2.1 Introduction

Acquired Immuno Deficiency Syndrome (AIDS) is caused by the Human Immuno Deficiency Virus (HIV), which passed on from primates into humans. Although isolated cases of infection in people might have appeared earlier, the first case of the current epidemic probably occurred in the year 1930, and the disease spread rapidly in the 1970’s. The prime targets of HIV are the T cells (CD4+T helper cells) leading to the collapse of the immune system of the body that results in death. Very early stages of HIV just after infection resemble the flu or other viral infections. This follows a latent stage (often years) with no symptoms and then an early AIDS stage (also often years) with various symptoms, many of them non-specific and not easy for diagnosis. AIDS becomes much
AIDS is an illness that damages a person’s ability to fight off disease, leaving the body open to attack from ordinarily innocuous infections and some form of cancer. HIV is the causative agent responsible for AIDS. This virus infects certain types of white blood cells, primarily CD4 cells and monocytes/macrophages. The HIV infects a person initially but does not produce any manifest symptoms of illness for a long period. The HIV once having entered the body of an uninfected person from infected, remains inside the receptor CD4 cells for a random period. This period is called the incubation period and according to [48] Fusaro et al. (1989) the incubation period is the time interval between the point of infection to the time point to onset of clinical symptoms of AIDS. The average incubation period is estimated to be around 8 to 10 years for adults, and afterwards the infected person develops the AIDS symptoms. The time interval between the manifestation of AIDS symptoms and the death of the individual is also a matter of concern and many researchers have investigated this aspect.

HIV is a member of the retrovirus family. It is a genetically diverse population of viruses that is responsible for causing AIDS worldwide. Two major types of HIV have been identified so far, namely, HIV-1 and HIV-2. The first type, i.e., HIV-1 is the worldwide epidemic and is commonly referred to be HIV. It is a highly notorious virus, mutating readily in human body and destroys certain white cells that are essential to the human
immune system. The second type, i.e., HIV-2 is much less pathogenic and occurs rarely, indicating that the period between initial infection and illness is longer. It is observed mostly in West Africa HIV-1 and HIV-2 are two types of viruses transmitted in the same way and are associated with similar possible infections and AIDS.

In 1993, the Center for Disease Control and Prevention (CDC) revised the AIDS case definition to include HIV - infected adults and adolescents who had a CD4+T cells count less than 200 cells/mm3 or whose CD4+T cells constituted less than 14 per cent of total lymphocytes. In 1993, the CDC also generated a comprehensive classification system that consists of a matrix of CD4+T cells status (levels 1, 2 and 3) and clinical manifestations (categories A, B and C). This classification system became effective on January 1 st, 1993 and is still in use (see Table 2.1.1).

Table 2.1.1: Center for disease control and prevention classification system for HIV infection

<table>
<thead>
<tr>
<th>CD4+T Cells Categories</th>
<th>Clinical Categories</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>A (Asymptomatic, first-degree HIV or PGL)</td>
</tr>
<tr>
<td>Category 1:</td>
<td></td>
</tr>
<tr>
<td>≥ 500 cells/mm³</td>
<td>A1</td>
</tr>
<tr>
<td>Category 2:</td>
<td></td>
</tr>
<tr>
<td>200 - 499 cells/mm³</td>
<td>A2</td>
</tr>
<tr>
<td>Category 3:</td>
<td></td>
</tr>
<tr>
<td>&lt; 200 cells/mm³</td>
<td>A3⁺</td>
</tr>
</tbody>
</table>

The World Health Organization recognizes four stages in HIV disease progression.
Stage one is asymptomatic infection, when the CD4 count is normally greater than 500 per \( mm^3 \) of blood. Stage two is when the count is between 350 and 499 per \( mm^3 \), and the symptoms might include some mild weight loss, fungal infections, and herpes zoster (shingles). When the CD4 cell count falls below 350, in stage three, a person has advanced immunosuppression with opportunistic infections, fever, severe weight loss, diarrhea, and possibly TB. Stage four, AIDS occurs when there are fewer than 200 CD4 cells per \( mm^3 \) of blood and the person is seriously ill with diseases such as TB which may spread beyond the lungs, Pneumocystis carinii and other pneumonias, the parasitic disease toxoplasmosis, and meningitis. A few people may experience symptoms of disease with CD4 counts above 200, while others may show no symptoms with CD4 counts below 200. Generally, infections will increase in frequency, severity, and duration until the person dies. The CD4 count is one of the measures used by physicians in deciding when to begin drug therapy.

### 2.2 Structure of HIV

HIV is a member of lentivirus family of retroviruses. When viewed under the electron microscope, HIV appears as spherical particles that are approximately 110 nm in diameter, with knob-like projections on the surface of the virus and a cone shaped viral core. HIV particles contain two types of an RNA genome, each of which is approximately 10,000 base pairs in length and encodes nine genes. Two different HIV species have been identified, namely, HIV-1 and HIV-2.
Figure 2.2.1: Structure of HIV

The outer coat of the virus, known as the viral envelope, is composed of two layers of fatty molecules called lipids, taken from the membrane of a human cell when a newly formed virus particle buds from the cell. Each viral particle contains 72 glycoprotein complexes, which are integrated into this lipid membrane, and are composed of trimmers of an external glycoprotein gp120 and a transmembrane spanning protein gp41. The bonding between gp120 and gp41 is only loose and therefore gp120 may be shed spontaneously within the local environment. Glycoprotein gp120 can be detected in the serum as well as within the lymphatic tissue of HIV-infected patients. The matrix protein p17 is anchored to
the inside of the viral lipoprotein membrane. The p24 core antigen contains two types of HIV-1 RNA. The HIV-1 RNA is part of a protein-nucleic acid complex, which is composed of the nucleoprotein p7 and the reverse transcriptase p66. The viral particle contains all the enzymatic equipments that are necessary for replication: a reverse transcriptase, an integrase p32 and a protease p11.

After the virus has entered the cell, the HIV reverse transcriptase enzyme converts the single-standard HIV RNA into DNA. Through a complex process, some of the viral DNA migrates into the nucleus, and the viral enzyme splices the viral DNA into the host cell DNA. Once HIV DNA becomes integrated into the human DNA, it is referred to as proviral DNA. Subsequently, the host can transcribe proviral DNA into messenger RNA (mRNA), a process controlled by the interaction of the promoters and enhancers in the viral genome with the Tat protein and cellular factors. Posttranscriptional processing of the viral mRNA takes place in the host cell nucleus. The viral mRNA is transported into the host cell cytoplasm; once there, viral mRNA is translated into viral proteins. The late stage of viral replication involves both the assembly of the viral particles, with each viral core incorporating two copies of the viral RNA genome and the budding, and release of the virus from the surface of the cell.
2.2. STRUCTURE OF HIV

2.2.1 Pathogenesis of HIV

HIV is a retrovirus, which has only RNA. The infection of HIV occurs through binding of the virus to a specific cellular receptor namely, the CD4+ molecule. The CD4+ T cells plays a central role in organizing immune responses. This is the very cell that HIV has chosen as its target of attack. It appears that HIV remains intracellular in these CD4 cells and creates a mucus membrane around itself, so that the macrophages are unable to identify the antigen and neutralizing its viral effect. The HIV once having entered the CD4 molecule starts the replication, which is the process of reproduction. In doing so it uses the DNA of the host cell. However, the newly produced HIV’s have the properties of the HIV itself.

The process of reproduction lasts for a random period, which is known as the latency period during which the assembly and replication of HIV takes place. After the latency period is over the newly produced HIV’s come out from the host cell and infects the other CD4 cells. It is true that the HIV need not wait till the latency period is over to come out of the host CD4 cells, killing the same. There is every possibility that a sufficient amount of HIV can come out of the host cell before the latency period itself. This can be found out by the presence of antibodies. So the very moment of identification of the antibodies, it is an indication of the seropositive nature of the affected individual, which means the seronegative period is over. Hence in some cases the seronegative period and the latency period may be the same.

Antigenic variation seems to be a major property of many parasitic infections. In
response to the ability of immune system to attack foreign antigens, parasites have evolved the capacity to escape immunological surveillance by mutating their immunodominant epitopes continuously during the time of an infection. The immune response is thus continually confronted with new targets. No sooner has the immune system generated cellular or humoral attack against these targets than the parasite has escaped with mutated antigens. This antigenic variation with the subsequent development of neutralizing antibodies to each emergent sub-population of the pathogen is a common feature of lentivirus infections in particular.

The various viral proteins associated with HIV act as antigens, that is foreign to the immune system. The host's immune mechanism is activated to produce antibodies to fight these antigens. Within two to four weeks after the infection, viral particles and or the major core antigen could be detected in the blood. During this phase the antibodies are detectable. This initial phase of HIV infection without manifest antibodies but with the virus transiently present in the blood is known as ‘window period’. The time interval between the point of infection to the time at which the antibodies specific to HIV are detected in the blood is called the ‘seronegative period’ or ‘latency period’. The onset of production of antibodies in the infected is referred to as ‘seropositive period’. The process of changeover from seronegative state to seropositive state is called seroconversion.

It may be noted that CD4 count is the number of CD4 cells in a cubic millimeter of blood. CD4 cells help to protect people from getting infections. HIV attacks and destroys
CD4 cells. The CD4 count in a healthy, HIV negative adult is usually 600-1200 CD4 cells per cubic millimeter of blood. The CD4 count of most people with HIV usually falls over time. If the CD4 count drops below 200 cells per cubic millimeter of blood, there is a high risk of serious infection.

2.3 Spread or Transmission of HIV/AIDS

The risk of HIV transmission is present, if an HIV-negative person comes into contact with the blood, semen or vaginal fluids of an HIV positive source person. HIV is found in all body fluids of an infected person, although in minimal quantities in sweat, tears and saliva. Exposure to blood or blood products carries the maximum risk of infection. This is why there is so much concern around blood safety and hygiene in health care settings, and why there are high levels of transmission among drug users who share syringes. However, sexual intercourse is the most common source of transmission: 75-85 percent of people are infected this way. This includes both homosexual and heterosexual intercourse, though globally heterosexual intercourse predominates. The virus can be passed from infected mothers to their infants by crossing the placenta, during the birth process, and through breast milk. Reducing the risk prior to or during birth is simple: in most resource-poor settings, the drug nevirapine is used, which lowers mother-to-child transmission between 8% and 17%.
2.4 Modes of Transmission

HIV infection spreads through the following four modes.

- Unprotected hetero/homo sexual contacts
- Infected blood transfusion
- Sharing of unsterile needles for drug abuse
- Prenatal transmission.

2.4.1 Unprotected Hetero / Homo Sexual Contacts

The majority of HIV infections are acquired through unprotected sexual contacts. HIV can enter the body through the mucous membranes that line the vagina, rectum, urethra, and possibly, on rare occasions, the mouth. Damage to a mucous membrane may increase the risk of transmission of HIV, but is not necessary for transmission to occur. Almost all cases of sexually transmitted HIV have been caused by vaginal intercourse without condom. In the United States and Europe, most cases of sexually transmitted HIV infection occur during homosexual contact, which is between two people of the same gender. In Africa, Asia and other parts of the world, HIV is transmitted primarily through heterosexual contact that is between two people of opposite genders.
2.4. MODES OF TRANSMISSION

2.4.2 Infected Blood Transfusion

Spread of HIV through blood occurs through specific identifiable practices and in specific identifiable situations. They include blood transfusion, sharing of blood and other skin-piercing instrument without proper cleaning and sterilization. Blood taken from one person with the infection and given to another person for medical treatment, would lead to infection to the latter. The tattooing and piercing the patients caused by the health care workers like doctors and nurses are of high-risk and such practices would lead to infection by HIV. Moreover HIV can spread in health care settings through accidental needle sticks or contact with contaminated fluids.

2.4.3 Sharing of Unsterilized Needles for Drug Abuse

Sharing unsterilized injection equipment that has been previously used for some one who is infected is an important route of HIV transmission in many countries with a high prevalence of intravenous drug users. HIV frequently spread among drug users who share needles or syringes that are contaminated with blood from an infected person. It is rare, however, for a patient to give HIV to a health care worker or vice-versa by accidental sticks with contaminated needles or other medical instruments.
2.4.4 Prenatal Transmission

HIV can be transmitted from an infected woman to her fetus during pregnancy and during delivery. This is referred to as vertical or perinatal transmission. Antiretroviral therapy used at the appropriate time in pregnancy significantly reduces the risk of transmission from mother to fetus. Additionally, using certain safe methods for delivery also helps to reduce transmission. Since breast milk can transmit HIV, avoiding breastfeeding further reduces vertical transmission.

2.5 The Different Stages of HIV Infection

HIV infection can generally be broken down into four distinct stages

- Primary infection
- Asymptomatic infection
- Symptomatic HIV infection
- Progression from HIV to AIDS

2.5.1 Primary Infection

HIV is disseminated to the brain, central nervous system and lymphatic tissue (lymph nodes, spleen, tonsils, and adenoids). Lymphatic tissue is the major reservoir of HIV.
Ten to thirty days after infection, about 80% to 90% of people develop what is called “acute retroviral syndrome” or “primary HIV infection”. This is an illness that resembles the flu and usually lasts about one to two weeks. Symptoms include fever, swollen glands, sore throat, faint rash that generally starts in the torso, sores on the mouth and sometimes around the anus, weight loss, and muscle or joint pain.

During the first two to three months of HIV infection, viral load may be high and the CD4 count drops below normal. After a few months, the CD4 count generally rises close to normal levels and viral load drops. Viral load stabilizes at about 3 to 9 months to what is known as a viral “set point”. A higher viral set point and more severe acute retroviral syndrome symptoms are considered predictors of more rapid progression to AIDS.

2.5.2 Asymptomatic Infection

Asymptomatic infection refers to an infection without symptoms. Many people with HIV may have few or no signs or symptoms of the disease for up to 10 years. However, some people may progress much faster, seeing their CD4 cells decline within a few years and experiencing symptoms in the first few years after infection.

During asymptomatic period, the only evidence of HIV infection may come from lab tests. Blood tests may show lower-than-normal numbers of CD4 cells and moderate levels of HIV. The amount of HIV in the blood is usually called the viral load. Although the immune system is able to fight HIV, it cannot get rid of the virus completely. Despite
2.5. The Different Stages of HIV Infection

lack of symptoms, HIV disease is progressing. On average, CD4 cells decline at a rate of approximately sixty points per year, while viral load gradually increases.

2.5.3 Symptomatic HIV Infection

This stage is said to be ‘Early and medium stage of HIV infection’. In this third stage, the immune system becomes so damaged by HIV that the symptoms begin to appear. The symptoms, mild at first, become more severe. The infection takes advantage of the vulnerable state of the immune system and affects almost all the systems of the body. After some years, a variety of medical symptoms may develop, often involving skin and gastrointestinal disorders. Viral load continues to rise and the CD4 count shows a more accelerated decline of about 1.5 to 2 years before development of AIDS - defining illness.

2.5.4 Progression from HIV to AIDS

HIV infection and a specific group of diseases or conditions are indicative of severe immunosuppression related to infection with the human immunodeficiency virus. This is the terminal stage of the disease; and it represents the irreversible break down of the immune mechanism; and this leads to the susceptibility of the patient to wide variety of opportunistic infections and malignancies. Without treatment, it appears that the majority of HIV infected people will develop AIDS within ten to fifteen years after being infected, though some people who have been infected longer than this remain healthy even without
2.6 Prevention

As there is no cure for AIDS, prevention of HIV infection becomes extremely important in controlling the disease. Efforts to prevent the spread of AIDS include:

(1) Restricting sexual activity to a single partner and practicing safer sex. Besides avoiding the risk of HIV infection, condoms are successful in reducing other sexually transmitted diseases and unwanted pregnancies. Before engaging in a sexual relationship with someone, getting tested for HIV infection is recommended.

(2) Avoiding needle sharing among intravenous drug users.

(3) It is mandatory to screen the blood before planned major surgery to prevent risk of infection from a blood transfusion, although blood supplies are extremely safe in the developed world.

(4) Practicing universal safety precautions when handling body fluids or needles. Healthcare professionals, first responders, and teachers, for example, are now trained in these precautions.

(5) Testing for HIV infection by anyone suspected of infection. If treated aggressively and early, the development of AIDS may be postponed. If HIV infection is confirmed, it
can be tested and receive medical attention.

2.6.1 HIV Test

A Human Immunodeficiency Virus (HIV) test detects antibodies to HIV or the genetic material (DNA or RNA) of HIV in the blood or another type of sample. This determines whether an HIV infection is present (HIV-positive). HIV infects white blood cells called CD4+ cells, which are part of the body’s immune system that help fight infection. HIV can progress to acquired immunodeficiency syndrome (AIDS). Several tests can find antibodies or genetic material (RNA) to the HIV virus. These tests include a number of different procedures used in the diagnosis and treatment of HIV-infected patients. Tests that measure antibodies to the human immunodeficiency virus (HIV) are called AIDS serology tests.

Serology is the branch of immunology that deals with the identification and measurement of antibodies in serum which indicate the presence of disease or immunity to disease. Serum is the normally clear light yellow no cellular portion of blood that forms after the sample is allowed to clot. Some AIDS tests measure HIV antigens or nucleic acid rather than antibodies produced in response to HIV infection. AIDS test evaluate the presence of HIV in blood serum and the effects of HIV infection on the patient’s immune system.
2.6.2 **ELISA Test**

Enzyme-Linked Immunosorbent Assay (ELISA) is a rapid immunochemical test that involves an enzyme (a protein that catalyzes a biochemical reaction). It also involves an antibody or antigen (immunologic molecules). ELISA tests are utilized to detect substances that have antigenic properties, primarily proteins (as opposed to small molecules and ions such as glucose and potassium). Some of these include hormones, bacterial antigens and antibodies. If antibodies to HIV are presented (positive), the test is usually repeated to confirm the diagnosis. If ELISA is negative, other tests are not usually needed. This test has a low chance of having a false result after the first few weeks that a person is infected.

2.6.3 **Western Blot Test**

The Western Blot Test is an antibody detection test. This test is more difficult than the ELISA to perform. But it ought to be done to confirm the results of two positive ELISA tests for an infected individual. In a generally healthy low-risk population, indeterminate results on western blot occur on the order of 1 in 5000 patients. There is no universal criterion for interpreting the western blot test.

2.6.4 **Polymerase Chain Reaction (PCR) Test**

PCR test detects either the RNA of the HIV virus or the HIV DNA in white blood cells infected with the virus. PCR testing is not done as frequently as antibody testing because
2.6. Prevention

it requires technical skill and expensive equipment. This test may be done in the days or weeks after exposure to the virus. Genetic material may be found even if other tests are negative for the virus. The PCR test is very useful to find a very recent infection, determine if an HIV infection is present when antibody test results were uncertain and screen blood or organs for HIV before donation. Testing is often done at 6 weeks, 3 months and 6 months after exposure to find out if a person is infected with HIV.

2.6.5 Indirect Florescent Antibody (IFA) Test

This is a serological test used to detect agent-specific antibodies. Both infected and uninfected cells are attached to the microscope slide. Test serum is placed over the cells and left to allow antibody binding to antigens; unbound antibodies are then washed off the slide. A fluorescent-labeled species-specific anti-immunoglobulin antibody is added to the slide and allowed to bind to the antibodies; unbound labeled antibody is washed off. Then a fluorescent microscope is used to observe specific fluorescence. These assays are rapid, inexpensive and highly sensitive, but their interpretation is subjective and requires a skilled observer.

2.6.6 AIDS-Related Complex (ARC) Test

A complex means the same thing as a syndrome, which is a group of signs and symptoms that collectively indicate or characterize a disease or other abnormal condition. When AIDS
was first recognized as a more or less complete breakdown of the immune system and not merely the coincidental onset of a number of unrelated infections, much inquiry ensued to identify the reason for the breakdown. These inquires revealed that preceding the syndrome of full-blown AIDS there occurred a lesser pattern of signs and symptoms like those of the chronic fatigue syndrome, which indicated the likely progression to AIDS so to differentiate between the two conditions which there was no clear demarcation, the lesser pattern was given the name of AIDS - related complex (ARC).

2.7 Treatment for HIV/AIDS

At the time of advent of AIDS in the United States, there were no vaccines of cure to the immune deficiency. However the scientist and health professionals developed some drugs to fight the HIV infection. The Food and Drug Administration (FDA) approved a number of drugs for testing HIV infection.

Nucleoside Reserve Transcriptase Inhibitors (NRTIs) were the first group of drugs used to treat HIV infection. The spread of HIV in the body may be delayed. This group of drugs called nucleoside analogs includes the following

- AZT (Azidothymidine)
- Ddc (Zalcitabine)
- ddI (dideoxyinosine)
2.7. Treatment for HIV/AIDS

- d4T (stavudine)
- 3TC (lamivudine)
- Abacavir (Ziagen)
- Tenofovir (viread)
- Emtriva (entricitabine)

Health care providers can prescribe Non-nucleoside Reverse Transcriptase Inhibitors (NNRTIs) such as

- Delavirdine (Rescriptor)
- Nevirapine (Viramune)
- Efravirenz (Sustiva)

A second class of drugs, called protease inhibitors, has also been approved by the FDA, for treating HIV infection. These drugs interrupt the virus from making copies of itself at a later stage in its life cycle.

The second class drugs include

- Ritonavir (Norvir)
- Saquinavi (Invirase)
- Indinavir (Crixivan)
- Amprenivir (Agenerase)
- Nelfinavir (Viracept)
- Lopinavir (Kaletra)
- Atazanavir (Reyataz)
- Fosamprenavir (Lexiva)

The third class of drugs, otherwise called ‘fusion inhibitors’, were introduced by the Food and Drug Administration (FDA), to HIV infection. The first fusion inhibitor, Fuzeon, works by interfering with HIV-1’s ability to enter into cells by blocking the merging of the virus with the cell membranes. These inhibitors block the HIV’s ability to enter and infect the human immune cells. Fuzeon combined with other anti-HIV treatment reduces the level of HIV infection in the blood and is found to be active against HIV.

When the multiple drugs are used in combination, it is said to be Highly Active Antiretroviral Therapy (HAART) and can be used by people who are newly infected with HIV and also by the people with AIDS.