IJRAR.ORG E-ISSN: 2348-1269, P-ISSN: 2349-5138

INTERNATIONAL JOURNAL OF RESEARCH AND ANALYTICAL REVIEWS (IJRAR) | IJRAR.ORG

An International Open Access, Peer-reviewed, Refereed Journal

Marine Anti-HIV Drugs

Prajakta Misal*, Dhanashri Nanaware, Smita Jadhav, Shivani Ashtage, Narendra Mangalpawar

Department Of Pharmacognosy

Sahyadri College of Pharmacy, Methwade-Sangola413307, Solapur, Maharashtra, INDIA.

ABSTRACT:

Nature has been crucial in the development of potent medicinal agents. In fact, when considering the development of medicine in relation to the evolution of humankind, terrestrially derived medicines have typically prevailed. More than 70% of the earth's surface is covered by the marine ecosystem, which accounts for 95% of the biosphere. Oceans and sea's contain various Marine Plants, creatures, organisms along with Secondary Metabolites. [1]

HIV-1 reverse transcriptase (HIV-1 RT) is a therapeutic target for the treatment of people with HIV or those who have already developed symptoms of AIDS. In this light, the discovery of novel inhibitors for this enzyme is crucial given the increasing viral resistance to available medications. Upto 2004, there was no any marine drug was approved by FDA, after 2004 Marine drug development has been started. [2]

Keywords: Marine Biosphere, Marine Environment, Anti-retroviral, Drug development, reverse transcriptase.

INTRODUCTION:

In the country as of 2020, there are 81,430 children and 23,18,737 adults who are HIV positive.[3] HIV attacks CD4 Helper T cells of lymphocytes which are related to immune cells, weakening the body's ability to fight against infections like pneumonia, TB well as some cancers. Acquired Immunodeficiency Syndrome (AIDS), the most advanced stage of HIV infection, can take two to 15 years to manifest depending on the person.

One of the top five deadliest diseases in the world is acquired immune deficiency syndrome (AIDS), the terminal stage of a pathogenic disease brought on by the human immunodeficiency virus (HIV). AIDS is a growing epidemic that continues to claim lives every year. 40 million individuals worldwide are now living with HIV or AIDS as a result of the 5 million new HIV infections that occurred in 2001. There have been an estimated 24.8 million deaths worldwide from AIDS and/or linked opportunistic diseases since the epidemic's inception just 20 years ago, and the death rate is not expected to go down anytime soon 3 million people died from AIDS in 2001. [4]

History Of Marine Drugs :

- The colour Tyrian purple, derived from marine molluscus by the Phoenicians in 1600 BC, is the oldest marine product in history. For a long time, the field of marine natural products was dominated by fish and algal metabolites. Marine biopolymers such as agar and carrageenan, vitamins A and D from fish liver oil, and polyunsaturated fatty acids such as eicosapentaenoic acid and docosahexaenoic acid are all examples.

- The discovery of spongothymidine and spongouridineq from the Caribbean sponge Tethya crypta in the 1950s marked the beginning of true marine drug development. Cephalosporin C, a fungus generated by *Acremonium chrysogenum* and identified from Mediterranean Sea water samples near Sardinia in the 1940s, was the catalyst for the invention of the cephalosporin antibiotic class. [5]

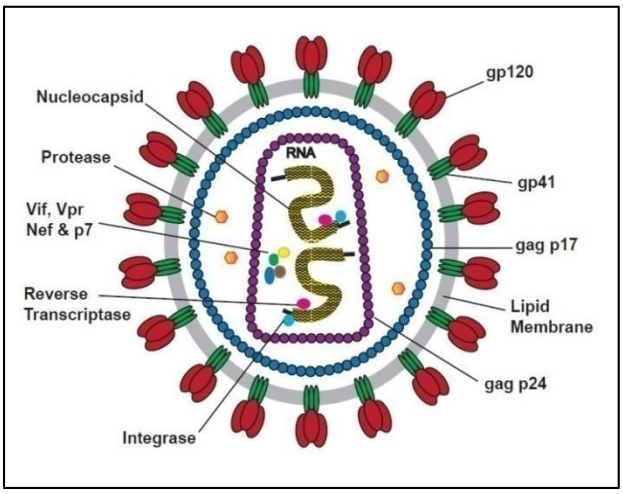


Fig. No. 1: HIV Virus Structure [6]

Marine Drugs used for HIV:

- Phlorotannins
- *Styllissa carteri* (Red Sponge)
- Lectins
- Snake Venom (Phospholipase A2)
- Bioactive Peptides
- Sulphated Polysachharide
- Chitin, Chitosan, Chito-oligosachharide derivatives
- Avarol (sponge)
- Lamellarins (Mollusks)
- Clathsterol (sponge)
- Neamphamide (sponge)
- Dehydrofurodendin (sponge) [7]

1) Phlorotannins:



Fig no. 2: *Durvillaea antarctica* [8]

Phlorotannins are a form of tannin found in brown algae such kelps and rockweeds/sargassacean species, as well as some red algae in smaller amounts. These substances are: oligomers of phloroglucinol, not hydrolysable or condensed tannins (polyphloroglucinols).Tannins have the potential to precipitate proteins, as their name suggests. Some phlorotannins have been discovered to have the ability to oxidise and form covalent connections with proteins.

Three forms of terrestrial tannins (procyanidins, profisetinidins, and gallotannins) did not appear to form covalent interactions with proteins under similar experimental settings. These phenolic compounds are important structural components of brown algal cell walls, but they also appear to serve a variety of other ecological roles, including UV protection and grazing defence.[9]

Phlorotannins are phloroglucinol-based (13,5-trihydroxybenzene) polyphenolic chemicals that make up 5 to 12 percent of the total dry mass of marine brown algae (Phacophyta). The phloroglucinol oligomer polymerization via the polyketide (acetate-malonate) route produces 14 phlorotannins. Phlorotannins with health-promoting biological activity have been found in a variety of marine brown algae-

i. *Hizikia fusiformis*
ii. *Ecklonia cava*
iii. *Laminaria japonica*
iv. *E kurome*
v. *Shige okamurae*
vi. *Sargassum thunbergii.*
vii. *Eisenia bicyclis*
viii. *Undaria pinnatifida*
ix. *Estolonifera*

Phlorotannins are concentrated and prominent in the epidermal cortex of brown seaweeds, but they have also been found linked to the cell walls of marine microalgae. Phlorotannins are hypothesised to aid plant defence by acting as a herbivore deterrent and possibly an appetite suppressant. [10]

They are located in the cell as microscopic vesicles called physodes, where the soluble, polar fraction is sequestered, and as an insoluble structural component of the cell wall. Because they
respond plastically to a range of environmental stimuli, their concentration is known to vary greatly between taxa and geographical areas. Phlorotannins are also produced by brown algae in the nearby saltwater. [11]

Phloroglucinol:

The edible marine brown alga Ecklonia cava (EC) belongs to the Laminariaceae family and has a wide range of bioactivities. NMR methods were used to isolate and describe the bioactive 6,6-hieckol, one of the most important phloroglucinol derivatives found naturally in this species. The antiviral activity of 6,6'-bieckol against HIV-1 caused syncytia formation, lytic effects, and viral p24 antigen production was demonstrated for the first time. Further research revealed that 6,6-hicckol suppressed HIV-1 reverse transcriptase (RT) enzyme activity and HIV-1 entrance with an EC(50) of 1.07 microM. Unlike most other tannins, 6,6'-bieckol showed no cytotoxicity at concentrations that effectively stopped HIV-1 replication. As a result, it's possible that the potentially effective 6,6'-bieckol could be used as a medication for the development of next-generation HIV therapies. [12]

Phloroglucinol compounds are a family of over 700 naturally occurring chemicals with synthetic or semi-synthetic moieties. This is a significant family of natural compounds with the basic component 1,3,5-trihydroxybenzene. These chemicals have a broad range of activities, including anti-HIV, anti-inflammatory, anti-cancer, anti-microbial, anti-allergic, enzyme inhibitory, neuro-regenerative, and antioxidant Antiviral compositions comprising phloroglucinol or its derivatives, either synthetic or derived from plants, as active ingredients or in combination with other antiviral components including AZT, DDC, and HIV protease inhibitors are also known. Compounds containing R1-R6 H alkyl or acyl chains of different lengths, aryl, halogen, nitro, alkylene, alkenylene, and so on are said to have a general formula.

Pro-drugs of phloroglucinol or its derivatives, such as phloroglucinal or derivatives of phloroglucinol coupled to other chemical groups that may improve stability or reduce toxicity. With a pharmaceutically appropriate carrier or diluent, the chemicals can be administered parenterally, orally, rectally, buccally, or transdermally. The chemicals are available in both solid and liquid forms. There are two Japanese patents on phloroglucinol compounds anti herpes virus activity. 2,4-diacetylphloroglucinol, for example, is made by reacting phloroglucinol with the appropriate acid in the presence of BF3. Infections with the herpesvirus can be treated with Et20.Oral or inhalation administration of phloroglucinol substances is possible. [11]

Pro-drugs of phloroglucinol or its derivatives, such as phloroglucinol or derivatives of phloroglucinol coupled to other chemical groups that may improve stability or reduce toxicity. With a pharmaceutically appropriate carrier or diluent, the chemicals can be administered parenterally, orally, rectally, buccally, or transdermally. The chemicals are available in both solid and liquid forms.

2) ***Styllissa carteri*** **(Red sponge)**



Fig.no.3*: Styllissa carteri* (Red sponge) [13]

Marine sponges are a valuable source of natural compounds with powerful biological properties and distinct structural traits. Several sponge-derived chemicals have been successfully transformed into FDA-approved pharmaceuticals for a variety of medicinal uses. Due to the inaccessibility of the Saudi Arabian coast, the bioactive potential of marine sponges un the Central Red Sea has not been thoroughly investigated. Based on their ability to demonstrate phenotypic comparable vities, they have antiviral potential.

Sponges are significant components of marine benthic ecosystems, and they're linked to microbial symbionts that perform ecologically important tasks. In the Red Sea, *Stylissa carteri* is a common species with a low microbiological abundance. The most actively expressed bacterial genes in *S. carteri* are being functionally and taxonomically characterised by researchers. Prokaryotic mRNA was isolated from sponge total RNA, sequenced with Illumina HiSeq, and annotated with the metagenomics Rapid Annotation using Subsystem Technology (MG-RAST) workflow. Synechococcus members have a high expression of archaeal ammonia oxidation and photosynthetic carbon fixation. Stress response and membrane transporters were among the functions that S. carteri symbionts expressed the highest. Surprisingly, gammaproteobacterial symbionts expressed a lot of genes associated to methylotrophy. [14]

Stylissa carteri i.e Elephant Ear sponge is a sponge species that can be found from the Red Sea to Australia. Its robust, yellowish-orange body with numerous spicules grows up to 50 cm and is normally free-standing.[15] Bioactivity-guided isolation supported by LC-HRESIMS metabolic profiling led to the isolation of two new compounds, a ceramide, stylissamide A (1), and a cerebroside, stylissoside A (2), from the methanol extract of the Red Sea sponge Stylissa carteri.[16]

By using mass spectrometry, the existence of probable HIV-1 inhibitory compounds in candidate HIV-1 inhibitory fractions was determined, leading to the identification of three previously identified compounds, namely : i) debromohymenialdisine (DBH)
ii) hymenialdisine (HD)
iii) oroidin.

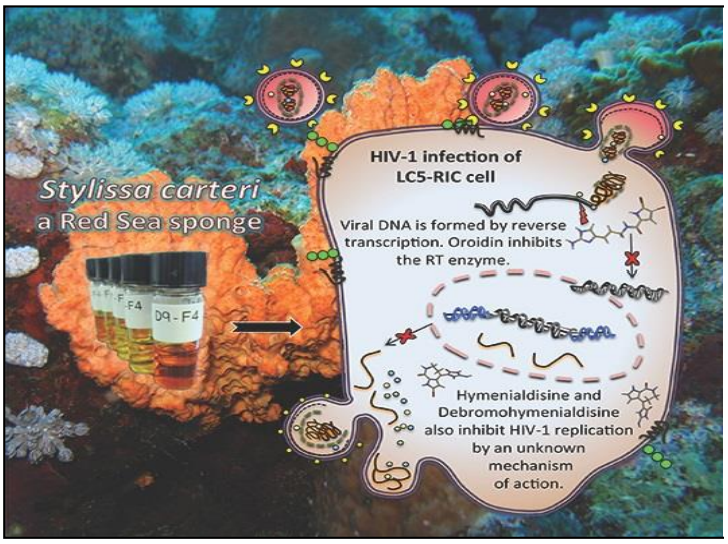


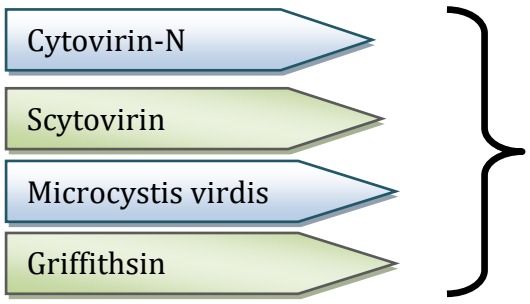
Fig. No.4: Mechanism of Action of *Stylissa cartera* against HIV 1 infection [17]

3) Lectins :

Some lectins have potent antiviral activity against the human immunodeficiency virus (HIV) and other enveloped viruses. As a result, they are highly appealing candidates for the creation of novel antiviral medicines. Lectins that can block HIV and other viruses include cyanovirin-N and griffithsin. Retroviruses such as HIV and many other viruses have envelopes that are made up of virally encoded glycoproteins. Gp41 and gp120, which are extensively glycosylated with glycans on the HIV envelope, are thought to comprise over half of the molecular weight of gp120. Agents that interact strongly and selectively with glycans may disrupt the interface between the host cells and the viral envelope proteins.

Sugar-binding proteins on the viral surface can crosslink with glycans and block other interactions with co-receptors. Antiviral lectins can inhibit viruses from infecting host cells. They keep HIV from infecting you by attaching to the sugars on the envelope's surface. The bulk of currently available antiviral drugs work by interrupting the viral life cycle. Antiviral lectins are suited for topical treatment and have lesser toxicity than other antiviral lectins. Low pH and high temperatures are commonly resistant to antiviral proteins. [18]

*Lectins derived from Marine



4) Snake venom (Phospholipases A2):



Fig. No.5: Marine Venom Snake [19]

Phospholipases family is widely distributed in nature and includes hydrolase enzymes, which are essential for phospholipid metabolism and for the regulation of membrane lipids, membrane composition, signaling, digestion, and inflammation.

Based on the cleavage location in the phospholipid molecule, these proteins are divided into four primary families (A, B, C, and D). Phospholipases A2 (PLA2s) are the best studied of the phospholipases family. These enzymes break down the 2-acyl ester link in 2-sn phospholipids, releasing free fatty acids and lysophospholipids in the process. Eicosanoids (prostaglandins, thromboxanes, prostacyclins, and leukotrienes) are metabolised from free fatty acids (arachidonic acid) and are linked to a variety of physiological and pathological effects, including inflammation and platelet activation.

In the virucidal model, toxins act directly on virus particles before infecting the cell monolayer; in the pre-infection model, uninfected monolayers are pre-treated with toxins before viral infection; and in the post-infection model, cell monolayers are adsorbed with the virus followed by toxin treatment. We summarise a putative mechanism of antiviral action mediated by sPLA2s from snake venom based on this information. sPLA2s have proven to be effective antiviral inhibitors by interfering with many stages of the virus replication cycle, including entrance, replication, and release (as shown in above figure). Current studies have reported that the antiviral action of $sPLA_2$s on steps of viral cycle can occur through a direct action upon viral particle and/or by an interaction with virus or host cell components.

The ability of catalytically-active sPLA2s to cleave glycerophospholipids in the virus lipid envelope is likely associated with their potent virucidal activity against enveloped viruses, and it is reasonable to speculate that sPLA2s also contain domains capable of interacting with viral envelope components, which could lead to viral envelope disruption, exposing viral content (viral inactivation), and compromising the early stages of infection. [20]

Table No.1: sPLA2s from snake venom with antiviral effects- [21]

Sr. No.	Species/ Source	mechanism of action (inhibition)
1.	*Bothrops asper*	Entry (virucidal activity)
2.	*Bothrops jararacussu*	Entry (virucidal activity)
3.	*Bothrops leucurus*	Replication (interfering in host cell components)
4.	*Crotalus durissus terrificus*	Entry (virucidal activity)
5.	*Naja mossambica mossambica*	Entry (virucidal activity)
6.	*Naja nigricollis*	Entry (interfering in host cell components)
7.	*Oxyuranus scutellatus*	Entry (interfering in host cell components)

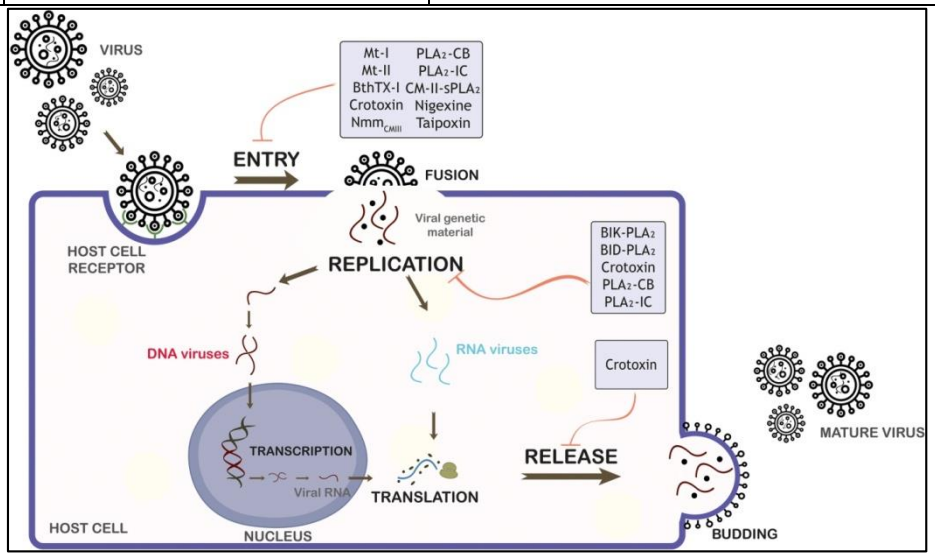


Fig No.6**: Schematic representation of mechanism of action of sPLAs from Snake venom on viral replicative cycle** [22]

5) Oscillatoria agardhii



Fig. No.7: *Oscillatoria agardhii* [23]

Oscillatoria agardhii agglutinin homologue (OAAH) proteins are lectins that were only recently found. When compared to other antiviral carbohydrate-binding drugs, the founding member OAA and a developed hybrid OAAH (OPA) identify comparable but distinct carbohydrate structures in Man-9 (CBAs). The inactivating capabilities of these two recently identified CBAs on HIV multiplication and transmission, as well as their potential as microbicides, were investigated. [24]

Anti-HIV activity Of *Oscillatoria agardhii :*

OAA and OPA block HIV replication, syncytium formation between HIV-1-infected and uninfected T cells, and DC-SIGN-mediated HIV-1 capture and transmission to CD4+ target T cells, rendering a wide range of HIV-1 and HIV-2 clinical isolates non-infectious, regardless of coreceptor usage. Flow cytometry and surface plasmon resonance analyses demonstrate that both CBAs prevent the binding of the Man(1-2)Man-specific 2G12 monoclonal antibody (mAb). These CBAs suppressed the HIV-1 NL4.32G12res, NL4.3MVNres, and IIIBGRFTres strains as well as the wild-type HIV-1 strains. With the exception of OPA/GRFT, combination studies show that OAA and OPA function synergistically with Hippeastrum hybrid agglutinin, 2G12 mAb, and griffithsin (GRFT).[24]

6) Chitin, Chitosan and Chitooligosaccharide Derivatives

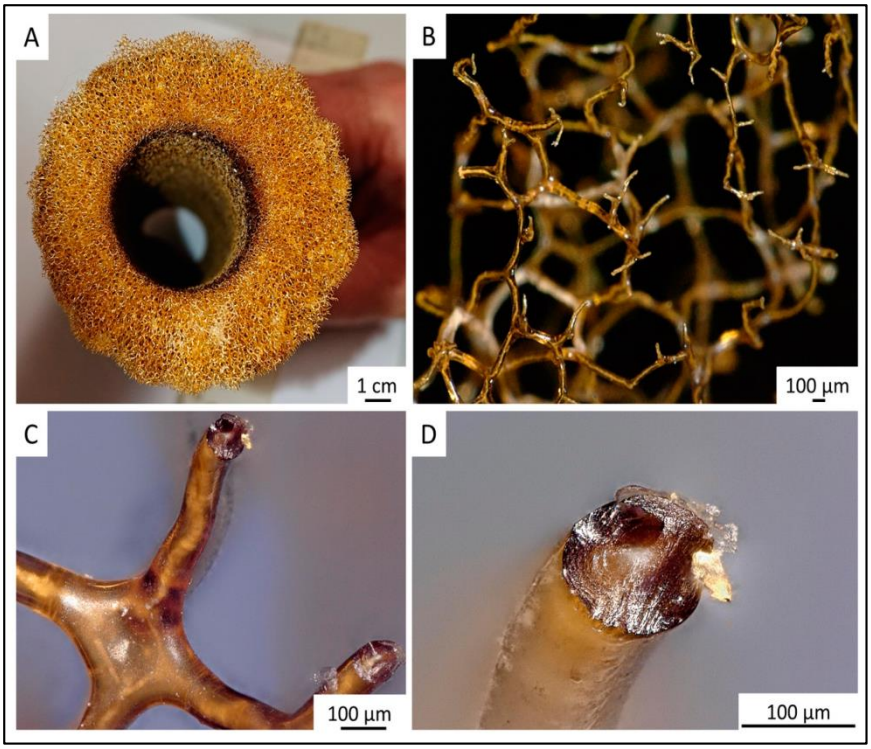


Fig. No.8: 3D Chitin Scaffold Marine Demosponge [25]

Chitin is a long-chain polymer of N-acetylglucosamine that is found in many living species, including insects, fungus, crustaceans, and invertebrates. It's one of the most common polysaccharides, and it's mainly made from crab and shrimp shells. Deacetylation of chitin produces chitosan, which is a partly deacetylated polymer of N-acetylglucosamine.

Chitin and chitosan are chemically modified to produce new biofunctional materials with desirable biological activity and physicochemical qualities. Anti-HIV-1, anti-oxidant, anti-microbial, blood anti-coagulant, and hemagglutination inhibition are among the biological actions of sulfated chitin and chitosan. Furthermore, some of these sulfated chitin and chitosan derivatives have applications in drug administration, metal ion adsorption, and cancer metastasis prevention.

Retrovirus replication has been particularly inhibited by sulfated chitosan derivatives. N-carboxymethyl chitosan-N,O-sulfate has been demonstrated to limit the synthesis of virus-specific proteins and reduce HIV-1 replication in cultured T-cells, as well as Rausher murine leukaemia virus proliferation in cultured mice fibroblasts. The interaction of the viral glycoprotein gp120 (the major viral coat protein) with its receptors on T-lymphocytes was discovered to be blocked by sulfated N-carboxymethyl chitosan. This activity reduced the virus's ability to infect CD4+ cells. Furthermore, the modified CH hindered virus-specific reverse transcriptase activity, most likely by interfering with the enzyme's binding to its polyA-oligo-dT template. These cell cultures revealed almost minimal cytotoxicity when exposed to the chitosan derivative.

The chitosan derivative sulfated at the O2 and/or O3 sites within the glucosamine residues had the maximum activity. In MT-4 cells, this derivative was effective in suppressing HIV-1 replication. An electrostatic interaction between the anionic polymer and the positively charged V3-loop in the gp120 protein prevented the virus from fusing with the cell membrane. Although the ability of anionic chitosan derivatives to inhibit retroviral infection was found to be similar to that of sulfated polysaccharides such as heparin, dextran sulphate, and others, the effectiveness and specificity of the inhibitors was found to be dependent on the position of the sulphate groups in the glucosamine residue.

Sulfated chito-oligosaccharide inhibits viral entry and prevents virus-cell fusion by blocking HIV-1gp120 from interacting to the CD4+ cell surface receptor. As a result, CH and its sulfated derivatives may be useful as antiviral medicines in the future. Gao et al. (2018) explored the anti-HPV activities of 3,6-O-sulfated CH to learn more about the antiviral characteristics of sulfated chitosans (36S). The anti-HPV action of 3,6-O-sulfated CH could be related to targeting the viral capsid protein as well as control of the

host PI3K/Akt/mTOR pathways, according to the researchers.[26]

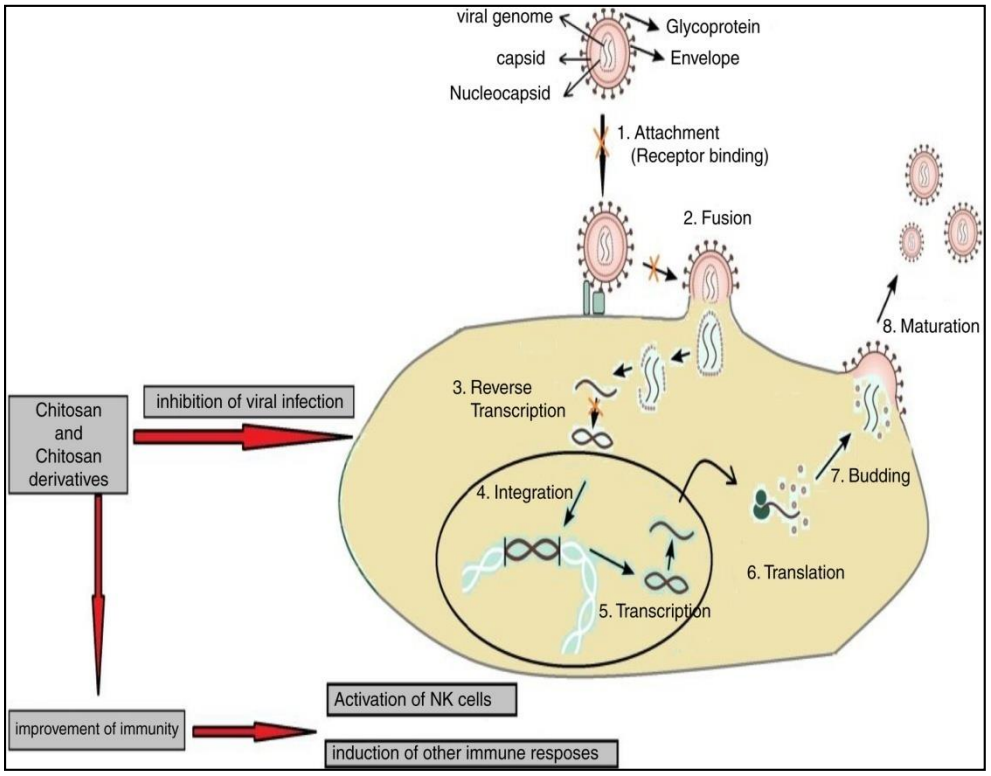


Fig. No.9: Mechanism Of action of Chitin as an anti-viral agent [26]

Chitosan Vehicles for Antiviral Drug Delivery :

Drug delivery systems (DDS) are used to administer drugs in a regulated and sustained manner as well as to specific cells and tissues. The fundamental benefit of these controlled-release systems is that they increase the therapeutic range between the lowest effective and hazardous amounts, reducing drug side effects. By reducing the number of injections, lowering the overall dose, and boosting the specificity for the afflicted region, this strategy can improve patient satisfaction. Nonetheless, DDS has some drawbacks, such as poor biocompatibility and the possibility of toxicity from the matrix or its breakdown products. The delivery route may be challenging, such as requiring a surgical procedure for implantation, and the cost may be considerable when compared to traditional formulations.[38]

Conclusion:

Recently, a great deal of interest has been expressed regarding marine-derived anti-HIV agents such as phlorotannins, sulfated chitooligosaccharides, sulfated polysaccharides, lectins and bioactive peptides. Anti-HIV therapy involving chemical drugs has improved the life quality of HIV/AIDS patients. However, emergence of HIV drug resistance, side effects and the necessity for long term anti-HIV treatment are the main reasons for failure of anti-HIV therapy. Therefore, it is essential to isolate novel anti-HIV therapeutics from marine resources.

References:

1. https://www.google.com/search?q=current+aids+cases+in+india&rlz=1C1UEAD_enIN967IN967&oq=current+Aids&aqs=chrome.0.0i512j69i57j0i512l3j0i20i263i512j0i512l4.7649j0j15&sourceid=chrome&ie=UTF8#:~:text=As%20of%202020%2C%20there%20are%2023%2C18%2C737%20people%20living%20with%20HIV%2C%20including%2081%2C430%20children%2C%20in%20the%20country.

2. Leonardo A. Miceli, Alessandra M.T. deSouza, Carlos R. Rodrigues et.al. HIV-1 Reverse Transcriptase: a potential target for marine products, Brazilian Journal of Pharmacognosy, Aug (2004), 22 (4), https://doi.org/10.1590/S0102-695X2012005000084

3. Karlo Wittine, Lara Saftic, Zeljka Persuric, and Sandra Kraljevic Pavelic et.al. Novel Antiretroviral Structures from Marine Organisms,(MDPI)(2019) Oct; 24(19): 3486.(2019). doi: 10.3390/molecules24193486 PMID: 31561445

4. D.J Gochfeld, K.A El. Sayed, M Yousaf et.al. Sept (2003),(researchgate) Mini Reviews in Medicinal Chemistry, 2003, 3, 451-475

5. Keremkleroo Jym Adil,Chilly Gay Remonde,Esdson Luck Gonzales et.al Biomolecules & Therapeutics (Sci)(Scopus) (PubMed Central);2022:30(4):320-327

6. https://images.app.goo.gl/YQ2ZAN8FNavkZVz16

7. https://images.app.goo.gl/z8CoT7TwPFR4yFMJ9

8. https://images.app.goo.gl/GSeCLnAiadxcSm5ZA

9. Moitinho-Silva L, Seridi L, Ryu T, Voolstra CR, Ravasi T, Hentschel U. Revealing microbial functional activities in the Red Sea sponge Stylissa carteri by metatranscriptomics. Environ Microbiol. 2014 Dec;16(12):3683-98. doi: 10.1111/1462-2920.12533. Epub 2014 Jul 9. PMID: 24920529.

10. Reda F.A. Abdelhameed, Eman S. Habib, fathy A. Mohammed et.al; New Cytotoxic Natural Products from Red Sea sponge Styllissa carteri,(MDPI),Marine. Drugs 2020, 18(5), 241; https://doi.org/10.3390/md18050241

11. O'Rourke A, Kremb S, Bader TM, Helfer M, Schmitt-Kopplin P, Gerwick WH, Brack-Werner R, Voolstra CR. Alkaloids from the Sponge Stylissa carteri Present Prospective Scaffolds for the Inhibition of Human Immunodeficiency Virus 1 (HIV-1). Mar Drugs. 2016 Feb 4;14(2):28. doi: 10.3390/md14020028. PMID: 26861355; PMCID: PMC4771981.

12. Aubrie Elise O'Rourke ,Bioprospecting of Red Sponges for Novel Antiviral pharmacophores,AubrieORourDisseratation,apr(2012),https://repository.kaust.edu.sa/bitstream/handle/10754/552842/Aubrie%200Rour%20Dissertation.pdf?sequence=2

13. https://www.google.com/imgres?imgurl=https%3A%2F%2Fonlinelibrary.wiley.com%2Fcms%2Fasset%2F42f3b04f-f4ac-4028-b41c-8246cd5f80ed%2Fece31511-toc-0001m.jpg&imgrefurl=https%3A%2F%2Fonlinelibrary.wiley.com%2Fdoi%2F10.1002%2Fece3.1511&tbnid=XQwm5o0EMJPHfM&vet=1&docid=ynUOoE8YR7DNfM&w=622&h=415&source=sh%2Fx%2Fim

14. Lucas Moitinho-silva,Loqumane Seridi,Taewoo Rue et. al; Revealing microbial functional activities in the Red Sea sponge Stylissa carteri by metatranscriptomics, 2014 Dec;16(12):3683-98. doi: 10.1111/1462-2920.12533. Epub 2014 Jul

15. https://en.wikipedia.org/wiki/Stylissa_carteri

16. Reda F.A. Abdelhameed, Eman S. Habib, fathy A. Mohammed et.al; New Cytotoxic Natural Products from Red Sea sponge Styllissa carteri,(MDPI),Marine. Drugs 2020, 18(5), 241; https://doi.org/10.3390/md18050241

17. https://www.google.com/imgres?imgurl=https%3A%2F%2Fwww.mdpi.com%2Fmarinedrugs%2Fmarinedrugs00028%2Farticle_deploy%2Fhtml%2Fimages%2Fmarinedrugs-14-00028-ag.png&imgrefurl=https%3A%2F%2Fwww.mdpi.com%2F1660-3397%2F14%2F2%2F28&tbnid=22qYgjB3tp7EAM&vet=12ahUKEwiAv-r5mLr5AhUKKbcAHTe_BwkQMygAegQIARBC..i&docid=pzPZuXcjiQ2BpM&w=550&h=486&q=stylissa%20carteri%20mechanism%20of%20action&ved=2ahUKEwiAv-r5mLr5AhUKKbcAHTe_BwkQMygAegQIARBC

18. Ouafae Akkouh, Tzi Bun Ng, Cuiming Yin et.al, Lectins with Anti-HIV activity; A Review, (MDPI)(PubMed Central), (Molecules) 2015 Jan; 20(1): 648–668; Published online 2015 Jan 6. doi: 10.3390/molecules20010648.

19. https://images.app.goo.gl/zPKurkBbvSrKUQCLA

20. S.C.Texiera,B.C.Borges et.al;Insights into the antiviral activity of phospholipases A_2 (PLA_2s) from snake venoms; Int J Biol Macromol. 2020 Dec 1; 164: 616–625.Published online 2020 Jul 19. doi: 10.1016/j.ijbiomac.2020.07.178

21. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7368918/table/t0005/?report=objectonly

22. https://images.app.goo.gl/emWE8KC7rBrtiaVK7
23. https://www.sciencephoto.com/media/16192/view/blue-green-alga-oscillatoria
24. Geoffrey Férir, Dana Huskens, Sam Noppen, Leonardus M. I. Koharudin, Angela M. Gronenborn,et.al; Broad anti-HIV activity of the *Oscillatoria agardhii* agglutinin homologue lectin family; Journal of Antimicrobial Chemotherapy, vol(69), Issue 10, October 2014, Pages 2746–2758, https://doi.org/10.1093/jac/dku220
25. https://images.app.goo.gl/g3JuK6PJLN3cRt1n9
26. Nisrein Jaber,Mayyas Al-Remawi,Faisal Al-Akayleh,Phillip J. Collier et.al;.review of the antiviral activity of Chitosan, including patented applications and its potential use against COVID-19; First published: 04 July 2021; https://doi.org/10.1111/jam.15202