, Manfredi KP Blunt JW, McMahon JB, Buckheit RW, Bringmann G, Schäffer M, Cragg GM, "Anti-HIV michellamines from Ancistrocladus korupensis", Journal of medicinal chemistry 1994;37(12):1740-5.

[13] Abu-Zeyad R, Khan AG and Khoo C, "Occurrence of arbuscular mycorrhiza in *Castanospermum australe* A Cunn. & C Fraser and effects on growth and production of castanospermine", Mycorrhiza, 1999, 9(2), 111-117.

[14] Singh IP, Bharate SB and Bhutani KK, "Anti-HIV natural products", Current Science, 2005, 89(2): 269-290