International Standard Serial Number (ISSN): 2249-6807

International Journal of Institutional Pharmacy and Life Sciences 5(2): March-April 2015

INTERNATIONAL JOURNAL OF INSTITUTIONAL PHARMACY AND LIFE SCIENCES

Pharmaceutical Sciences **Review Article……!!!**

Received: 24-03-2015; Revised: 29-03-2015; Accepted: 30-03-2015

HAART TO NANO MEDICINE IN HIV TREATMENT

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Keywords:

HIV/AIDS, Antiretroviral therapy, Nanomedicine , Nanotechnology, Drug delivery

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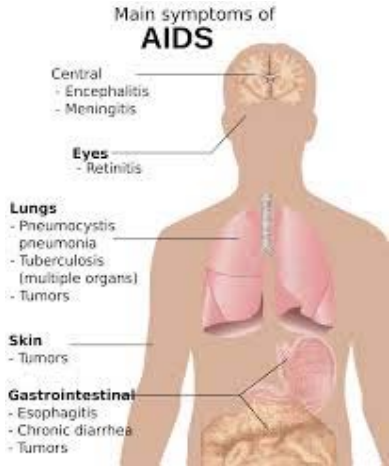
ABSTRACT

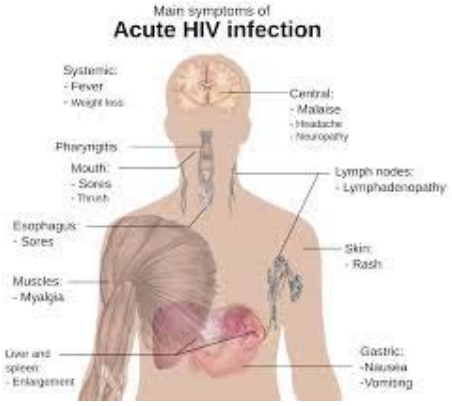
HIV/AIDS is a global pandemic and is the leading infectious cause of death among adults. Although antiretroviral (ARV) therapy has dramatically improved the quality of life and increased the life expectancy of those infected with HIV, life-long suppressive treatment is required and a cure for HIV infection remains elusive. Furthermore, preventative measures such as a vaccine or microbicide are urgently needed to curb the rate of new infections. This article reviews the potential for the multidisciplinary field of nanotechnology to advance the fields of HIV treatment and prevention. Nanotechnology is an emerging multidisciplinary field that is revolutionizing medicine in the 21st century. It has a vast potential to radically advance the treatment and prevention of HIV/AIDS. Here we discuss the challenges with the current treatment of the disease and shed light on the remarkable potential of nanotechnology to provide more effective treatment and prevention for HIV/AIDS. Indeed, a lot of assignments left behind for researchers to overcome the challenges hindering the wider application of nanomedicines in treatment of HIV/AIDS. The introduction of highly active antiretroviral therapy (HAART) in 1996 has transformed a lethal disease to a chronic pathology with a dramatic decrease in mortality and morbidity of AIDS-related symptoms in infected patients. However, HAART has not allowed the cure of HIV infection, the main obstacle to HIV eradication being the existence of quiescent reservoirs. Besides these new strategies aiming to eliminate the virus, efforts must be made to improve current HAART.

INTRODUCTION

Human immunodeficiency virus (HIV)/acquired immunodeficiency syndrome (AIDS) is a global pandemic and is the leading infectious disease resulting in significant morbidity and mortality and consequently devastating socioeconomic effects. With the advent of multidrug, highly active antiretroviral therapy (HAART), the prognosis for HIV-infected patients has significantly improved; however, it has not eradicated HIV infection, particularly in sequestered, anatomically privileged sites, such as the brain, testes, gut, liver, kidney, and secondary lymphoid tissue. Additionally, emergence of resistant viral strains and the adverse side effects associated with prolonged use continue to slow down the application of effective antiviral therapies.



A diagram of a human labeled with the most common symptoms of AIDS (figure 1)[2]



A diagram of a human labeled with the most common symptoms of an acute HIV(figure 2)[2]

Nanotechnology is an emerging multidisciplinary field that has the potential to advance the treatment and prevention of HIV/AIDS radically. The use of nanotechnology for numerous biomedical applications has become an area of intense research over the last decade.[1]

The emergence of AIDS was first reported in 1981 followed by the identification of HIV as the cause of the disease in 1983 [1-4]. HIV/AIDS is now a global pandemic that has become the leading infectious killer of adults worldwide [5]. By 2006, more than 65 million people had been infected with the HIV virus worldwide and 25 million had died of AIDS [6]. At the end of 2007, around 33 million people were living with the virus, with 2.7 million new infections and 2 million deaths each year [7]. This has caused tremendous social and economic damage worldwide, with developing countries, particularly Sub-Saharan Africa, heavily affected.

A cure for HIV/AIDS has been elusive in almost 30 years of research. Early treatments focused on anti retroviral drugs that were effective only to a certain degree. The first drug, zidovudine, was approved by the US FDA in 1987, leading to the approval of a total of 25 drugs to date, many of which are also available in fixed-dose combinations and generic formulations for use in resource-limited settings (to date, only zidovudine and didanosine are available as true generics in the USA) [8,9]. However, it was the advent of a class of drugs known as protease inhibitors and the introduction of triple-drug therapy in the mid-1990s that revolutionized HIV/AIDS treatment [10,11]. This launched the era of highly active antiretroviral therapy (HAART), where a combination of three or more different classes of drugs are administered simultaneously [11]. The use of the HAART regimen, particularly in the developed world, has resulted in tremendous success in improving the expectancy and quality of lives for patients [12]. However, some HAART regimens have serious side effects and, in all cases, HAART has to be taken for a lifetime, with daily dosing of one or more pills. Some patients also develop resistance to certain combinations of drugs, resulting in failure of the treatment. The absence of complete cure under current treatment underscores the great need for continued efforts in seeking innovative approaches for treatment of HIV/AIDS.

In addition to treatment, the best way to fight global infections is through preventive strategies, vaccines being the most effective agents. Vaccines have historically been very effective at controlling other major infectious diseases such as measles, mumps, rubella and polio, with smallpox completely eradicated. There have been enormous efforts to develop a safe and effective vaccine for HIV/AIDS. However, the pursuit has been very daunting so far, with recent failures of clinical trials for major candidate vaccines [13-15]. This has raised a debate over which path to take in HIV/AIDS vaccine research. Despite this debate, it is clear

that novel approaches for identifying new antigens and adjuvants as well as better delivery systems are necessary. Another preventive strategy that has been under investigation is the development of effective intravaginal microbicides that can be used by women. There has been remarkable progress in the understanding and design of technologies for microbicide development. However, recent clinical trials failed to show efficacy, indicating the need for more research and development to design better systems [16,17].

The complexity of HIV/AIDS as a disease: challenges to complete eradication of the virus from the body:

If left untreated, HIV infection is associated with very high viral load in the body leading to progressive fall in immune cells particularly CD4+ T cells. This can be interrupted by treatment with highly active antiretroviral therapy (HAART), which should contain at least three drugs regimens made up of at least two classes of antiretroviral agents. However, the great challenge is that immediately after initial infection, this virus is able to establish reservoirs where it escapes from the effect of drugs and keeps releasing the viral progeny to the blood as long as the patient lives. This makes it one of the chronic and lifelong diseases especially with the introduction of HAART. There are two types of viral reservoirs within tissues that serve this role. The anatomical reservoirs; these are tissues inaccessible to optimal levels of antiviral drugs due to due to the presence of barriers, such as the blood–brain barrier (BBB), blood-cerebrospinal fluid barrier, and blood-testes barrier. and the others are cellular reservoirs; cells in which this virus remains latent and hence escaping the action of antivirals due to the presence of efflux proteins such as P-glycoprotein and multidrug resistance protein on the cell surface preventing the drugs from attaining therapeutic intracellular concentrations [10,11].

Dendritic cells within lymphoid tissue trap a large number of extracellular virions on their surface to protect virus from antiretroviral drugs. On top of this, latently infected CD4+ T cells help the HIV to persist despite the presence of effective antiretroviral therapy as it is not replicating at this stage. Last but not least, monocytes/macrophages that are specifically found in brain, pulmonary alveoli, spleen and lymph nodes are relatively long-lived cells since HIV has very low cytopathic effects on them making them a persistent reservoir of HIV regardless of the presence of highly active antiretroviral therapy [11-13]. Therefore, it is the existence of these persistent and stable reservoirs for the virus that makes it difficult to efficiently eradicate HIV from the body even with the advent of HAART. This is due to the

fact that these drugs in free form have poor local bioavailability and low residence time in these reservoirs when administered systemically [14] highlighting the need for new drug or delivery system with the potential of averting these problems so as to achieve a cure from HIV/AIDS.

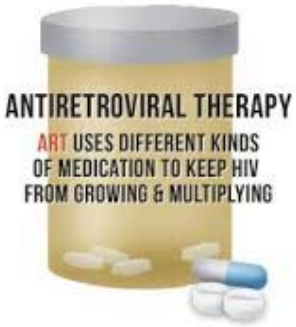
The need to advance HIV/AIDS clinical therapies:

The HIV/AIDS epidemic is one of the major public health threats especially in sub-Sahara countries. Globally, about 35.3 million people were living with HIV in 2012 which is an increase from previous years as more people are receiving the life-saving anti retroviral therapy. The prevalence of HIV/AIDS continues to increase and it is expected that over 90 million people will ultimately be infected in Africa alone. On the other hand, there were 2.3 million new HIV infections and 1.6 million AIDS deaths in 2012 globally. In 2012, 9.7 million people in low- and middle-income countries received antiretroviral therapy, representing 61% of all who were eligible under the 2010 World Health Organization (WHO) HIV treatment guidelines. However, under the 2013 WHO guidelines, the HIV treatment coverage in low- and middle-income countries represented only 34% of the 28.3 million people eligible in 2013. Anti retroviral therapy not only prevents AIDS-related illness and death: it also has the potential to significantly reduce the risk of HIV transmission and the spread of tuberculosis. From 1996 to 2012, anti retroviral therapy averted 6.3 million AIDS-related deaths worldwide, including 5.2 million deaths in low- and middle-income countries [18].

However, despite the clear advantages of HAART in management and prevent of HIV, there are several significant shortcomings. Significant drug interactions, additional and or synergistic toxicity as a result of combination, low adherence rate secondary to pill burden and frequent dosing, and potential for development of drug resistant virus which is even more difficult to treat. An additional important issue is that upon discontinuation of treatment or when resistance develops, even with HAART, the viral load rebounds in the blood [19]. Lastly, systemically available drug needs to cross biological barriers for delivery to cellular and anatomical sites which most currently available anti retroviral formulations couldn't. CNS availability of most anti-retroviral agent is very low due to poor permeability across the blood–brain barrier. The consequence of these interdependent processes is insufficient concentrations and very short residence time of the anti-retroviral agents at the cellular and anatomical sites [20,21].

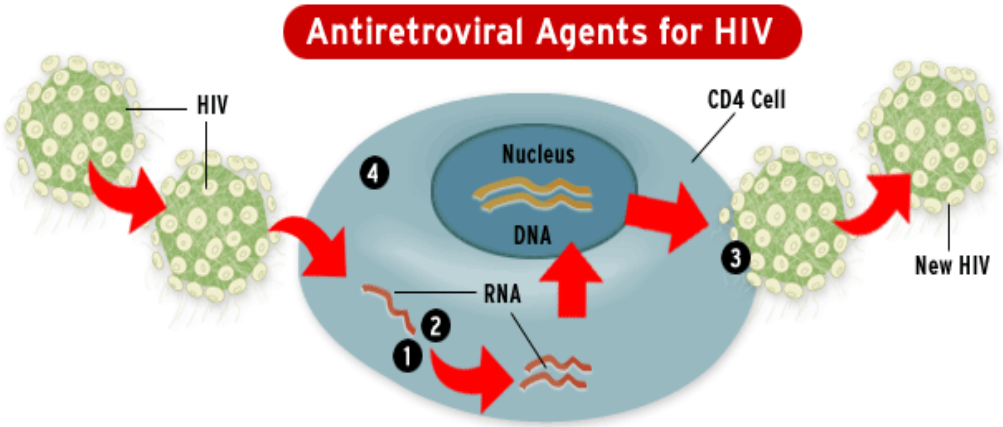
Current HIV/AIDS Treatment:

The current state-of-the-art treatment modality for HIV/AIDS is HAART, where three or more antiretroviral drugs are given to patients simultaneously. The drugs used in combination are in most cases from different classes that work based on different mechanisms. Despite the remarkable successes with the current HAART treatment for HIV/AIDS, there are still various challenges remaining. The major difficulty has been the failure of the treatment, typically due to poor patient compliance [23]. Due to the need to take the medication daily for a lifetime, patients fail to adhere to the treatment schedule, leading to ineffective drug levels in the body and rebound of viral replication [22,24,25].



Overview Of HIV Treatments (figure 3)[24]

Moreover, in some patients, the virus develops resistance to particular combinations of drugs even with good adherence. Drug resistance is mainly caused by the high genetic diversity of HIV-1 and the continuous mutation it undergoes [26]. This problem is being addressed with individualized therapy, whereby resistance testing is performed to select a combination of drugs that is most effective for each patient [26]. In addition, side effects due to toxicities of the drugs are also a concern. There are reports that patients taking HAART experience increased rates of heart disease, diabetes, liver disease, cancer and accelerated aging [27]. Most experts agree that these effects could be due to the HIV infection itself or co-infection with another virus, such as co-infection with hepatitis C virus resulting in liver disease. However, the toxicities resulting from the drugs used in HAART could also contribute to these effects.

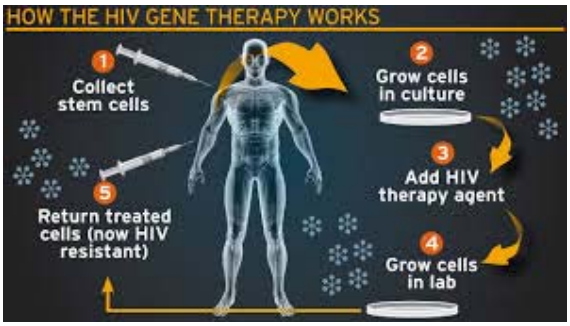


Anti retroviral Therapy For HIV/AIDS(figure 4)[25]

Under current treatment, complete eradication of the virus from the body has not been possible. The major cause for this is that the virus resides in 'latent reservoirs' within memory $CD4^+$ T cells and cells of the macrophage–monocyte lineage [25,27]. A major study recently found that, in addition to acting as latent reservoirs, macrophages significantly contribute to the generation of elusive mutant viral genotypes by serving as the host for viral genetic recombination [28]. The cells that harbor latent HIV are typically concentrated in specific anatomic sites, such as secondary lymphoid tissue, testes, liver, kidney, lungs, gut and the CNS [29–31]. The eradication of the virus from such reservoirs is critical to the effective long-term treatment of HIV/AIDS patients. Therefore, there is a great need to explore new approaches for developing nontoxic, lower-dosage treatment modalities that provide more sustained dosing coverage and effectively eradicate the virus from the reservoirs, avoiding the need for lifetime treatments.

Gene Therapy For HIV/AIDS:

In addition to improving existing antiretroviral therapy, there are ongoing efforts to discover alternative approaches for treatment of HIV/AIDS [31,32]. One promising alternative approach is gene therapy, in which a gene is inserted into a cell to interfere with viral infection or replication. Other nucleic acid-based compounds, such as DNA, siRNA, RNA decoys, ribozymes and aptamers or protein-based agents such as fusion inhibitors and zinc-finger nucleases can also be used to interfere with viral replication [33].



In order for human cells to make the HIV-inhibitory protein in the laboratory, he had to insert a new gene - a process known as gene therapy.(figure 5)[35]

Early efforts in gene therapy for HIV/AIDS have been focused on viral vectors as the delivery agents with various clinical trials in progress [34,35-38]. In one of these studies, Benitec Ltd and City of Hope are collaborating in an ongoing clinical trial to study the safety and feasibility of a gene therapy strategy based on the combination of three different inhibitory genes in a single lentiviral vector that utilizes stem cells in the delivery process [36]. Recently, scientists from UCLA reported that a Phase II gene therapy clinical trial showed that cell-derived gene transfer is safe and biologically active in HIV-infected individuals [37]. These efforts are encouraging and support the growing excitement around gene therapy for the treatment of HIV/AIDS. However, lessons learned over the past two decades indicate that the use of viral vectors for gene delivery poses fundamental problems such as toxicity, immunogenicity, insertion mutagenesis and limitations with scale-up procedures [39,40]. These problems have encouraged the investigation of nonviral vectors for gene delivery, where nanotechnology platforms are showing great promise [40-42].

In recent years, the Nobel prize-winning discovery of RNA interference (RNAi) in 1998 by Fire, Mello and colleagues has gained much attention in the clinical therapeutics field and is generating billion dollar investments in therapeutic applications [43,44]. Ongoing clinical trials for the treatment of age-related macular degeneration and respiratory syncytial virus have provided data that are creating tremendous excitement in the field [44]. RNAi is also considered to have therapeutic potential for HIV/AIDS [31,32,45]. Gene silencing is induced by double stranded siRNA, which targets for destruction the mRNA of the gene of interest. For HIV/AIDS, RNAi can either target the various stages of the viral replication cycle or various cellular targets involved in viral infection such as CD4, CCR5, and/or CXCR4, the major cell surface co-receptors responsible for viral entry. HIV replicates by reverse transcription to

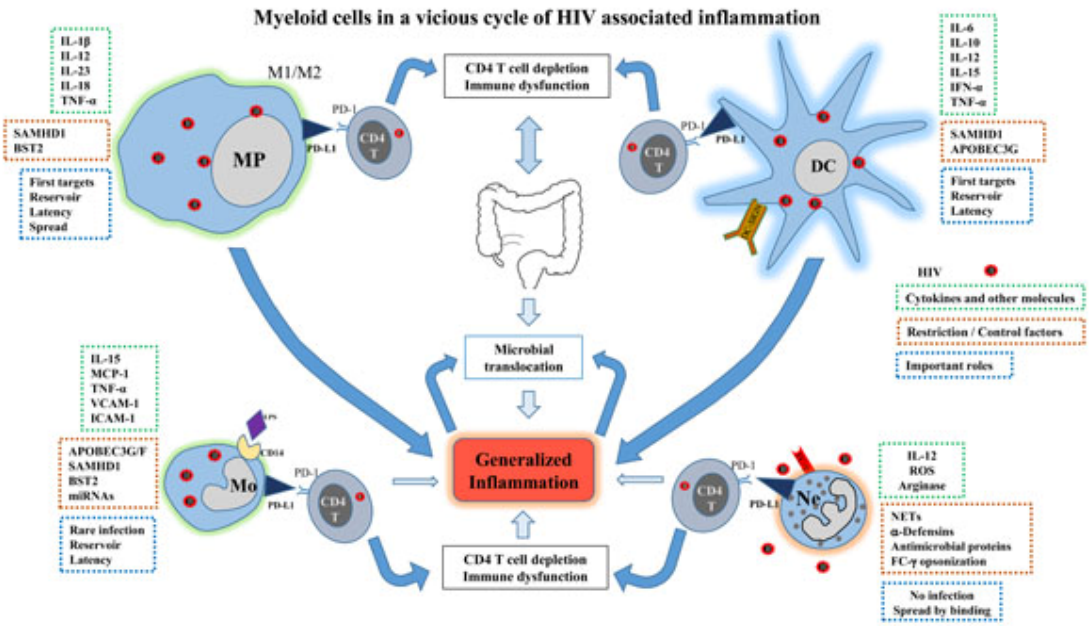
form DNA and uses the DNA to produce copies of its mRNA for protein synthesis; siRNA therapy could be used to knock down this viral mRNA.

As with other gene therapy techniques, delivery of siRNA to specific cells and tissues has been the major challenge in realizing the potential of RNAi [44]. New nanotechnology platforms are tackling this problem by providing nonviral alternatives for effective and safe delivery. The first nontargeted delivery of siRNA in humans via self-assembling, cyclodextrin polymer-based nanoparticles for cancer treatment have recently entered Phase I clinical trials [45].

Although at an early stage, nonviral delivery of siRNA for treatment of HIV infection is also gaining ground. A fusion protein, with a peptide transduction domain and a double stranded RNA-binding domain, was used to encapsulate and deliver siRNA to T cells ***in vivo*** [46]. CD4- and CD8-specific siRNA delivery caused RNAi responses with no adverse effects such as cyto-toxicity or immune stimulation. Similarly, a protamine-antibody fusion protein-based siRNA delivery demonstrated that siRNA knockdown of the ***gag*** gene can inhibit HIV replication in primary T cells [47].

Immunotherapy For HIV/AIDS:

The various treatment approaches described above focus on treating HIV/AIDS by directly targeting HIV at the level of the host cell or the virus itself. An alternative approach is immunotherapy aimed at modulating the immune response against HIV. $CD8^+$ cytotoxic T-cell responses to acute HIV infection appear to be relatively normal, while neutralizing antibody production by B cells is delayed or even absent [48]. Over time, viral mutation leads to loss of the $CD8^+$ T cell cytotoxic function. However, the major effect of an infection by HIV is the loss of $CD4^+$ T cells. These 'helper' T cells are responsible for a number of supportive functions for other immune populations and their loss leads to profound immunosuppression, manifested by the presence of dysfunctional B-cells, natural killer cells and the macrophages in chronically HIV-infected patients [48]. In recent years, there has been increasing interest in the therapeutic use of immune responses to restore the regular function of the immune system as an effective way to treat HIV/AIDS [49-51]. There has been increasing evidence that the immune system is capable of controlling HIV in certain individuals [49]. Hence, strategies to rebuild or allow the reconstitution of immune function could be one of the best approaches for effective treatment.



Immune Suppression by Myeloid Cells in HIV Infection: New Targets for Immunotherapy (figure 6)[50]

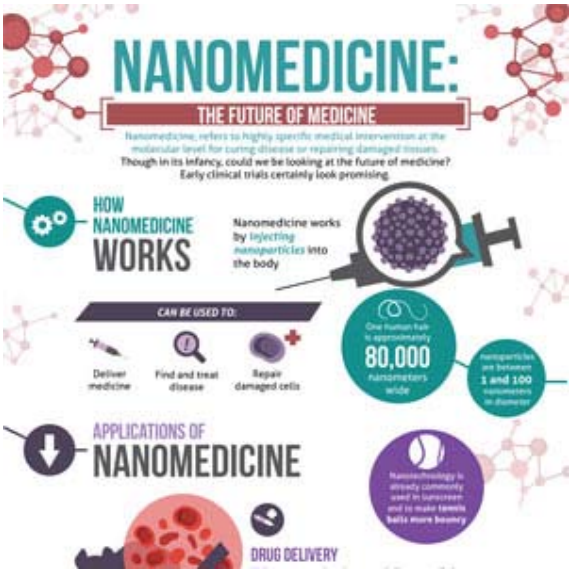
Immunotherapy is a treatment approach involving the use of immunomodulatory agents to modulate the immune response against a disease. Similar to vaccines, it is based on immunization of individuals with various immunologic formulations; however, the purpose is to treat HIV-infected patients as opposed to protect healthy individuals (preventive vaccines will be discussed in an upcoming section). The various immunotherapy approaches for HIV/AIDS could be based on delivering cytokines (such as IL-2, IL-7 and IL-15) or antigens [52,53]. The development of cellular immunity, and to a large degree humoral immunity, requires antigen-presenting cells (APCs) to process and present antigens to $CD4^+$ and $CD8^+$ T cells. Dendritic cells (DCs) are the quintessential professional APCs responsible for initiating and orchestrating the development of cellular and humoral (antibody) immunity [54,55]. Protein/peptide antigens or DNA immunogens (which lead to endogenous protein expression) could then be delivered through viral vectors to endogenous or ***ex vivo***-generated DCs.

Nanotechnology approaches in HIV/AIDS management:

Nanotechnology is a new discipline of science and engineering that is advancing many areas of medicine. It involves the understanding, design, engineering and fabrication of materials at the atomic and molecular level. The National Nanotechnology Initiative defines nanotechnology as the study of structures with roughly 1–100 nm in size in at least one dimension but structures up to several hundred nanometers are also considered under

nanotechnology applications [28]. The application of nanotechnology to medicine, commonly referred to as nanomedicine, involves the use of nanoscale materials for preventive, therapeutic and diagnostic purposes [29]. There have been major advances in nanomedicine over the last few decades, particularly in cancer diagnosis and therapy [30-32]. Although at an earlier stage, applications of nanotechnology for prevention and treatment of HIV/AIDS have also gained attention in recent years. There are emerging novel approaches in which nanotechnology can enhance current treatment as well as advance new therapeutic strategies, such as gene therapy and immunotherapy. Moreover, some nanomaterials have therapeutic effects by themselves. Nanotechnology can also play a major role in preventive strategies for developing vaccines and microbicides.

Nano-medicine is a term implying the application of nanotechnology (the technology that uses nanosized particles) for therapy and diagnosis of diseases. Nanoparticles have improved pharmacokinetics and tissue distribution of therapeutic agents there by diminishing toxicity by their preferential accumulation at the target site. In addition, they improve therapeutic potential of drugs by facilitating intracellular delivery and prolonging their retention time either inside the cell or in blood circulation [56].



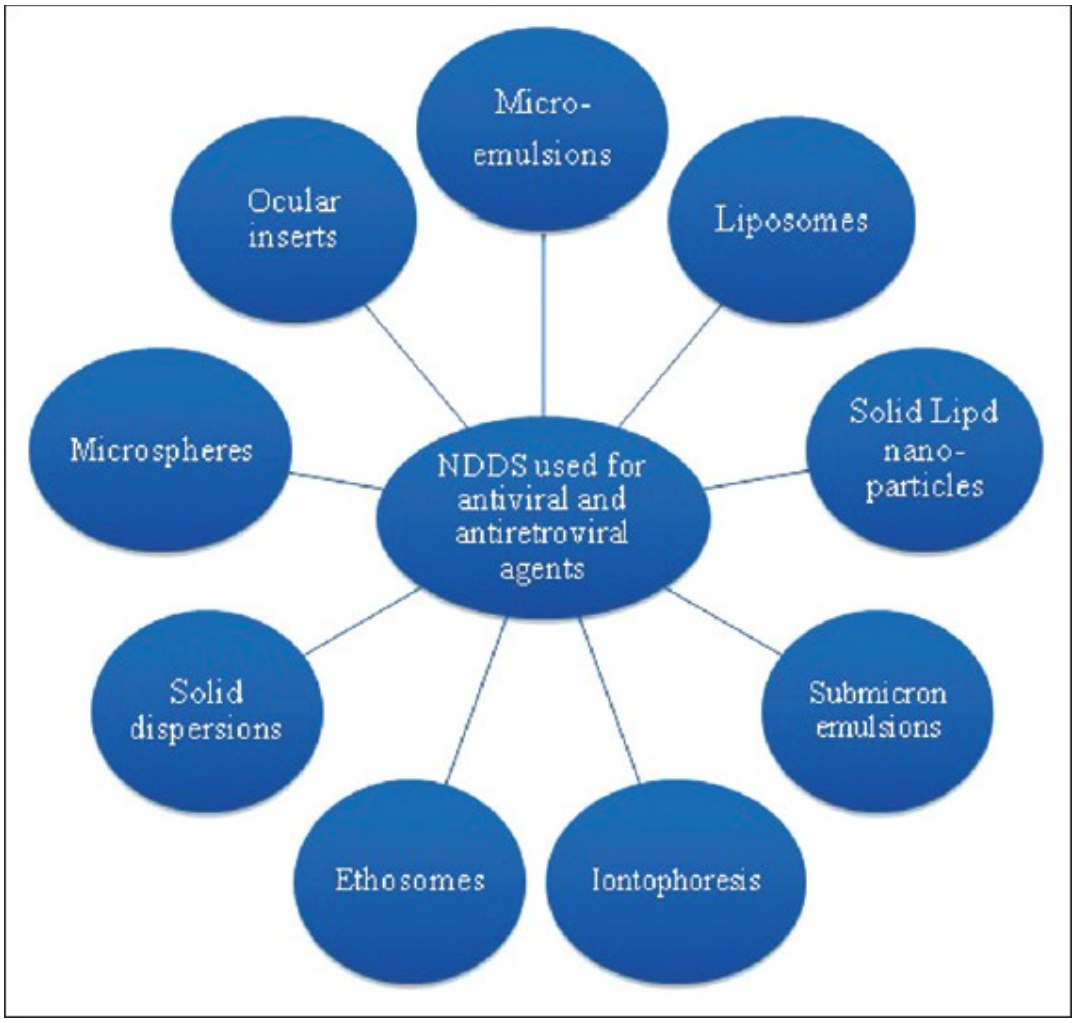
NanoMedicine: Application of Nanotechnology in Medicine(figure 7)[56]

In general, nanomedicines particularly polymer based drug delivery systems are highly fascinating and hence attracting the attention of scientists from different corners of the world. This is especially true for the diagnosis, prevention and treatment of intracellular infections like hepatitis, tuberculosis and HIV/AIDS [57] which can be considered as a triple plague in

developing countries like Ethiopia. The underlined fact for this is that one can easily manipulate their properties like molecular weight to adapt to the drug delivery requirements.

Nanotechnology for antiretroviral drug delivery:

The use of nanotechnology platforms for delivery of drugs is revolutionizing medicine in many areas of disease treatment [58]. Cancer patients have been the biggest beneficiaries of this revolution so far, with significant advances in the last few decades. Many nanoscale systems for systemic cancer therapy are either FDA approved or in clinical trials [43,59]. This tremendous success has been due to the unique features that nanotechnology imparts on drug delivery systems. Using nanotechnology, it has become possible to achieve improved delivery of poorly water-soluble drugs, targeted delivery of drugs to specific cells or tissues and intracellular delivery of macromolecules [38,56].



Anti retroviral Agents in Treatment Of HIV/AIDS (figure 8)[59]

Nanotechnology-based platforms for systemic delivery of antiretroviral drugs could have similar advantages. Controlled-release delivery systems can enhance their half-lives, keeping them in circulation at therapeutic concentrations for longer periods of time. This could have major implications in improving adherence to the drugs. Nanoscale delivery systems also enhance and modulate the distribution of hydrophobic and hydrophilic drugs into and within different tissues due to their small size. This particular feature of nanoscale delivery systems appears to hold the most promise for their use in clinical treatment and prevention of HIV. Specifically, targeted delivery of antiretroviral drugs to $CD4^+$ T cells and macrophages as well as delivery to the brain and other organ systems could ensure that drugs reach latent reservoirs [49,54]. Moreover, by controlling the release profiles of the delivery systems, drugs could be released over a longer time and at higher effective doses to the specific targets.

Nanomaterials as therapeutic agents:

In addition to being used as delivery agents, nanomaterials have also been shown to have therapeutic effects of their own. Studies have shown that the capsid of HIV could be a target for structure-based drug design for inhibiting viral replication [60,61]. As a result, both computational and experimental studies have identified compounds that could inhibit the assembly of the HIV capsid. Various nanomaterials have been found to inhibit viral replication ***in vitro*** and it is suggested that these effects are based on structural interference with viral assembly.

Various fullerene (C-60)-based structures, dendrimers and inorganic nanoparticles, such as gold and silver, have been shown to have anti-HIV activity ***in vitro.*** [50-60] While these efforts have not yet progressed beyond ***in vitro*** studies, they illustrate the potential of therapeutic nanomaterials to inhibit HIV replication.

Vaccine delivery:

The search for a safe and effective HIV/AIDS vaccine has been challenging in the almost three decades since the discovery of the disease. Recently, high-profile clinical trial failures have prompted great debate over the vaccine research, with some suggesting the need for a major focus on fundamental research, with fewer efforts on clinical trials [33-35,61].

HIV vaccine (figure 9)[61]

Nanoparticles have potential as adjuvants and delivery systems for vaccines. Over the past few decades, controlled-release systems have been used for sustained release of various agents. This has great advantages for vaccine delivery since the release of antigens in a controlled manner could lead to a prolonged and stronger initiation of the immune response. By protecting the delivered antigen from body fluids (e.g., lymph, serum and mucus) nanoparticle antigen encapsulation can increase the half-life of an immunizing antigen. Nanoparticles can also be designed to effectively target APCs [62]. Targeting antigen delivery to DCs with surface-functionalized nanoparticles presents a major opportunity for delivery of antigen and initiation of immune responses. Another major benefit of nanoparticle vaccines is that they can be optimized for various routes of administration. Conventional vaccines are mostly administered intramuscularly, but nanoparticles provide expanded opportunity for oral and nasal vaccinations where mucosal immunity could be induced [63].

Various lipid-based systems have been investigated for HIV/AIDS vaccine delivery. In an earlier study, nasal immunization of mice with the HIV gp160 protein encapsulated in a liposome induced high titers of gp160-specific neutralizing antibody responses [64]. The liposomes were made from a mixture of cholesterol, sphingomyelin, phosphatidylethanolamine, phosphatidylcholine and phosphatidylserine. The HIV gp 41 protein was also delivered through a variety of liposomes (110–400 nm) eliciting strong antibody responses in mice and rabbits [65-67]. To date, there have been no vaccines that can elicit a broadly neutralizing antibody response to HIV; therefore, it is doubtful that an antibody response to a monovalent encapsulated antigen alone will be enough for a sterilizing HIV vaccine [66]. These nanodelivery systems therefore need to be improved to include a variety of HIV epitopes and potentially even epitopes engineered to enhance access to and generation of antibody responses to areas of the HIV glycoproteins not elicited by acute HIV infection [66].

Intravaginal microbicides:

Although vaccines that induce sterilizing immunity are the most ideal way to prevent the spread of HIV/AIDS, other approaches are also being pursued until a safe and effective vaccine is developed. Since sexual transmission is the major route of infection, prevention methods aimed at behavioral changes as well as the use of personal protection such as condoms have helped in reducing the spread of the disease in some countries [67]. However, effective protection methods that can be utilized by women have not been readily available, making women more vulnerable to the disease. Among people infected with HIV/AIDS, women account for nearly 50% of infections worldwide and 60% in Sub-Saharan Africa [67]. As a result, there are major efforts focused on developing effective microbicides for HIV/AIDS prevention. Microbicides are preventive agents that are topically applied into the vagina to prevent the transmission of HIV/AIDS or other sexually transmitted diseases.

Vaginal microbicides are being designed in many forms (figure 10)[67]

There are currently over 50 drug candidates in preclinical development and 12 candidates in clinical trials for use as microbicides [66]. These microbicide candidates work by different mechanisms that either target the virus or inhibit viral binding to the target cell. Most of the current microbicides in development are based on gels formed from anionic polymers and polysaccharides, whereas microbicides based on antiretroviral drugs have also gained attention [66,67,68]. However, similar to the difficulties faced in vaccine development, major candidate microbicides for HIV/AIDS failed in recent efficacy clinical trials suggesting the need for further studies and new approaches [36,37]. Here, we discuss the most recent nanotechnology-based approaches that focus on using dendrimers, siRNA and nanoparticles in microbicides for HIV/AIDS.

VivaGel is a microbicide gel formed from the L-lysine dendrimer that has a polyanionic outer surface developed by the company Starpharma [68]. After comparing the antiviral effects of various dendrimers based on L-lysine, poly(amido amine) and poly(propylene imine), the

product SPL7013, based on the dendrimer L-lysine, was identified as the best microbicide candidate [68]. In earlier works, SPL7013 applied as a topical microbicide in female pigtailed macaques showed a dose-dependent resistance to viral challenge [69]. Recently, it was shown in a Phase I safety trial that the dendrimer solution is safe in humans [65].

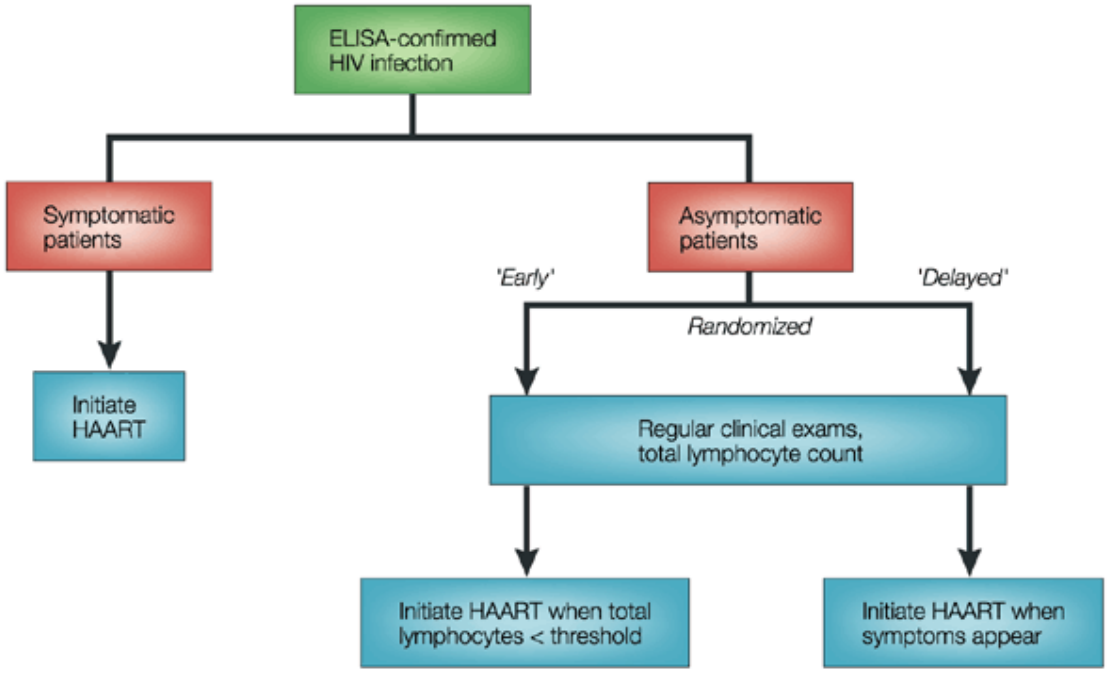
Challenges to widely apply nanomedicine for HIV/AIDS therapy:

Even though, nanomedicines are the promising future of HIV/AIDS prevention and treatment, several hurdles remain unresolved, including but not limited to toxicity, unwanted biological interactions and the difficulty and cost of large-scale synthesis of nanopharmaceuticals [39]. Another challenge is the fact that targeted delivery of antiretroviral drugs using nano polymers to viral reservoir sites may lead to HIV drug resistance. This could be because of two reasons. Firstly, the targeted delivery of an antiretroviral drugs which if not accompanied by systemic HAART administration will lead to suboptimal doses of the drug in non-targeted tissues, with the potential to select out drug resistant mutations there.

Furthermore, most studies involving nanocarriers use a single antiretroviral drug, which would effectively select out resistant virus in targeted tissues [48,57] highlighting the need to combine at least three drugs for use with nanocarriers as in conventional HAART in future researches.

Why is it important to improve HAART

There are several reasons why HAART should be improved. One is the existence of a residual viraemia in patients undergoing HAART. The origin of this viraemia is still debated. There are two theories explaining this residual viraemia: (i) long-lived cells containing latent HIV provirus that can produce HIV at low levels following reactivation; and (ii) low-level cryptic on-going replication despite therapy. Latency is best described as a lack of proviral gene expression. In contrast, on-going replication requires continuous viral gene expression without cytopathic effects. Ineffective treatment in cells supporting on-going replication could result from poor drug penetration into sanctuaries such as the brain, where infected microglial cells are located,[70] or from cell-to-cell transfer of the virus.

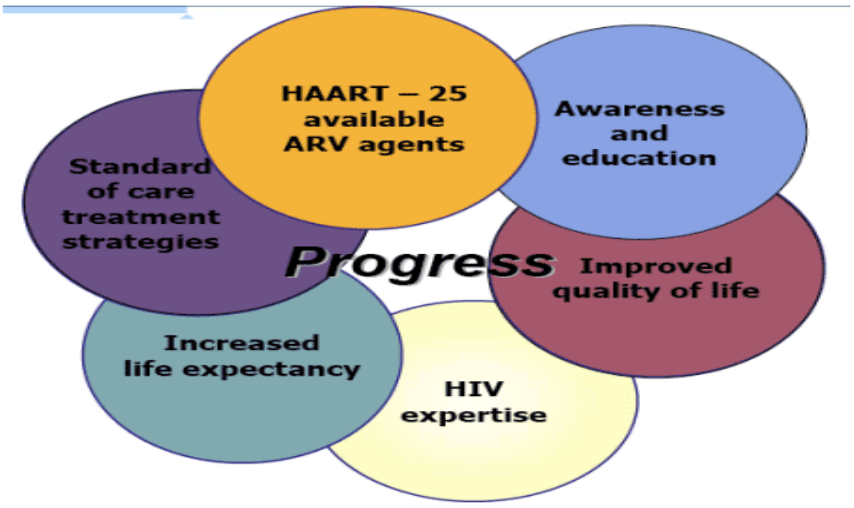


All symptomatic HIV-infected participants would be treated with highly active antiretroviral therapy (HAART) (figure 11)[69]

It is important to distinguish between these two theories, since the therapeutic approaches they suggest are essentially different. The theory of on-going replication suggests that drug resistance to treatments might develop. In this case treatment intensification and the design of new anti HIV-1 molecules are needed in the long term. On the other hand, if viruses are released in bursts from stable reservoirs, multidrug resistance does not develop, however, HAART alone is ineffective as well. Several studies have looked at the efficiency of such intensification of HAART on residual viraemia and only one failed to reduce it.[71] The second reason to improve HAART is related to the 'shock and kill' strategy discussed above. HAART by itself is not able to achieve a cure, but is still needed (to kill) in association with HIV reactivation from quiescent cells (to shock). Finally, emergence of drug resistances, toxicity and compliance with treatment are all obstacles to the current management of HIV-1 infection and therefore need improvement of HAART.[72]

How can we improve HAART:

Current management of HIV-1 treatment is based on seven classes of antiretrovirals: nucleoside/nucleotide reverse transcriptase inhibitors (NRTIs), nucleotide reverse transcriptase inhibitors (NtRTIs), non-nucleoside reverse transcriptase inhibitors (NNRTIs), protease inhibitors (PIs), entry/fusion inhibitors (EIs), coreceptor inhibitors (CRIs) and integrase inhibitors (INIs).[72] The therapy of HIV-1-infected patients is based on a combination of three or more drugs from two or more classes.[73] We believe that new drugs should target other steps of the HIV-1 cycle such as transcription, since there is no drug currently available targeting this step. An increasing number of studies suggest that inhibitors of cellular LTR-binding factors, such as NF-κB and Sp1, repress LTR-driven transcription.[74] Recently it has been shown that proteins of the DING family are good candidates to repress HIV-1 gene transcription. Indeed, the inhibitory effect of the human DING protein named HPBP (human phosphate binding protein) on HIV-1 replication is very strong, even compared with other canonical drugs currently used in HAART.[75] HPBP is also a potent anti-HIV-1 drug in peripheral blood lymphocytes and in primary macrophages, which is not true for several other anti-HIV-1 drugs. Very interestingly, HPBP, which targets transcription, is as effective against drug-resistant HIV strains as wild-type strains, highlighting the potential therapeutic advantage of HPBP. Moreover, such drugs could also be used to cope with chronic inflammation, which leads to non-AIDS events.[75] We believe that this protein or its derivatives are potentially interesting molecules and deserve further study. As suggested for X-DING-CD4,[72] proteins belonging to the DING protein family might have a role in the innate response to infections, including HIV-1.



Perspectives in HIV Drug Resistance (figure 12)[73]

Finally, the use of nanotechnology involving structures 1–100 nm in size is an exciting approach since it will make it possible to reduce toxicity and facilitate treatment adherence. Indeed, these nano-delivery systems will permit: (i) modulation of drug release; (ii) protection of drugs from metabolism; and (iii) specific targeting of infected cells, even those located in sanctuaries. In corollary, this approach will allow improved bioavailability and therefore reduce toxicity.[76] Among new nanotechnology-based drug delivery systems are liposomes, polymeric micelles, dendrimers and nanosuspensions.

CONCLUSIONS

In this review, we have gone through many works done on the application of nano-polymers for the treatment and prevention of HIV infections. This paper showed that nanopolymers of different quality are designed and evaluated for delivery of ARVs. Of all, surprising thing is the fact that some polymers like dedrimers have an inherent antiviral activity beside their carrier roles which further makes them more appropriate to use them for delivery of ARVs in medicine.

In this article, we have also discussed very promising features for HIV therapy such as improving residence time of the drugs at the targeted sites, several fold increase in the uptake of the drugs to those previously less accessible viral reservoir tissues, improved antiviral efficacy and significantly reduced toxicity of antiretroviral drugs. These findings clearly suggest the hope that HIV can be completely eradicated from the body through the application of nano-polymers for delivery of ARVs in the near future.

However, a lot of assignments left behind for researchers to overcome the challenges hindering the wider application of nanomedicines in treatment of HIV/AIDS. Beside this, clinical trials involving the use of nano-polymers to deliver HAART regimens should be designed to investigate both the beneficial and drawbacks of this technology.

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