GENETIC POLYMORPHISM IN THE CCR5-DELTA32 ALLELE AND THE ROLE OF MOBILE TECHNOLOGY IN EVALUATING HIV DRUG RESISTANCE

BEATRICE ATIENO ONGADI

(HND IMIS, BSc. IT, and MSc. Molecular Biology & Bioinformatics)

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DECLARATION

I declare that this thesis is my original work and has not been submitted elsewhere for examination, award of a degree or publication. Where other people's work or my own work has been used, this has properly been acknowledged and referenced in accordance with the University of Nairobi's requirements.

Signature: ..... Date: 9/6/2023

Beatrice Atieno Ongadi

REG. NO. I80/52029/2017

Department of Biochemistry

University of Nairobi

This thesis is submitted for examination with our approval as research supervisors.

	Signature	Date
Prof. George Obiero, Department of Biochemistry, University of Nairobi, P.O Box 30197 - 00100 Nairobi - Kenya	.....	9/6/2023
Dr. Raphael Lihana, Centre for Virus Research, Kenya Medical Research Institute. P.O Box 54840 - 00200 Nairobi - Kenya	.....	12th June 2023
Dr. John Ndemi Kiiru, Centre for Microbiology Research, Kenya Medical Research Institute P.O Box 54840 - 00200 Nairobi - Kenya	.....	12/6/2023

DEDICATION

This thesis is dedicated to my family, the source of my inspiration. Without their support, patience, understanding and love; the completion of this work would not have been possible. To my beloved husband and children Ian and Joy may God reward you richly for the immense support. To my late father Mr. Eliashib W. Ongadi your passion for academic excellence and last words are still as clear; and a major source of success in my academic life and finally to my loving mum thank you very much for your interminable Prayers.

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ABSTRACT

More than 37 million individuals are infected with the human immunodeficiency virus (HIV), and numerous people die from HIV-related illnesses annually. Although HIV prevalence and patterns vary by nation, the occurrences of HIV in high-risk populations vary by category. CCR5Δ32 is a non-destructive gene caused by deletion of 32 base pairs on Cysteine-Cysteine Chemokine Receptor 5 (CCR5). The allele protects its homozygous carriers from HIV infection while slows disease progression to its heterozygous carriers. This study aimed at determining the genetic polymorphism in the CCR5-Delta32 allele through *in silico* approach and promotes the use of mobile technology in evaluating HIV drug resistance. A systematic review and meta-analysis of published studies was carried out and results used in this study to show a comparison of the allele spread between different populations. CCR5Δ32 related sequences were downloaded and grouped according to geographical regions for further analysis. Analysis, visualization and presentation was done using Unipro Ugene bioinformatics software in combination with other free online bioinformatics tools. For mutation detection, SNP discovery, allele identification, and sequence confirmation, the core receptor regions of HIV sequences were subjected to a similarity search through the Basic Local Alignment Search Tool and assembled against HIV-1 reference sequences. The *pol* protein region was trimmed and compared to a template in the Stanford database (https://hivdb.stanford.edu/), then used to create an Android mobile application (*ARVPredictor*) capable of interpreting HIV drug resistance and anti-retroviral drug interactions. Three geographical regions; Europe, Africa, and Asia were categorized from the 37 studies reviewed with a total of 17,535 participants. Caucasians made up 44.7% of the population, Africans 17.8% and Asians 19.3%. With a pooled Odds Ratio (OR) of 0.08 (95% CI, 0.06 - 0.10, P < 0.00001), test of subgroup differences at I^2 = 47.0%, and a P value of 0.13, the distribution of the CCR5Δ32 allele among different populations in comparison to its heterozygosity showed a significant association. Caucasians had a subtotal OR of 0.07 (95% CI, 0.05 - 0.10, I2 = 82%) and a significant P value of < 0.00001, indicating high level of CCR5Δ32 homozygosity, compared to Asians with an OR of 0.14 (95% CI, 0.05 - 0.37, I^2 = 31%) and Africans with an OR of 0.27 (95% CI, 0.04 – 1.69).A strong indication that race can be a factor in determining CCR5Δ32 homozygosity or heterozygosity, and that Caucasians are more likely to be homozygous of the allele. Similarly, when compared to the Stanford HIV

Database, the developed *ARVPredictor* identified similar HIV subtypes in 98/100 sequences during test performance (kappa - 0.98 – near perfect agreement). *ARVPredictor* identified 89/100 major NNRTI and NRTI mutations that were similar to those found in the Stanford HIV Database (kappa - 0.89 – near perfect agreement). This study reports a novel tool that accurately identifies HIV-1 drug resistance mutations that targets the HIV pol gene, and provides appropriate antiretroviral drugs for use at the point of care. It takes advantage of and utilizes today's high-speed data networks as well as smartphone accessibility.

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ADCC	Antibody-Dependent Cellular Cytotoxicity
AIDS	Acquired Immune Deficiency Syndrome
ART	Antiretroviral Treatment
ARVs	Antiretroviral
AZT	Azidothymidine
CCR5	Chemokine Receptor Type 5
CXCR4	CXC Chemokine Receptor 4
CCR5Δ32	CCR5 Delta 32
D32	Delta 32
GP160	Envelope Glycoprotein 160
HIV	Human Immune Deficiency Virus
HIVDR	HIV Drug Resistance
IMAP	Internet Message Access Protocol
KAIS	Kenya AIDS Indicator Survey
KEMRI	Kenya Medical Research Institute
MySQL	My Structured Query Language
NRTI	Nucleoside Reverse Transcriptase Inhibitor
NNRTI	Non-nucleoside Reverse Transcriptase Inhibitors
POP3	Post Office Protocol version 3
RAD	Rapid Application Development
RES	Resistance
SDLC	Software Development Life Cycle
SIV	Simian Immunodeficiency Virus
SNP	Single Nucleotide Polymorphisms
SUS	Susceptible
UNAIDS	Joint United Nations Programme on HIV/ Acquired Immune Deficiency Syndrome
W.H.O	World Health Organization

CHAPTER ONE

INTRODUCTION

1.1 Background Information

Mammalian cell membranes are made of cysteine-cysteine chemokine receptor 5 (CCR5) also referred to as CD195 and are popularly known for allowing in chemokines that signal the body to respond during inflammation and damages (Ekere et al., 2020; Rottman et al., 1997). CCR5 is coded by a gene located in the human chromosome3(Ellwanger, Kulmann-Leal, Wolf, et al., 2020). Several known mutations of CCR5 results into damage of the expressed receptor either by deletion, insertion and/or omission (Lopalco, 2010). The CCR5-Delta32 is a damage resulting from a deletion of 32 base pairs in CCR5 resulting into a mutant form of a non-destructive gene. This is unlike other genes that cause serious and harmful damages such as sickle cell anemia, cystic fibrosis, diabetes among others when knocked out; nevertheless carriers of CCR5-Delta32 mutation can enjoy its' documented benefits (Ellwanger, Kulmann-Leal, Kaminski, et al., 2020). CCR5 receptor allows entry of HIV into the body; during initial stages of infection (Caruso & Swift, 2023; Kawamura et al., 2003). For individuals enjoying the presence of CCR5-Delta32 mutation in their body, acquisition of HIV resulting to Acquired Immune Deficiency Syndrome (AIDS) for its homozygous carriers is prevented and the disease progression for its heterozygous carries delayed (Liu et al., 1996; Zajac, 2018).

In human, Delta 32 mutation of the CCR5 locus (CCR5Δ32) is an advantageous allele and highly geographically diverse indicating adaptive traits and the co-evolution between pathogens and their respective hosts (Samson et al., 1996; Zajac, 2018). This mutant gene results into a shortened protein that cannot be expressed on the surface hence giving resistance to HIV-1 infection and hinder progression to AIDS to infected persons (A. Gupta & Padh, 2015; Kleinman et al., 2022).

In the global AIDS epidemic report 2012; the United Nations Joint Programme on HIV/AIDS indicated that the variances between the rise and drop among HIV infected persons globally is varied, with 34.0 million (31.4 million – 35.9 million) people living with HIV by the end of

2011. This was estimated at 0.8% of adults aged between 19 – 49 years worldwide. Despite the diverse variance between regions and countries; the most affected area was sub-Saharan Africa with almost one (1) in every twenty (20) adults living with HIV and AIDS hence taking care of the 69% of people living with HIV globally. In comparing HIV prevalence across regions; sub-Saharan Africa (SSA) now leads with close to 21 Million people affected by the virus (Kloek et al., 2022). Kenya is listed among top countries affected by HIV/AIDS (Waruru et al., 2021). The infection rate targets vulnerable groups including; commercial female sex workers, men who have sex with men (MSM), transgender women and men, individuals who inject illicit drugs and cisgender male sex workers (Kloek et al., 2022). However, Kenya has consistently implemented effective HIV prevention programs, leading to a significant reduction in the number of new infections. (*UNAIDS*, 2019).

In 2017, it was estimated that between 1.6 million and 2.1 million persons tested positive and learnt of their HIV status globally (Waruru et al., 2021). During the same time, the UNAIDS's call for the probable fast track HIV epidemic control strategy 90-90-90 was revised. This target aimed at 90% of persons living with HIV knowing their HIV status, 90% of those who already knew their status receiving antiretroviral therapy (ART) and 90% of those on treatment being virally suppressed by the year 2020 (UNAIDS, 2021). The importance of genetic understanding of the Human Immunodeficiency Virus type 1 (HIV-1) became a focus of attention soon after it was identified as the primary cause of AIDS (Acquired immune Deficiency Syndrome). In contrast to isolates from Europe and North America, the entire genomes of two isolates recovered from Zairian patients were cloned and sequenced, indicating a common evolutionary origin (Clavel et al., 1986; Laure et al., 1987). Nevertheless, a further check on the homologous proteins obtained from these isolates showed a lot more genetic polymorphism than before, creating great interest in critically studying the conserved regions like the envelope with the aim of diagnostic development (Stephenson & Barouch, 2013). The envelope heterogeneity has also hampered the development of HIV vaccines by making it harder to introduce neutralizing antibodies against its proteins. For development of an effective vaccine against HIV-1 strain it has become important to fully understand the HIV-1specific binding antibodies and the epitope diversity of antibody responses (Araújo & Almeida, 2013). Several studies have also shown that the gp120 protein changes significantly

in the course of the HIV diseases progression allowing it to strategically escape immune responses (Jacquemard et al., 2021; Myers et al., 1997; Stephenson & Barouch, 2013).

1.2 Statement of the Problem

Although HIV still has no known cure; the affected population can access effective antiretroviral treatment to control the virus and help prevent further transmission as they enjoy long and more productive lives. UNAIDS, 2020 reported that 28.2 million people were successfully accessing antiretroviral therapy as of 30th June 2021. Despite the development and usage of highly effective combination of antiretroviral throughout the period; the main challenge has been the high rate of mutation of the HIV virus. The virus quickly develops resistance to existing combination of antiretroviral drugs; a threat to life-long use of the multi-drug combination.

Studies have shown that treatment failures, costly second and third line treatments as well as transmission of emerging drug resistant virus strains are some of the main causes of resistance. This calls for the availability of an accurate resource to interpret the genetically modified HIV strains for appropriate drug choices to clinicians, health care givers, and research scientists during treatment of their patients. Several studies have so far shown that knowledge of the genotypic resistance before start of new treatment improves the likelihood of positive response to that treatment.

The viral *env* glycoprotein compound together with CCR5 and sometimes CXCR4 coupled with CD4 antigen facilitate the entry of HIV/AIDS infection into individuals. The chemokine receptors located at the surface of the host cell express at different of infection times with CCR5 during the early stages of HIV infection and CXCR4 in late stages (Michael *et al*., 1998)**.** These chemokine receptors have been studied to reasonable conclusions and documented that a mutation in the CCR5 gene known as CCR5Δ32 resists HIV infection to its homozygous carriers. However, by studying and understanding the distribution of the resistance allele it would be possible to advice on the best method to counter the spread of HIV across different populations and also to formulate a much better way of channeling prevention and treatment strategies.

Due to frequent travels across the globe and possible cross infections among travelers; this study on comparison of genetic diversity, genomic distribution and the distribution of CCR5Δ32 resistance allele among varied populations may also help perceive the possible outcome of a more virulent strain of HIV and or unforeseen resistant allele that can trigger further research in the HIV field despite the frequent uptake of ARV.

1.3 Justification of the study

Human Immunodeficiency Virus (HIV) remains a major public health problem throughout the globe; having claimed well beyond 36.3 million lives by 2020 and leaving over 37 million people living with the disease in the same year (UNAIDS, 2021). Other documented details by the same Global HIV & AIDS statistics - Fact sheet of 2021 indicate that over 1.5 million people became newly infected with HIV in 2021 alone. Notably, since the beginning of the epidemic, 79.3 million [55.9 million–110 million] persons have been infected with HIV. With over 25.6 million individuals living with the disease in Africa, UNAIDS identified the continent as the one most afflicted in 2017. This number accounted for roughly two thirds of all new infections worldwide in 2017. The susceptibility to AIDS as a disease is highly contributed to by most affected populations having legal and social issues in relation to their behaviors. This kind of lifestyle reduces their access to now available rapid diagnostic tests (RDTs) which are able to detect the absence and or presence of HIV antibodies within the same day. The HIV prevalence and pattern also tend to vary from country to country however in high risk population (such as commercial sex workers, men having sex with other men, injecting illicit drug users) the occurrence may vary depending on the specific population.

1.4 Hypothesis

There is no significant in genetic variation of HIV-1 envelope glycoprotein (gp160) amongst Caucasians, Africans and Asians.

1.5 Research Questions

a) How is the CCR5Δ32 allele distributed among Caucasians, Africans, and Asians?

b) What is the genetic variation of HIV-1 envelope glycoprotein (gp160) among the Caucasians, Africans and Asians?
c) Can HIV-1 target gene be used in mobile technology to assess HIV drug resistance?

1.6 Objective of the study

1.6.1 Main Objective

To determine the genetic polymorphism in the CCR5Δ32 allele and use of mobile technology to evaluate HIV drug resistance.

1.6.2 Specific Objectives

(i) To determine the distribution of CCR5Δ32 allele among Caucasians, Africans and Asians.
(ii) To determine the genetic variation of HIV-1 envelope glycoprotein (gp160) among Caucasians, Africans and Asians
(iii) To develop and evaluate an Android Mobile based application to detect HIV subtype and HIV-1 Drug Resistance mutations targeting the HIV pol gene.

CHAPTER TWO

LITERATURE REVIEW

2.1 HIV and AIDS

2.1.1 History

Human Immunodeficiency Virus (HIV) is believed to have originated around 1920 in the Democratic Republic of Congo. This is after a successful cross species from Chimpanzees to human beings (Faria et al., 2014). Not much is known on the number of individuals affected by the disease up until 1980s; although scarce cases of AIDS were recorded before 1970 (Hong et al., 2020; Worobey et al., 2016). Available phylogenetic reports indicate that the now experienced epidemic could have started between mid and late 70s and by 1980 the spread already covered five continents including Africa, Europe, North America, Australia and South America (AVERT, 2015). The disease then progressively spread among different populations. From the beginning of the epidemic up to the time of writing this document over 76 Million individuals have been infected by the virus and about 38 Million people dead of AIDS related illnesses globally (UNAIDS, 2020, 2021; WHO, 2019).

2.1.2 Biology and Taxonomy of HIV

Human Immunodeficiency Virus (HIV) is classified as a genus lentivirus in the family of retroviridae and sub family Orthoretrovirinae (Blut & Blood, 2016; Mozhi & Ganapathy, 2021) (Table 2.1). This classification was agreed upon by the International Committee for the Taxonomy and Classification of Viruses (ICTV) and the HIV research community after several informal meetings before the final formal one in September 1999 (Lapointe & Harrigan, 2020; Robertson et al., 2000)

Table 2.1: Taxonomic Classification of Human Immunodeficiency Virus (ASHM, 2003)

Virus	HIV
Realm	Riboviria
Kingdom	Pararnavirae
Phylum	Artevervicota
Class	Ortervirales
Order	Revtravirocetes
Family	Retroviridae
Subfamily	Orthoretrovirinae
Genus	Lentivirus

Genetically it is grouped into two: HIV-1 and HIV- 2. Both HIV1 and HIV2 are believed to have descended from Simian Immunodeficiency Virus (SIV) (Greenwood et al., 2014); HIV2 from [*SIVsmm*] in sooty mangabey monkeys (AVERT, 2015; Cho et al., 2022; Sanchez-Pescador et al., 1985) and HIV 1 from chimpanzees (Hirsch et al., 1989; Huet et al., 1990; Myers et al., 1997) (Figure 2.1).

Both HIV-1 and HIV-2 are thought to have crossed over to humans and eventually developed into diseases in a similar manner. It probably involved consumption of raw monkey meat. Mode of transmission and pathogenesis of both HIV-1 and HIV-2 are almost similar; however HIV-2 is most prevalent in Western part of Africa (Clavel et al., 1986; Esbjörnsson et al., 2019; Visseaux et al., 2019) where only two of its transfers resulted into major human spread.

Human Immunodeficiency Virus 1 (HIV 1) strain is documented to have evolved very fast as compared to HIV-2 which was not discovered until 1986. This is associated to its rapid reproductive rate (Araújo & Almeida, 2013; Bulanda et al., 2020). By 1992 there was already a clear indication of differences in HIV-1 groups. Strains of HIV-1 got classified into four main groups namely M, N, O and P. All these groups are detectable through antibody tests. Groups N, O and P are very rare; while group O is believed to cause up to 5% of HIV infections in Central and West African countries, groups N and P have been scantily identified in Cameroon (Abongwa et al., 2019; Araújo & Almeida, 2013). Researchers have shown that

HIV-1 groups M and N are closely linked to those of chimpanzees sub-species *Pan troglodytes troglodytes* from Cameroon (Abongwa et al., 2019; Keele et al., 2006). Groups O and P, on the other hand, are shown to be closely linked to the Western lowland gorillas that can still be found in Cameroon (D'arc et al., 2015; Kouanfack et al., 2020). Group M which is responsible for the highest infection world over displays a distribution of ten (10) HIV-1 subtypes ranging from A to K plus additional Unique Recombinant Forms (URF) and Circulating Recombinant forms (CRF) (Abongwa et al., 2019; Myers et al., 1997).

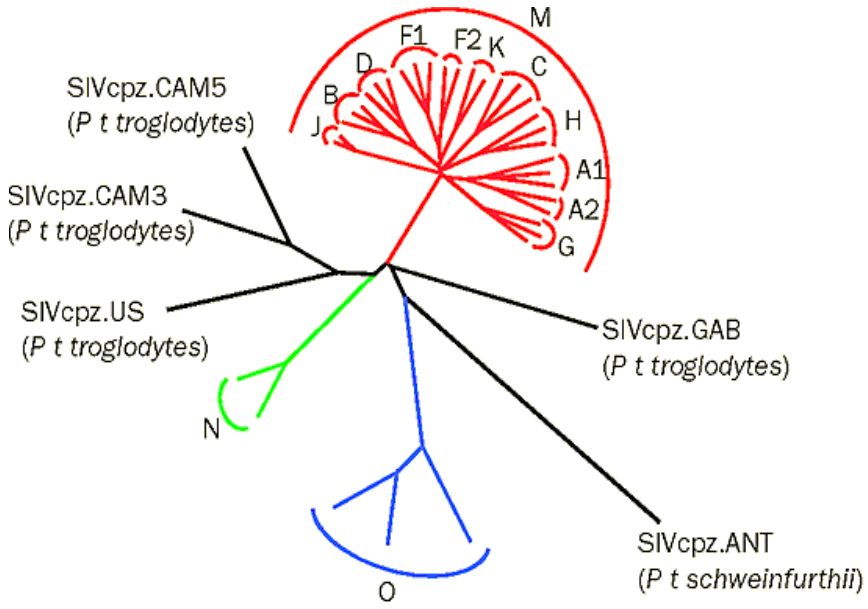


Figure 2.1: Phylogenetic Tree of SIV and HIV Viruses**:** Presenting HIV-1 group M with Subtypes A-J: Group N and Group O: Simian SIVcpz from Chimpanzee *Pan Troglodytes Troglodytes* and SIVcpz from Chimpanzee *Pan Troglodyte shweinfuthii* (Thomson et al., 2002).

2.1.3 Structure of HIV

Human Immunodeficiency Virus (HIV) is a single stranded virus which is spherical in shape. It is approximated to be 100nm in diameter. It has a lipid envelop covered in spikes of the glycoproteins gp120 and gp41 (Figure 2.2). The HIV genome has nine (9) genes: *gag, pol, env, tat, rev, nef, vif, vpr and vpu/vpx* encoding a total of nineteen (19) proteins, both HIV1 and HIV2 contain similar proteins, however some differ in molecular weight (van Heuvel et al., 2022). The two viral proteins (gp120 and gp41) critical in this study are encoded by *env*

gene and are responsible for attachment of the virus to the host cell by locking onto the CD4 receptor on CD4 T cells and another co-receptor CCR5 or CXCR4 (Engelman & Cherepanov, 2012).

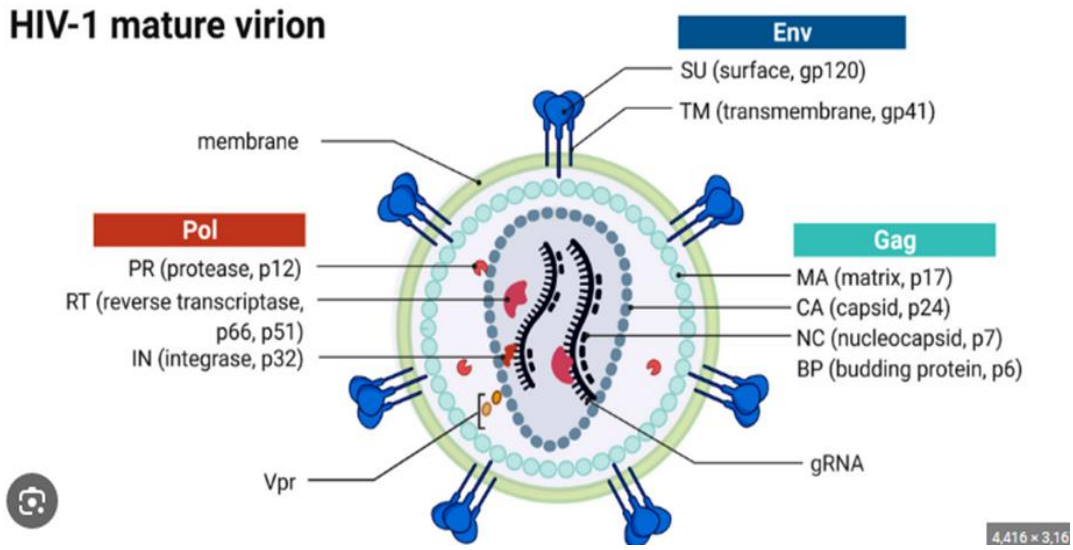


Figure 2.2: Structure of Human Immunodeficiency Virus (HIV**)** (van Heuvel et al., 2022)**.**

2.1.4 HIV Replication Cycle

Human Immunodeficiency Virus (HIV) is categorized as a retrovirus and integrates with the host cells deoxy ribonucleic acid (DNA) during infection. It targets the T-helper cells also known as CD4 cells (CD4+ T cell) which is a type of white blood cells found in the body's immune system. The intended purpose of CD4 cells is to keep the human body healthy through fighting out infections. The count of CD4 cells in a cubic millimeter of blood in the body gives a clear indication of its immune classification (AIDSMAP, 2015). The HIV life cycle (Figure 2.3) begins during attachment and fusion onto the CD4 cells, and progresses to maturation and multiplication of HIV in the blood; resulting into weakened immune system. This process largely depends on the general body health and attempted mitigation measures (Cichocki, 2020).

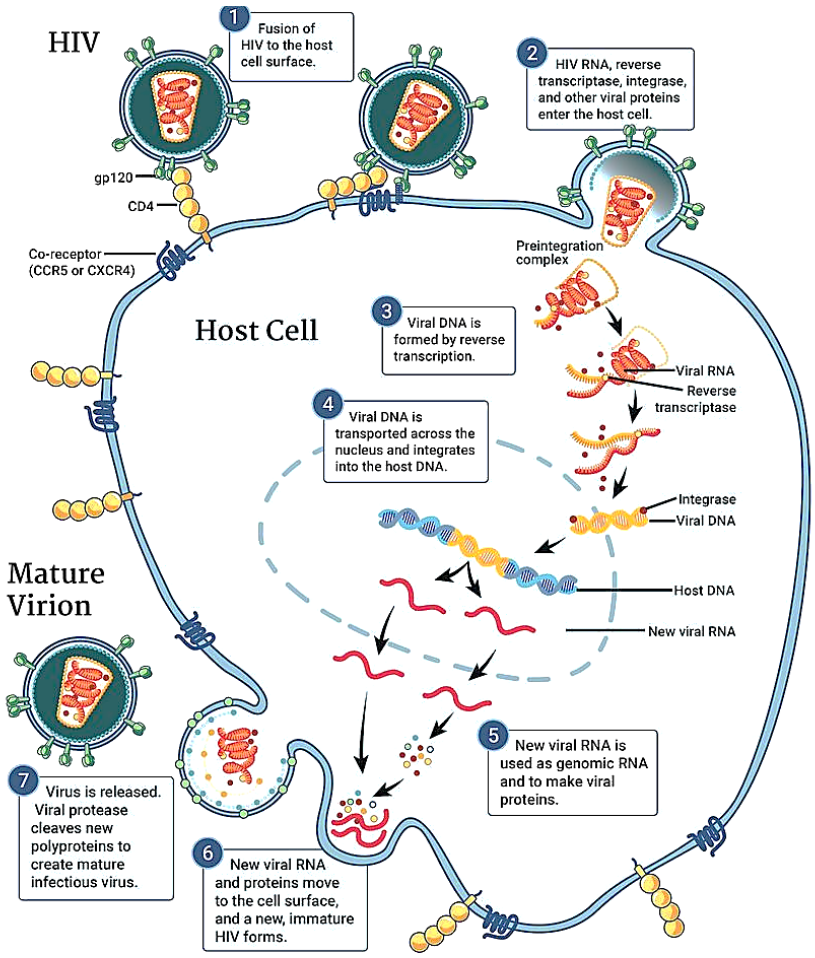


Figure 2.3: Seven stages of HIV Life Cycle**:** a) Binding b) Fusion c) Reverse transcription d) Integration e) Replication f) Assembly g) Budding

The replication cycle of HIV can be classified into seven main stages: a) Binding b) Fusion c) Reverse transcription d) Integration e) Replication f) Assembly g) Budding (*HIV Replication Cycle | NIH*, 2018).

During the initial stages of HIV replication, the virus binds to the receptors on the host CD4 cells through its external glycoproteins. It uses two co-receptors CCR5 and CXCR4. Cysteine-Cysteine Chemokine Receptor 5 (CCR5), also referred to as CD195, is a component of the mammalian cell membrane and is receptor for chemokines that are activated during cell damage (B. A. Ongadi et al., 2018). Once binding is complete the HIV RNA, reverse transcriptase, integrase, and other viral proteins fuses with the cell and enters into it. Different HIV medications that are categorized as entry inhibitors have an impact on this early binding

and infusion process. Rukobia, also known as fostemsavir, accomplishes this by connecting to the HIV gp120 env glycoprotein and preventing it from latching to CD4 receptors, whereas ibalizumab, also known as Trogarzo, binds to CD4 itself. Another medicine, Maraviroc, also known as Selzentry, accomplishes this by blocking the CCR5 receptors (Muccini et al., 2022).

The virus then uses the reverse transcriptase enzyme to concert HIV RNA into DNA enabling the virus gain entry into the cell nucleus; hence stage three of reverse transcription. Basically in this stage the virus uses the RNA as the prototype and a second strand of DNA created by adding the appropriate nucleotide (A, T, C, and G).

HIV medications called reverse transcriptase inhibitors are designed to halt the conversion of RNA to DNA. Replication is halted during reverse transcription by nucleoside reverse transcriptase inhibitors (NRTI), such as (Deeks, 2018) and retrovir, since the enzyme views them as defective building blocks. Rilpivirin and doravirine are examples of non-nucleoside reverse transcriptase inhibitors (NNRTIs), which bind to the reverse transcriptase enzyme and interfere with its function.

An integrase enzyme inserts the newly generated and fresh HIV DNA into the human DNA during integration. In the absence of medications that target it, this proviral DNA template can begin creating new viruses, or it can instead stay latent in CD4 cells that are at rest when treatment is followed. To stop the integration process, HIV medications known as integrase inhibitors were developed, including bictegravir, cabotegravir, and dolutegravir (Ndashimye et al., 2021).

The HIV DNA inserts itself into the host chromosome during the integration stage and serves as a template for the production of viral proteins. All the structural proteins, envelope proteins, and enzymes required to construct new virus particles, known as virions, are produced as a result of this process. This is made possible by messenger RNA (mRNA), which the proviral DNA acts as a template for and which, through transcription, transforms into new protein particles (Winans & Goff, 2020).

HIV protease enzyme cuts the viral proteins that were previously translated from mRNA into little pieces during the maturation and assembly stages. They are then put together to create harmless virions that can emerge from the host cell (Kucharska et al., 2020).

Protease inhibitor medications, such as darunavir, are created to stop the enzyme from slicing the newly produced viral protein for use elsewhere. Lenacapavir, an HIV capsid inhibitor, prevents the virus from assembling(Reed et al., 2021).

Combination therapies where different drugs that work in different ways are used together have proven to be more effecting in HIV management. This was originally realized during mid-1990s upon introduction of protease inhibitors such as; NNRTIs and NRTIs(Chesney et al., 2000; LaMont et al., 2022).

2.1.5 Global Distribution of HIV and AIDS

In 2021, several years after first discovery of HIV, it continues to be a major public health problem globally. An estimate of over 76 million people have been infected with HIV; out of which 36.3 million people have died of HIV/AIDS related sicknesses while over 37.0 million are living with the disease. This number includes an estimated 1.8 million children and 32.3 million adults (UNAIDS, 2020). However, continuous and rigorous research has continued in understanding the HIV viral pathogenesis. By the end of 2019, about 25.4 million people were accessing antiretroviral treatment and the global prevalence rate at 0.8% (Figure 2.4). Global distribution of the disease showing that majority of people living with HIV and AIDS are in low and medium income countries with the highest number of infections in sub-Saharan Africa.

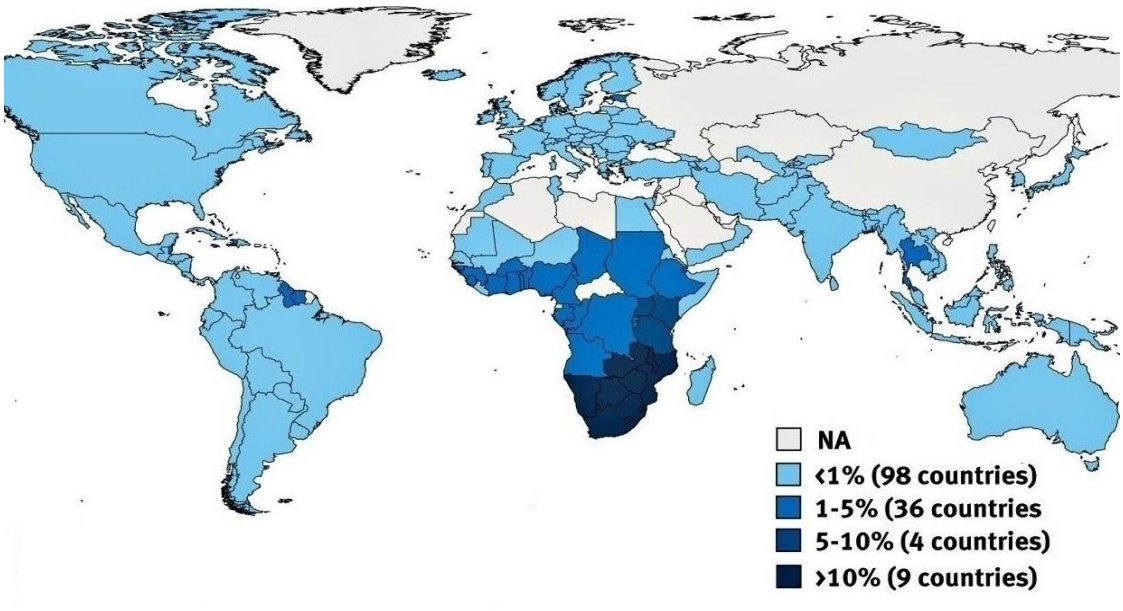


Figure 2.4: HIV Global Prevalence of 0.8% in 2019**:** The estimate based on UNAIDS report and include adults of ages 15-19 sourced from Kaiser Foundation, www.GlobalHeatlhFacts.org.

2.1.6 Epidemiology of HIV Subtypes

Over the years, both HIV-1 subtypes and HIV-2 groups have displayed a complex worldwide spread pattern in distinct directions. The diversity of HIV-1 M group makes it more complicated and resulting into numerous studies targeting the origin and spread of the disease (Li *et al*., 2012). The origins of the O and M groups are nearly identical. In comparison to the O group, the M group has a much wider global distribution and multiple changing subtypes. It's highly likely that the limited distribution of group O versus M was due to variables connected with the early stages of the HIV-1 epidemic (Faria *et al*., 2014).

Human Immunodeficiency Virus (HIV) isolates from patients in Africa and other parts of the world differed significantly in early research. However, over time, different subtypes have been isolated from various parts of the world. Using subtype prevalence data by Hemelaar and colleagues and infection prevalence data from UNAIDS Data 2019 (Hemelaar et al., 2019; UNAIDS, 2020), Figure 2.5 representing the universal representation of HIV-1 group M subtypes was realized as a final graphical summary. Globally, in 2019 HIV-1 subtype C

accounted for approximately 50% of the infections and had shown dominance in Southern Africa, Eastern Africa, India, Nepal, and parts of China (Goudsmit, 1997).

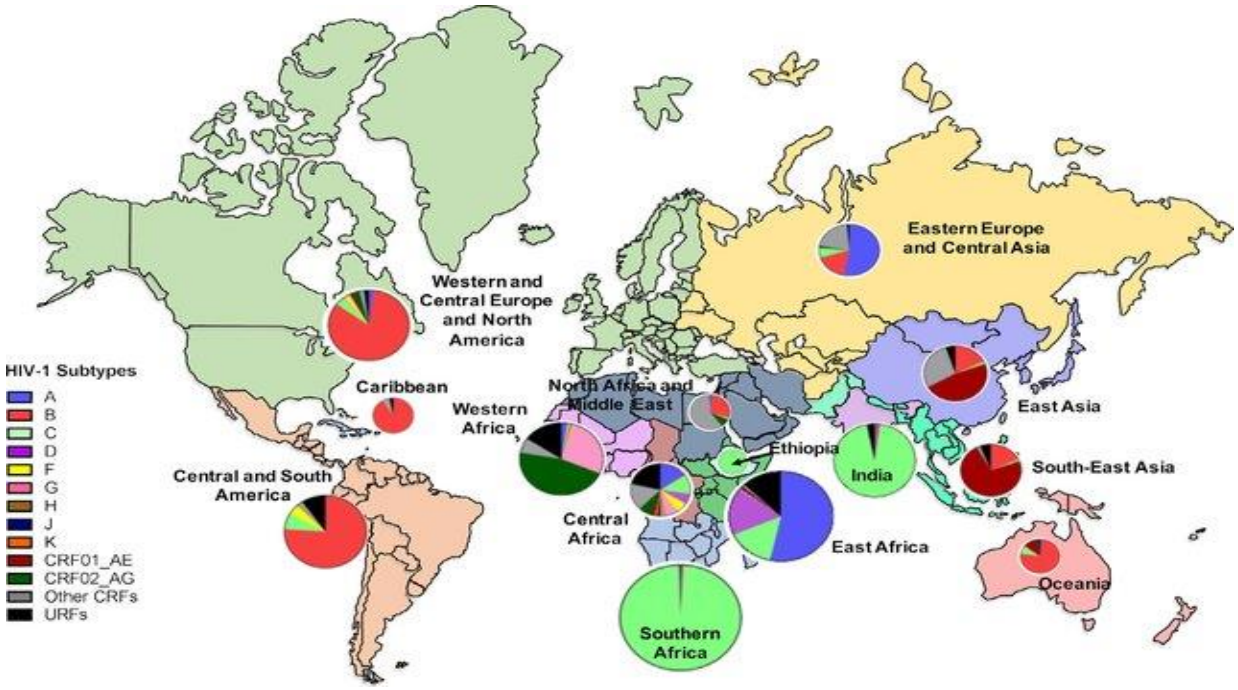


Figure 2.5: Global representation of HIV-1 group M subtypes**.**

(Each region has its own color scheme. Respective pie charts indicate the percentage of subtypes that circulate within a given region. Size of the chart represents total number of infection within the region.)
Adapted from: (Gartner *et al*., 2020)

Since its initial isolation, HIV has had varying degrees of impact on the lives of numerous people, numerous families, and communities.. It has equally impacted negatively on the economic growth worldwide leaving several orphaned children and empty homes. This pandemic has greatly affected the global workforce together with their families and increased the healthcare expenditures to very high levels (Gayle & Hill, 2001). In a previous report (Bakilana et al., 2005), the World Bank HIV/AIDS team estimated that the AIDS will account for 40% of infectious deaths by 2020. This estimate was surpassed practically immediately after this publication.

2.1.7 Key Population affected by HIV and AIDS

Globally, Certain group of individuals have been classified as the key populaion mostly affected by HIV and AIDS. They usually account for higher rates of new infections across the world. They include among others: (a) Commercial sex workers and their clients, (b) Gay men and men who have sex with other men, (c) Individuals who inject drugs and (d) Transgender population

According to a UNAIDS report from 2020, the risk of contracting HIV is 35 times greater among drug injectors, 34 times higher for transgender women, 26 times higher among sex workers, and 25 times higher among gay men and men who have sex with men (UNAIDS, 2020). The same report shows that these set of individuals accounted for 65% new HIV infecions that occured in 2020 both outside and within sub-Saharan Africa.. Generally, known risk factors associated with acquisition of HIV infection include among other things men who have sex with men, unsafe sexual practices, the use of intravenous drugs, vertical transmission, blood transfusions and unsafe contact with blood products (Justiz Vaillant & Gulick, 2022).

2.1.8 Clinical Manifestation and Diagnosis of AIDS

HIV effects on the immune system of a body may result into life-threating opportunistic infections. This may present a destructive clinical effect, resistance to therapy and increased rate of relapse (AIDS, 1986). Opportunistic infections tend to be more frequent and severe in individuals with compromised immunity. Some of the documented HIV related opportunistic infections include but may not be limited to: Herpes simplex virus 1 infection (HSV), Salmonella Infection, Candidiasis and Toxoplasmosis. Suitable treatment is highly dependent on the opportunistic infection in question and its percentage of manifestation.

Active antiretroviral therapy helps prevent many common opportunistic infections, but also causes a variety of side effects. Global pattern of HIV opportunistic diseases is varied and is highly dependent on local prevalence of latent and acquired infections and on the survival of HIV infected patients (Lucas, 2002).

Three types of HIV diagnosis exist; the tests can be carried out through saliva or blood of the patient. Nucleic Acid test tends to look for the actual virus in the blood of the patient and it’s the first test to become positive after exposure to the disease. Antibody tests look for HIV antibodies in the blood and or saliva. It may not be accurate up to until three to twelve weeks

after exposure. Antigen/antibody tests tend to look for both HIV antibodies and antigen. In this case antigens are substances of HIV while antibodies are produced by body's immune system exposed to HIV infection. A combination of both antigen as well as antibody tests may also be detectable between two to six weeks of exposure (Mayoclinic, 2020).

2.1.9 Methods of Controlling Spread of HIV

In the recent years, the UNAIDS 90-90-90 initiative has been seen as a hope for many infected and diagnosed as well as yet to be diagnosed populations. It aims at having 90% of persons living with HIV know their diagnosis results; 90% of those diagnosed put on antiretroviral therapy (ART) and 90% of individuals on ART be virologically suppressed by the year 2026 (UNAIDS, 2014). Towards achieving this 90-90-90 strategy; by the end of 2020, 84 percent of people living with HIV already knew their HIV status, 87 percent of those knowing their status were receiving treatment, and 90 percent of those on treatment were virally suppressed (UNAIDS, 2020).

Although there is a lot of commitment to limiting the pandemic around the world, it has expanded dramatically since the initial detection. However, a few independent HIV treatment and control regimens and success stories have been documented all over the world. (Bertozzi *et al*., 2006).

HIV can be acquired or spread through known human actions like as sex and injectable drug use, rather than through casual contact such as a handshake or embrace. It can spread from one HIV-positive individual to the next by blood, sperm, pre-seminal fluids, rectal fluids, vaginal fluids, and breast milk (HIVInfo, 2020). Pre-exposure prophylaxis (PrEP) can protect HIV-negative people who are at risk of contracting the disease, while regular condom use can lower the chance of infection. Women and girls accounted for 63 percent of all new HIV infections in Sub-Saharan Africa in 2020, showing extremely high rates of infection (UNAIDS, 2020).

2.2 IMMUNOLOGY OF HIV

Researchers have made great progress since the disease was originally identified in understanding the whole spectrum of infectious agents and its immunopathogenesis.. It is now clear that HIV infection induces deep immunologic abnormalities (Table 2.2) on every aspect of the immune system (Chinen & Shearer, 2002). Macrophages and dendritic cells, both of

which are important in controlling adaptive immunity, bind the virus and carry it into lymph nodes with significant numbers of CD4 T cells. HIV has evolved a method of bypassing antiviral immunity by using this mode of transport. The entry of the virus triggers the production of anti-HIV antibodies as well as cytotoxic T cells. However after some times the immune system is weakened through destruction of memory T cells ($CCR5^{+}$ and CD 4+) below critical levels and loss of cell mediated immunity (AVERT, 2015; Chinen & Shearer, 2002).

Table 2.2: HIV induced Immunologic abnormalities **(Zunich & Lane,, 1991)**

Cellular/Hormonal	Immunologic abnormality
Lymphocytes	Decreased CD4+ T lymphocyte and dysfunction Increased CD8+ T lymphocyte levels EventualCD4+/CD8+ imbalance
Monocytes	Innate immune activation Decreased phagocytosis, chemotaxis Intracellular killing Cytokine expression
Neutrophils	Neutropenia Pancytopenia
Natural Killer (NK) Cells	Down-regulated cytokine production. Reduced ability of NK cells to perform ADCC due to a reduction in the number of the cytolytic $CD56^{dim}CD16^{+}$NK cells population and reduction in the intracellular stores of perforin and granzyme A.
B-cell	Decreased B-cell number; polyclonal activation of B cells Increased production of nonspecific immunoglobulins IgG, IgA and IgM

The pathogenic illustration of HIV in (Figure 2.6) shows the complexity of the disease. It is documented that even before identification of HIV an irony existed where immune system was abnormally activated at the same time that individual was experiencing immune deficiency (Fauci, 2003)

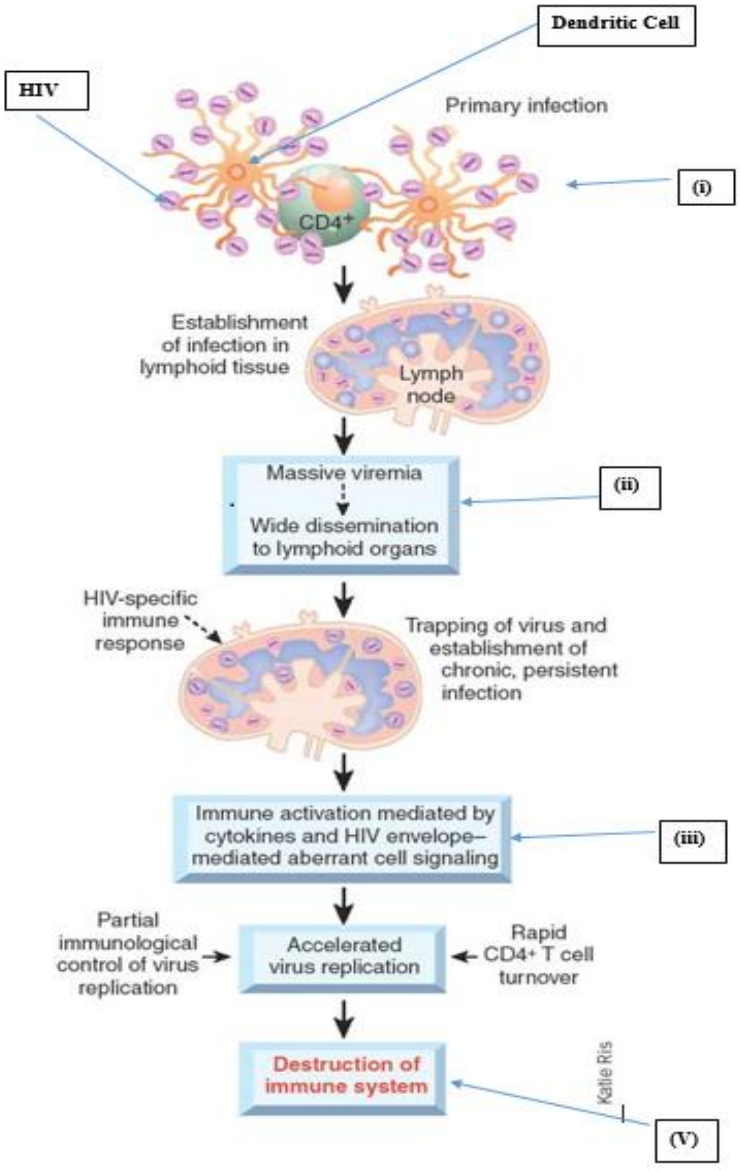


Figure 2.6: Pathogenic effect of untreated HIV mediated disease**:** (Fauci, 2003)

2.2.1 Immune response to HIV infection

The HIV virus infects a host cell by utilizing a receptor (CD4) and one of the co-receptors (CCR5 or CXCR4) to gain entry. The trimeric gp120 envelop complex protruding from the lipoprotein bilayer interacts with CD4 cell-surface receptors and either CCR5 or CXCR4 chemokine co-receptors to gain this entry (Dybowski et al., 2010).

Cysteine-Cysteine Chemokine Receptor 5 (CCR5), also referred to as CD195, is a component of the mammalian cell membrane and is receptor for chemokines that are activated during cell damage and inflammations. This receptor is coded by a gene located in the human chromosome 3. Presence of CCR5 on the surface of the cell enable HIV to latch on it and finally infects the cell, A Mutation on this CCR5 through deletion of 32 base pairs results into a non-destructive gene CCR5Δ32. It enables protection against HIV infection to its homozygous carriers and slows progression of the disease to heterozygous carriers (P. K. Gupta & Saxena, 2021; Liu et al., 1996).

Few individuals have the CCR5 genetic mutation world over. At the time of CCR5Δ32 discovery; scientists believed that it was as a result of the survivors of the bubonic plague also referred to as "Black Death" that occurred several centuries ago in Europe (Glatter & Finkelman, 2021; Stephens et al., 1998). However, other studies disputed the original theory by indicating that the mutation was present way before the bubonic plague (Cohn & Weaver, 2006; Sebbane & Lemaître, 2021). The main reason behind the scientific dispute is that the bubonic plague of 1346-52 and the resistance to HIV 1 were not supported heavily by the available historical evidence. Two hypothesis presented show that the "Black Death" did not affect Europe alone but was spread to China, North America, and the Middle East. However, this mutation seem unique to European descendants. It's documented that the level of Black Dearth mortality does not respond to topographical distribution of CCR5Δ32 among European descendants, but showing the most affected are from the Mediterranean with the lowest HIV resistance allele (Cohn & Weaver, 2006).

It is now believed that the origin of CCR5Δ32 is traced over 2500 years ago and is expected to fall gradually by genetic drift over the next 300 years. It's linked to the persistent epidemic of hemorrhagic fever which is believed to have assisted in reinforcing the frequency to about $5x10^{(-5)}$ at the time of the Black Death around 1347 (Duncan *et al*., 2005). This disputes the theory of sole responsibility of the HIV resistant mutation to bubonic plague.

2.2.2 Oxidative Stress and HIV Infection

Regulated Reactive Oxygen Species (ROS) supports various and important natural cell procedures. However, increased levels beyond defense mechanism neutralization are considered dangerous. It often results into functional alterations and damaged biological

molecules. Likewise HIV induced oxidative stress plays significant roles in development of wide spectrum of virus related pathologies (Ivanov *et al*., 2016). These include but are not strictly limited to dementia, neurotoxicity (alteration of normal nervous system activity), cardiovascular and lung disorders, exhaustion of $CD4^+/CD8^+$ T-cells as well as antiretroviral side effects (Ivanov *et al*., 2016) .

Research evidence indicates activation of oxidative stress with the presence of HIV in both laboratory models and HIV infected bodies. Therefore, when a person has HIV, their monocytes produce more reactive oxygen species, and their plasma contains significantly more oxidized nucleic acids and produces more alkaline gas (Allard et al., 1998; Awodele et al., 2012; Elbim et al., 1999; Musey et al., 1999).

2.2.3 Immune Evasion Mechanism in HIV

Human Immunodeficiency Virus (HIV) displays unique ways of evading the body's immune system and causes progressive and deadly deterioration to it. It logically manipulates the natural defense mechanism to its own advantage leaving the host to struggle for survival. If HIV is transmitted through sexual activity; it uses mucous membranes lining the vagina, rectum and mouth to enter the blood stream. This progression makes mucosa the first line in attempting to physically defend the pathogens. HIV evades this first line defense by crossing the mucosal surface through seizing intraepithelial dendritic cells (Malim & Emerman, 2008). It then targets the CD4 T cells by establishing acute immune cells infection within the mucosa through mucosal breaking steps (Mona Sadat *et al.*, 2018). It is capable of defeating the host defense mechanism through activation of a classical pathway of a complement system through envelop protein gp41 binding. Viral capsid protein (p24) acts as immune invasion through hiding HIV-1 nucleic to impersonate cellular protein (Keele et al., 2008; Zhang et al., 2018) Presence of HIV1 gp120 in a host triggers production of CC chemokine receptor 5 (CCR5) which in turn induces the movement of monocytes as well as other Natural Killer cells and dendritic cells. It's known that cytokines and chemokines play an important role in HIV 1 pathogenesis (Mona Sadat *et al*., 2018). One of the induced monocytes in this process is monocyte chemoattractant protein-1 (MCP-1 or CCL2) responsible for encouraging monocytes to leave the blood stream to become tissue macrophages while HIV1 makes use of its accessory products to evade laid cytokine networks (Frankel & Young, 1998). Some of the

accessory products used by HIV for its invasion are HIV-1 *tat* (Trans-Activator of Transcription), a regulatory protein which imitates β-chemokines and triggers viral transcription and infection. HIV-1 *nef* (Negative Regulatory Factor) a myristoylated protein leading to increased production of stimulatory function of IL-12 and IL-15 cytokines as well as down-regulating HLA class I gene products (HLA-A and HLA-B) on target cells (Rolland, 2016). A therapeutic approach towards overcoming this disease still calls for a deeper understanding on how it manages to defeat the host's immune system.

2.3 Antiretroviral Treatment for HIV

Management of HIV-1 infection in the early years consisted mainly of treatment against common opportunistic infections and related sicknesses. After 1996, the treatment lines changed with the development and introduction of inhibitors of the reverse transcriptase and protease enzymes (Arts & Hazuda, 2012). This revolution brought into place a drug regimens now commonly referred to as anti-retroviral (ARVs) and has proven to enhance overall effectiveness in HIV management. Antiretroviral therapies (ART) are drugs that can be used to manage HIV. It therefore means that they are not a cure to the disease but can be used as a control measure hence regulating the effect of HIV and maintains the hosts' health status. Antiretroviral (ARV) medication tends to work by attempting to stop the HIV from replicating in the host system and enables the body's immune system to be repaired after any slight damage. With good adherence to ARV uptake; HIV positive individuals are likely to live a near normal long life just like HIV negative people. By sustaining low levels of HIV in the body to undetectable viral load levels the risk of passing on the disease is equally decreased.

2.3.1 Current Classes of ARVs

Currently available ARVs can be classified into seven main classes; cell fusion inhibitors for preventing the HIV envelop from merging with the host CD4 cell. The two categorized classes of HIV reverse transcriptase inhibitors, non-nucleoside reverse transcriptase inhibitors (NNRTIs) and nucleoside reverse transcriptase inhibitors (NRTIs). The NNRTIs binds to the HIV enzyme (reverse transcriptase) hindering the viral replication process while NRTIs tends to mimic nucleosides also blocking the reverse transcription by preventing reverse

transcriptase from converting RNA into DNA (Naswa et al., 2012). The integrase strand transfer inhibitors (INSTIs) for preventing the virus from inserting its genetic material into host chromosome and protease inhibitors (PIs) which prevents the HIV from maturing to a new virus capable of infecting other CD4 cells (Naswa et al., 2012). CCR5 antagonists, for blocking the CCR5 coreceptor on the cell surface to prevent entry. While Post-attachment inhibitors operates by binding to the CD4 receptors on the host CD4 cell (*Types of Antiretroviral Medications*, 2021).

A combination of ARV therapy has greatly reduced HIV related deaths since its discovery. The UNAIDS 90-90-90 initiative has been seen as a hope for many infected, yet not diagnosed populations (UNAIDS, 2014).

2.3.2 Challenges of ARV Treatment

Two key interventions towards HIV treatment by ARVs were recommended by the September, 2015 WHO decision. They involved immediate antiretroviral treatment to all people living with HIV irrespective of gender, age and CD4 cell count and introduction of pre-exposure prophylactic (PrEP) treatment to high risk population (Bharat Bhushan Rewari *et al*., 2017) . At this point, it was anticipated that, if correctly followed, the 2015 guidelines would drastically alter the trajectory of the HIV epidemic and reduce its burden.

Although, a lot has been achieved towards HIV management, some challenges have been experienced over the years especially in the implementation of the WHO 2015 guidelines. The challenges were previously classified into four categories as below (Bigna *et al*., 2016)*:*

a) Health System Challenges: They are challenges related to financial availability in the implementation of the new guidelines. They spread out to enough health workers for the increased work load, adequate stock of antiretroviral treatment and suitable infrastructure for treatment process.
b) Universal Testing challenges: These are challenges surrounding discrimination and stigmatization of those affected by HIV. They by extension involve fear of no privacy and confidentiality during testing both at home and in the facility. Some individuals may also feel affected by whom to be present during testing.

c) Immediate Initiation to Antiretroviral Therapy: These challenges revolve around how soon a patient can be introduced to perfect treatment regimen after successful testing. They describe a full connection between health care facility for antiretroviral initiation and the testing entity.
d) Retention and adherence to antiretroviral treatment: These challenges surround availability of long term retention plan on ARV treatment by infected individuals. A lot needs to be covered on an early signs or indicators of an HIV drug resistance.

However, some of these barriers have been resolved as a result of excellent research and revised HIV medicine implementation strategies. .

2.4 Community Perception of HIV and AIDS

Emergence of HIV in 1980s brought with it a series of social challenges described by panicking, fear, discrimination and stigmatization. To date enormous effort has been put world over towards reducing the element of HIV associated discrimination and stigmatization. (Bogart et al., 2008; Herek, 1999). A declaration of commitment was issued in June 2001 emphasizing that discrimination on the basis of HIV is a form of human rights violation and impedes efficient prevention and treatment of the disease. It highlighting the necessity of eradicating HIV-related stigma around the world (WHO, 2001).

2.5 Role of CCR5 in the control of HIV infection

It has been more than 40 years since HIV was identified as the main cause of AIDS. Contrary to the expectations of many, no vaccination that is effective against it has yet been discovered. Hutter and colleagues published a research paper on stem cell bone marrow transplant from a CCR5-Delta32 homozygous donor to a HIV positive individual (Hütter et al., 2009). It is further documented that despite having stopped anti-retroviral (ARV) therapy, the HIV positive person turned HIV negative and remained virus-free following the transplant. CCR5-Delta32 is a genetic mutation that inhibits the cell from HIV infection (Liu et al., 1996). It is a non-destructive gene which promotes protection against HIV infection to its homozygous carriers and slows the disease progression to its heterozygous carriers. The origin of CCR5Delta32 is estimated to have occurred around 700 years ago. This is by applying coalescence theory to the current haplotype genealogy and utilizing an estimated range of 275-1,875 years. (P. K. Gupta & Saxena, 2021; Stephens et al., 1998). According to some

geneticists, the CCR5-Delta32 mutation was present as long ago as 2,500 years ago. The fact that the CCR5-Delta32 mutation is unique to Europe raises the possibility that the middle ages' plagues significantly increased the mutation's frequency. These plagues had a 100% case death rate, they were only found in Europe, and continued for more than 300 years (*Biologists Discover Why 10 Percent Of Europeans Are Safe From HIV Infection*, 2005).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Distribution of CCR5Δ32 allele among Caucasians, Africans and Asians.

This was a systematic review and meta-analysis assessment, in which organized methods were used to locate, assemble and evaluate titles and literature of published papers on HIV/AIDs and CCR5Δ32 from 1984 to 2017. The study period believed to cover the three documented decades of HIV pandemic. The search targeted online and freely available resources including but not limited to Hinari specifically PubMed Central, English database of Google scholar, Science Direct, Research4Life, National Center for Biotechnology Information (NCBI), OVID databases, AIDS Journal and Google. The main search strategy used in this study was majorly borrowed from the documented Cochrane guidelines number seven (Higgins, 2011) for appropriate and permissible low bias. All data utilized in this systematic review and meta-analysis met the requirements for ethical research and is publicly available through various databases.

Valid explanatory simulation method was used by selecting topics such "*global distribution of CCR5*" and (PICO) Population, Intervention, Comparison, Outcome method to formulate research questions and objectives (Tawfik et al., 2019). An example of PICO research question used in this study is: How is the distribution of CCR5Delta32 among Africans? (P: Africans, I: CCR5Δ32, C: CCR5, O: distribution). The (SPIDER) Sample, Phenomenon of Interest, Design, Evaluation, and Research type technique for systematic reviews was also recommended for qualitative and mixed method searches. General flow and output summary followed the Preferred Reporting Items for Systematic Review and Meta-analysis statement (PRISMA checklist 2009) (Moher et al., 2009). PRISMA diagram has been used to summarize details on data identification, screening and eligibility.

PICO and SPIDER are tools used in systematic reviews and meta-analysis of mainly clinical studies; PICO focuses on quantitative articles while SPIDER focuses on qualitative and mixed methods searches (Higgins, 2011).

Additional search strategy using key words from a combination of Medical Subject Heading (MeSH) and free text including terms related to CCR5, CCR5Δ32 and HIV was also performed in Medical Literature Analysis and Retrieval System Online (MEDLINE) through Ovid Open Access.

3.1.1 Preliminary investigation and Objective validation

A preliminary search was agreed upon by the review team in order to categorize important papers and eliminate duplicate articles. This stage was also critical in ensuring that sufficient papers for evaluation were available.

3.1.2 Inclusion and exclusion criteria

The eligibility criteria of this study were based on published and original articles on HIV infected individuals from various countries globally. It targeted articles from the general population, both HIV positive and HIV negative persons, exposed seronegatives (ESN), exposed seropositives (ESP), and highly exposed seronegatives (HESN). The exclusion criteria included letters of correspondences, conference presentations, papers with missing relevant data such as abstracts and papers without translation link.

To reduce reporting bias only studies with participants successfully genotyped for CCR5Δ32 and results accurately recorded were included. A predetermined and comprehensive inclusion and exclusion criteria was arrived at to facilitate objective screening of different articles. For the final review, only published and original publications on the distribution of the CCR532 allele in HIV-1 infected people from various countries were included.

3.1.3 Data Abstraction and Statistical Analysis

This search strategy targeted specifically two most important and often used databases in HIV genetic research. The first being the HIV Sequence Database in Los Alamos (http://www.hiv.lanl.gov/) (Rhee, 2003) which conserves HIV sequences and precisely focuses on annotation and data analysis. The second is the reserve transcriptase/protease sequence in Stanford (http://hivdb.stanford.edu/) (Mellors, 1996) storing all sequences related to the development of viral resistance against ARVs (Anti-retroviral drugs). Stanford database equally focuses on the analysis of different sequences and matches them with appropriate antiretroviral therapy for a treatment cycle. It formed the replicated worksheet and the

prototype from which an android application (ARVPredictor) was developed as part of this study. BLAST (basic local alignment search tool) was the main algorithm used for comparing primary biological sequence information retrieved against the wild type sequences. Alignment was done by use of MAFFT (Multiple Alignment using Fast Fourier Transform) (Katoh & Standley, 2013) and further viewed in MegaX (Kumar et al., 2018a) and Unipro Ugene (Okonechnikov et al., 2012) bioinformatics softwares.

The search process was conducted in two main phases; first a group of three reviewers independently cataloguing articles as per the agreed criteria. The result of the initial phase was cross checked by an independent reviewer to ensure agreement accuracy of 90% and above. The second phase involved full text review and confirmation for inclusion suitability. Uncertainties and conflict of opinions were discussed and resolved in consensus by the reviewers. An excel sheet was utilized as the ultimate storage tool for correct data abstraction. It listed all of the authors' names, publication year, journal, URL, DOI, and abstract. All records were stored in a secure Mendeley library, which helped to eliminate duplicate records with the same title, authors, year of publication, and journal of publication. The final 37 articles with 17,353 participants were accepted for inclusion in the meta-analysis as per the Prisma flow diagram in Figure 3.1. This is after the team carried out a manual check of the excel sheet to lessen any unseen human errors and bias.

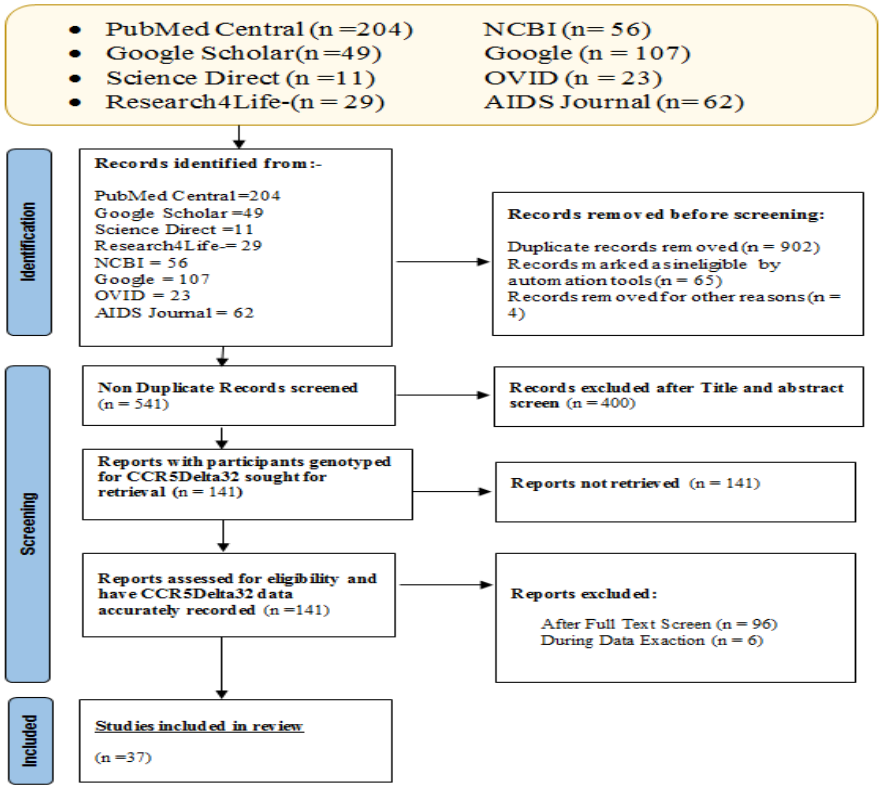


Figure 3.1: Summary of studies selected for analysis by PRISMA flow diagram.

3.2 Genetic variation of HIV-1 envelope glycoprotein (gp160) among Caucasians, Africans and Asians

Molecular Evolutionary Genetics Analysis (MEGA X) software, was used to evaluate selected HIV env genetic sequence data received from HIV databases from different parts of the globe (Kumar et al., 2018a). With the review period ranging from 1997 to 2016 covering the three decades of HIV infectivity; a total of 57 sets of near full length *env* sequences were retrieved and rooted against Simian immunodeficiency virus strain (SIVcpz) from a sub-species of chimpanzees; Pan troglodytes troglodyte. Twenty-three (23) sets were downloaded from Asians, twenty (20) from Africans and fourteen (14) from the Europeans. Studies with incomplete sequences (whole of HIV *env*) were excluded during analysis. Using the Maximum Likelihood approach and the General Time Reversible model, the evolutionary history of the collected *env* sequences was inferred. (Thomas, 2001). The tree with the highest log likelihood (-82484.14) was then generated. A matrix of pairwise distances calculated using the Maximum

Composite Likelihood (MCL) approach was used to automatically generate the initial tree(s) for the heuristic search. The topology with the best log likelihood value was then chosen. To represent the variations in evolutionary rates between the five categories (+G, parameter = 0.6429) of sites, a discrete Gamma distribution was used. Some sites ([+I], 19.43% sites) could be evolutionary invariable according to the rate variation paradigm. Branch lengths were calculated using the number of substitutions per site, and the tree was drawn to scale. The resulting dataset had 2913 locations over 94 nucleotide sequences for the analysis. Based on the analysis outputs of the HIV-1 envelop; the study aimed to understand the importance of the underlying biological consequences of different subtypes. Additionally, sequences extracted from the highly immunogenic V3 loop were also extracted and analyzed.

This study aimed at analyzing the viral superficial protein (gp120) and the trans-membrane protein (gp41) which plays a great role in the HIV virus replication cycle. They both work together as part of the envelop protein to facilitate the viral particle's fusion with the target cells during entrance, and are collectively referred to as gp160 (Figure 3.2)

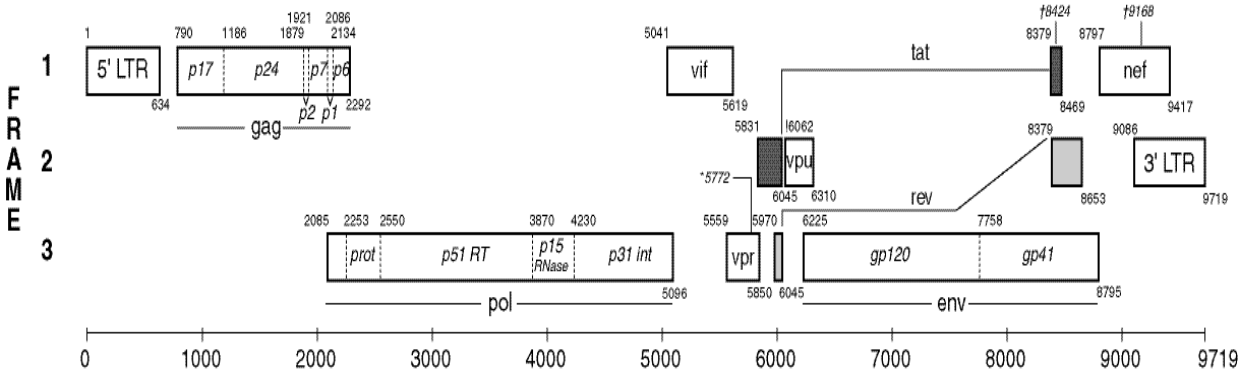


Figure 3.2: HIV Gene Map: Open reading frames shown as rectangles. The gene start, indicated by the small number in the upper left corner of each rectangle while the number in the lower right records the last position of the stop codon (Korber et al., 1997).

Further analysis covered the V3 loop region of the HIV-1 gp120 envelope protein, which has shown to be very efficient in preventing virus infectivity and cell fusion independent of the initial gp120-CD4 binding (Figure 3.3). This particular region is found to be essential for HIV virus infectivity and it is where the resistance mutations to cysteine-cysteine chemokine receptor 5 (CCR5) antagonists are located.

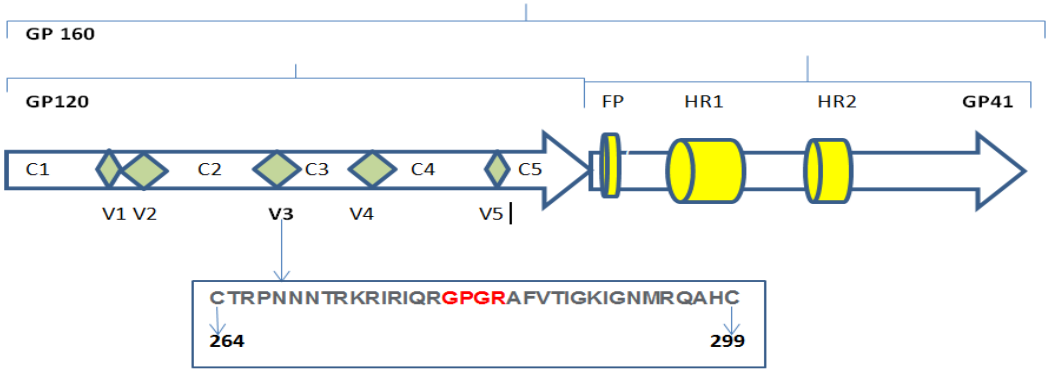


Figure 3.3: The V3 loop region of the HIV-1 gp120 envelop

3.3 Development and evaluation of an Android Mobile based application to detect HIV subtype and HIV-1 Drug Resistance mutations targeting the HIV pol gene.

Four distinct stages of application development were used to achieve this objective: pre-design associated activities, design, development, and support related steps. The final result being a smartphone application that can evaluate antiretroviral resistance levels by obtaining reverse transcriptase, protease, and integrase sequences or mutations belonging to HIV patients from users.

3.3.1 Application Design and Setup

From various drug-resistance databases, the research team obtained a duplicated spreadsheet for constructing the app (ARVPredictor). The list included mutations existing in the ANRS (the French National Agency for Research on AIDS), HIVdb, IAS–USA (International Antiviral Society–USA), Los Alamos, and Rega algorithm lists (Bennett *et al*., 2009). The final register was summarized to a total of 19 normalized core tables, 10 lookup tables, and up to 20 derived tables. This was in line with other simulated databases mostly implemented using MySQL on Linux platforms. The end result was a set of ordered relationships connecting the major components, such as the history of antiretroviral medication, the results of an isolated drug trial, and patient plasma HIV-1 RNA levels. The

final database allows users to retrieve and analyze different sets of sequences that meet particular criteria. Common queries envisioned included retrieval of sequences of HIV-1 isolates containing mutations at specific positions, patients receiving a specific drug regimen, and drug-susceptibility data on HIV-1 isolates containing combinations of mutations. Every designed query provided the following category of data: hyperlinks to MEDLINE and GenBank records, a list of mutations in the sequence, a classification of the sequence, drug-susceptibility results, and some technical data. The design availed options for downloading or viewing raw sequence data at the back end; each predesigned table returned eight or more columns of data.

3.3.2 Participants

To test the app's usability, a random population comprising 100 health practitioners actively involved in HIV/AIDS management, app developers, and information and communications technology (ICT) students was recruited. The health practitioners enrolled in the usability survey included 10 HIV experts from the Kenya HIVDR Technical Working Group on drug-resistance approval and regimen guidance, 25 HIV scientists based at the Kenya Medical Research Institute and various universities in Kenya. Others included 20 medical and pharmacy students from the University of Nairobi, Kenya, 20 graduates of the National Advanced HIV Clinical Course (NAHCC) class of 2015, 10 app developers, and 15 ICT students from Jomo Kenyatta University of Agriculture and Technology, Kenya. All the enrolled health practitioners were required to own or have access to Android-based smartphones; those who needed intensive hands-on training on downloading and using their smartphones were automatically excluded in the survey. The health practitioners were invited to participate in this survey using open invites through email to selected human health research institutions, universities (especially ICT and virology departments), and HIV comprehensive care centers. The survey also invited other participants directly via phone calls. Participants willing to be engaged recorded their interest through replying to our email by filling out a short acceptance/consenting online form and were enrolled on a first-come-first-enrolled basis. Those selected received a link on how to download and use our *ARVPredictor* Android app for test purposes.

3.3.3 App Development Process

ARVPredictor was designed and developed from models of a combination of software development life cycle methodology (Choudhury, 2011) and rapid application development (RAD) (Beynon-Davies *et al*., 1999). RAD is an agile strategy for developing software that has proven to be fast and helps complete a project within a shorter timeframe. It achieves this by reducing the time spent in planning and maximizing prototyping development. The RAD enables faster communication between a developer and the end user; hence, its high efficiency is achieved by following 4 main phases at a reasonably lower cost The initial working prototype is developed and is improved gradually through discussions until a satisfying output is reached (Figure 3.4).

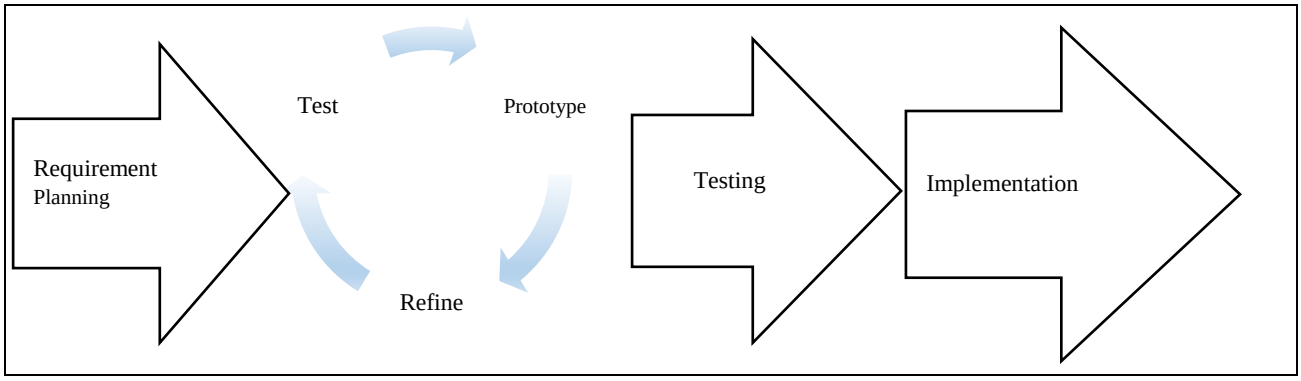


Figure 3.4: ARVPredictor: Rapid Application Development (RAD) Model.

3.3.4 Gap Identification and Requirements Gathering

A combination of 3 main activities was carried out at the initial stages of the development to meet both functional and nonfunctional requirements of the app. A brief questionnaire was randomly administered to consented selected HIV/AIDS doctors/clinicians and health care providers to determine and understand the current treatment process. This was followed by a face-to-face Key informant chat with a few health care providers who were very keen in using their smartphones to support their daily services to their patients. By performing a quick analysis of the results and with reference to relevant literature, a glaring gap was identified in the turnaround time and availability of point-of-care resources for interpreting the genetically modified HIV strains for appropriate antiretroviral drug choices. It was also understood that with the

current mobile telephony systems, different forms of data can now be shared easily among various devices (Chung & Ko, 2015). To create the initial prototype of the app, the output identified during this stage was translated into modules for better understanding and ownership of the whole app (Paetsch *et al*., 2003).

3.3.5 Use Case Modeling

Use case diagrams show the interaction between users and the system (Gomaa & Mason, 2011). In this app, the main actors and their respective interactions with the app were first identified. Each identified role was assigned relevant access rights and hence classified as patient, health care givers, and administrators of the system (Figure 3.5). Administrators have the capability to manage the critical functions of the app, whereas a normal user has very limited access to the back end functionality. By signing in using an existing email address and an active phone number, a normal user can query the database using mutations/sequences. A willing patient also has the capability to view the analysis output with predefined limitations.

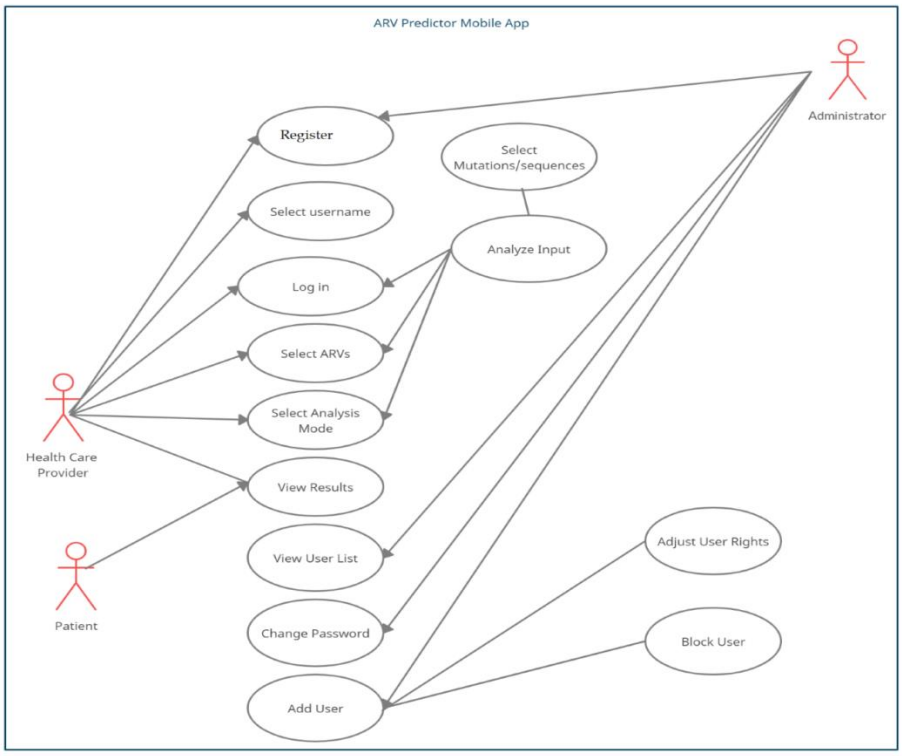


Figure 3.5: *ARVPredictor* use case diagram illustrating graphical interaction between users and the app

3.3.6 Operational Process of the App

A simple plain text prototype identified several app components, including mobile clients, web application clients, real-time servers, and databases. Following that, the development environment was set up for full development of the mobile app based on the various hardware and software specifications:

3.3.6.1 Android Studio 4.1 and Java 10

The team developed this Android app using Android Studio 4.1 (Smyth, 2020) and Java version 10 (Potts & Friedel, 1996) as the programming language. Android Studio is the official Integrated Development Environment (IDE) for Android development and usually includes several aspects required for building various Android apps. It is based on IntelliJ IDEA (IntelliJ IDEA Handbook, 2017) with powerful code editors and other developer-preferred tools. Integrated aspects of IntelliJ IDEA are essential in maximizing productivity with intelligent coding support. This aspect of the tool proved very handy in code management and provided coding hints during the development of ARVPredictor. The Java programming language used in this app was developed in early 1990 by Sun Microsystems . It is simple and efficient and can be used for various programming purposes. The combined and ultimate strength of these tools provided a suitable development platform for this app.

3.3.6.2 Nginx Server Version 1.17. 0

The development of this app involved Nginx Server (Dusch, 2012) written in C programming language. This is an open source and a high-performance development platform. It is characterized by reasonable resource consumption, ease of configuration, stability, and a comprehensive feature set. Nginx supports both a high-performance HTTP server and a reverse proxy together with POP3/IMAP proxy servers. The developed app (*ARVPredictor*) is hosted on the Nginx server with DigitalOcean Droplets (DigitalOcean, 2011) offering the cloud hosting platform.

3.3.6.3 MySQL

Massive growth of data in ARVPredictor was projected and MySQL which is an open source document database written in C++ (*MySQL*, 1995) used. It is capable of creating and deploying a highly scalable database with high performance capability. It was selected due to its ability to work across platforms, high querying capability, availability, and predictable online professional support. The MySQL database management system has the capability to store data from a single record to a large amount of information. User data in *ARVPredictor* are stored in the MySQL database hosted on the same Nginx Server version 1.17.0 (Dusch, 2012). The actual arrangement of the development heap comprising mobile phones, servers, and databases is presented in Figure 3.6.

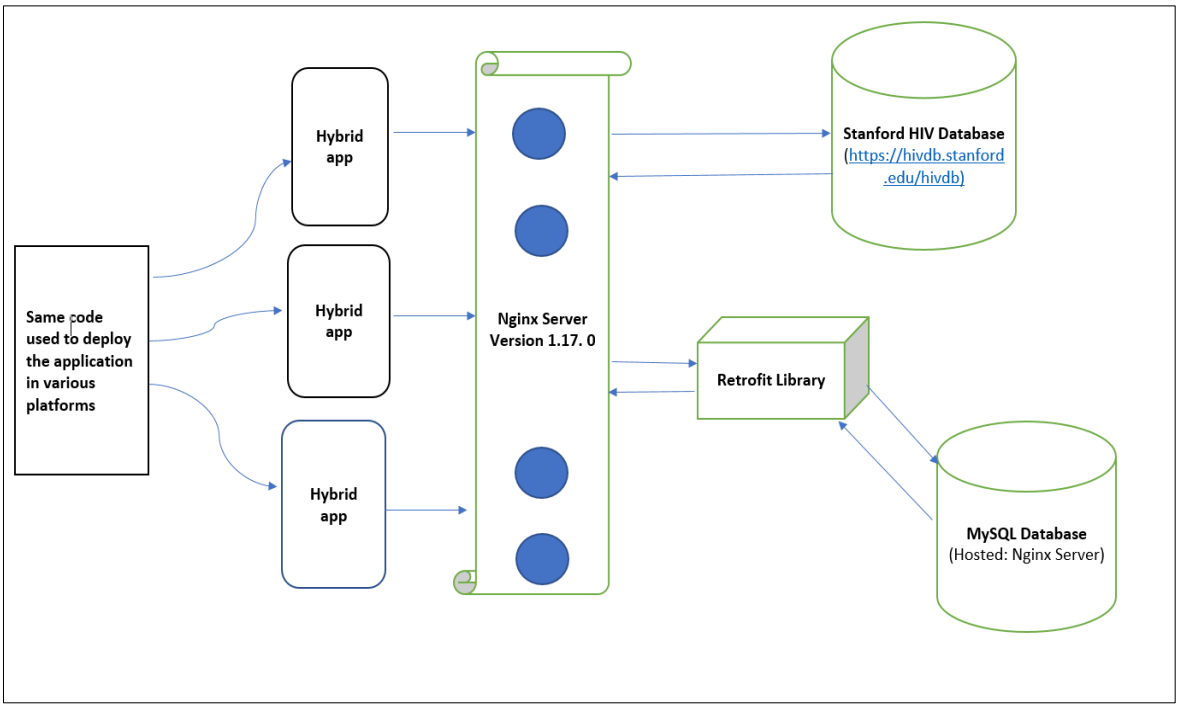


Figure 3.6: App development stack: back end arrangement of numerous connecting devices, main server, and active connection between the app (*ARVPredictor*) (B. Ongadi et al., 2022) and Stanford HIVdb (Ongadi, 2021; Rhee, 2003).

3.3.6.4 Apollo Android

The *ARVPredictor* app was developed to connect to the Stanford Database (Rhee, 2003) query language (GraphQL) through the Apollo Android library (GraphQL, 2021). This

platform converts and transfers data between the HIV Stanford Database and the *ARVPredictor* user interface.

3.3.6.5 Retrofit Android Library

This is a rest client library (Drohan D, 2017); in this app, retrofitting was used to handle all network calls from the app's back end built on PHP's Laravel framework.

3.3.6.6 Mobile Client Environment

To ensure proper functionality and accurate returning of results, the app requires phone-free space of not less than 50 MB, touch h screen display of 3.0 inches or higher, Android OS version 3.2 or later, and adequate data bundles.

3.3.7 App System Design

The design of the system was created and subjected to a simple objective versus output evaluation to ensure that all aspects of the app requirement were captured (Figure 3.6). The health care provider signs up using a valid and authenticated email address and uses a phone number and a password to access the app. A real-time server is set to build and send sequence or mutation files to the Stanford Database and return the expected analysis outcome. Different variables are captured immediately into an Excel sheet (Microsoft) and available for statistical analysis of the app.

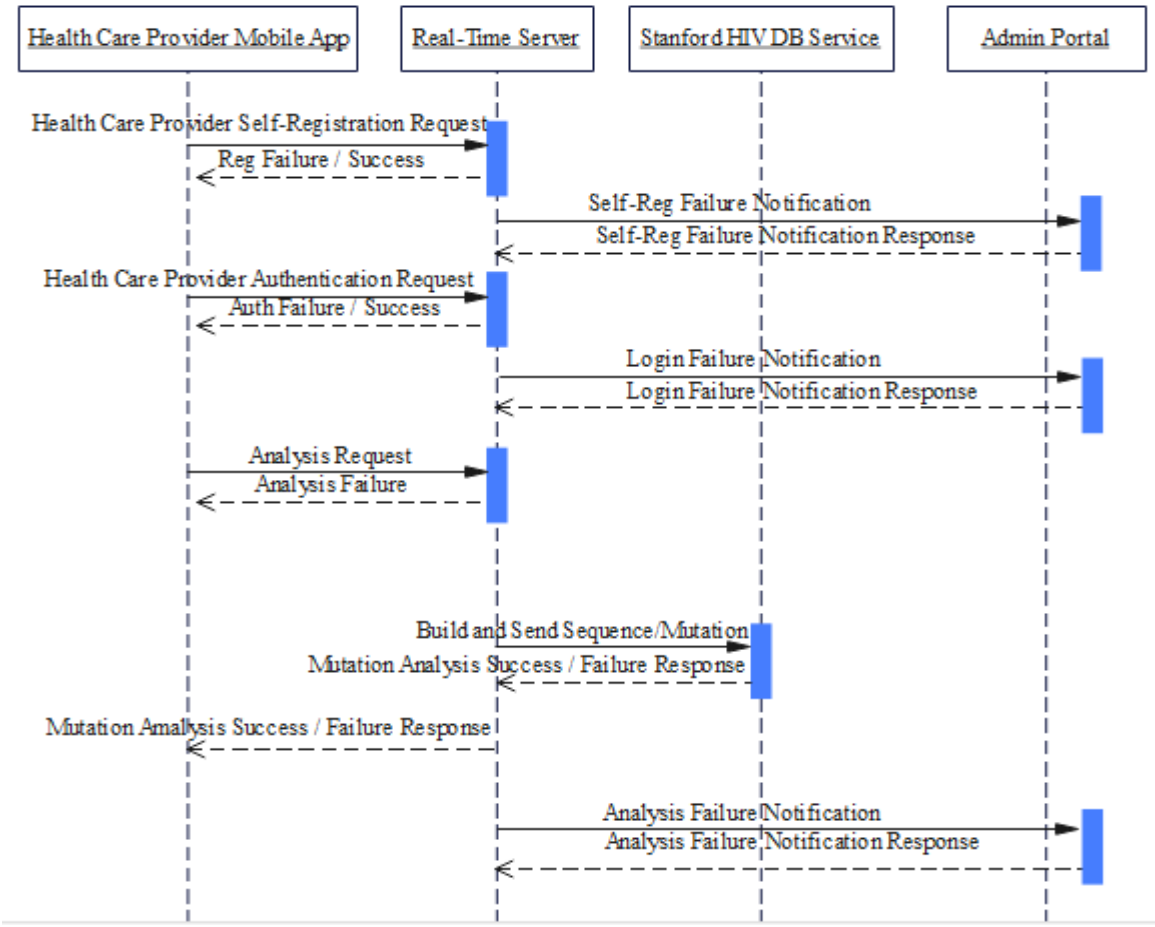


Figure 3.7: Application system design and preliminary testing procedure: Model indicating all probable activities by the user of *ARVPredictor* and predictable respective responses by both real-time server and Stanford HIVdb (B. Ongadi et al., 2022).

3.3.7.1 *ARVPredictor* Deployment and Testing

ARVPredictor is hosted on the Nginx server with DigitalOcean Linux Droplets offering the cloud hosting platform. For security, public access rights were set to be limited for the MySQL database. The Android app is freely distributed through Google Play (Google P lay Store). To use *ARVPredictor*, users first need to create an account with very limited personal details (e.g. valid email addresses, passwords, and preferred active phone numbers; see Multimedia Appendix 1). Data gathered during the sign-in process was deemed as very important for future growth of the application and would be kept under secure password protection and can only be accessed by the administrator. The fully

developed mobile app was then subjected to 2 levels of evaluation and testing, first through random entry of data to ensure system component interactivity and proper functionality. The second is to guarantee accurate analysis and result output. Black-box testing was used where the functionality of each simple app was subjected to testing without minding its internal structures or workings. This preliminary testing helped to reduce the cost and time spent in the final testing stages of the app development.

3.3.7.2 *ARVPredictor* Maintenance

Regular and administrative monitoring steps were set up to assist in the continuous review of the app's system logs. This information was considered useful in understanding and maintaining the app's service health status. The output of this process informs maintenance-related needs such as any faults, downtimes, and any unauthorized activities. It helps in debugging and resolving any issues that arise as well as provides a platform for future upgrades.

3.3.7.3 Input of Actual Data for Analysis

Two types of data, namely, sequences and point mutations, are usable for analysis in *ARVPredictor.* The said sequences can be keyed in or pasted directly onto the screen of the phone or alternatively uploaded from a separate file. HIV point mutations are preconfigured and are selectable based on the WHO 2009 listing (Bennett *et al*., 2009); both can be analyzed and confirmed according to the latest available version of the Stanford Database (Rhee, 2003).

3.3.7.4 Test performance

The test performance of *ARVPredictor* against the Stanford HIV Database to determine HIV subtypes and both major and minor proteases, reverse transcriptase, and integrase mutations was determined using kappa statistics (McHugh, 2012). Accuracy of the *ARVPredictor* was then determined. A set of 100 sequences (Appendix 1.2) was blasted using both platforms, and the similarity was identified . The subtypes and minor and major mutations were noted for both platforms. To test for method accuracy, each sequence was blasted three times, and in each case, any variation (if any) in the subtype, base pairs, and mutations recorded.

CHAPTER FOUR

RESULTS

4.1 Distribution of CCR5Δ32 allele among Caucasians, Africans and Asians

The search generated thirty seven (37) suitable and complete publications for analysis. Nineteen (19) major and comprehensive papers were analyzed from among the Caucasian population over the three HIV decades. Nine (9) studies with complete genotyped individuals were available for analysis from Asia while five (5) studies covered Africa population and four (4) under the ungrouped (others) category. In 1997, Martinson and his colleagues completed an extensive study that served as the foundation for our investigation (Martinson et al., 1997). It included 3324 unrelated people from a worldwide scattered population and is the largest study in this review thus far; subsequent studies are smaller and less internationally constituted. Table 4.1 summarizes the complete list of all studies examined in this systematic review. The large amount of Caucasian-related data that was retrieved and evaluated strongly suggests that the risk of publication bias was not entirely eradicated. An ad hoc evaluation of unpublished studies in this field revealed that the situation was likely to stay the same even if the review period was extended by an additional five years to 2022. This skewed dataset exposes the general weakness of this type of analysis.

Table 4.1: Characteristics of all studies selected and grouped for review and meta-analysis

GP—General Population, SN—Seronegatives, SP—Seropositives, ESN—Exposed Seronegatives, HESN—Highly Exposed Seronegatives

									CCR5 Heterozygous			CCR5 Homozygous		
Publication	**Year**	**Population genotyped**	**Study Category**	**Sample Size**	**Case Group**	**HIV +ve**	**HIV -ve**	**General Pop(GP)**	**HIV + (Wt/Δ32)**	**HIV - (Wt/Δ32)**	**GP (Wt/Δ32)**	**HIV+ (Δ32/Δ32)**	**HIV- (Δ32/Δ32)**	**GP (Δ32/Δ32)**
(Martinson *et al*., 1997)	1997	Europeans	Caucasians	788	GP			788			134			7
(Martinson *et al*., 1997)	1997	African	Africans	598	GP			598			1			0
(Martinson *et al*., 1997)	1997	Asians	Asians	837	GP			837			13			2
(Yudin *et al*., 1998)	1998	Russian	Caucasians	531	GP			*531*			59			*12*
(Kostrikis *et al*., 1999)	1999	African-Americans	Other	1,442	HIV +ve and ve-	1,235	207			5			0	
(Pereira *et al*., 2000)	*2000*	*Brazilian*	Caucasians	907	GP			907			93			2
(John *et al*., 2001)	2001	Kenya	Africans	276	SP	276			1			0		
(Munerato *et al*., 2003a)	2002	Brazilian	Caucasians	*68*	*Both SP and SN*	*29*	*39*		*1*	*1*		*0*	*1*	
(Munerato *et al*., 2003b)	2003	Brazilian	Caucasians	*298*	HIV Positive	*183*		*115*	*21*		*15*	*0*		*0*
(Ryabov *et al*., 2004)	2004	Russian	Caucasians	171	GP			171			31			*0*
(Sidoti *et al*., 2005)	2005	Sicilian	Caucasians	1015	HIV +ve and ve-	114	901		5	70		0	5	
(Trecarichi *et al*., 2006)	2006	Italians	Caucasians	150	ESN and HIV +ve	120	30 ESN	120	9	6	12	0	0	0
(Smoljanović *et al*., 2006)	2006	Dalmatia, Croatia	Caucasians	200	GP			200			13			1
(Vargas *et al*., 2006)	2006	Brazilian	Caucasians	*103*	*GP*			*103*			*7*			*1*
(Angelis *et al*., 2007)	2007	Brazilian	Caucasians	51	HIV Positive		51		2			0		
	2007	Cameroon	Africans	1390	GP			1390			0			*0*
(Oh et al., 2008; Torimiro et al., 2007)	2008	German	Caucasians		HIV +	595	352		115	75		1	1	

(Oh *et al*., 2008)	2008	Africans	Africans		HIV +	35	25		1	1		0	0	
(Salem *et al*., 2009)	2009	*Bahraini*	Asians	*304*	*HIV -ve*		*304*				*15*			*1*
(Zapata *et al*., 2013)	2011	*Colombia*	Caucasians	65	Both	28	37		1	1		0	1	
(Zapata *et al*., 2013)	2011	*Colombia*	Caucasians	80	Both	33	47		3	2		0	1	
(Valadez-González *et al*., 2011)	2011	*Mexico*	Caucasians	*355*	*HIV+ and HIV-*	*62*	*51*	*242*	*11*	*7*	*15*	*0*	*2*	*0*
(Al-Mahruqi *et al*., 2014)	2013	Omani	Asian	115	GP			115			0			0
(Chavhan Ab *et al*., 2013)	2013	Indians	Asians	108	GP			108			2			0
(Nkenfou *et al*., 2013)	2013	Cameroon	Africans	179	HIV +ve and -ve	32	147		0	0		0	0	
(Zapata *et al*., 2013)	2013	Colombia	Caucasians	239	SP and HESN	57	70	112 HC				0	0	
(Al-Jaberi *et al*., 2013)	2013	Emiratis	Asians	253	GP			253			0			*0*
(Al-Jaberi *et al*., 2013)	2013	Tunisians	Africans	150	GP			150			0			*0*
(Lopes *et al*., 2014)	2014	Afro-Brazilian	Other	1042	SCD GP			1042			809			0
(Rahimi *et al*., 2014)	2014	Iranian	Asians	570	HIV +ve and -ve	530	40			6			1	
(Bharti *et al*., 2015)	2015	India	Asians	200	Seronegatives		200			0			0	
(Corado *et al*., 2016)	2016	Brazilian	Caucasians	*249*	*Both SP and SN*	*110*	*139*		*6*	*0*		*0*	*0*	
(Roy & Chakrabarti, 2016)	2016	Indians	Asians	571	HIV +ve and -ve	181	568					0	0	
(Mehlotra *et al*., 2015)	2016	Papua New Guinea	Other	620	GP			620			0			0
(Corado *et al*., 2016)	2016	Brazilian	Caucasians	177	HIV1 positive		177		11			0		
(Heydarifard *et al*., 2017)	2017	Iranian	Asians	400	HIV +ve and -ve	140	300		1	9		0	0	

Cumulatively, Caucasians accounted for 44.7 percent of the 17,353 participants in this study Asians 3363 accounted for 19 percent, and Africans 3094 at 17 percent (Figure 4.1).

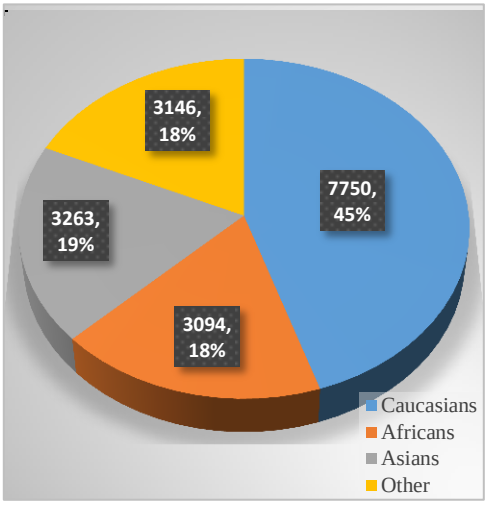


Figure 4.1: Distribution of participants with CCR5Δ32 genotyped by category.

Total number of studies accessed and reviewed per predetermined category is Caucasians (19, 51%), Asians (9, 24%), Africans (5, 13%) and others (4, 10%) summarized in (Figure 4.2).

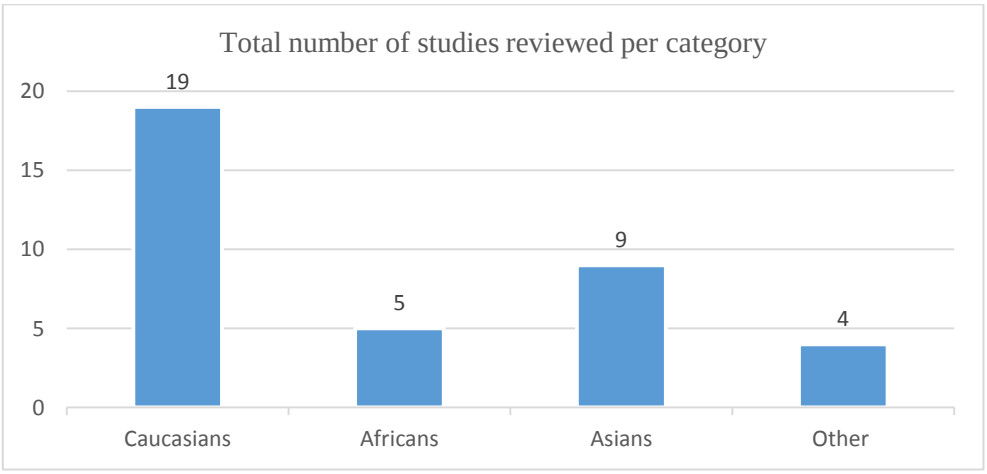


Figure 4.2: Distribution of reviewed studies and sample size by category.

This accounted for 91 percent of CCR5Δ32 homozygotes among Caucasians and 7% percent, among the Asian community (Figure 4.3). In this study, there were 52 Caucasian and 7 Asian homozygotes.

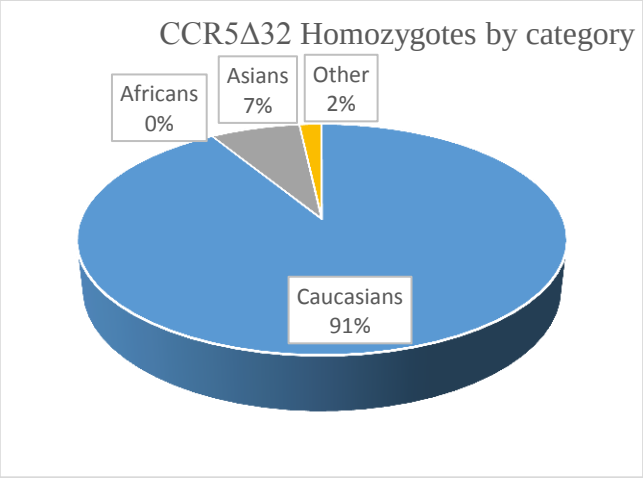


Figure 4.3: Percentage of CCR5Δ32 Homozygotes by category.

From the forest plot analysis results in Figure 4.4, there is a clear indication that most studies reviewed and meta-analyzed were from Caucasians population. These studies carried a lot of weight and significance as evidenced by the size and visibility of the small squares in the plot. Both pooled effect and 95% confidence Interval did not cross the line of effect hence result herein are significant.

The cumulative odds ratio (OR) of CCR5Δ32 allele is 0.08 (0.06; 0.10), with a P value of <0.00001 implying that CCR532 and HIV infectivity have a substantial relationship. Again, this P value of way below the predetermined cutoff (0.5 or P<0.5) this association is considered statistically very significant.

With heterogeneity at 62%, it is observable that being a Caucasian is a factor for CCR5Δ32 homozygosity hence it can be assumed that this is a protected population in terms of the allele. With heterogeneity at 31% being an Asian is not a sure factor for CCR5Δ32 homozygosity hence no protection from HIV infection; few cases presenting the allele here could be as a result of gene flow or descendants of admixture with Caucasians.

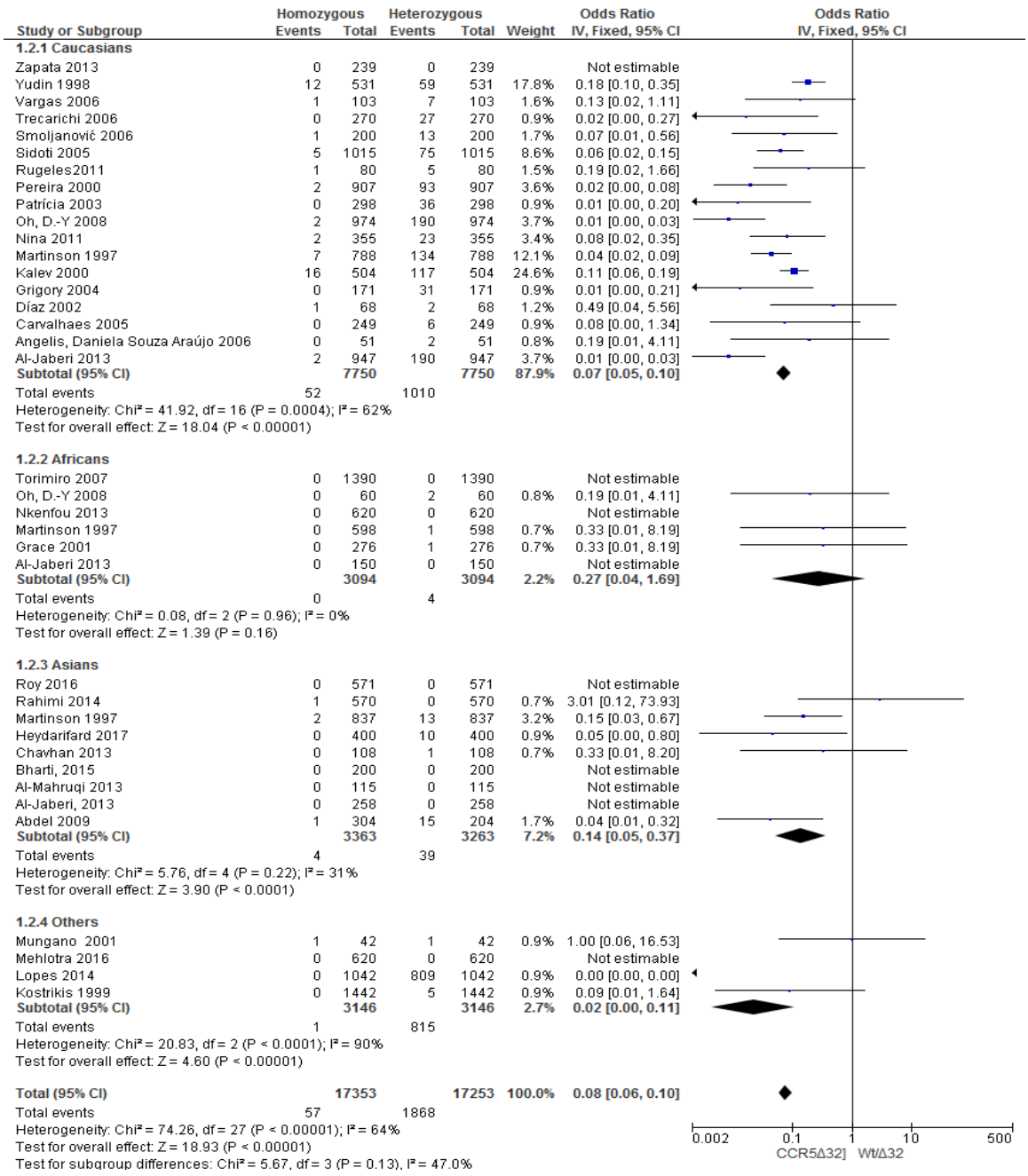


Figure 4.4 Forest plot showing comparison of CCR5 Homozygosity and Heterozygosity among various populations (Caucasians, Asians, Africans and Others).

The forest plot summarized in Figure 4.5 equally indicates a strong association between CCR5 Homozygosity and Heterozygosity among HIV positive and HIV negative individuals with an odds ratio (OR) of 0.10 (0.06; 0.19) and a P value of <0.00001. In this case, also both pooled effect and 95% confidence Interval did not cross the line of effect hence significant. However

with an the I^2 value which is is >50% it might mean the studies are inconsistent due to a reason other than chance.

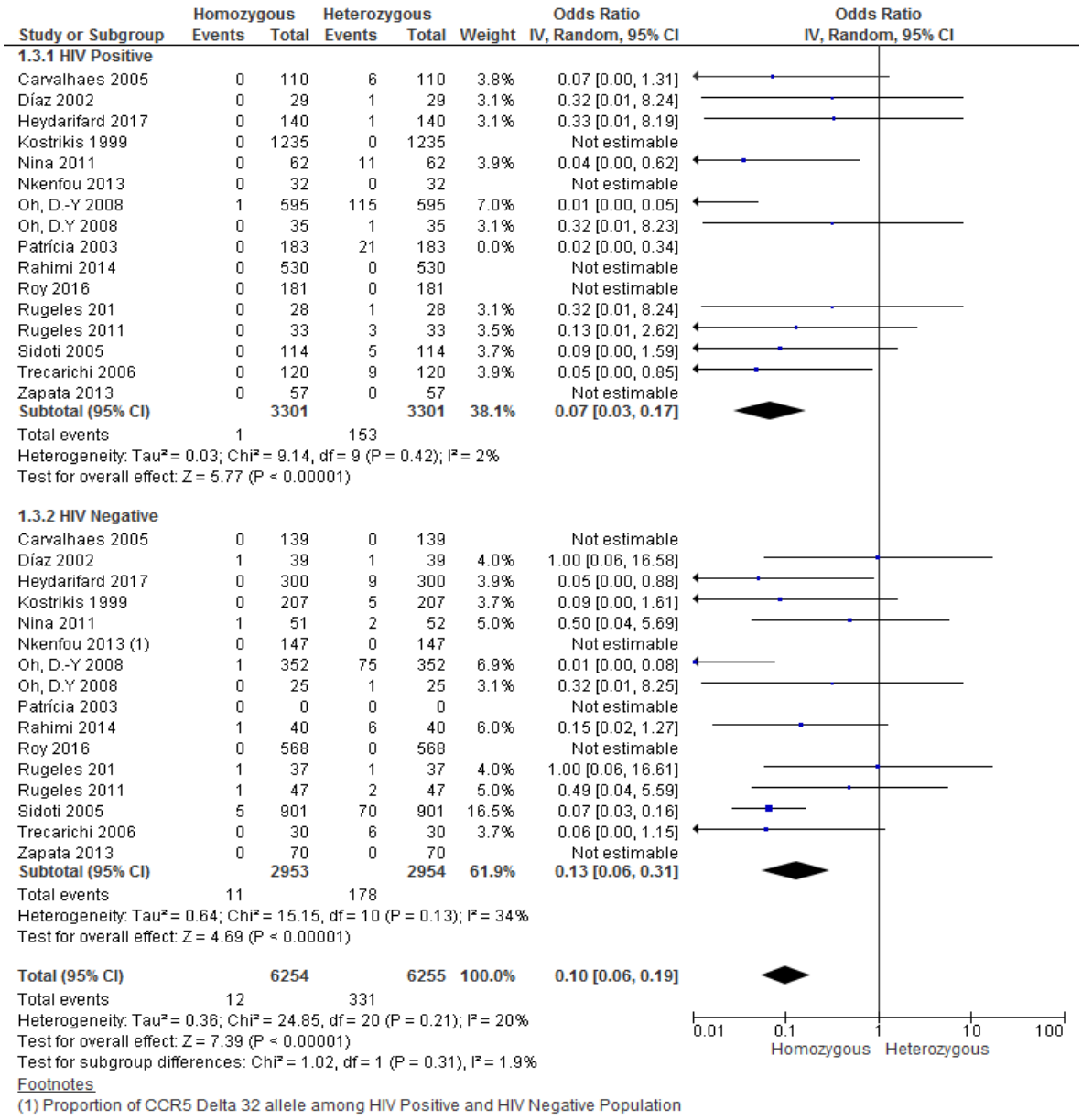


Figure 4.5: Forest Plot for comparison of CCR5 Homozygosity and Heterozygosity among HIV positive and HIV negative individuals in the reviewed data.

4.2 Genetic variation of HIV-1 envelope glycoprotein (gp160) among Caucasians, Africans and Asians

From this study, a dendrogrammatic representation of different HIV-1 subtypes shows irregular distribution within different geographical locations globally (Figure 4.6). However, some subtypes are seen to be predominant within specific areas. Findings indicating that subtypes A and D are primarily found in East and Central Africa in nations like the Democratic Republic of the Congo, Rwanda, Kenya, Uganda, and Tanzania, while sub-subtypes of A are also seen to be concentrated in the eastern region of the former Soviet Union's member states of Europe. HIV-1 Subtype B is predominant in North and Latin America (Jamaica, Dominican Republic, Paraguay, Ecuador, Venezuela, Mexico, and Haiti) and Asia (Japan, Hong Kong, Philippines and South Korea). Although Subtype C is common in Southern part of Africa (South Africa, Botswana, Malawi) its presence is also seen in India, Israel, Nepal and Georgia while Subtype G has been identified in Cuba and Guinea Bissau. In line with previous studies (Buonaguro *et al*., 2007), sub-Saharan Africa has a comparable distribution of almost all the available HIV-1 subtypes. The recombinants of subtypes A, G, K and J are common in the West African countries (Mali, Niger, Gambia, Senegal, and Burkina Faso). Phylogenetic results showing that SIVcpz (pan troglodytes troglodytes) genetic combination is thought to be the ancestral sequence and can be seen to group separately from any available HIV-1 subtype.

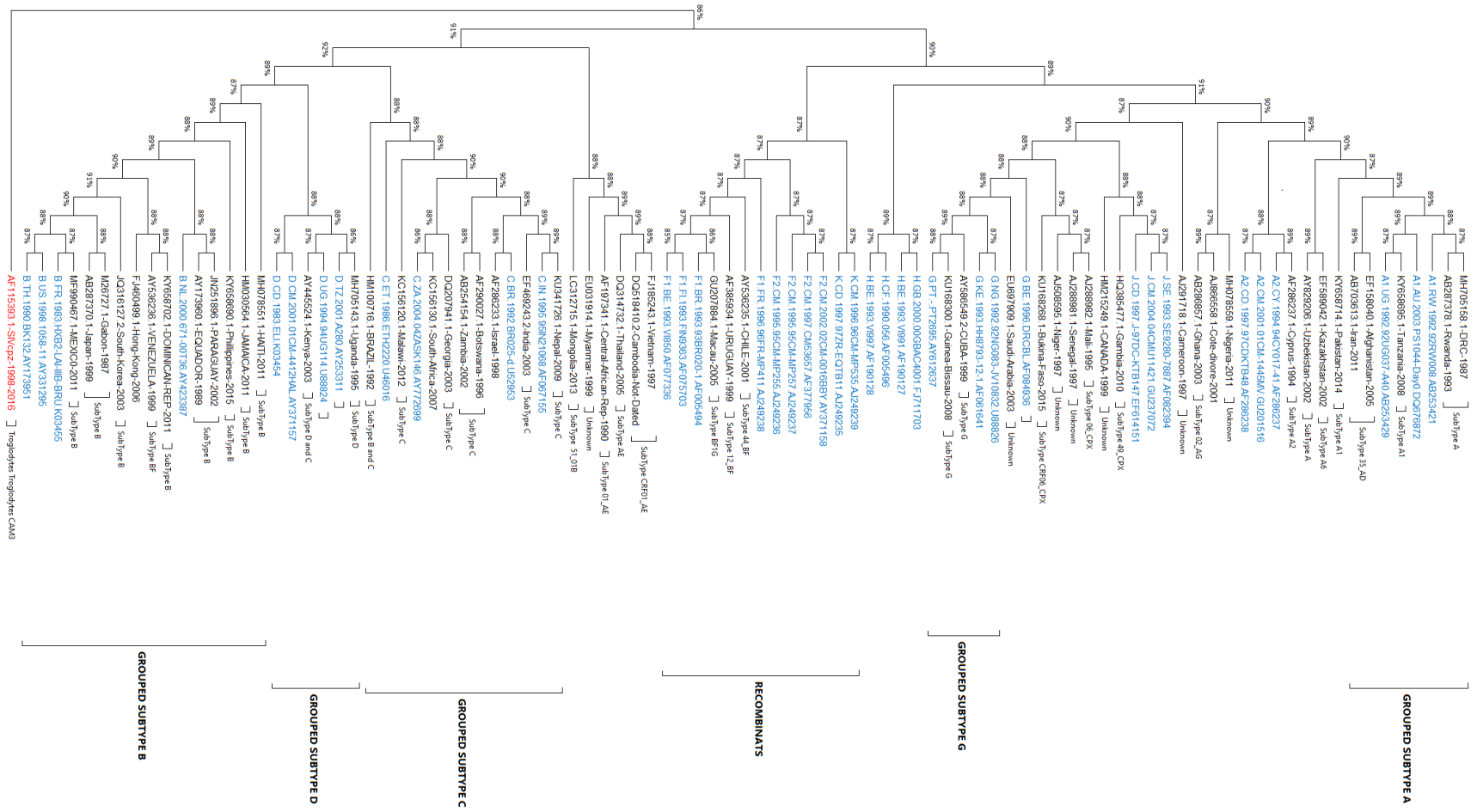
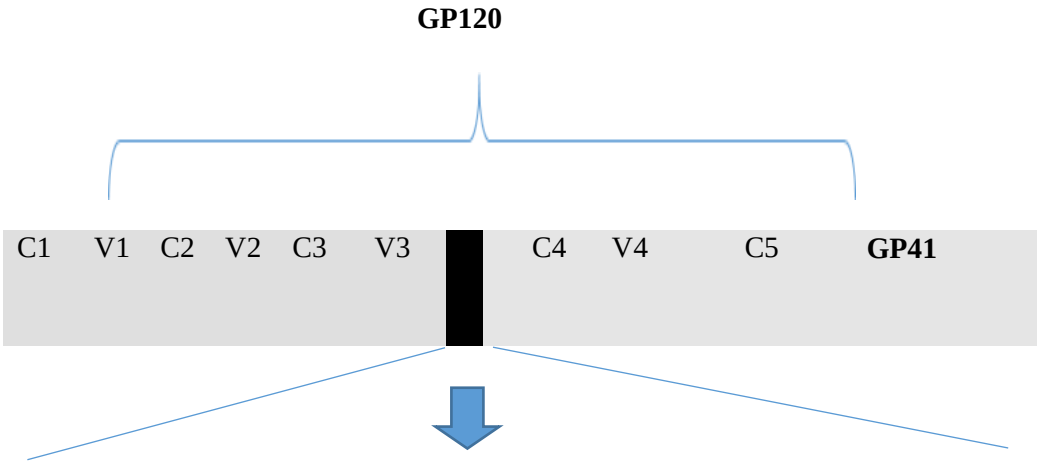


Figure 4.6: Phylogenetic analysis of env sequences inferred using the Maximum Likelihood method and General Time Reversible Model**.** *Summary:* The highest log likelihood is -82484.14). Preliminary trees generated automatically by using the Maximum Composite Likelihood (MCL) method and the Neighbor-Join and BioNJ algorithms on a matrix of pairwise distances. Different evolutionary rates among sites were modeled using discrete Gamma distribution (5 categories (+G, parameter = 0.5010)). Some sites ([+I], 19.43% sites) could be evolutionarily invariable according to the rate variation paradigm. There were 94 nucleotide sequences in this analysis. Codon positions 1st+2nd+3rd+Non-coding were included. The final dataset contained 2913 locations altogether. HIV-1 M group references sequences represented in Blue.

Variant representation of the highly conserved V3 loop sequences (Figure 4.7) indicates a visible difference within same HIV 1 subtype and across different subtypes. The study approximated the percentage value between 12% - 20% within a subtype and goes up to 35% across subtypes in this study. The output reflects a high mutability of the loop and shows that the region is under selective pressure for length of about thirty-five (35) residues.



Name	C	I	R	P	N	N	N	T	R	K	S	V	R	-	-	I	G	P	G	Q	T	F	Y	A	T	G	E	-	-	I	I	G	D	I	R	K	A	H	C
1. A1.AU.2003.PS1044-Day0.DQ676872	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
2. A1.RW.1992.92RW008.AB253421	.	T	.	.	.	.	.	.	T	.	I	.	.	-	-	.	.	.	.	.	S	.	H	.	.	.	D	-	-	.	.	.	.	.	.	Q	.	.	.
3. A1.UG.1992.92UG037-A40.AB253429	.	T	.	.	.	.	.	.	R	.	.	.	.	-	-	.	.	.	.	.	.	.	.	.	.	.	D	-	-	.	.	.	.	.	.	Q	.	.	.
4. A2.CD.1997.97CDKTB48.AF286238	.	T	.	.	.	.	.	.	.	.	.	I	.	-	-	F	.	.	.	.	A	.	.	T	N	N	N	-	-	.	.	.	.	.	.	Q	.	.	.
5. A2.CM.2001.01CM-1445MV.GU201516	.	T	.	.	.	.	.	.	.	.	.	I	.	-	-	F	.	.	.	.	A	.	.	T	N	N	K	-	-	.	.	.	.	.	.	Q	.	.	.
6. A2.CY.1994.94CY017-41.AF286237	.	.	.	.	.	.	.	.	.	.	.	I	.	-	-	F	.	.	.	.	A	.	.	-	.	N	.	-	-	.	.	.	.	.	.	Q	.	.	.
7. AB097872.1 Myanmar 1999	I	T	F	Q	P	P	S	G	G	D	L	E	I	T	M	H	H	F	N	C	K	G	E	F	F	Y	C	N	T	S	R	L	F	N	N	T	C	I	T
8. AB254154.1 Zambia 2002	I	F	A	.	H	S	G	G	D	L	E	I	T	T	H	S	F	N	C	R	G	E	F	F	Y	C	N	T	S	K	L	F	N	N	T	L	V	N	S
9. AB286857.1 Ghana 2003	I	.	F	A	.	H	S	G	G	D	V	E	I	T	T	H	S	F	N	C	G	G	E	F	F	Y	C	N	T	S	G	L	F	N	S	T	W	E	N
10. AB287370.1 Japan 1999	V	A	K	K	L	R	E	Q	F	N	K	T	T	I	V	F	T	S	S	S	G	G	D	P	E	I	V	M	H	S	F	N	C	G	G	E	F	F	Y
11. AB287378.1 Rwanda 1993	L	E	I	T	T	H	S	F	N	C	G	G	E	F	F	Y	C	N	T	S	G	L	F	N	S	T	W	T	N	E	S	M	Q	V	N	S	T	E	S
12. AB703613.1 Iran 2011	N	F	N	S	S	S	G	G	D	I	E	I	T	T	H	S	F	N	C	G	G	E	F	F	Y	C	N	T	S	S	L	F	N	S	T	W	Y	N	N
13. AF086817.1 Taiwan 1994	.	N	A	Q	F	*	L	W	.	G	I	F	L	L	*	C	N	T	T	V	*	*	.	L	E	C	Y	Q	Y	F	E	C	Y	*	*	R	K	*	K
14. AF115393.1 SIVcpz 1998/2016	P	G	G	D	P	E	V	.	N	M	M	F	N	C	G	G	E	F	F	Y	C	N	T	T	.	L	F	N	Y	T	W	K	N	N	N	I	T	K	G
15. AF197341.1 Central African Rep 1990	I	Q	F	K	P	P	Y	S	G	G	D	L	E	I	T	M	H	H	F	N	C	R	G	E	F	F	Y	C	N	T	T	R	L	F	N	I	S	T	N
16. AF286233.1 Israel 1998	H	S	F	N	C	R	G	E	F	F	Y	C	N	T	S	K	L	F	N	S	.	Y	L	S	N	S	T	S	Y	N	S	T	S	N	S	T	I	I	L
17. AF286237.1 Cyprus 1994	T	N	S	S	G	G	D	P	E	I	T	T	T	L	S	F	N	C	A	.	E	F	.	.	C	N	T	T	G	L	F	N	.	T	W	W	N	N	G	T
18. AF290027.1 Botswana 1996	A	P	S	S	G	G	D	L	E	I	T	T	H	S	F	N	C	R	.	E	F	.	.	C	N	T	S	N	L	F	N	K	T	Y	W	P	N	G	T
19. AF385934.1 URUGUAY 1999	S	S	S	G	G	D	P	E	I	T	M	H	S	F	N	C	R	G	E	F	F	F	Y	C	N	.	S	K	L	F	T	S	T	V	P	N	N	G	T	I
20. AF408632.1 ARGENTINA 1997	S	G	G	D	P	E	I	.	M	H	.	F	N	C	R	G	E	F	F	Y	C	N	T	S	S	L	F	N	N	T	G	T	N	D	T	I	T	L	P
21. AJ288981.1 Senegal 1997	G	G	D	L	E	I	T	.	H	S	F	N	C	G	G	E	F	F	Y	C	N	T	S	Q	L	F	N	S	S	N	L	N	N	N	S	S	D	N	N
22. AJ288982.1 Mali 1995	G	D	L	E	V	T	T	H	S	F	N	C	G	G	E	F	F	Y	C	N	.	S	Q	L	F	N	N	N	I	T	D	S	N	E	T	T	N	F	T
23. AJ291718.1 Cameroon 1997	S	F	N	C	R	G	E	F	F	Y	C	N	T	S	G	L	F	N	N	T	W	L	F	N	S	T	W	N	S	S	Q	E	L	N	G	T	E	P	N
24. AJ508595.1 Niger 1997	T	T	H	S	F	.	C	R	G	E	F	F	Y	C	N	T	S	G	L	F	N	M	S	G	S	S	N	I	S	G	N	S	N	E	T	I	T	L	P
25. AJ866558.1 Cote divore 2001	E	.	T	T	H	S	F	.	C	G	G	E	F	F	Y	C	S	T	S	A	L	.	N	S	.	W	M	M	V	N	G	T	W	S	D	T	.	N	D
26. AY037270.1 BOLIVIA 1999	H	S	F	N	C	G	G	E	F	F	Y	C	N	T	T	E	L	F	N	S	.	W	N	F	.	S	T	W	N	M	T	S	G	S	N	Y	S	A	E
27. AY173960.1 EQUADOR 1989	Q	S	S	G	G	D	P	E	I	V	M	H	S	F	N	C	.	G	E	F	F	F	Y	C	D	.	T	N	L	F	N	S	T	W	N	E	T	Q	L	F
28. AY445524.1 Kenya 2003	P	H	S	G	G	D	P	E	I	T	T	H	S	F	N	C	.	G	E	F	F	F	Y	C	N	.	S	.	L	F	N	S	T	W	N	D	T	S	Y	S
29. AY536235.1 CHILE 2001	T	T	H	S	F	.	C	R	G	E	F	F	F	C	N	T	T	R	L	F	N	D	T	G	F	N	D	T	K	S	N	D	T	D	T	I	T	L	P
30. AY536236.1 VENEZUELA 1999	S	A	G	G	D	P	E	I	I	M	H	S	F	N	C	G	.	E	F	F	Y	C	N	T	A	Q	L	F	N	S	T	W	K	R	D	N	N	N	T
31. AY586549.2 CUBA 1999	R	E	K	L	I	E	S	F	P	N	K	T	I	N	F	N	A	S	S	G	G	D	L	E	I	A	P	H	S	F	N	C	G	G	E	F	F	Y	.
32. AY829206.1 Uzbekistan 2002	P	G	G	D	L	E	V	.	T	H	.	F	N	C	G	G	E	F	F	Y	C	N	T	T	D	L	F	N	S	T	W	D	N	V	T	T	V	T	N
33. B.FR.1983.HXB2-LAI-IIIB-BRU.K03455	.	T	.	.	.	.	.	.	.	.	R	I	.	I	Q	R	.	.	.	R	A	.	V	T	I	.	-	-	-	K	.	.	N	M	.	Q	.	.	.
34. B.NL.2000.671-00T36.AY423387	.	T	.	.	.	.	.	.	.	.	.	I	H	-	-	.	.	.	R	A	.	.	.	.	.	.	-	-	.	.	.	.	.	.	.	Q	.	.	.
35. B.TH.1990.BK132.AY173951	.	T	.	.	.	.	Y	.	.	.	R	I	T	-	-	M	.	.	.	R	V	Y	.	T	.	.	.	-	-	.	.	.	.	.	.	.	R	.	.	.
36. B.US.1998.1058-11.AY331295	.	.	.	.	.	.	.	.	.	.	R	I	T	-	-	M	.	.	.	K	V	Y	.	T	.	.	Q	-	-	.	.	.	.	.	.	.	Q	.	.	.
37. C.BR.1992.BR025-d.U52953	.	T	.	.	.	.	.	.	.	.	.	I	.	-	-	.	.	.	.	.	A	.	.	.	.	.	.	-	-	.	.	.	.	.	.	.	Q	.	.	.

Figure 4.7: Diagrammatic representation of HIV-1 envelop. Gray partitions showing constant and variable regions observed for resistance to inhibitors for entry. Aligned sequences obtained from HIV Los Alamos database including HIV-1 M group subtype references sequences. The tip of V3 loop designated by the blue arrow is also the location of CCR5 coreceptor antagonists.

The result of the sequence alignment output of the V3 region of gp120 summarized in Figure 4.8 shows an almost near identical set of sequences for the subtypes A and C. They are both characterized by a distinct GPGQ motif. However, there is a discrete difference between subtypes A and B where subtype B has predominance of GPGR/K motif.

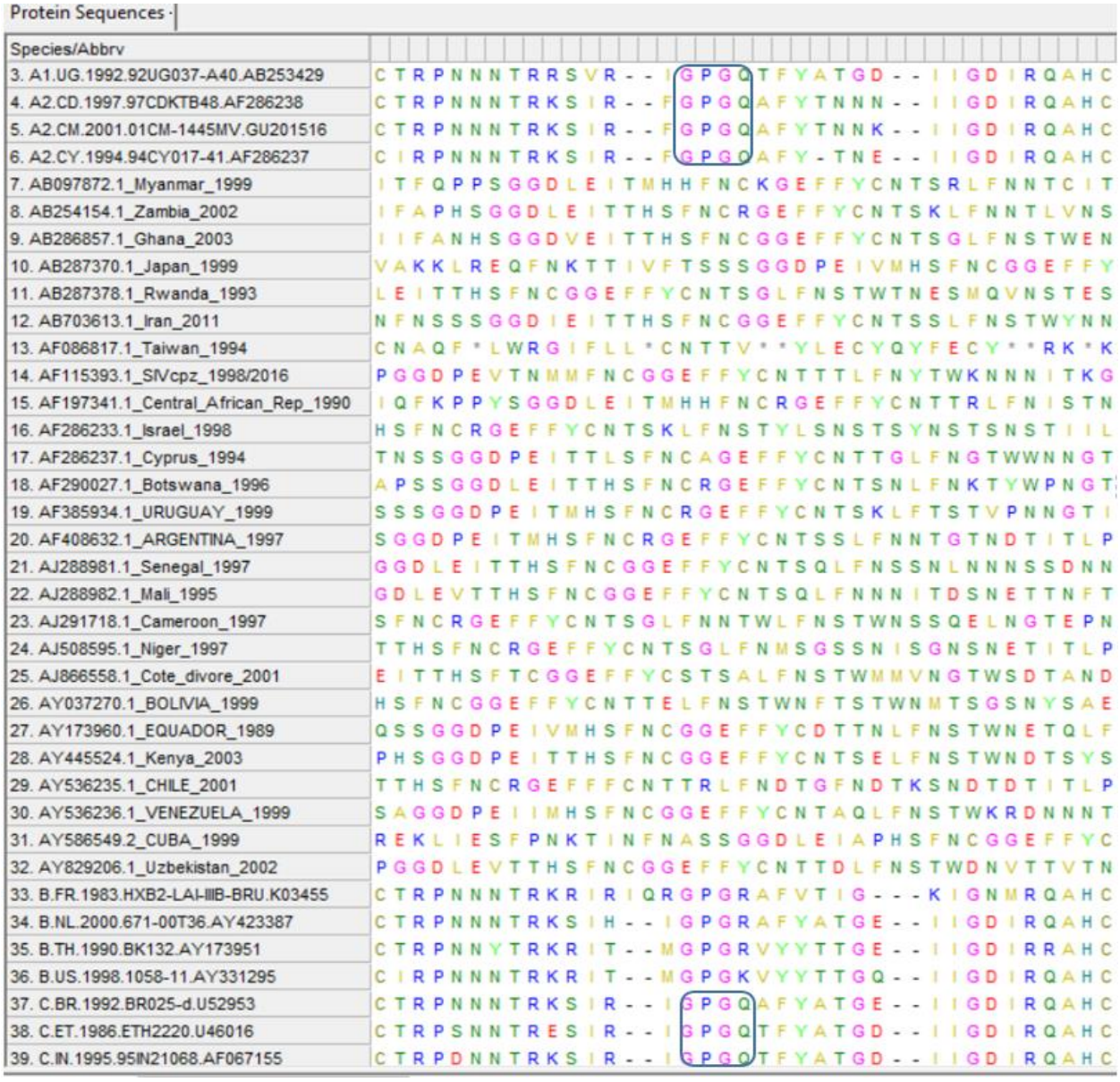


Figure 4.8: Distribution of highly conserved V3 loop tip among HIV-1 M group subtypes: GPGQ (Gly-Pro-Gln) seen among varied subtypes and GPGR (Gly-Pro-Arg) common among subtype B. Output showing near identical sequences of subtype A and C. Aligned sequences retrieved from HIV databases.

However, the phylogenetic representation of the V3 region shown in Figure 4.9 in this study shows a non-uniform distribution of subtypes. The Simian immunodeficiency virus of chimpanzees (SIVcpz) sequences remains the out-group hence still the probable ancestor.

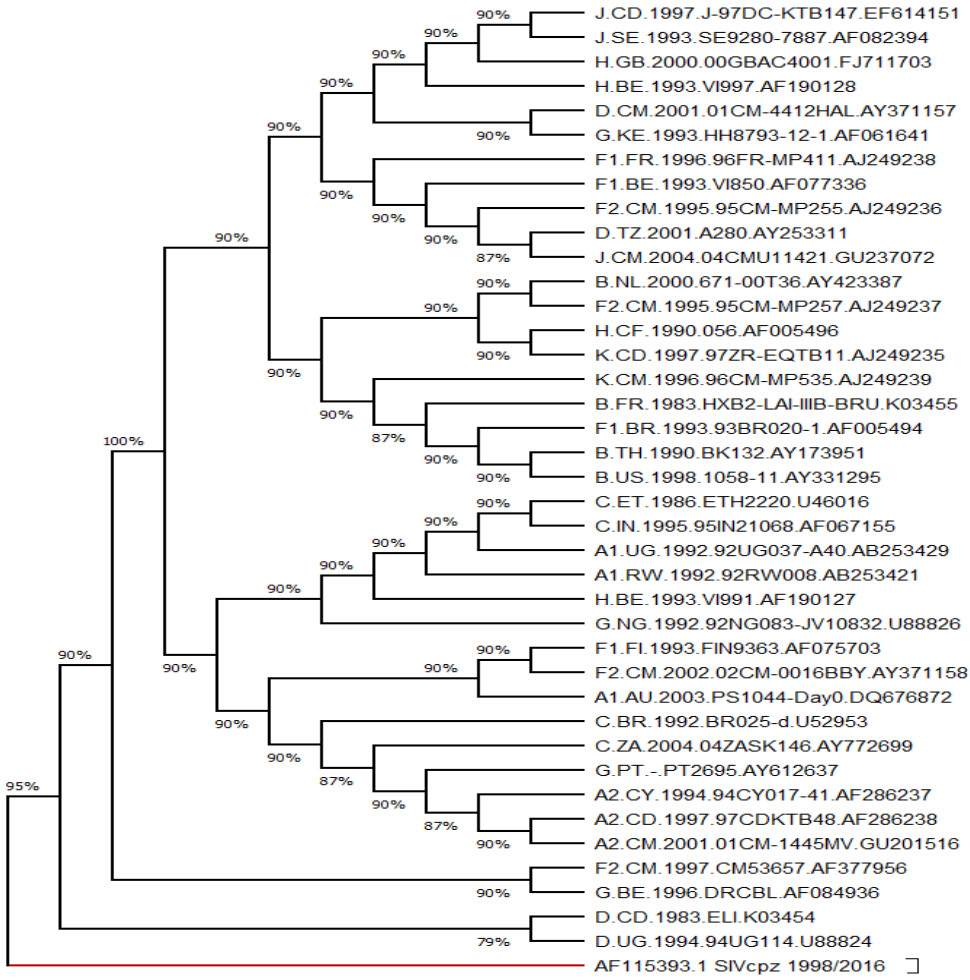


Figure 4.9: Dendrogrammatic representation of the V3 loop sequences. Inferred by maximum likelihood method and JTT matrix based model (Jones *et al*., 1992). Tree with highest likelihood of -991.01 is shown. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using a JTT model. A discrete Gamma distribution was used to model evolutionary rate differences among sites (5 categories (+G, parameter = 1.0308)). Rate variation model allowed for some sites to be evolutionarily invariable ([+I], 1.91% sites). Final tree drawn to scale with branch lengths measured in the number of substitutions per site. Analysis involved 40 amino acids sequences from Los Alamos HIV database using Mega X (Kumar et al., 2018b).

On further analysis on the significance and difference in the two categories of highly conserved motifs of the V3 loop tip i.e. Gly-Pro-Gly-Arg and Gln (GPGR and Q). The outcome was plotted and ratios compared in terms of synonymous (*dS*) and non-synonymous *(dN*) in different subtypes (Figure 4.10). With a P-value threshold of 0.1 a total number of 31 sites were analyzed using Data monkey software (Weaver *et al*., 2018). The output displayed a significant positive selection at five (15) sites and negative selections at eight (18) sites. The HIV-1 subtype B displayed higher ratios of dN/dS as well as higher measures of disorder as compared to other subtypes. The same subtype B was also found to be common for GPGR motif at the V3 loop. From our findings there could be a positive assumption that different subtypes experience varied pressures hence contributing to various classifications of HIV- M group subtypes.

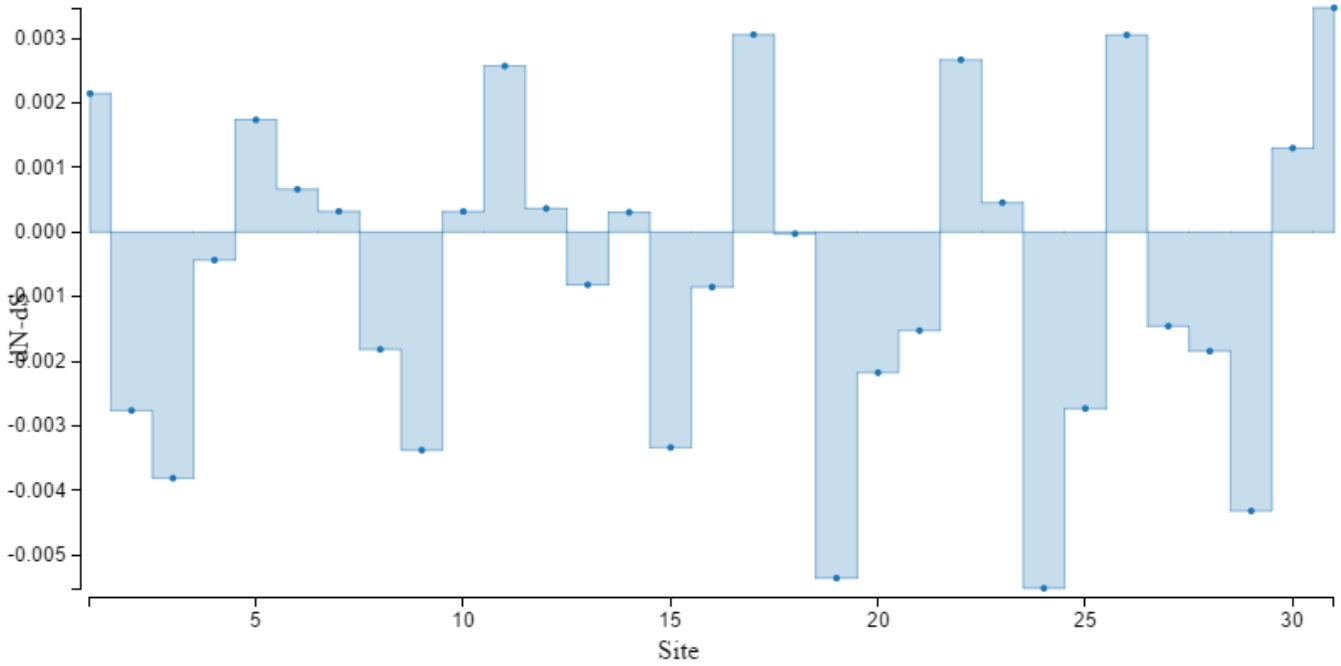


Figure 4.10: Graph showing the ration of dN-dS per site in the analyzed V3 loop sequences**.** Output showing Positive Selection at five sites and Negative Selection at 8 sites

4.3 Android Mobile based application to detect HIV subtypes and HIV-1 Drug Resistance mutations targeting the HIV pol gene

ARVPredictor was designed to take in data on protease, reverse transcriptase, and integrase mutations. It returns inferred levels of resistance to selected proteases, nucleosides, non-nucleosides, and integrase inhibitors for accurate HIV management at the point of care.

By evaluating several sets of reference sequences from the National Center for Biotechnology Information (NCBI) and other related studies, the tool demonstrates its precise significance and usability. This is based on multiple test results using a test sequence dataset (Macharia, 2016) as presented in Table 4.2.(Rhee, 2003).

Table 4.2: Test sequence data set: Accession No: KX505361.1: HIV-1 isolates 5873 from Kenya pol protein (pol) gene, partial cds.

AATGGCCATTGACRGAAGAAAAAATAAAGGCATTGATAGAAATTTGTACAGAGATGGAAAAGGAAGGAA AAATTTCAAGAATTGGGCCTGAGAATCCATACAATACTCCAGTATTTGCCATAAAAARGAAAGACAGTACT AAGTGGAGAAAATTAGTAGATTTCAGGGAACTCAATAAAAGAACCCAAGACTTTTGGGAAGTTCAATTAG GRATACCACACCCAGCAGGGTTAAAARAGAAAAAATCAGYGACAGTACTAGATGTGGGGGATGCRTATTT TTCAGTWCCTTTAGATGAAAGCTTCAGGAAATATACTGCATTYACCATACCRAGTRTAAACAATGAGACA CCAGGAATCAGRTATCAGTACAATGTGCTTCCACAAGGATGGAAAGGATCACCRGCAATATTCCAAGCTA GCATGACAAAAATYCTGGAACCTTTTAGGAAACAAAATCCAGAAATGATTATCTATCAATACATGGATGA TTTGTATGTAGGATCTGACTTAGAAATAGGGCAACATAGAGCAAAAATAGAGRAATTAAGGGAACACCT GTTAAAGTGGGGGTTTACTACACCAGACAAAAAGCATCAGAAAGAACCTCCAYTCCTTTGGATTGGTTAT

In Figure 4.11, there is a pictorial comparison of sampled results for the *ARVPredictor* against the gold-standard Stanford HIV Database.

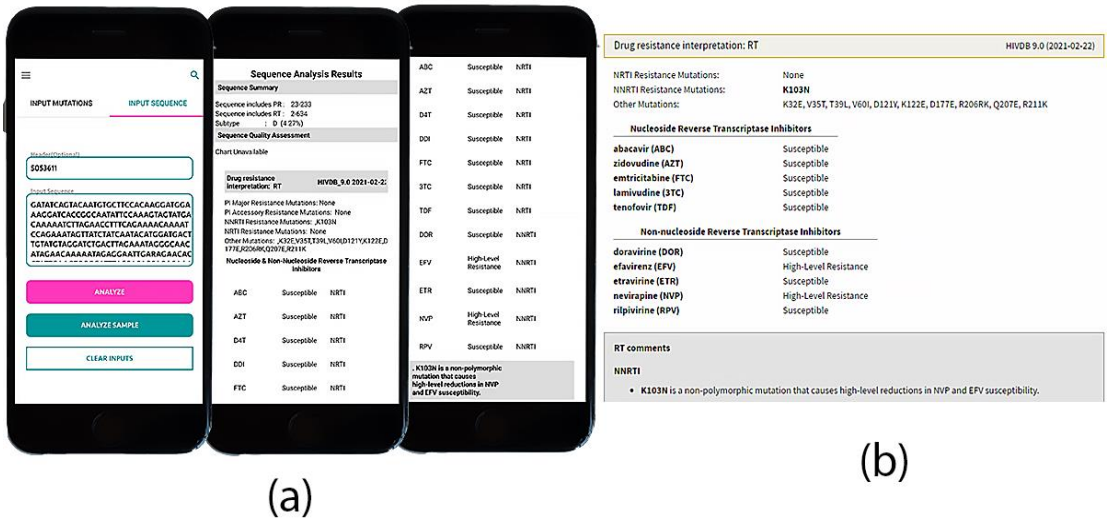


Figure 4.11: Sequence analysis results: Demonstrating similarity between sequence analysis results output for both (a) *ARVPredictor* and (b) Stanford HIV Database.

For mutation analysis output, the study tabulates the analysis results of the predefined HIV-1 mutation M184V as per the WHO Major HIV-1 Drug Resistance Mutations list (Bennett et al., 2009). For both the ARVPredictor and Stanford Database, the most prevalent resistance mutations occurred in nucleoside reverse transcriptase inhibitors (NRTIs) in vitro. Likewise, there is a high degree of resistance to emtricitabine (FTC) and lamivudine (3TC), as well as a potentially low level of resistance to didanosine (DDI), with susceptibility to zidovudine (ZDV) and tenofovir (TEN) (TDV). The figures in Figure 4.12 exhibit equivalent results.

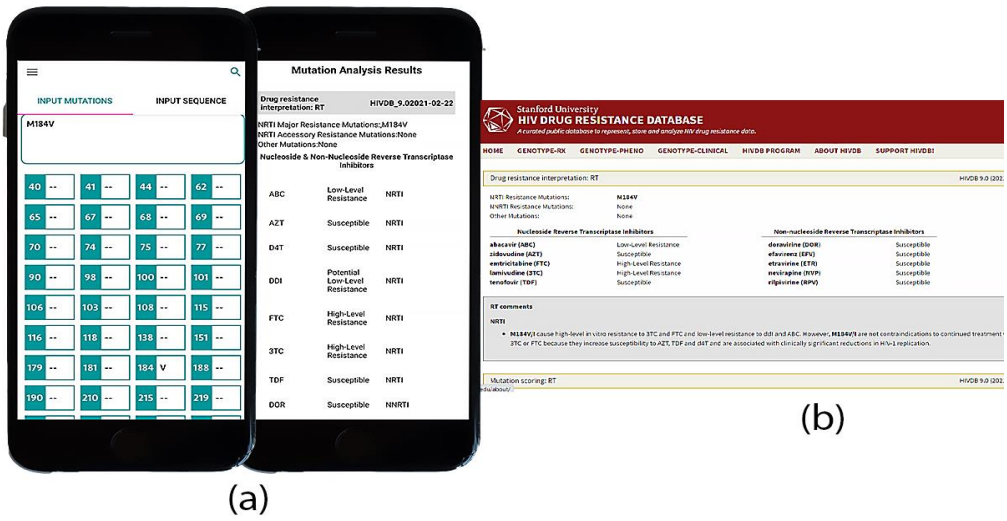


Figure 4.12: Mutation Analysis Results. Demonstrating similarity between mutation analysis results for (a) *ARVPredictor* and (b) Stanford HIV Database.

4.3.1 Test Performance and Agreement

Table 4.3 summarizes sequences with variation in subtype and mutations as determined by both the *ARVPredictor* app and the Stanford Database during test performance. The *ARVPredictor* app identified similar HIV subtypes in 98/100 sequences compared with the Stanford HIV Database (κ=0.98, indicating near perfect agreement). There were 89/100 major NNRTI and NRTI mutations identified by *ARVPredictor*, similar to the Stanford HIV Database (κ=0.89, indicating near perfect agreement). Seven mutations classified as major mutations by the Stanford HIV Database were classified as other mutations by *ARVPredictor*. This further indicates that the Stanford-confirmed GraphQL web service works fairly well, and all the results are in sync with most parts of the web version. Both tools found several minor/other mutations, but depending on small phone display window, some may be hidden from view.

Table 4.3: Test Performance Output: Performance results of *ARVPredictor* using the Stanford HIV database as the gold standard in identification of HIV subtypes and mutations **(B. Ongadi et al., 2022)**.

	Stanford HIV Database						©ARVPredictor					
			RT Mutation Output						RT Mutation Output			
SeqID	HIV Subtype	Susceptibility	NNRTI	NRTI		Other Mutations	HIV Subtype	Susceptibility	NNRTI	NRTI		Other Mutations
				TAMs	Non-TAMs					TAMs	Non-TAMs	
1	A	Susceptible	**V179T**	None	None	E28EA, V35T, E40D, V60I, K122E, D123G, F214L	A	Susceptible	**V179T**	None	None	F214L
6	A	**NNRTI**	**K103KN**	None	None	K32KN, V35T, K43E, V60I, K122E, D123G, I135T, K173I, Q174K, V179I, T200A, Q207A, R211S	A	**NNRTI**	**K103KN**	None	None	R211S
9	B	**NNRTI, NRTI**	**V106VA, F227FL, M230I**	None	**K65KR**	V35I, K49R, K102KE, K122E, D123S, I135IV, S162A, D177E, I178M, V179I, T200A, E204EK, Q207E, R211K	C+D	**NNRTI, NRTI**	**V106VA, F227FL**	None	**K65KR**	M230I
10	C + D	**NNRTI, NRTI**	**V106A**	None	**M184V**	V35I, K49R, K102E, K122E, D123S, S162A, T165K, D177E, I178M, V179I, T200A, Q207G, R211K	C+D	**NNRTI, NRTI**	**V106A**	None	**M184V**	R211K
14	D	**NNRTI, NRTI**	**A98G, Y181C, H221HY**	None	**M184V**	V35T, E40D, K43R, K49R, V60I, D121Y, K122E, D123E, I135T, D177E, I178M, I195L, T200I, E203EK, E204K, Q207E, R211K	D	**NNRTI, NRTI**	**A98G, Y181C**	None	**M184V**	H221HY
18	A	Susceptible	None	None	None	V35T, T39N, V60I, K122E, D123S, S162A, K173S, Q174K, D177E, T200I, I202V, Q207D, R211K	A	Susceptible	None	None	None	R211K
30	A	**NNRTI, NRTI**	**G190A**	None	**M184V**	V35T, T39E, I142T, T165A, K173S, D177G, V179I, I202V, Q207A, R211S, P226PR	A	**NNRTI, NRTI**	**G190A**	None	**M184V**	P226PR
31	B	**NNRTI**	**V179VD**	None	None	V35T, E40ED, V60I, K122E, K173A, D177E, T200A, Q207E, R211RK	B	**NNRTI**	**V179VD**	None	None	R211RK
32	D	**NNRTI, NRTI**	**Y181YC**	None	**V75VM**	V35T, T39K, K49R, V60I, K64R, K122E, D123G, I135IMR, D177E, I178M, T200I, E203G, Q207E, R211K	C+D	**NNRTI, NRTI**	**Y181YC**	None	**V75VM**	R211K
33	A	Susceptible	None	None	None	V60I, K122E, D123N, K173T, Q174K, D177E, V179VI, T200A, Q207A, R211S	A	Susceptible	None	None	None	R211S
34	B	Susceptible	None	None	None	V35T, E40D, K49R, V60I, Q174K, D177E, Q207E	B	Susceptible	None	None	None	Q207E
37	A	**NNRTI**	**Y181YFIN**	None	None	V35T, V60I, V90VI, K122E, D123S, I135T, I142V, K173S, Q174K, D177E, V179I, Q207A, R211S	A	**NNRTI**	**Y181YFIN**	None	None	R211S
40	G	**NNRTI**	**Y181C, H221Y**	None	None	V35I, V60I, K122E, D123N, I135V, K173R, Q174E, D177E, I178M, T200A, Q207K, R211N	G	**NNRTI**	**Y181C**	None	None	H221Y
41	A	Susceptible	None	None	None	V35T, T39I, V60I, D121H, I135T, K173S, Q174N, D177E, I195L, T200A, Q207A, R211S, F214L	A	Susceptible	None	None	None	F214L
45	A	Susceptible	None	None	None	V35T, T39L, V60I, D121H, K122E, I135T, K173L, Q174K, D177E, V179I, Q207A, R211S	A	Susceptible	None	None	None	R211S
46	A	Susceptible	**V179T**	None	None	V35T, K49R, V60I, K122E, D123G, I135T, K173L, Q174K, D177E, I178V, I202V, Q207D, R211S, F214L	A	Susceptible	None	None	None	F214L
47	D	Susceptible	None	None	None	V35T, T39I, E40D, K49R, V60I, D121Y, K122E, I135T, F171Y, K173L, Q174K, D177E, V179I, Q207A, R211S	D	Susceptible	None	None	None	R211S
50	CRF02_AG	**NNRTI**	**K103N, F227FL**	None	None	V60I, V90I, I135A, I142V, S162A, K173KT, Q174E, T200A, Q207E, L228Q, V245Q, D250E	CRF02_AG	**NNRTI**	**K103N, F227FL**	None	None	D250E
51	A	Susceptible	None	None	None	V35T, T39Q, S105A, K122E, D123N, I135T, K173A, D177G, V179I, T200A, I202V, Q207A, R211Q	A	Susceptible	None	None	None	R211Q
52	A	**NNRTI, NRTI**	**K101E, E138A, G190A**	**F77FL, T215TAS**	M184V	Q85P, K122E, D123N, I135T, I142V, K173S, Q174K, V179I, T200A, Q207A, R211S, F214FL	A	**NNRTI, NRTI**	**K101E, E138A, G190A**	**F77L**	M184V	T215TAS
53	D	Susceptible	None	None	None	V35T, E40D, I47L, K49R, V60I, D121C, K122E, D123E, R125RK, K173E, D177E, T200A, Q207E	D	Susceptible	None	None	None	Q207E
54	C+D	Susceptible	None	None	None	V35I, K49R, K102KE, K122E, D123S, I135IV, S162A, D177E, I178M, V179I, T200A, E204EK, Q207EG, R211K	C+D	Susceptible	None	None	None	R211K
55	A	Susceptible	None	None	None	V35T, T39Q, S105A, K122E, D123N, I135T, K173A, D177G, V179I, T200A, I202V, Q207A, R211Q	A	Susceptible	None	None	None	R211Q
56	A	**NNRTI, NRTI**	**A98G, Y181C**	**L210*W, T215HNY**	**M184V,**	K122E, D123S, T128TP, I135T, T139R, K173L, Q174*K, N175NFIY, P176PFLS, D177*E, I178IK, V179I, T200A, I202IM, E203A, Q207A, R211S	A	**NNRTI, NRTI**	**A98G, Y181C**	**L210W**	None	**T215HNY**
57	A	Susceptible	None	None	None	V35T, K104R, D121H, I135T, S162Y, K173L, Q174K, D177E, Q207A, R211S	A	Susceptible	None	None	None	R211S
58	A	**NNRTI, NRTI**	**A98G, K101E, V106VI, V179T, Y181C, G190S**	None	**M184V**	D121H, K122E, K173A, Q174K, D177E, I178M, G196E, T200A, I202IM, E203EDGV, Q207A, R211N	A	**NNRTI, NRTI**	**A98G, K101E, V106VI, V179T, Y181YC**	None	**M184V**	R211N
61	D	**NNRTI, NRTI**	**V108I, Y181YC**	**T215TN**	None	K32E, V35T, T39L, V60I, D121Y, K122E, D177E, R206RK, Q207E, R211K	D	**NNRTI**	**V108I, Y181YC**	None	None	**T215TN**
62	A	**NNRTI**	**G190A**	None	None	V35T, V60I, K122E, D123S, I135T, I142V, F171Y, K173S, Q174K, D177E, V179I, T200A, Q207A, R211S	A	**NNRTI**	**G190A**	None	None	R211S
85	D	Susceptible	None	None	None	V35T, K49R, V60I, D121Y, K122E, K173KQR, D177E, I180IV, E194D, T200TI, Q207E, R211K	D	Susceptible	None	None	None	R211K
88	A	Susceptible	None	None	None	V35T, V60I, K122E, D123NS, I135IT, K173S, Q174K, D177E, V179I, I202V, Q207D, R211S	A	Susceptible	None	None	None	R211S
89	B	**NNRTI**	**G190A, F227FL**	None	None	S48T, V60I, K122E, D123S, I135T, T139A, D177E, Q207E, R211K	B	**NNRTI**	**G190A**	None	None	F227FL
90	A	Susceptible	None	None	None	V35T, T39M, V60I, K64R, D121H, K122E, I135T, A158S, S162A, K173S, Q174K, D177E, I178L, I180IM, T200A, Q207A, R211S	A	Susceptible	None	None	None	R211S
91	D	**NNRTI, NRTI**	**V108I, Y181YC**	**T215TN**	None	V35T, T39I, E40D, K49R, V60I, W88WG, D123E, D177E, T200A, E204Q, Q207A	D	**NNRTI, NRTI**	**V108I, Y181YC**	None	None	**T215TN**
92	A	Susceptible	None	None	None	E28A, V35T, T39L, K122E, D123E, T131TP, I135T, K173S, Q174K, D177E, V179I, T200A, Q207A, R211S	A	Susceptible	None	None	None	R211S
100	C	**NNRTI**	**K103N, V106I**	None	None	V35T, T39R, K43E, S48T, K102R, K122E, I142V, K166R, K173A, Q174R, D177E, I195L, G196K, T200A, Q207E, R211K	C	**NNRTI**	**K103N, V106I**	None	None	R211K

4.3.2 ARVPredictor Availability

ARVPredictor is currently distributed freely through the Google Play Store and App Store (Apple Inc.), with basic rules requiring users to create an account and provide very limited personal details such as email addresses, passwords, and preferred

telephone numbers. Different user-friendly interfaces viewable through the usage of the app are shown in Figures 4.13 through to Figure 4.17. The source code for *ARVPredictor* is also available under the MIT permissive license in a GitHub repository (Bongadi, 2020/2020).

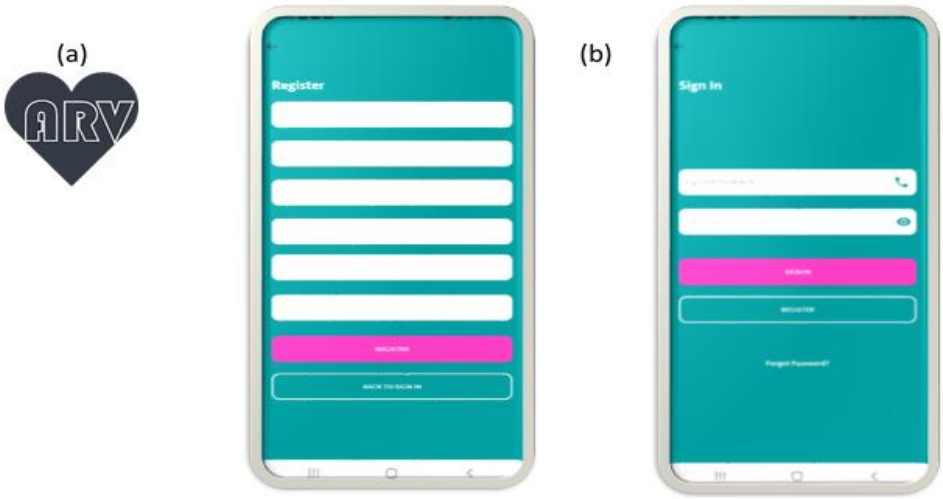


Figure 4.13: *ARVPredictor* Preliminary User Interfaces. The pictorial icon (a) shows how *ARVPredictor* appears ready for download from the app/play store. Potential users must register in order to use the application, registered users are required to sign-in (b).

4.3.3 User Friendly Interfaces

4.3.3.1 User Registration and User login

Registration is required for first-time users of the app. This procedure helps protect the app from illegal access and use. An SMS text message alert containing a one-time activation code is then delivered to the provided phone number. The system can be registered in one of two ways: through the app itself as shown in Figure 4.13 or by the administrator remotely on request. Figure 4.14 shows the backend code that was used to create this registration process. Only authenticated and valid email addresses/phone numbers are allowed in this process for registration.

```
call.enqueue(new Callback<Register>() {
    @Override
    public void onResponse(@NotNull Call<Register> call, @NotNull Response<Register> response) {
        Register respo = response.body();

        if (respo != null) {
            UserData userData = respo.getData();

            if (respo.getToken().equals("NONE")) {
                Message.makeToast(activity, activity, respo.getMessage());
            } else {

                if (userData != null) {
                    userDataBox.removeAll();
                    userDataBox.put(userData);
                    Session.sessionStoreData(respo.getToken(), userData.getFirstName(), userData.getSecondName(),
                            userData.getPhoneNumber(), userData.getEmail(), userData.getStatus().toString(), activity, activity);
                    StaticVariables.first_name = userData.getFirstName();
                    StaticVariables.email = userData.getEmail();

                }
                onLoginFormActivityListener.doLogin();
                Message.makeToast(activity, activity, respo.getMessage());
                PreferenceManager preferenceManager = new PreferenceManager(activity);
                preferenceManager.setLoginStatus(true);

            }

        } else {
            Message.makeToast(activity, activity, message: "Response is null!");
        }
```

Figure 4.14: User Registration and Sign in source code: (A Set of computer logical instructions aiding registration and sign in process before using the app).

4.3.3.2 Antiretroviral Drug Options/Input Window

The next button opens the known and common antiretroviral drug display window (Figure 4.15). This only occurs after successful registration and verification. Some antiretroviral drugs will be premarked by default, but checkboxes can be used to add or remove any drug. A "save" button at the bottom of the phone screen allows one to save and use his/her final choice

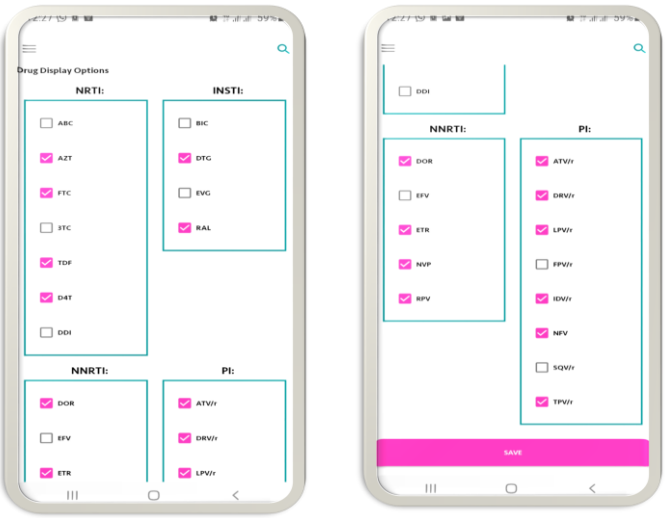


Figure 4.15: Antiretroviral Display Options: Displaying all current ARVs and categorizes them as Nucleoside reverse transcriptase inhibitors (NRTIs), Non-nucleoside reverse

transcriptase inhibitors (NNRTIs), Integrase Strand Transfer Inhibitor (INSTIs) and Protease Inhibitor (PI).

Figure 4.16 illustrates the back end code used in the development of this app's preferred drug selection process.

```
try{
    holder.drug.setText(data.get(position).getName());
    holder.drug.setChecked(data.get(position).getSelected());
    holder.drug.setTag(position);
    holder.drug.setOnClickListener(new View.OnClickListener(){

        @Override
        public void onClick(View view) {
            Integer pos = (Integer) holder.drug.getTag();
            String name = data.get(pos).getName();
            if (preferenceManager.getDrugStatus(name)){
                preferenceManager.setDrugSelected( selected: false,name);
            }
            else {
                preferenceManager.setDrugSelected( selected: true,name);
            }
        }
    });
}catch (Exception e){
    e.printStackTrace();
}
```

Figure 4.16: ARV Drug Selection code by category. (A Set of back-end computer related logical instructions for selection of different ARV drugs available for use by various HIV Patients).

4.3.3.3 HIV Mutation Selection Window

The subsequent screen displays two (2) options of the input type for analysis: point mutations or sets of sequences (Figure 4.17). The first group is a list of all the registered major HIV-1 drug-resistance mutations as per WHO 2009 data (Bennett *et al*., 2009). Three mutation categories, namely, reverse transcriptase, protease, and integrase, can be selected from a scroll down window and analyzed at the bottom of the screen. The second part of this figure is a sequence input window. It provides an option to either upload a set of sequences from a separate source or manually key them in.

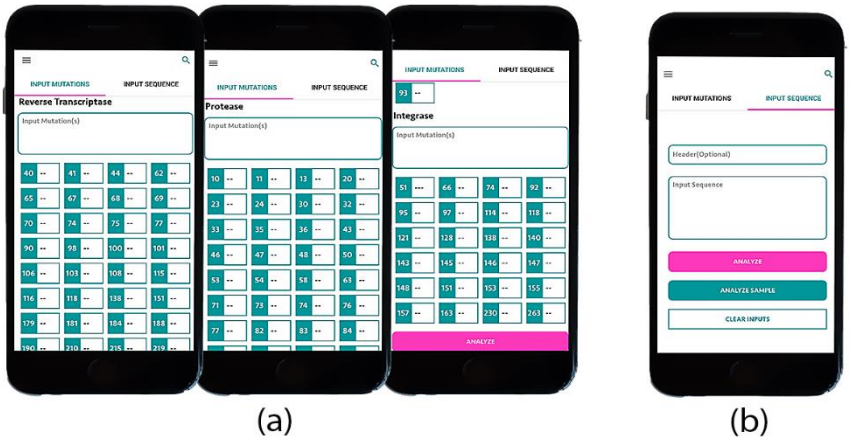


Figure 4.17: *ARVPredictor* (a) Major HIV-1 drug resistance mutation list. (b) Sequence entry option window.

4.3.3.4 ARVPredictor Mutation Analysis Results Window

The results are analyzed and sent back in comparison to the latest version of the Stanford HIV Database. The output packaged in terms of the region targeted, for example, protease, reverse transcriptase, and integrase, and preferred antiretroviral listed alongside Figure 4.18.

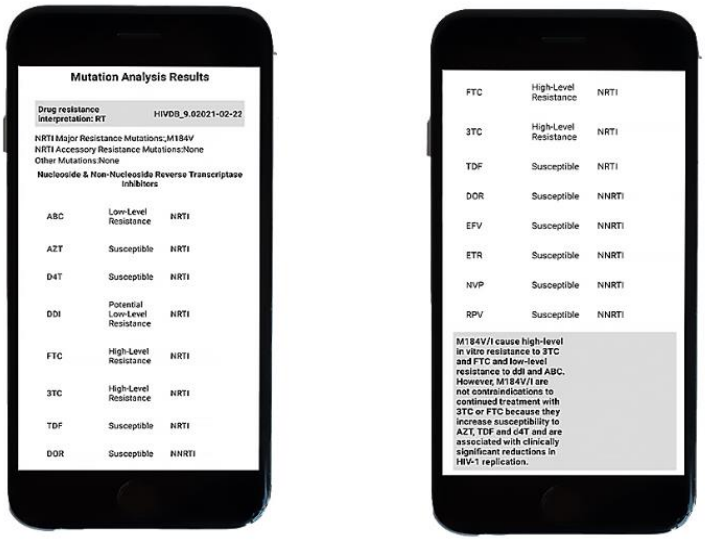


Figure 4.18: *ARVPredictor* Mutation Analysis Results: **(**Including the latest version of Stanford db. referenced, Major/Minor Mutations identified and ARVs susceptibility levels)

The development code for displaying the mutation analysis output in this app is shown in Figure 4.19.

```
myApolloClient.query(MutationAnalysisRequestQuery.builder().mutations(myMutations).build())
        .enqueue(new ApolloCall.Callback<MutationAnalysisRequestQuery.Data>() {
            @Override
            public void onResponse(@NotNull Response<MutationAnalysisRequestQuery.Data> response) {
                getActivity().runOnUiThread(
                        new Runnable() {
                            @Override
                            public void run() {
                                List<MutationAnalysisRequestQuery.DrugResistance> load = response.getData().mutationsAnalysis().drugResistance();
                                progDialog.dismiss();
                                if (load != null){
                                    Paper.book().write("MUTRESULT",load);
                                    Paper.book().write("DATA",response.getData());
                                }
                                myMutations.clear();
                                Intent intent = new Intent(getActivity(), MutationResultActivity.class);
                                startActivity(intent);
                                  getActivity().finish();
                            }
                        }
                );
            }
            @Override
            public void onFailure(@NotNull ApolloException e) {
                getActivity().runOnUiThread(
                        new Runnable() {
                            @Override
                            public void run() {
                                myMutations.clear();
                                progDialog.dismiss();
                                errorDialog.show();
                            }
                        }
                );
            }
        });
```

Figure 4.19: *ARVPredictor* Mutation Analysis Code: Set of back-end computer related logical instructions for analyzing selected set of HIV mutations.

Figures 4.20 and 4.21 display the analyzed sequences and respective back end development codes.

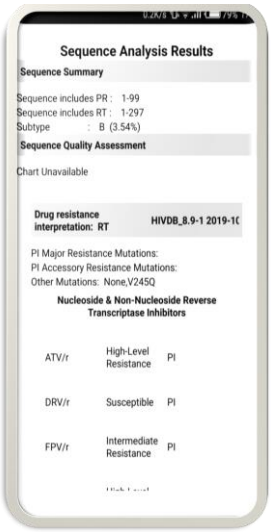


Figure 4.20: *ARVPredictor* Sequence Analysis Results. (Including the latest version of Stanford db. referenced, Position sequenced, Major/Minor Mutations identified, HIV Subtype and ARVs susceptibility levels)

```
myApolloClient.query(SequenceAnalysisRequestQuery.builder().sequences(actualInputs).build())
        .enqueue(new ApolloCall.Callback<SequenceAnalysisRequestQuery.Data>() {
            @Override
            public void onResponse(@NotNull Response<SequenceAnalysisRequestQuery.Data> response) {
                getActivity().runOnUiThread(new Runnable() {
                    @Override
                    public void run() {
                          sequence.setText(response.getData().toString());
                        Paper.book().write("SEQDATA",response.getData().sequenceAnalysis());
                        Paper.book().write("RAWDATA",response.getData());
                        progDialog.dismiss();
                        header.setText("");
                        sequence.setText("");
                        actualInputs.clear();
                        Intent intent = new Intent(getActivity(), SequenceAnalysisResults.class);
                        startActivity(intent);
                    }
                });
            }
            @Override
            public void onFailure(@NotNull ApolloException e) {
                  sequence.setText(e.getMessage());
                getActivity().runOnUiThread(new Runnable() {
                    @Override
                    public void run() {
                        header.setText("");
                        sequence.setText("");
                        header.setText("");
                        sequence.setText("");
                        progDialog.dismiss();
                        errorDialog.show();
                    }
                });
            }
        });
```

Figure 4.21: *ARVPredictor* Sequence Analysis code. : Set of back-end computer related logical instructions for analyzing HIV sequences.

CHAPTER FIVE

DISCUSSION

The ultimate aim of this study was to determine the genetic polymorphism in the CCR5Δ32 allele through *in silico* approach and also use mobile technology to evaluate HIV drug resistance. It largely intended to figure out how the CCR532 allele is distributed among Caucasians, Asians, and Africans across the globe. The research herein correlated data from studies on the distribution of CCR5Δ32 which is a natural selection allele acting in humans against HIV as previously demonstrated (Martinson *et al*., 1997). The result of this analysis involving 37 articles with 17,535 participants from diverse backgrounds sheds great lights on the distribution of this allele globally as well its association with HIV-1 epidemiology. The study demonstrates that there is a wide knowledge gap on CCR5Δ32 especially in African where HIV burden is highest but further confirms that CCR5Δ32 heterozygosity does not protect individuals against HIV-1 infection but rather slows progression of the disease (Mehlotra *et al*., 2015; Rahimi *et al*., 2014). High concentration of studies on CCR5Δ32 is seen among the Caucasians. Globally, 51% of the publications accessible for review were on Caucasians, 24% were on Asians, and five on Africans, accounting for meager 13%. The overall outcome for all the populations meta-analyzed indicates that race can be a factor that determines CCR5Δ32 homozygosity or heterozygosity and it highly favors the Caucasians. This was demonstrated by the fact that 52 out of the 57 individuals' found to be positive of the allele were Caucasians. However the few homozygotes seen among the Asians could have been as a result of gene flow and may need further analysis (Martinson *et al*., 1997). Notably, no one in the African population was positive for the allele, whereas one individual was found in the remaining un-grouped population in Nigeria, according to Martinson and group. (Martinson *et al*., 1997). Based on the data available online, it's probable that the findings of this study are regionally and or racially biased. Another factor that may have influenced the findings is the language bias, particularly on articles published in languages other than English without a translating link.

The research team equally determined the divergence in geographical distribution of HIV-1 group M subtypes globally in this investigation, which is consistent with earlier studies (Buonaguro et al., 2007). Studies have shown that some HIV-1 subtypes poses phenotypic differences from refined changes in the envelop structure especially within the third hypervariable domain (V3 loop) (Lynch et al., 2009). This is possible since single genome amplification sequencing of HIV-1envelop gene mostly within the variable (V) is used as a maker for HIV 1 genome diversity (Rao et al., 2013) (Gall et al., 2013).

HIV-1 group M is characterized by ten (10) known clades ranging from A to K besides additional Unique Recombinant Forms (URF) and Circulating Recombinant forms (CRF). Group M is listed as being responsible for HIV pandemic word over. However, some subtypes are seen to be predominant within specific areas. Subtypes A and D are mostly found in East and Central part of Africa. Subtype A is also common among some Asian community especially in Pakistan, Kazakhstan and Cyprus. Subtype B is leading in North and Latin America (Jamaica, Dominican Republic, Paraguay, Ecuador, Venezuela, Mexico, and Haiti) and Asia (Japan, Hong Kong, Philippines and South Korea). Although Subtype C is most frequent in southern Africa (South Africa, Botswana, and Malawi), it has also been found in India, Israel, Nepal, and Georgia, while Subtype G has been identified in Cuba and Guinea Bissau. Sub-Saharan Africa has a comparable distribution of almost all the available HIV-1 subtypes. The recombinants of subtypes A, G, K and J are common in the West African countries (Mali, Niger, Gambia, Senegal, and Burkina Faso). The result of these studies opens up a non-conclusive debate on the origin of HIV-1. The subsequent dendrogram described herein may not be conclusive due to limitation of data retrieved from the Los Alamos HIV database with complete sequences covering the entire HIV env and by extension the HIV -1 gp160 protein. However, it gives a clear indication that there is no visible connection between a particular subtype and its transmission pattern. Nevertheless, some studies have shown that various risk behaviors support HIV transmission modes within and across regions and generated multi transmission routes also fuel the spread of the HIV epidemic (Buonaguro *et al*., 2007).

The alignment of the highly conserved V3 loop sequences equally confirms documented diversity that exists within HIV-1 group M subtypes (Araújo & Almeida, 2013). The greatly volatile loop under selective pressure of thirty-five (35) residues is believed to be largely responsible for the differences within and across subtypes. An estimated percentage variation of between 12 and 20 percent within specific subtypes and 35 percent across some subtypes was also determined. The differences seen within these subtypes may be a factor contributing to HIV-1 recombinants, the rate of HIV infections and transmission hosts. Studies should be carried out to explore possible relationship between subtypes and host specific transmissions for potential antiviral interventions. This is because the V3 loop participates directly in HIV viral entry and therefore has the possibility of displaying more insight on HIV-1 inter-subtype prevalence among different populations. According to the HIV sequence compendium from 2018, it is relatively conserved, with no significant deletions, insertions, or glycosylation alterations (Foley et al., 2018)

This study shows a close similarity between consensus sequences of HIV-1 subtypes A and C in comparison to others. These near identical subtypes also pose a higher threat of HIV infection and are seen to be highly predominant among the African populace. However, high levels of genetic variation have been displayed by HIV-1 subtypes of group M after identification of sites changing under positive or negative selection in the gp160. The phylogenetic representation of the V3 region does not show a uniform distribution of HIV-1 subtypes globally as compared to the whole *env* sequences. However the Simian immunodeficiency virus of chimpanzees (SIVcpz) sequences still remains the out-group in both cases hence the probable ancestor. This remains in line with previously documented studies on perceived origin of HIV-1 (Sharp & Hahn, 2011) (Hirsch et al., 1989; Huet et al., 1990).The study equally displays a significant difference in the two categories of highly conserved motifs of the V3 loop tip i.e. Gly-Pro-Gly-Arg and Gln (GPGR and Q). HIV-1 subtype B has higher ratios of dN/dS as well as higher measures of disorder as compared to other subtypes. The same subtype B was also found to be common for GPGR motif at the V3 loop. Different subtypes experience varied pressures hence contributing to various classifications of HIV-1 group M subtypes and their global distribution among different population.

Finally, providing timely and accurate help to health care providers in the management of HIV patients has proven difficult, particularly in resource-constrained settings. It is believed that with the release of *ARVPredictor*, a much-needed solution for health-care practitioners at the point of care will be realized. The desired work environment and data output were taken into consideration when planning and selecting tools for the creation of this mobile application. This choice supported quick analysis of the mutations or sequences to be uploaded by the health care provider. The overall aim is to provide a solution that offers accurate and easily accessible management strategy to HIV health care providers with a short turnaround time. For accuracy and time management, every attempt was taken to reduce the predicted input variables. The mutation screen achieves this by presenting a dropdown and choice-enabled entry that includes a comprehensive list of all currently documented mutations. The sequence analysis window allows keying in, pasting, and uploading from a remote file. The mobile application, which is now freely downloadable from Google Play Store or App Store and enlisted as *ARVPredictor*, is free for use and technically supported by developers. Its security is enhanced through controlled and authenticated log-in process, while critical and back end data are only accessible to authorized individuals. The application administrator will handle any anticipated mobile application-related errors or challenges after reviewing various levels of the application error logs.

In evaluating the functionality of the *ARVPredictor* in comparison to the replicated Stanford HIV Database in this study, there is strong evidence that several benefits, including but not limited to concurrence, convenience, and simplicity, are realized. *ARVPredictor* can therefore be used to determine HIV-1 drug-resistance mutations in the HIV pol gene with ease and convenience in mobile devices. The app equally has the advantages of high-speed data networks and smartphone accessibility.

It therefore adds to other available and upcoming mobile health (mHealth) interventions in the area of HIV and antiretroviral use among different health care providers. Mobile health technology has enabled faster and efficient communication among health care service providers and their patients as well as reducing frequent and high cost of hospitals visits. However, it's additional benefits, such as improved

diagnosis, accuracy, and better coordination, will still contribute highly to the biomedical field.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 General Conclusions

In this study two forest plots were drawn; the first one comparing the distribution of CCR5 homozygosity and heterozygosity among various populations and the second detailing the comparison of CCR5 homozygosity and heterozygosity among HIV positive and HIV negative individuals. Both plots demonstrate higher concentration of studies on CCR5Δ32 among the Caucasians followed by Asians and lastly Africans. Further details on the output reveals that there is a higher likelihood of being homozygous of the allele when a Caucasian than other. Most CCR5Δ32 homozygotes identified in this study were Caucasians.

While comparing the CCR5 homozygosity and heterozygosity among HIV positive and HIV negative individuals, we conclude that; Caucasians as compared to other populations are less susceptible to HIV virus infection due to the expression rates of CCR5Δ32 while the rest of the populations experience a much higher prevalence of the disease. However, due to a wide knowledge gap there could be a possibility that the results in our study are regionally and or racially biased based on the available data.

In order to achieve its stated goals, this study also agrees with other research on two key points: HIV-1 has a number of subtypes, some of which are shown to be more prevalent than others in different parts of the world. The diversity that has been seen within HIV 1 group M subtypes is supported by the highly conserved V3 loop sequences.

Finally, this study reports a novel innovation of a mobile application known as *ARVPredictor* which can be installed freely from the PlayStore. This novelty can accurately define HIV-1 drug resistance mutations targeting the HIV *pol* gene and provides appropriate antiretroviral drugs for use at the point of care. It taps into the benefits of current high-speed data networks and smartphone accessibility. *ARVPredictor* adds to

other available and upcoming mobile health interventions in the area of HIV and antiretroviral use among different care providers. The application can also be scaled up to include other life threatening diseases and aids in reducing drug resistance.

6.2 General Recommendations

1. Further collaborative studies on population genetics and epidemiology can also be explored to help understand more about the global distribution of CCR5Δ32 in Africa.
2. More studies on the near minimal percentage difference between subtypes A and C with the aim of any possible antiretroviral or vaccine development hints should be explored.
3. Additional and continuous modification of the mobile application (*ARVPredictor*) should be explored to take care of the first evolving mobile technology and diversity of HIV/AIDS and cover other diseases.

REFERENCES

Abongwa, L. E., Nyamache, A. K., Torimiro, J. N., Okemo, P., & Charles, F. (2019). Human immunodeficiency virus type 1 ((HIV-1) subtypes in the northwest region, Cameroon. *Virology Journal*, *16*(1), 103. https://doi.org/10.1186/s12985-019-1209-6

AIDS, A. (1986). Clinical Manifestations of HIV Infection. In *Confronting AIDS: Directions for Public Health, Health Care, and Research*. National Academies Press (US). https://www.ncbi.nlm.nih.gov/books/NBK219125/; Accessed 2020-09-22

AIDSMAP. (2015). *CD4 cell counts*. Aidsmap.Com. https://www.aidsmap.com/about-hiv/cd4-cell-counts; Accessed on 2020-09-22

Al-Jaberi, S. A., Ben-Salem, S., Messedi, M., Ayadi, F., Al-Gazali, L., & Ali, B. R. (2013). Determination of the CCR5Δ32 frequency in Emiratis and Tunisians and the screening of the CCR5 gene for novel alleles in Emiratis. *Gene*, *529*(1), 113–118. https://doi.org/10.1016/j.gene.2013.07.062

Allard, J. P., Aghdassi, E., Chau, J., Salit, I., & Walmsley, S. (1998). Oxidative stress and plasma antioxidant micronutrients in humans with HIV infection. *The American Journal of Clinical Nutrition*, *67*(1), 143–147. https://doi.org/10.1093/ajcn/67.1.143

Al-Mahruqi, S. H., Zadjali, F., Beja-Pereira, A., Koh, C. Y., Balkhair, A., & Al-Jabri, A. A. (2014). Genetic diversity and prevalence of CCR2-CCR5 gene polymorphisms in the Omani population. *Genetics and Molecular Biology*, *37*(1), 7–14. https://doi.org/10.1590/S1415-47572014000100004

Angelis, D. S. A. de, Freire, W. S., Pannuti, C. S., Succi, R. C. de M., & Machado, D. M. (2007). CCR5 genotypes and progression to HIV disease in perinatally infected children. *Brazilian Journal of Infectious Diseases*, *11*(2), 196–198. https://doi.org/10.1590/S1413-86702007000200004

Araújo, L. A. L., & Almeida, S. E. M. (2013). HIV-1 Diversity in the Envelope Glycoproteins: Implications for Viral Entry Inhibition. *Viruses*, *5*(2), 595–604. https://doi.org/10.3390/v5020595

Arts, E. J., & Hazuda, D. J. (2012). HIV-1 Antiretroviral Drug Therapy. *Cold Spring Harbor Perspectives in Medicine*, *2*(4), a007161. https://doi.org/10.1101/cshperspect.a007161

ASHM. (2003). The taxonomy of HIV and primate immunodeficiency viruses. *HIV Management Guidelines*. https://hivmanagement.ashm.org.au/basic-hiv-virology/the-taxonomy-of-hiv-and-primate-immunodeficiency-viruses/

AVERT. (2015, July 20). *History of HIV and AIDS overview*. Avert. https://www.avert.org/professionals/history-hiv-aids/overview; Accessed on 2020-09-21

Awodele, O., Olayemi, S. O., Nwite, J. A., & Adeyemo, T. A. (2012). Investigation of the levels of oxidative stress parameters in HIV and HIV-TB co-infected patients. *The Journal of Infection in Developing Countries*, *6*(01), Article 01. https://doi.org/10.3855/jidc.1906

Bennett, D., Camacho, RJ., Otelea, D., Kuritzkes, DR., Herve, F., Kiuchi, M., Heneine, W., Kantor, R., Jordan, MR., & Schapiro, JM. (2009). *Drug Resistance Mutations for Surveillance of Transmitted HIV-1 Drug-Resistance: 2009 Update*,.

Bertozzi, S., Padian, N. S., Wegbreit, J., DeMaria, L. M., Feldman, B., Gayle, H., Gold, J., Grant, R., & Isbell, M. T. (2006). HIV/AIDS Prevention and Treatment. In D. T. Jamison, J. G. Breman, A. R. Measham, G. Alleyne, M. Claeson, D. B. Evans, P. Jha, A. Mills, & P. Musgrove (Eds.), *Disease Control Priorities in Developing Countries* (2nd ed.). World Bank. http://www.ncbi.nlm.nih.gov/books/NBK11782/

Beynon-Davies, P., Carne, C., Mackay, H., & Tudhope, D. (1999). Rapid application development (RAD): An empirical review. *European Journal of Information Systems*, *8*. https://doi.org/10.1057/palgrave.ejis.3000325

Bharat Bhushan Rewari, Reshu Agarwal, Suresh Shastri, Sharath Burugina Nagaraja, & Abhilakh Singh Rathore. (2017). Adoption of the 2015 World Health Organization guidelines on antiretroviral therapy: Programmatic implications for India. *WHO South-East Asia Journal of Public Health*, *6*(1), 90–93. https://apps.who.int/iris/handle/10665/329608

Bharti, D., Kumar, A., Mahla, R. S., Kumar, S., Ingle, H., Yadav, T., Mishra, A., Raut, A. A., & Kumar, H. (2015). Low prevalence of CCR5-Δ32, CCR2-64I and SDF1-3′A alleles in the Baiga and Gond tribes of Central India. *SpringerPlus*, *4*. https://doi.org/10.1186/s40064-015-1238-6

Bigna, J. J. R., Plottel, C. S., & Koulla-Shiro, S. (2016). Challenges in initiating antiretroviral therapy for all HIV-infected people regardless of CD4 cell count. *Infectious Diseases of Poverty*, *5*(1), 85. https://doi.org/10.1186/s40249-016-0179-9

Biologists Discover Why 10 Percent Of Europeans Are Safe From HIV Infection. (2005). ScienceDaily. https://www.sciencedaily.com/releases/2005/03/050325234239.htm

Blut, G. A. C. B. (Arbeitskreis, & Blood, S. 'Assessment of P. T. by. (2016). Human Immunodeficiency Virus (HIV). *Transfusion Medicine and Hemotherapy*, *43*(3), 203. https://doi.org/10.1159/000445852

Bogart, L. M., Cowgill, B. O., Kennedy, D., Ryan, G., Murphy, D. A., Elijah, J., & Schuster, M. A. (2008). HIV-Related Stigma among People with HIV and their Families: A Qualitative Analysis. *AIDS and Behavior*, *12*(2), 244–254. https://doi.org/10.1007/s10461-007-9231-x

Bongadi. (2020). *Bongadi/ARV_Predictor_App_OngadiBA* [Java]. https://github.com/bongadi/ARV_Predictor_App_OngadiBA; Accessed on 2021-01-02 (Original work published 2020)

Bulanda, B. I., Bongenya, B. I., Chatte, A., Kateba, E. T., Kabasele, J.-Y. D., Omakoy, M. O., Chuga, D., Tshibumbu, C., Mwanaut, I., & Kamangu, E. N. (2020). Molecular Diversity of the Human Immunodeficiency Virus Type 1 in Metropolitan Cities in Central Africa: An

Update of Data. *World Journal of AIDS*, *10*(2), Article 2. https://doi.org/10.4236/wja.2020.102007

Buonaguro, L., Tornesello, M. L., & Buonaguro, F. M. (2007). Human Immunodeficiency Virus Type 1 Subtype Distribution in the Worldwide Epidemic: Pathogenetic and Therapeutic Implications. *Journal of Virology*, *81*(19), 10209–10219. https://doi.org/10.1128/JVI.00872-07

Caruso, J. R., & Swift, C. J. (2023). Florida HIV Safety for Florida Clinical Laboratory Personnel. In *StatPearls*. StatPearls Publishing. http://www.ncbi.nlm.nih.gov/books/NBK589667/

Chavhan Ab, Pawar Ss, Jadhao Rg, & Patil Kg. (2013). Distribution of CC-chemokine receptor-5-Δ32 allele among the tribal and caste population of Vidarbha region of Maharashtra state. *Indian Journal of Human Genetics*, *19*(1), 65–70. https://doi.org/10.4103/0971-6866.112894

Chesney, M. A., Morin, M., & Sherr, L. (2000). Adherence to HIV combination therapy. *Social Science & Medicine (1982)*, *50*(11), 1599–1605. https://doi.org/10.1016/s0277-9536(99)00468-2

Chinen, J., & Shearer, W. T. (2002). Molecular virology and immunology of HIV infection. *Journal of Allergy and Clinical Immunology*, *110*(2), 189–198. https://doi.org/10.1067/mai.2002.126226

Cho, M., Min, X., & Son, H. S. (2022). Analysis of evolutionary and genetic patterns in structural genes of primate lentiviruses. *Genes & Genomics*, *44*(7), 773–791. https://doi.org/10.1007/s13258-022-01257-6

Choudhury, A. (2011). *Waterfall Model/SDLC [online] SDLC Tutorials*.

Chung, M., & Ko, I. (2015). Data-Sharing Method for Multi-Smart Devices at Close Range. *Mobile Information Systems*, *2015*, e931765. https://doi.org/10.1155/2015/931765

Cichocki, M. (2020). *Understanding the HIV Life Cycle*. Verywell Health. https://www.verywellhealth.com/the-hiv-life-cycle-47876

Clavel, F., Guyader, M., Guétard, D., Sallé, M., Montagnier, L., & Alizon, M. (1986). Molecular cloning and polymorphism of the human immune deficiency virus type 2. *Nature*, *324*(6098), 691–695. https://doi.org/10.1038/324691a0

Cohn, S. K., & Weaver, L. T. (2006). The Black Death and AIDS: CCR5-Δ32 in genetics and history. *QJM: An International Journal of Medicine*, *99*(8), 497–503. https://doi.org/10.1093/qjmed/hcl076

Corado, A. de L. G., da Silva, G. A. V., Leão, R. A. C., Granja, F., & Naveca, F. G. (2016). Frequency of CCR5 genotypes in HIV-infected patients in Roraima, Brazil. *The Brazilian Journal of Infectious Diseases*, *20*(3), 314–315. https://doi.org/10.1016/j.bjid.2016.01.001

D'arc, M., Ayouba, A., Esteban, A., Learn, G. H., Boué, V., Liegeois, F., Etienne, L., Tagg, N., Leendertz, F. H., Boesch, C., Madinda, N. F., Robbins, M. M., Gray, M., Cournil, A., Ooms, M., Letko, M., Simon, V. A., Sharp, P. M., Hahn, B. H., … Peeters, M. (2015). Origin of the HIV-1 group O epidemic in western lowland gorillas. *Proceedings of the National Academy of Sciences of the United States of America*, *112*(11), E1343-1352. https://doi.org/10.1073/pnas.1502022112

Deeks, E. D. (2018). Bictegravir/Emtricitabine/Tenofovir Alafenamide: A Review in HIV-1 Infection. *Drugs*, *78*(17), 1817–1828. https://doi.org/10.1007/s40265-018-1010-7

DigitalOcean. (2011). *DigitalOcean – The developer cloud*. https://www.digitalocean.com/; Accessed on 2020-12-18

Drohan D. (2017). *Android.retrofit*. 39. http://www.wit.ie; Accessed on 2020-12-20

Duncan, S. R., Scott, S., & Duncan, C. J. (2005). Reappraisal of the historical selective pressures for the CCR5-Delta32 mutation. *Journal of Medical Genetics*, *42*(3), 205–208. https://doi.org/10.1136/jmg.2004.025346

Dusch, V. (2012). *Nginx—The Webserver you might actually like*. https://www.researchgate.net/publication/231400849_Nginx; Accessed on 2020-12-18

Dybowski, J. N., Heider, D., & Hoffmann, D. (2010). Prediction of Co-Receptor Usage of HIV-1 from Genotype. *PLOS Computational Biology*, *6*(4), e1000743. https://doi.org/10.1371/journal.pcbi.1000743

Ekere, E. F., Useh, M. F., Okoroiwu, H. U., & Mirabeau, T. Y. (2020). Cysteine-cysteine chemokine receptor 5 (CCR5) profile of HIV-infected subjects attending University of Calabar Teaching Hospital, Calabar, Southern Nigeria. *BMC Infectious Diseases*, *20*(1), 5. https://doi.org/10.1186/s12879-019-4737-1

Elbim, C., Pillet, S., Prevost, M. H., Preira, A., Girard, P. M., Rogine, N., Matusani, H., Hakim, J., Israel, N., & Gougerot-Pocidalo, M. A. (1999). Redox and Activation Status of Monocytes from Human Immunodeficiency Virus-Infected Patients: Relationship with Viral Load. *Journal of Virology*, *73*(6), 4561–4566. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC112496/

Ellwanger, J. H., Kulmann-Leal, B., Kaminski, V. de L., Rodrigues, A. G., Bragatte, M. A. de S., & Chies, J. A. B. (2020). Beyond HIV infection: Neglected and varied impacts of CCR5 and CCR5Δ32 on viral diseases. *Virus Research*, *286*, 198040. https://doi.org/10.1016/j.virusres.2020.198040

Ellwanger, J. H., Kulmann-Leal, B., Wolf, J. M., Michita, R. T., Simon, D., Lunge, V. R., & Chies, J. A. B. (2020). Role of the genetic variant CCR5Δ32 in HBV infection and HBV/HIV co-infection. *Virus Research, 277*, 197838. https://doi.org/10.1016/j.virusres.2019.197838

Engelman, A., & Cherepanov, P. (2012). The structural biology of HIV-1: Mechanistic and therapeutic insights. *Nature Reviews Microbiology*, *10*(4), 279–290. https://doi.org/10.1038/nrmicro2747

Esbjörnsson, J., Jansson, M., Jespersen, S., Månsson, F., Hønge, B. L., Lindman, J., Medina, C., da Silva, Z. J., Norrgren, H., Medstrand, P., Rowland-Jones, S. L., & Wejse, C. (2019). HIV-2 as a model to identify a functional HIV cure. *AIDS Research and Therapy*, *16*(1), 24. https://doi.org/10.1186/s12981-019-0239-x

Faria, N. R., Rambaut, A., Suchard, M. A., Baele, G., Bedford, T., Ward, M. J., Tatem, A. J., Sousa, J. D., Arinaminpathy, N., Pépin, J., Posada, D., Peeters, M., Pybus, O. G., & Lemey, P. (2014). HIV epidemiology. The early spread and epidemic ignition of HIV-1 in human populations. *Science (New York, N.Y.)*, *346*(6205), 56–61. https://doi.org/10.1126/science.1256739

Fauci, A. S. (2003). HIV and AIDS: 20 years of science. *Nature Medicine*, *9*(7), Article 7. https://doi.org/10.1038/nm0703-839

Frankel, A. D., & Young, J. A. T. (1998). HIV-1: Fifteen Proteins and an RNA. *Annual Review of Biochemistry*, *67*(1), 1–25. https://doi.org/10.1146/annurev.biochem.67.1.1

Gall, A., Kaye, S., Hué, S., Bonsall, D., Rance, R., Baillie, G. J., Fidler, S. J., Weber, J. N., Mcclure, M. O., Kellam, P., & Investigators, T. (2013). *Restriction of V3 region sequence divergence in the HIV-1 envelope gene during antiretroviral treatment in a cohort of recent seroconverters*. 1–15.

Gartner, M., Roche, M., Churchill, M., Gorry, P., & Flynn, J. (2020). Understanding the mechanisms driving the spread of subtype C HIV-1. *EBioMedicine*, *53*, 102682. https://doi.org/10.1016/j.ebiom.2020.102682

Gayle, H. D., & Hill, G. L. (2001). Global Impact of Human Immunodeficiency Virus and AIDS. *Clinical Microbiology Reviews*, *14*(2), 327–335. https://doi.org/10.1128/CMR.14.2.327-335.2001

Glatter, K. A., & Finkelman, P. (2021). History of the Plague: An Ancient Pandemic for the Age of COVID-19. *The American Journal of Medicine*, *134*(2), 176–181. https://doi.org/10.1016/j.amjmed.2020.08.019

Gomaa, H., & Mason, G. (2011). Software Modeling and Design: UML, Use Cases, Patterns, and Software Architectures. *Cambridge University Press, Cambridge.*

Goudsmit, J. (1997). *Viral sex: The nature of AIDS*. New York : Oxford University Press. http://archive.org/details/viralsexnatureof00goudrich; Accessed on 2022-03-03

GraphQL. (2021). *GraphQL (microservices) architecture by Apollo | by Dev by RayRay | ITNEXT*. https://itnext.io/graphql-microservices-architecture-by-apollo-8b6eb557c5e2; Accessed on 2021-06-02

Greenwood, E. J. D., Schmidt, F., & Heeney, J. L. (2014). Chapter 5—Simian Immunodeficiency Virus Infection of Chimpanzees (Pan troglodytes). In A. A. Ansari & G. Silvestri (Eds.), *Natural Hosts of SIV* (pp. 85–101). Elsevier. https://doi.org/10.1016/B978-0-12-404734-1.00005-X

Gupta, A., & Padh, H. (2015). Frequency Distribution of Mannose Binding Lectin-2 and Vitamin D Receptor Gene Variants: Putative Markers for Tuberculosis. *Genetics Research International*, *2015*, 1–7. https://doi.org/10.1155/2015/264120

Gupta, P. K., & Saxena, A. (2021). HIV/AIDS: Current Updates on the Disease, Treatment and Prevention. *Proceedings of the National Academy of Sciences, India. Section B*, *91*(3), 495–510. https://doi.org/10.1007/s40011-021-01237-y

Hemelaar, J., Elangovan, R., Yun, J., Dickson-Tetteh, L., Fleminger, I., Kirtley, S., Williams, B., Gouws-Williams, E., Ghys, P. D., & WHO–UNAIDS Network for HIV Isolation Characterisation. (2019). Global and regional molecular epidemiology of HIV-1, 1990-2015: A systematic review, global survey, and trend analysis. *The Lancet. Infectious Diseases*, *19*(2), 143–155. https://doi.org/10.1016/S1473-3099(18)30647-9

Herek. (1999). *AIDS and Stigma*. https://journals.sagepub.com/doi/10.1177/0002764299042007004; Accessed on 2020-09-30

Heydarifard, Z., Tabarraei, A., & Moradi, A. (2017, October 25). *Polymorphisms in CCR5Δ32 and Risk of HIV-1 Infection in the Southeast of Caspian Sea, Iran* [Research Article]. Disease Markers; Hindawi. https://doi.org/10.1155/2017/4190107

Hirsch, V. M., Olmsted, R. A., Murphey-Corb, M., Purcell, R. H., & Johnson, P. R. (1989). An African primate lentivirus (SIV sm closely related to HIV-2. *Nature*, *339*(6223), Article 6223. https://doi.org/10.1038/339389a0

HIV Replication Cycle | NIH: National Institute of Allergy and Infectious Diseases. (2018, June 19). https://www.niaid.nih.gov/diseases-conditions/hiv-replication-cycle

HIVInfo. (2020). *The Basics of HIV Prevention | HIVINFO*. https://hivinfo.nih.gov/understanding-hiv/fact-sheets/basics-hiv-prevention; Accessed on 2020-09-23

Hong, S. L., Dellicour, S., Vrancken, B., Suchard, M. A., Pyne, M. T., Hillyard, D. R., Lemey, P., & Baele, G. (2020). In Search of Covariates of HIV-1 Subtype B Spread in the United States-A Cautionary Tale of Large-Scale Bayesian Phylogeography. *Viruses*, *12*(2), 182. https://doi.org/10.3390/v12020182

Huet, T., Cheynier, R., Meyerhans, A., Roelants, G., & Wain-Hobson, S. (1990). Genetic organization of a chimpanzee lentivirus related to HIV-1. *Nature*, *345*(6273), Article 6273. https://doi.org/10.1038/345356a0

Hütter, G., Nowak, D., Mossner, M., Ganepola, S., Müssig, A., Allers, K., Schneider, T., Hofmann, J., Kücherer, C., Blau, O., Blau, I. W., Hofmann, W. K., & Thiel, E. (2009). Long-term control of HIV by CCR5 Delta32/Delta32 stem-cell transplantation. *The New England Journal of Medicine*, *360*(7), 692–698. https://doi.org/10.1056/NEJMoa0802905

IntelliJ IDEA Handbook. (2017). 73. https://www.javacodegeeks.com/; Accessed on 2020-12-20

Ivanov, A. V., Valuev-Elliston, V. T., Ivanova, O. N., Kochetkov, S. N., Starodubova, E. S., Bartosch, B., & Isaguliants, M. G. (2016, October 13). *Oxidative Stress during HIV Infection: Mechanisms and Consequences* [Review Article]. Oxidative Medicine and Cellular Longevity; Hindawi. https://doi.org/10.1155/2016/8910396

Jacquemard, C., Koensgen, F., Colin, P., Lagane, B., & Kellenberger, E. (2021). Modeling of CCR5 Recognition by HIV-1 gp120: How the Viral Protein Exploits the Conformational Plasticity of the Coreceptor. *Viruses*, *13*(7), 1395. https://doi.org/10.3390/v13071395

Java Programming Language—An overview | ScienceDirect Topics. (2003). https://www.sciencedirect.com/topics/computer-science/java-programming-language

John, G. C., Bird, T., Overbaugh, J., Nduati, R., Mbori-Ngacha, D., Rostron, T., Dong, T., Kostrikis, L., Richardson, B., & Rowland-Jones, S. L. (2001). CCR5 Promoter Polymorphisms in a Kenyan Perinatal Human Immunodeficiency Virus Type 1 Cohort: Association with Increased 2-Year Maternal Mortality. *The Journal of Infectious Diseases*, *184*(1), 89–92. https://doi.org/10.1086/321006

Jones, D. T., Taylor, W. R., & Thornton, J. M. (1992). The rapid generation of mutation data matrices from protein sequences. *Computer Applications in the Biosciences: CABIOS*, *8*(3), 275–282. https://doi.org/10.1093/bioinformatics/8.3.275

Justiz Vaillant, A. A., & Gulick, P. G. (2022). HIV Disease Current Practice. In *StatPearls*. StatPearls Publishing. http://www.ncbi.nlm.nih.gov/books/NBK534860/

Katoh, K., & Standley, D. M. (2013). MAFFT Multiple Sequence Alignment Software Version 7: Improvements in Performance and Usability. *Molecular Biology and Evolution*, *30*(4), 772–780. https://doi.org/10.1093/molbev/mst010

Kawamura, T., Gulden, F. O., Sugaya, M., McNamara, D. T., Borris, D. L., Lederman, M. M., Orenstein, J. M., Zimmerman, P. A., & Blauvelt, A. (2003). R5 HIV productively infects Langerhans cells, and infection levels are regulated by compound CCR5 polymorphisms.

Proceedings of the National Academy of Science, *100*, 8401–8406. https://doi.org/10.1073/pnas.1432450100

Keele, B. F., Giorgi, E. E., Salazar-Gonzalez, J. F., Decker, J. M., Pham, K. T., Salazar, M. G., Sun, C., Grayson, T., Wang, S., Li, H., Wei, X., Jiang, C., Kirchherr, J. L., Gao, F., Anderson, J. A., Ping, L.-H., Swanstrom, R., Tomaras, G. D., Blattner, W. A., … Shaw, G. M. (2008). Identification and characterization of transmitted and early founder virus envelopes in primary HIV-1 infection. *Proceedings of the National Academy of Sciences of the United States of America*, *105*(21), 7552–7557. https://doi.org/10.1073/pnas.0802203105

Keele, B. F., Van Heuverswyn, F., Li, Y., Bailes, E., Takehisa, J., Santiago, M. L., Bibollet-Ruche, F., Chen, Y., Wain, L. V., Liegeois, F., Loul, S., Ngole, E. M., Bienvenue, Y., Delaporte, E., Brookfield, J. F. Y., Sharp, P. M., Shaw, G. M., Peeters, M., & Hahn, B. H. (2006). Chimpanzee reservoirs of pandemic and nonpandemic HIV-1. *Science (New York, N.Y.)*, *313*(5786), 523–526. https://doi.org/10.1126/science.1126531

Kleinman, A. J., Pandrea, I., & Apetrei, C. (2022). So Pathogenic or So What?—A Brief Overview of SIV Pathogenesis with an Emphasis on Cure Research. *Viruses*, *14*(1), 135. https://doi.org/10.3390/v14010135

Kloek, M., Bulstra, C. A., van Noord, L., Al-Hassany, L., Cowan, F. M., & Hontelez, J. A. C. (2022). HIV prevalence among men who have sex with men, transgender women and cisgender male sex workers in sub-Saharan Africa: A systematic review and meta-analysis. *Journal of the International AIDS Society*, *25*(11), e26022. https://doi.org/10.1002/jia2.26022

Korber, B., Foley, B., & Leitner, T. (1997). *Human retroviruses and AIDS 1997* (LA-UR-98-1268). Los Alamos National Lab. (LANL), Los Alamos, NM (United States). https://doi.org/10.2172/607510

Kostrikis, L. G., Neumann, A. U., Thomson, B., Korber, B. T., McHardy, P., Karanicolas, R., Deutsch, L., Huang, Y., Lew, J. F., McIntosh, K., Pollack, H., Borkowsky, W., Spiegel, H. M. L., Palumbo, P., Oleske, J., Bardeguez, A., Luzuriaga, K., Sullivan, J., Wolinsky, S. M., … Moore, J. P. (1999). A Polymorphism in the Regulatory Region of the CC-Chemokine Receptor 5 Gene Influences Perinatal Transmission of Human Immunodeficiency Virus Type 1 to African-American Infants. *Journal of Virology*, *73*(12), 10264–10271. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC113080/

Kouanfack, C., Unal, G., Schaeffer, L., Kfutwah, A., Aghokeng, A., Mougnutou, R., Tchemgui-Noumsi, N., Alessandri-Gradt, E., Delaporte, E., Simon, F., Vray, M., Plantier, J.-C., & ANRS 12168 DynaMO Study. (2020). Comparative Immunovirological and Clinical Responses to Antiretroviral Therapy Between HIV-1 Group O and HIV-1 Group M Infected Patients. *Clinical Infectious Diseases: An Official Publication of the Infectious Diseases Society of America*, *70*(7), 1471–1477. https://doi.org/10.1093/cid/ciz371

Kucharska, I., Ding, P., Zadrozny, K. K., Dick, R. A., Summers, M. F., Ganser-Pornillos, B. K., & Pornillos, O. (2020). Biochemical Reconstitution of HIV-1 Assembly and Maturation. *Journal of Virology*, *94*(5), e01844-19. https://doi.org/10.1128/JVI.01844-19

Kumar, S., Stecher, G., Li, M., Knyaz, C., & Tamura, K. (2018a). MEGA X: Molecular Evolutionary Genetics Analysis across Computing Platforms. *Molecular Biology and Evolution*, *35*(6), 1547–1549. https://doi.org/10.1093/molbev/msy096

Kumar, S., Stecher, G., Li, M., Knyaz, C., & Tamura, K. (2018b). MEGA X: Molecular Evolutionary Genetics Analysis across computing platforms. *Molecular Biology and Evolution*, *35:*, 1547-1549.

LaMont, C., Otwinowski, J., Vanshylla, K., Gruell, H., Klein, F., & Nourmohammad, A. (2022). Design of an optimal combination therapy with broadly neutralizing antibodies to suppress HIV-1. *ELife*, *11*, e76004. https://doi.org/10.7554/eLife.76004

Lapointe, H. R., & Harrigan, P. R. (2020). Human Immunodeficiency Virus Phylogenetics in the United States-and Elsewhere. *The Journal of Infectious Diseases*, *222*(12), 1939–1940. https://doi.org/10.1093/infdis/jiaa108

Laure, F., Leonard, R., Mbayo, K., Lurhuma, Z., Kayembe, N., Brechot, C., Sarin, P. S., Sarngadharan, M., Wong-Staal, F., & Gallo, R. C. (1987). Genomic diversity of Zairian HIV isolates: Biological characteristics and clinical manifestation of HIV infection. *AIDS Research and Human Retroviruses*, *3*(4), 343–353. https://doi.org/10.1089/aid.1987.3.343

Li, Y., Ndjango, J.-B., Learn, G. H., Ramirez, M. A., Keele, B. F., Bibollet-Ruche, F., Liu, W., Easlick, J. L., Decker, J. M., Rudicell, R. S., Inogwabini, B.-I., Ahuka-Mundeke, S., Leendertz, F. H., Reynolds, V., Muller, M. N., Chancellor, R. L., Rundus, A. S., Simmons, N., Worobey, M., … Hahn, B. H. (2012). Eastern Chimpanzees, but Not Bonobos, Represent a Simian Immunodeficiency Virus Reservoir. *Journal of Virology*. https://journals.asm.org/doi/abs/10.1128/JVI.01498-12

Liu, R., Paxton, W. A., Choe, S., Ceradini, D., Martin, S. R., Horuk, R., MacDonald, M. E., Stuhlmann, H., Koup, R. A., & Landau, N. R. (1996). Homozygous Defect in HIV-1 Coreceptor Accounts for Resistance of Some Multiply-Exposed Individuals to HIV-1 Infection. *Cell*, *86*(3), 367–377. https://doi.org/10.1016/S0092-8674(00)80110-5

Lopalco, L. (2010). CCR5: From Natural Resistance to a New Anti-HIV Strategy. *Viruses*, *2*(2), 574–600. https://doi.org/10.3390/v2020574

Lopes, M. P., Santos, M. N. N., Faber, E. W., Bezerra, M. A. C., Hatzlhofer, B. L. D., Albuquerque, D. M., Zaccariotto, T. R., Ribeiro, D. M., Araújo, A. da S., Costa, F. F., & Sonati, M. de F. (2014, November 11). *The CCR5Δ32 Polymorphism in Brazilian Patients with Sickle Cell Disease* [Research Article]. Disease Markers; Hindawi. https://doi.org/10.1155/2014/678246

Lucas, S. (2002). The pathology of HIV infection. *Leprosy Review*, *73*(1), 64–71.

Lynch, R. M., Shen, T., Gnanakaran, S., & Derdeyn, C. A. (2009). *Appreciating HIV Type 1 Diversity: Subtype Differences in Env*. *25*(3), 237–248.

Malim, M. H., & Emerman, M. (2008). HIV-1 accessory proteins—Ensuring viral survival in a hostile environment. *Cell Host & Microbe*, *3*(6), 388–398. https://doi.org/10.1016/j.chom.2008.04.008

Martinson, J. J., Chapman, N. H., Rees, D. C., Liu, Y. T., & Clegg, J. B. (1997). Global distribution of the CCR5 gene 32-basepair deletion. *Nature Genetics*, *16*(1), 100–103. https://doi.org/10.1038/ng0597-100

Mayoclinic. (2020). *HIV/AIDS - Diagnosis and treatment—Mayo Clinic*. https://www.mayoclinic.org/diseases-conditions/hiv-aids/diagnosis-treatment/drc-20373531; Accessed on 2020-09-22

McHugh, M. (2012). Interrater reliability: The kappa statistic. *Biochemia Medica : Časopis Hrvatskoga Društva Medicinskih Biokemičara / HDMB*, *22*, 276–282. https://doi.org/10.11613/BM.2012.031

Mehlotra, R. K., Hall, N. B., Bruse, S. E., John, B., Zikursh, M. J. B., Stein, C. M., Siba, P. M., & Zimmerman, P. A. (2015). CCR2, CCR5, and CXCL12 variation and HIV/AIDS in Papua New Guinea. *Infection, Genetics and Evolution : Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases*, *36*, 165–173. https://doi.org/10.1016/j.meegid.2015.09.014

Mellors, J. W., & et al. (1996). Mutations in retroviral genes associated with drug resistance. *Theoretical Biology and Biophysics, Los Alamos National Laboratory, Los Alamos*.

Michael, N. L., Nelson, J. A. E., KewalRamani, V. N., Chang, G., O'Brien, S. J., Mascola, J. R., Volsky, B., Louder, M., White, G. C., Littman, D. R., Swanstrom, R., & O'Brien, T. R. (1998). Exclusive and Persistent Use of the Entry Coreceptor CXCR4 by Human

Immunodeficiency Virus Type 1 from a Subject Homozygous for CCR5 Δ32. *Journal of Virology*, *72*(7), 6040–6047. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC110409/

Moher, D., Liberati, A., Tetzlaff, J., Altman, D. G., & PRISMA Group. (2009). Preferred reporting items for systematic reviews and meta-analyses: The PRISMA statement. *PLoS Medicine*, *6*(7), e1000097. https://doi.org/10.1371/journal.pmed.1000097

Mona Sadat, L., Seyed Mehdi, S., & Amitis, R. (2018). HIV-1 Immune evasion: The main obstacle toward a successful vaccine. *Archives of Asthma, Allergy and Immunology*, *2*(1), 013–015. https://doi.org/10.29328/journal.aaai.1001013

Mozhi, P. K., & Ganapathy, D. (2021). AWARENESS OF STRUCTURAL BIOLOGY OF HIV AMONG DENTAL STUDENTS. *European Journal of Molecular and Clinical Medicine*, *8*(1), 491–504. https://go.gale.com/ps/i.do?p=AONE&sw=w&issn=25158260&v=2.1&it=r&id=GALE%7CA698747685&sid=googleScholar&linkaccess=abs

Muccini, C., Canetti, D., Castagna, A., & Spagnuolo, V. (2022). Efficacy and Safety Profile of Fostemsavir for the Treatment of People with Human Immunodeficiency Virus-1 (HIV-1): Current Evidence and Place in Therapy. *Drug Design, Development and Therapy*, *16*, 297–304. https://doi.org/10.2147/DDDT.S273660

Munerato, P., Azevedo, M. L., Sucupira, M. C. A., Pardini, R., Pinto, G. H. N., Catroxo, M., Souza, I. E., & Diaz, R. S. (2003a). Frequency of polymorphisms of genes coding for HIV-1 co-receptors CCR5 and CCR2 in a Brazilian population. *The Brazilian Journal of Infectious Diseases: An Official Publication of the Brazilian Society of Infectious Diseases*, *7*(4), 236–240. https://doi.org/10.1590/s1413-86702003000400002

Munerato, P., Azevedo, M. L., Sucupira, M. C. A., Pardini, R., Pinto, G. H. N., Catroxo, M., Souza, I. E., & Diaz, R. S. (2003b). Frequency of polymorphisms of genes coding for HIV-

1 co-receptors CCR5 and CCR2 in a Brazilian population. *Brazilian Journal of Infectious Diseases*, *7*(4), 236–240. https://doi.org/10.1590/S1413-86702003000400002

Musey, L. K., Krieger, J. N., Hughes, J. P., Schacker, T. W., Corey, L., & McElrath, M. J. (1999). Early and Persistent Human Immunodeficiency Virus Type 1 (HIV-1)–Specific T Helper Dysfunction in Blood and Lymph Nodes following Acute HIV-1 Infection. *The Journal of Infectious Diseases*, *180*(2), 278–284. https://doi.org/10.1086/314868

Myers, G., Foley, B., Korber, B., Mellors, J. W., Jeang, K. T., & Wain-Hobson, S. (1997). *Human retroviruses and AIDS 1996. A compilation and analysis of nucleic acid and amino acid sequences* (LA-UR-97-31). Los Alamos National Lab., NM (United States). https://doi.org/10.2172/463607

MySQL. (1995). https://www.mysql.com/; Accessed on 2020-12-18

Naswa, S., Marfatia, Y. S., & Prasad, T. L. N. (2012). Microbicides and HIV: A Review and an update. *Indian Journal of Sexually Transmitted Diseases and AIDS*, *33*(2), 81–90. https://doi.org/10.4103/2589-0557.102098

Ndashimye, E., Li, Y., Reyes, P. S., Avino, M., Olabode, A. S., Kityo, C. M., Kyeyune, F., Nankya, I., Quiñones-Mateu, M. E., Barr, S. D., & Arts, E. J. (2021). High-level resistance to bictegravir and cabotegravir in subtype A- and D-infected HIV-1 patients failing raltegravir with multiple resistance mutations. *Journal of Antimicrobial Chemotherapy*, *76*(11), 2965–2974. https://doi.org/10.1093/jac/dkab276

Nkenfou, C. N., Mekue, L. C. M., Nana, C. T., & Kuiate, J. R. (2013). Distribution of CCR5-Delta32, CCR5 promoter 59029 A/G, CCR2-64I and SDF1-3'A genetic polymorphisms in HIV-1 infected and uninfected patients in the west region of Cameroon. *BMC Research Notes*, *6*, 288. https://doi.org/10.1186/1756-0500-6-288

Oh, D.-Y., Jessen, H., Kücherer, C., Neumann, K., Oh, N., Poggensee, G., Bartmeyer, B., Jessen, A., Pruss, A., Schumann, R. R., & Hamouda, O. (2008). CCR5Δ32 Genotypes in a German

HIV-1 Seroconverter Cohort and Report of HIV-1 Infection in a CCR5Δ32 Homozygous Individual. *PLOS ONE*, *3*(7), e2747. https://doi.org/10.1371/journal.pone.0002747

Okonechnikov, K., Golosova, O., Fursov, M., & UGENE team. (2012). Unipro UGENE: A unified bioinformatics toolkit. *Bioinformatics (Oxford, England)*, *28*(8), 1166–1167. https://doi.org/10.1093/bioinformatics/bts091

Ongadi. (2021, January 2). *ARVPredictor—Apps on Google Play*. https://play.google.com/store/apps/details?id=co.ke.ikocare&hl=en&gl=KE

Ongadi, B. A., Obiero, G., Lihana, R. W., & Kiiru, J. N. (2018). Distribution of Genetic Polymorphism in the CCR5 among Caucasians, Asians and Africans: A Systematic Review and Meta-Analysis. *Open Journal of Genetics*, *08*(03), 54–66. https://doi.org/10.4236/ojgen.2018.83006

Ongadi, B., Lihana, R., Kiiru, J., Ngayo, M., & Obiero, G. (2022). An Android-Based Mobile App (ARVPredictor) for the Detection of HIV Drug-Resistance Mutations and Treatment at the Point of Care: Development Study. *JMIR Formative Research*, *6*, e26891. https://doi.org/10.2196/26891

Paetsch, F., Eberlein, A., & Maurer, F. (2003). Requirements Engineering and Agile Software Development. *12th IEEE International Workshops on Enabling Technologies : Infrastructure for Collaborative Enterprises , Linz, , 11 June 2003*, 308-313.

Pereira, R. W., Pires, E. R., Duarte, A. P. M., Moura, R. P. de, Monteiro, E., Torloni, H., Proietti, A. B., Simpson, A. J. G., & Pena, S. D. J. (2000). Frequency of the CCRdelta32 allele in Brazilians: A study in colorectal cancer and in HTLV-I infection. *Genetics and Molecular Biology*, *23*(3), 523–526. https://doi.org/10.1590/S1415-47572000000300003

Potts, A., & Friedel, D. H. (1996). *Java: Programming language handbook*. Coriolis Group Bks. ; Accessed on 2020-12-18

Rahimi, H., Farajollahi, M. M., & Hosseini, A. (2014). Distribution of the mutated delta 32 allele of CCR5 co-receptor gene in Iranian population. *Medical Journal of the Islamic Republic of Iran*, *28*, 140. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4322332/

Rao, M., Peachman, K. K., Kim, J., Gao, G., Alving, C. R., Nelson, L., & Rao, V. B. (2013). HIV-1 variable loop 2 and its importance in HIV-1 infection and vaccine development. *Curr HIV Res.*, *11*(5), 11(5): 427-438.

Reed, J. C., Solas, D., Kitaygorodskyy, A., Freeman, B., Ressler, D. T. B., Phuong, D. J., Swain, J. V., Matlack, K., Hurt, C. R., Lingappa, V. R., & Lingappa, J. R. (2021). Identification of an Antiretroviral Small Molecule That Appears To Be a Host-Targeting Inhibitor of HIV-1 Assembly. *Journal of Virology*, *95*(3), e00883-20. https://doi.org/10.1128/JVI.00883-20

Rhee, S.-Y. (2003). Human immunodeficiency virus reverse transcriptase and protease sequence database. *Nucleic Acids Research*, *31*(1), 298–303. https://doi.org/10.1093/nar/gkg100

Robertson, D., Anderson, J., Bradac, J. A., Carr, J. K., Foley, B., Funkhouser, R. K., Gao, F., Hahn, B. H., Kalish, M. L., Kuiken, C., Learn, G., Leitner, T., McCutchan, F., Osmanov, S., Peeters, M., Pieniazek, D., Salminen, M., Sharp, P., Wolinsky, S., & Korber, B. (2000). HIV-1 Nomenclature Proposal. *Science*, *288*, 55–56. https://doi.org/10.1126/science.288.5463.55d

Rolland, M. (2016). HIV-1 immune evasion-a threat to effective vaccines? *Nature Medicine*, *22*(6), 580–581. https://doi.org/10.1038/nm.4119

Rottman, J. B., Ganley, K. P., Williams, K., Wu, L., Mackay, C. R., & Ringler, D. J. (1997). Cellular localization of the chemokine receptor CCR5. Correlation to cellular targets of HIV-1 infection. *The American Journal of Pathology*, *151*(5), 1341–1351.

Roy, P., & Chakrabarti, S. (2016). Mutation in AIDS restriction gene affecting HIV infection and disease progression in a high risk group from Northeastern India. *Medical Journal, Armed Forces India*. https://doi.org/10.1016/j.mjafi.2015.07.005

Ryabov, G. S., Kazennova, E. V., Bobkova, M. R., & Bobkov, A. F. (2004). Prevalence of Alleles Associated with HIV Resistance in Russia. *Genetic Testing*, *8*(1), 73–76. https://doi.org/10.1089/109065704323016067

Salem, A. H., Farid, E., Fadel, R., Abu-Hijleh, M., Almawi, W., Han, K., & Batzer, M. A. (2009). Distribution of Four HIV Type 1-Resistance Polymorphisms (CCR5-Δ32, CCR5-m303, CCR2-64I, and SDF1-3′A) in the Bahraini Population. *AIDS Research and Human Retroviruses*, *25*(10), 973–977. https://doi.org/10.1089/aid.2009.0066

Samson, M., Libert, F., Doranz, B. J., Rucker, J., Liesnard, C., Farber, C. M., Saragosti, S., Lapoumeroulie, C., Cognaux, J., Forceille, C., Muyldermans, G., Verhofstede, C., Burtonboy, G., Georges, M., Imai, T., Rana, S., Yi, Y., Smyth, R. J., Collman, R. G., … Parmentier, M. (1996). Resistance to HIV-1 infection in caucasian individuals bearing mutant alleles of the CCR-5 chemokine receptor gene. *Nature*, *382*(6593), 722–725. https://doi.org/10.1038/382722a0

Sanchez-Pescador, R., Power, M. D., Barr, P. J., Steimer, K. S., Stempien, M. M., Brown-Shimer, S. L., Gee, W. W., Renard, A., Randolph, A., Levy, J. A., Dina, D., & Luciw, P. A. (1985). Nucleotide Sequence and Expression of an AIDS-Associated Retrovirus (ARV-2). *Science*, *227*(4686), 484–492. https://doi.org/10.1126/science.2578227

Sebbane, F., & Lemaître, N. (2021). Antibiotic Therapy of Plague: A Review. *Biomolecules*, *11*(5), Article 5. https://doi.org/10.3390/biom11050724

Sharp, P. M., & Hahn, B. H. (2011). Origins of HIV and the AIDS Pandemic. *Cold Spring Harbor Perspectives in Medicine:*, *1*(1). https://doi.org/10.1101/cshperspect.a006841

Sidoti, A., D'Angelo, R., Rinaldi, C., De Luca, G., Pino, F., Salpietro, C., Giunta, D. E., Saltalamacchia, F., & Amato, A. (2005). Distribution of the mutated Delta32 allele of the CCR5 gene in a Sicilian population. *International Journal of Immunogenetics*, *32*(3), 193–198. https://doi.org/10.1111/j.1744-313X.2005.00507.x

Smoljanović, M., Ristić, S., & Hayward, C. (2006). Historic Exposure to Plague and Present-day Frequency of CCR5del32 in Two Isolated Island Communities of Dalmatia, Croatia. *Croatian Medical Journal*, *47*(4), 579–584. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2080457/

Smyth, N. (2020). *Android Studio 4. 1 Development Essentials—Java Edition Developing Android 11 Apps Using Android Studio 4. 1, Java and Android Jetpack.* Payload Media. http://public.eblib.com/choice/PublicFullRecord.aspx?p=6379904

Stephens, J. C., Reich, D. E., Goldstein, D. B., Shin, H. D., Smith, M. W., Carrington, M., Winkler, C., Huttley, G. A., Allikmets, R., Schriml, L., Gerrard, B., Malasky, M., Ramos, M. D., Morlot, S., Tzetis, M., Oddoux, C., di Giovine, F. S., Nasioulas, G., Chandler, D., … Dean, M. (1998). Dating the origin of the CCR5-Delta32 AIDS-resistance allele by the coalescence of haplotypes. *American Journal of Human Genetics*, *62*(6), 1507–1515. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC1377146/

Stephenson, K. E., & Barouch, D. H. (2013). A global approach to HIV-1 vaccine development. *Immunological Reviews*, *254*(1), 295–304. https://doi.org/10.1111/imr.12073

Tawfik, G. M., Dila, K. A. S., Mohamed, M. Y. F., Tam, D. N. H., Kien, N. D., Ahmed, A. M., & Huy, N. T. (2019). A step by step guide for conducting a systematic review and meta-analysis with simulation data. *Tropical Medicine and Health*, *47*, 46. https://doi.org/10.1186/s41182-019-0165-6

Thomas, R. H. (2001). Molecular Evolution and Phylogenetics. Masatoshi Nei and Sudhir Kumar. Oxford University Press, Oxford. 2000. Pp. 333. Price £65.00, hardback. ISBN 0 19 513584 9. *Heredity*, *86*(3), 385–385. https://doi.org/10.1046/j.1365-2540.2001.0923a.x

Thomson, M. M., Pérez-Álvarez, L., & Nájera, R. (2002). Molecular epidemiology of HIV-1 genetic forms and its significance for vaccine development and therapy. *The Lancet Infectious Diseases*, *2*(8), 461–471. https://doi.org/10.1016/S1473-3099(02)00343-2

Torimiro, J. N., Wolfe, N. D., Thomas, A., Martin, M. P., Mpoudi-Ngole, E., Tamoufe, U., Birx, D. L., Carrington, M., Burke, D. S., & Carr, J. K. (2007). Frequency of CCR5 variants among rural populations with low HIV-1 prevalence in Cameroon. *AIDS*, *21*(4), 527–528. https://doi.org/10.1097/QAD.0b013e328045c4bd

Trecarichi, E. M., Tumbarello, M., Donati, K. de G., Tamburrini, E., Cauda, R., Brahe, C., & Tiziano, F. D. (2006). Partial protective effect of CCR5-Delta 32 heterozygosity in a cohort of heterosexual Italian HIV-1 exposed uninfected individuals. *AIDS Research and Therapy*, *3*(1), 22. https://doi.org/10.1186/1742-6405-3-22

Types of antiretroviral medications. (2021, May 19). Aidsmap.Com. https://www.aidsmap.com/about-hiv/types-antiretroviral-medications

UNAIDS. (2014). *90–90–90—An ambitious treatment target to help end the aids epidemic*. http://www.unaids.org/en/resources/documents/2014/90-90-90

UNAIDS. (2019). https://aidsinfo.unaids.org/

UNAIDS. (2020). *Global HIV & AIDS statistics—2020 fact sheet*. https://www.unaids.org/en/resources/fact-sheet; Accessed on 2020-09-22

UNAIDS. (2021). *90-90-90: Treatment for all | UNAIDS*. https://www.unaids.org/en/resources/909090 Accessed on 2020-09-17 10:12:09

Valadez-González, N., González-Martínez, P., Lara-Perera, D., Vera-Gamboa, L., & Góngora-Biachi, R. (2011). Implicación del alelo CCR5-Δ32 en la progresión clínica de pacientes VIH-1+ en Yucatán, México. *Salud Pública de México*, *53*(6), Article 6. https://www.saludpublica.mx/index.php/spm/article/view/7092

van Heuvel, Y., Schatz, S., Rosengarten, J. F., & Stitz, J. (2022). Infectious RNA: Human Immunodeficiency Virus (HIV) Biology, Therapeutic Intervention, and the Quest for a Vaccine. *Toxins*, *14*(2), Article 2. https://doi.org/10.3390/toxins14020138

Vargas, A. E., Marrero, A. R., Salzano, F. M., Bortolini, M. C., & Chies, J. a. B. (2006). Frequency of CCR5delta32 in Brazilian populations. *Brazilian Journal of Medical and Biological Research*, *39*(3), 321–325. https://doi.org/10.1590/S0100-879X2006000300002

Visseaux, B., Le Hingrat, Q., Damond, F., Charpentier, C., & Descamps, D. (2019). [Physiopathology of HIV-2 infection]. *Virologie (Montrouge, France)*, *23*(5), 277–291. https://doi.org/10.1684/vir.2019.0789

Waruru, A., Wamicwe, J., Mwangi, J., Achia, T. N. O., Zielinski-Gutierrez, E., Ng'ang'a, L., Miruka, F., Yegon, P., Kimanga, D., Tobias, J. L., Young, P. W., Cock, K. M. D., & Tylleskär, T. (2021). Where Are the Newly Diagnosed HIV Positives in Kenya? Time to Consider Geo-Spatially Guided Targeting at a Finer Scale to Reach the "First 90". *Frontiers in Public Health*, *9*. https://doi.org/10.3389/fpubh.2021.503555

Weaver, S., Shank, S. D., Spielman, S. J., Li, M., Muse, S. V., & Kosakovsky Pond, S. L. (2018). Datamonkey 2.0: A Modern Web Application for Characterizing Selective and Other Evolutionary Processes. *Molecular Biology and Evolution*, *35*(3), 773–777. https://doi.org/10.1093/molbev/msx335

WHO. (2001). *WHO | 2001 Declaration of Commitment on HIV/AIDS*. WHO; World Health Organization. https://www.who.int/hiv/pub/advocacy/ungass_2001/en/; Accessed on 2020-09-30

WHO. (2019). *2019_summary-global-hiv-epi.png (682×486)*. https://www.who.int/hiv/data/2019_summary-global-hiv-epi.png?ua=1; Accessed on 2020-09-21

Winans, S., & Goff, S. P. (2020). Mutations altering acetylated residues in the CTD of HIV-1 integrase cause defects in proviral transcription at early times after integration of viral DNA. *PLoS Pathogens*, *16*(12), e1009147. https://doi.org/10.1371/journal.ppat.1009147

Worobey, M., Watts, T. D., McKay, R. A., Suchard, M. A., Granade, T., Teuwen, D. E., Koblin, B. A., Heneine, W., Lemey, P., & Jaffe, H. W. (2016). 1970s and 'Patient 0' HIV-1 genomes illuminate early HIV/AIDS history in North America. *Nature*, *539*(7627), 98–101. https://doi.org/10.1038/nature19827

Yudin, N. S., Vinogradov, S. V., Potapova, T. A., Naykova, T. M., Sitnikova, V. V., Kulikov, I. V., Khasnulin, V. I., Konchuk, C., Vloschinskii, P. E., Ivanov, S. V., Kobzev, V. F., Romaschenko, A. G., & Voevoda, M. I. (1998). Distribution of CCR5-delta 32 gene deletion across the Russian part of Eurasia. *Human Genetics*, *102*(6), 695–698. https://doi.org/10.1007/s004390050764

Zajac, V. (2018). Evolutionary view of the AIDS process. *The Journal of International Medical Research*, *46*(10), 4032–4038. https://doi.org/10.1177/0300060518786919

Zapata, W., Aguilar-Jiménez, W., Pineda-Trujillo, N., Rojas, W., Estrada, H., & Rugeles, M. T. (2013). Influence of CCR5 and CCR2 genetic variants in the resistance/susceptibility to HIV in serodiscordant couples from Colombia. *AIDS Research and Human Retroviruses*, *29*(12), 1594–1603. https://doi.org/10.1089/aid.2012.0299

Zhang, Y., Li, J., & Li, Q. (2018). Immune Evasion of Enteroviruses Under Innate Immune Monitoring. *Frontiers in Microbiology*, *9*, 1866. https://doi.org/10.3389/fmicb.2018.01866

Zunich, K. M., & Lane, H. C. (1990). The immunology of HIV infection. *Journal of the American Academy of Dermatology*, *22*(6, Part 2), 1202–1205. https://doi.org/10.1016/0190-9622(90)70163-C

Zunich, K. M., & Lane, H. C. (1991). Immunologic Abnormalities in HIV Infection. *Hematology/Oncology Clinics of North America*, *5*(2), 215–228. https://doi.org/10.1016/S0889-8588(18)30437-4

APPENDICES

Appendix 1.1: Major HIV-1 Drug Resistance Mutations Updated March 9, 2015

Major HIV-1 Drug Resistance Mutations

Updated March 9, 2015

Updated summary from the HIV Drug Resistance Database. This document can be downloaded from the http://hivdb.stanford.edu home page. Detailed and referenced versions of each drug class summary can be found at http://hivdb.stanford.edu/pages/drugSummaries.html

Major Nucleoside RT Inhibitor (NRTI)-Resistance Mutations

	Non-TAMs					TAMs						MDR	
	184	65	70	74	115	41	67	70	210	215	219	69	151
Cons	M	K	K	L	Y	M	D	K	L	T	K	T	Q
3TC	VI	R	E									Ins	M
FTC	VI	R	E									Ins	M
ABC	VI	R	E	VI	F	L			W	YF		Ins	M
TDF	***	R	E		F	L			W	YF		Ins	M
ZDV	***	***	*	*		L	N	R	W	YF	QE	Ins	M

Bold/underline: High-level reduced susceptibility or virological response. Bold: reduced suceptibility or virological response. Plain text: reduced susceptibility in combination with other NRTI-resistance mutations. Asterisk: increased susceptibility. **Additional NRTIs:** Stavudine (d4T) and didanosine (ddI) are no longer recommended. **TAMs:** Thymidine analog mutations. Selected by AZT and d4T and facilitate primer unblocking. Non-TAMs prevent NRTI incorporation. **MDR:** Multidrug resistance mutations. T69 insertions occur with TAMs. Q151M occurs with non-TAMs and accessory mutations A62V, V75I, F77L, and F116Y. **M184VI:** Although they cause high-level *in vitro* resistance to 3TC/FTC, they are not contraindications to 3TC/FTC because they increase TDF and AZT susceptibility and decrease viral replication fitness. **Additional mutations:** K65N is similar but weaker than K65R. K70GQ is similar to K70E. T69D and V75MT reduce susceptibility to d4T and ddI. T215SCDEIV (T215 revertants) evolve from T215YF in the absence of NRTIs. E40F, E44DA, D67GE, V118I, and K219NR are accessory TAMs. T69 deletions occur in combination with K65R and/or Q151M. With K65R (but not Q151M) they increase AZT susceptibility. **References:** http://hivdb.stanford.edu/DR/NRTIResiNote.html.

Major Non-Nucleoside RT Inhibitor (NNRTI)-Resistance Mutations

	100	101	103	106	138	179	181	188	190	227	230
Cons	L	K	K	V	E	V	Y	Y	G	F	M
NVP	I	PEH	NS	AM		DEF	CIV	LCH	ASEQ	LC	L
EFV	I	PEH	NS	AM		DEF	C	LCH	ASEQ	LC	L
ETR	I	PEH					CIV		EQ	C	L
RPV	I	PEH			KAGQ	DEF	CIV	L	EQ	C	L

Bold/underline: High-level reduced susceptibility or virological response. Bold: reduced suceptibility or virological response. Plain text: reduced susceptibility in combination with other NNRTI-resistance mutations. Asterisk: increased susceptibility. **Abbreviations**: nevirapine (NVP), efavirenz (EFV), etravirine (ETR), rilpivirine (RPV). **Synergistic combinations:** V179D+K103R reduce NVP and EFV susceptibility >10-fold. Y181C+V179F cause high-level ETR and RPV resistance. **ETR genotypic susceptibility score (GSS):** Y181IV (3.0); L100I, K101P, Y181C, M230L (2.5); V90I, E138A, V179F, G190S (1.5); A98G, K101EH, V106I, V179DT, G190A (1.0); <2.5 susceptible; 2.5 to 3.0 intermediate; >3.0 high-level. V90I, A98G, V106I, E138A, V179DT, G190A/S have little effect on ETR susceptibility unless they occur with a bolded mutations. **Additional accessory mutations:** V90I (ETR), A98G (NVP, EFV, ETR, RPV), V108I, V179T (ETR), V179L (RPV), P225H (EFV), K238T (NVP, EFV), L318F (NVP). **References:** http://hivdb.stanford.edu/DR/NNRTIResiNote.html.

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Major Protease Inhibitor (PI) Resistance Mutations

	24	32	46	47	48	50	54	76	82	84	88	90
Cons	L	V	M	I	G	I	I	L	V	I	N	L
ATV/r		**I**	IL	V	**VM**	**L**	**VTAM**		**ATSF**	**V**	**S**	M
DRV/r		**I**		**VA**		**V**	**LM**	**V**	**F**	V		
LPV/r	I	**I**	IL	**VA**	VM	**V**	**VTALM**	**V**	**ATSF**	**V**		M

Bold/underline: High-level reduced susceptibility or virological response. Bold: reduced suceptibility or virological response. Plain text: reduced susceptibility in combination with other PI-resistance mutations. **Abbreviations:** atazanavir (ATV), darunavir (DRV), lopinavir (LPV). Administered with ritonavir for pharmacokinetic boosting (/r). **Additional PIs:** Fosamprenavir (FPV), indinavir (IDV), saquinavir (SQV), and tipranavir (TPV) are rarely used. Nelfinavir (NFV) is no longer recommended. FPV/r and IDV/r are never more active than DRV/r and rarely if ever more active than LPV/r vs resistant viruses. TPV/r is occasionally useful for salvage therapy as it can be active vs LPV/r and DRV/r-resistant viruses with mutations that increase TPV susceptibility. Expert consultation +/- phenotypic testing should be obtained prior to using FPV, FPV/r, IDV/r, SQV/r, and TPV/r. **Additional mutations:** D30N and N88D are major NFV-resistance mutations. L10F, V11I, K20TV, L23I, K43T, F53L, Q58E, A71IL, G73STCA, T74P, N83D, and L89V are common nonpolymorphic accessory mutations. L10RY, V11L, L24F, M46V, G48ASTLQ, F53Y, I54S, V82CM, I84AC, N88TG are rare nonpolymorphic variants. **Hypersusceptibility:** I50L (each PI except ATV); L10F, L24I, I50V, I54L (TPV); L76V (ATV, SQV, TPV); I47A (SQV); N88S (FPV). **References:** http://hivdb.stanford.edu/DR/PIResiNote.html.

Major Integrase Inhibitor (INI)-Resistance Mutations

	66	92	138	140	143	147	148	155
Cons	T	E	E	G	Y	S	Q	N
RAL	A	**Q**	**KA**	**SAC**	**CRH**		**HRK**	**H**
EVG	**IAK**	**Q**	**KA**	**SAC**		**G**	**HRK**	**H**
DTG		Q	KA	SAC			**HRK**	

Bold/underline: High-level reduced susceptibility or virological response. Bold: reduced suceptibility or virological response. Plain text: reduced susceptibility in combination with other INI-resistance mutations. Asterisk: increased susceptibility. **Abbreviations**: raltegravir (RAL), elvitegravir (EVG), dolutegravir (DTG). **Additional mutations:** H51Y, L74M, T97A, S153YF, G163RK, S230R, and R263K are relatively nonpolymorphic INI-selected accessory resistance mutations. E92GV, E138T, Y143KSGA, Q148N, and N155ST are unusual variants at the positions listed above. P145S and Q146P are rare EVG-resistance mutations. G118R and F121Y are rare nonpolymorphic INI-resistance mutations. **References:** http://hivdb.stanford.edu/DR/INIResiNote.html.

HIV-1 RT and Protease Mutations for Drug-Resistance Surveillance*

NRTIs				NNRTIs		PIs			
M41	L	**Q151**	M	**L100**	I	**L23**	I	**G73**	S,T,C,A
K65	R	**M184**	V,I	**K101**	E,P	**L24**	I	**L76**	V
D67	N,G,E	**L210**	W	**K103**	N,S	**D30**	N	**V82**	A,T,S,F,L,C,M
T69	D,Ins	**T215**	Y,F,S,C,D,E,I,V	**V106**	A,M	**V32**	I	**N83**	D
K70	R,E	**K219**	Q,E,N,R	**V179**	F	**M46**	I,L	**I84**	V,A,C
L74	V,I			**Y181**	C,I,V	**I47**	V,A	**I85**	V
V75	M,T,A,S			**Y188**	L,C,H	**G48**	V,M	**N88**	D,S
F77	L			**G190**	A,S,E	**I50**	V,L	**L90**	M
Y115	F			**P225**	H	**F53**	F,Y		
F116	Y			**M230**	L	**I54**	V,L,M,T,A,S		

*Bennett DE, Camacho RJ, Otelea D,et al. Drug Resistance Mutations for Surveillance of Transmitted HIV-1 Drug-Resistance: 2009 Update, PLoS One 2009;4:e4724.Criteria for mutations on this list: (i) Cause/contribute to resistance. (ii) Nonpolymorphic (≤ 0.5% in ARV-naive persons) in 8 most common group M subtypes. http://hivdb.stanford.edu/cgi-bin/MutPrevBySubtypeRx.cgi.

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Appendix 1.2: *ARVPredictor*; Test Performance Sequences (Accession Numbers: KX505314-KX505372 and MK588680-MK588752)

PID	SEQUENCES
001	CAATGGCCATTGACAGMAGAAAAAATAAAAGCATTAACAGAAATTTGCACAGATATGGAAAAGGAA GGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAAAAA GACAGTACTAAATGGAGGAAATTAGTAGATTTCAGGGAGCTCAATAAAAGAACACAAGACTTTTGG GAAGTTCAATTAGGGATACCGCATCCAGCGGGCTTAAAAAAGAAGAAATCAGTAACAGTACTAGAT GTGGGGGACGCATATTTTTCAGTTCCTTTAGATGAAGGCTTTAGGAAATATACTGCGTTCACCATA CCTAGTATAAACAATGAGACACCAGGAATCAGATATCAGTATAATGTGCTCCCACAGGGATGGAAA GGATCACCAGCAATATTCCAGAGTAGYATGACAAAAATCTTAGAGCCCTTCAGATCAAAAAATCCA GAAATAACTATTTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAACAT AGAGCAAAAATAGAGGAGCTAAGAGAACATCTATTARGGTGGGGATTAACCACACCAGATAAGAAA CATCAGAAAGAACCCCCGTTTCTTTGGATGGGTTATGAACTA SUBTYPE A:SUS
002	AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTATGGAAATGGAGAAG GAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACACCAATATTTGCAATAAAGAAA AAGGATAGCACTAAATGGAGGAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAACACAAGACTTT TGGGAAGTTCAGCTAGGAATACCGCATCCAGCGGGTCTAAAAAAGAAAAAATCAGTAACAGTACTA GATGTGGGGGACGCATATTTTTCAGTTCCTTTACATGAAGGCTTTARAAAATATACTGCATTCACC ATACCTAGTACAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCCACAGGGATGG AAAGGATCACCATCAATATTCCAGAGTAGCATGATAAAAATTTTAGAACCTTTCAGATCAAAAAAT CCAGAAATAATTATCTATCAATATATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGAGCAA CATCGAGCAAAAATAGAAGAGTTGAGAGCTCATCTATTGAGCTGGGGATTTACTACACCAGACAAA AAGCATCAGAAAGAACCTCCATTCCTTTGGATGGGWTATGAACTA SUBTYPE A:SUS
003	AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTATGGAAATGGAGAAG GAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACACCAATATTTGCAATAAAGAAA AAGGATAGCACTAAATGGAGGAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAACACAAGACTTT TGGGAAGTTCAGCTAGGAATACCGCATCCAGCGGGTCTAAAAAAGAAAAAATCAGTAACAGTACTA GATGTGGGGGACGCATATTTTTCAGTTCCTTTACATGAAGGCTTTAGAAAATATACTGCATTCACC ATACCTAGTACAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCCACAGGGATGG AAAGGATCACCATCGATATTCCAGAGTAGCATGATAAAAATTTTAGAACCTTTCAGATCAAAAAAT CCAGAAATAATTATCTATCAATATATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGAGCAA CATCGAGCAAAAATAGAAGAGTTGAGAGCTCATCTATTGAGCTGGGGATTTACTACACCAGACAAA AAGCATCAGAAAGAACCTCCATTCCTTTGGATGGGTTATGAACTA SUBTYPE A:SUS

004

GTTAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGAA
AAGGAAGGAAAAATTTCAAAAATTGGACCTGAAAATCCATACAATACTCCAATATTTGCTATAAAG
AAAAAGGACAGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAACTCAAGAT
TTTTGGGAAGTTCAATTAGGAATACCGCATCCAGCGGGCCTAAAAAAGAAAAAATCAGTAACAGTA
CTGGATGTGGGGGATGCATATTTTTCAGTACCTTTAGATGAAAGCTTTAGAAAATATACTGCATTC
ACCATACCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGA
TGGAAAGGGTCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAA
AATCCAGAAATAGTTATCTACCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGG
CAGCATAGAGCAAAAATAGAAGAATTAAGAGCTCATCTGTTGAGCTGGGGATTTACTACCCCMGAC
AAAAAACATCAGAAAGAACCGCCATTTCTTTGGATGGGTTATGAAC

SUBTYPE A:SUS

005

AGAAATTTGTACAGATATGGAAAAGGAAGGAAAACTATCAAGGATTGGGCCTGAAAATCCATATAA
CACTCCAATATTTGCTATAAAGAAAAAAGACAGTACCAAGTGGAGAAAATTAGTAGATTTCAGGGA
ACTTAATAAGAGAACTCAAGATTTCTGGGAAGTTCAATTAGGAATACCACACCCGGCAGGGCTAAA
AAAGAAAAAATCAGTAACAGTACTGGATGTGGGTGATGCCTATTTTTCAGTTCCCTTATGTGAAGA
GTTTARAAAATATACTGCATTTACCATACCTAGTATAAACAATGAGACACCAGGAATTAGATATCA
GTACAATGTGCTTCCACAGGGATGGAAAGGATCACCAGCAATATTCCAAAGTAGCATGACAAAAAT
CTTAGAACCCTTTAGAGAACAAAATCCAGAAATAGTTATCTATCAATACATGGATGATTTGTATGT
AGGATCTGACTTAGAAATAGGGCAGCATAGAGCAAAAATAGAGGAACTAAGAGAACATCTATTGAG
GTGGGGATTTACCACACCAGATAAAAAACATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGA
ACTAAA

SUBTYPE D:SUS

006

CAATGGCCATTGACAGAAGAAAAAATAAAMGCATTAACAGAAATCTGTACAGAAATGGAGGAAGAA
GGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCGATAAAGAAAAAA
GATAGCACTAAATGGAGGAAATTAGTAGATTTTAGAGAGCTTAATAAAAGAACTCAAGACTTTTGG
GAGGTTCAATTAGGAATACCGCATCCAGCAGGTTTAAAAAAGAAMAAATCAGTAACAGTACTAGAT
GTGGGGGACGCATATTTTTCAGTTCCTTTAGATGAAGGCTTTAGAAAGTATACAGCGTTCACCATA
CCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGAAA
GGATCACCGGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAGCCCTTTAGAATAAAAAATCCA
GACATAATTATCTATCAATACATGGATGACTTATATGTAGGATCTGATTTAGAGATAGGGCAGCAT
AGAGCAAAAATAGAAGAGTTGAGAGCTCACCTATTGAGCTGGGGATTCACTACACCAGACAAAAAG
CATCAGAAAGAACCTCCATTCCTTTGGATGGGTTATGAACTA

SUBTYPE A:NNRTI RES EFV,NFP

007

AAACAATGGCCATTGACAGAAGAAAAGATAAAAGCATTGACAGAAATTTGTACAGACATGGAAAAG
GAAGGAAAAATTTCAAGAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAA
AAAGACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGAGAGCTTAATAAAAGAACTCAAGACTTC
TGGGAAGTTCAACTAGGAATACCACATCCTGCAGGGCTAAAGAAGAAAAAGTCAGTAACAGTACTA
GATGTGGGTGATGCATATTTTTCAGTTCCCTTATATGAAGATTTTAGAAAATATACCGCATTCACC
ATACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTTCCACAGGGATGG
AAAGGATCGCCGGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAACCTTTTAGAAAACGAAAT
CCAGAAGTGGTTATCTATCAATACATGGATGATTTGTATGTAGGGTCTGACTTAGAGATAGGGCAG
CATAGAATAAAAATAGAGGAATTAAGGGAACACCTATTGAAGTGGGGATTTACCACACCAGACAAA
AAGCATCAGAAAGAACCTCCATTTCTTTGGATGG

SUBTYPE D:SUS

008

CAATGGCCATTGACAGAAGAAAAAATAAAGGCATTGATAGAAATTTGTACAGAGATGGAAAAGGAA
GGAAAAATTTCAAGAATTGGGCCTGAGAATCCATACAATACTCCAGTATTTGCCATAAAAAAGAAR

GACAGTACWAAGTGGAGAAAATTAGTAGATTTCAGGGAACTCAATAAAAGRACCCARGACTTTTGG
GAAGTTCAATTAGGRATACCACACCCAGCAGGGTTAAAARAGAAAAAATCAGTGACAGTACTAGAT
GTGGGGGATGCRTATTTTTCAGTWCCTTTAGATGAAAGCTTCAGGAAATATACTGCATTCACCATA
CCAAGTATAAACAATGAGACACCAGGAATCAGRTATCAGTACAATGTGCTTCCACAAGGATGGAAA
GGATCACCAGCAATATTCCAAGCTAGCATGACAAAAATYCTGGAACCTTTTAGGAAACAAAATCCA
GAAATGATTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGGCAACAT
AGAGCAAAAATAGAGAAATTAAGGGAACACCTGTTRAAGTGGGGGTTTACTACACCAGACAAAAAG
CATCAGAAAGAACCTCCATTCCTTTGGATGGGTTATGAACTA

SUBTYPE B:SUS

009 AATGGCCATTGACRGAAGAAAAAATAAAGGCATTGATAGAAATTTGTACAGAGATGGAAAAGGAAG
GAAAAATTTCAAGAATTGGGCCTGAGAATCCATACAATACTCCAGTATTTGCCATAAAAARGAAAG
ACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGGGAACTCAATAAAAGAACCCAAGACTTTTGGG
AAGTTCAATTAGGRATACCACACCCAGCAGGGTTAAAARAGAAAAAATCAGYGACAGTACTAGATG
TGGGGGATGCRTATTTTTCAGTWCCTTTAGATGAAAGCTTCAGGAAATATACTGCATTYACCATAC
CRAGTRTAAACAATGAGACACCAGGAATCAGRTATCAGTACAATGTGCTTCCACAAGGATGGAAAG
GATCACCRGCAATATTCCAAGCTAGCATGACAAAAATYCTGGAACCTTTTAGGAAACAAAATCCAG
AAATGATTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGGCAACATA
GAGCAAAAATAGAGRAATTAAGGGAACACCTGTTAAAGTGGGGGTTTACTACACCAGACAAAAAGC
ATCAGAAAGAACCTCCAYTCCTTTGGATTGGTTAT

SUBTYPE B:RES NRTI, NNRTI

010 AACAATGGCCATTGACAGAAGAAAAAATAAAGGCATTGATAGAAATTTGTACAGAGATGGAAAAGG
AAGGAAAAATTTCAAGAATTGGGCCTGAGAATCCATACAATACTCCAGTATTTGCCATAAAAAAGA
AAGACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGGGAACTCAATAAAAGAACCCAAGACTTTT
GGGAAGTTCAATTAGGGATACCACACCCAGCAGGGTTAAAAGAGAAAAAATCAGCGACAGTACTAG
ATGTGGGGGATGCGTATTTTTCAGTACCTTTAGATGAAAGCTTCAGGAAATATACTGCATTCACCA
TACCAAGTATAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCCACAAGGATGGA
AAGGATCACCAGCAATATTCCAAGCTAGCATGAAAAAAATTCTGGAACCTTTTAGGAAACAAAATC
CAGAAATGATTATCTATCAATACGTGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGGCAAC
ATAGAGCAAAAATAGAGGAATTAAGGGGACACCTGTTGAAGTGGGGGTTTACTACACCAGACAAAA
AGCATCAGAAAGAACCTCCATTCCTTTGGATGGGTTATGAAC

SUBTYPE D:RES NNRTI,NRTI

011 CAGAAGARAAAATAAAAGCATTAACAGCAATTTGTGAAGARATGGAGAAGGAAGGAAAAATTACAA
AAATTGGGCCTGAAAATCCATATAACACTCCAGTATTTGCCATAAAAAAGAAGGACAGTACTAAGT
GGAGAAAATTAGTAGATTTCAGGGAACTCAATAAAAGAACTCAAGACTTTTGGGAAGTTCAATTAG
GGATACCACACCCAGCAGGTTTAAAGAAAAACAAATCAGTGACAGTACTAGATGTGGGGGATGCAT
ATTTTTCAGTCCCTTTAGATGAAAATTTCAGGAAGTATACAGCATTCACCATACCTAGTAYAAACA
ATGAGAMACCAGGGATTAGATATCAATACAATGTGCTTCCACAGGGATGGAAAGGATCACCAGCAA
TATTCCAAAGTAGTATGACAAAAATCTTAGAACCCTTTAGGGCACAAAATCCAGAAATGGTTATCT
ATCAATATGTGGATGATTTGTATGTAGGGTCTGACTTAGAAATAGGGCAACATAGAGCAAAAATAG
AGGAGTTGAGAAACCATCTATTGAAGTGGGGATTTACCACACCAGACAAAAAACATCAGAAAGAAC
CCCCATTTCTTTGGATGGGTTATGAACT

SUBTYPE C:RES NRTI,NNRTI

012 AATGGCCATTGACAGAAGAGAAAATAAAAGCAATAACAGCAATTTGTGAAGAAATGGAGAAGGAAG
GAAAAATTACAAAAATTGGGCCTGAAAATCCATATAACACTCCAGTATTTGCCATAAAAAAGAAGG
ACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGGGAACTCAATAAAAGAACTCARGASTTTTGGG
AAGTTCAATTAGGGATACCACACCCAGCAGGTTTAAAGAAAAACAAATCAGTGACAGTACTAGATG

TGGGGGATGCATATTTTTCAGTCCCTTTAGATGAAAATTTCAGGAAGTATACAGCATTCACCATAC
CTAGTATAAACAATGAGAAACCAGGGATTAGATATCAATACAATGTGCTTCCACAGGGATGGAAAG
GATCACCAGCAATATTCCAAAGTAGTATGACAAAAATCTTARAACCCTTTAGGGCACAAAATCCAG
AAATGGTTATCTATCAATATGTGGATGATTTGTATGTAGGGTCTGACTTAGAAATAGGGCAACATA
GAGCAAAAATAGAGGAGTTGAGAAACCATCTATTGAAGTGGGGATTTACCACACCAGACAAAAAAC
ATCAGAAAGAACCCCCATTTCTTTGGATGGGTTATGAAC

SUBTYPE C:RES NRTI,NNRTI

013 CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTATGGAAATGGAGAAAGAA
GGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCCATAAAGAAAAAG
GACAGTACAAAATGGAGAAAATTGGTAGATTTCAGAGAACTTAACAAGAGAACGCAAGATTTCTGG
GAAGTTCAATTAGGAATACCGCATCCTGCAGGGCTAAAAAARAARAAATCAGTAACAGTACTGGAT
GTGGGTGATGCATATTTTTCAGTTCCCTTATATGAARATTTTARGAAGTATACTGCATTCACCATA
CCCAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTACTTCCACAGGGATGGAAA
GGATCACCGGCAATATTCCAAAGTAGTATGACAAAAATCTTAGAACCCTTTAGGAAGAAAAATCCA
GAAATGGTCATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGACAGCAT
AGAACAAAAATAGAAGAATTAAGGGAACATTTATTGAGGTGGGGATTTACCACACCAGACAAAAAA
CATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE D:SUS

014 GACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGACATGGAAAGGGAAGGAAAAATTTC
AAGAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCCATAAARAAAAAAGACAGTACTAA
GTGGAGAAAATTAGTAGATTTCAGAGAACTTAATAAGAGAACTCAAGACTTTTGGGAAGTTCAGCT
AGGAATACCACATCCTGGAGGGCTAAAAAAGAAGAAATCAGTAACAGTATTGGATGTGGGTGATGC
ATATTTTTCAGTTCCCTTATATGAAGAATTTAGAAAATATACTGCATTCACCATACCTAGTACAAA
CAATGAGACACCAGGGATTAGATATCAGTACAATGTGCTTCCACAAGGATGGAAAGGATCACCAGC
AATATTCCAAAGTAGCATGACAAAAATCTTAGAACCTTTTAGAAAACAAAATCCAGAAATGGTTAT
CTGTCAATACGTGGATGATTTGTATGTAGGATCTGACTTAGAACTAGGGCAGCATAGAATAAAAAT
ARAAAAATTAAGAGAACACCTGTTAAAGTGGGGATTTACCACACCAGACAAAAAAYATCAGAAAGA
ACCTCCATTTCTTTGGATGGGTTATGAACT

SUBTYPE D:RES NRTI,NNRTI

015 AACAATGGCCATTGACAGAAGAGAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGAAAAGG
AAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCAATAAAGAAAA
AAGATAGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAACACAAGACTTTT
GGGAAGTTCAATTAGGGATACCGCATCCAGCGGGCTTAAAAAAGAAAAAATCAGTAACAGTGCTAG
ATGTGGGGGACGCATATTTCTCAGTTCCTTTACATGAAAGTTTCAGGAAGTATACTGCGTTCACCA
TACCTAGTATAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGA
AGGGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAAAATC
CAGAAATAGTTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAGC
ATAGAGCAAAAATAGAAGAGTTAAGARCTCATCTATTGAGCTGGGGGTTTACTACACCAGACAAAA
AGCATCAGAAAGAACCTCCATTCCTTTGGATGGGTTATGAA

SUBTYPE A:SUS

016 AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTGCAGATATGGAAAAA
GAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAACACCCCAATATTTGCAATAAAGAAA
AAAGATAGCACTAAATGGAGGAAATTAGTAGATTTCAGAGAGCTCAATAAGAGAACACAAGACTTC
TGGGAAGTTCAATTAGGAATACCACATCCAGCGGGCCTAAAAAAGAAAAAATCAGTAACAGTACTA
GATGTGGGGGATGCATATTTTTCAGTTCCTTTACATGAGGACTTTAGAAAGTATACTGCATTCACC
ATACCTAGTACAAACAATGAGACACCGGGAATCAGATACCAGTACAATGTGCTTCCACAAGGATGG

AAAGGATCACCAGCAATATTCCAGAGCAGCATGACAAAGATCTTAGAGCCCTTTAGATCAAAAAAT
CCACAAATAATCATCTACCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGCCAG
CATAGAGCAAAAATAGAAGAGCTGAGAGCTCATCTATTGAGCTGGGGGTTTACTACACCAGACAAA
AAGCATCAGAAAGAACCTCCATTCCTTTGGATGGGG

SUBTYPE A:SUS

017 CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTAATGAGATGGAAAAGGAA
GGAAAAATTTCAAAAATTGGGCCTGAGAATCCATACAATACTCCAATATTTGCTATAAAGAAAAAA
GACAGCACTAAATGGAGAAAATTAGTAGATTTTAGAGAGCTCAATAAAAGAACTCAAGACTTTTGG
GAAGTTCAATTGGGAATACCGCATCCTGCAGGTTTAAAAAAGAAAAAATCAGTAACAGTATTAGAT
GTGGGGGACGCCTATTTTTCAGTTCCCTTAGATGAAAGCTTTAGAAAGTATACTGCATTCACCATA
CCTAGTATAAACAATGAGACACCAGGGATCAGATATCAGTACAATGTGCTTCCACAGGGATGGAAA
GGATCACCGGCAATATTCCAGGCTAGCATGACAAAAATATTAGAACCCTTTAGATCAAAAAATCCA
GAAATAGTTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAGCAT
AGAATAAAAGTAGAGGAGTTGAGAGATCATCTATTGAAGTGGGGATTTACTACACCAGACAAAAAG
CATCAAAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE A:SUS

018 AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTAATGAGATGGAAAAG
GAAGGAAAAATTTCAAAAATTGGGCCTGAGAATCCATACAATACTCCAATATTTGCTATAAAGAAA
AAAGACAGCACTAAATGGAGAAAATTAGTAGATTTTAGAGAGCTCAATAAAAGAACTCAAGACTTT
TGGGAAGTTCAATTGGGAATACCGCATCCTGCAGGTTTAAAAAAGAAAAAATCAGTAACAGTATTA
GATGTGGGGGACGCCTATTTTTCAGTTCCCTTAGATGAAAGCTTTAGAAAGTATACTGCATTCACC
ATACCTAGTATAAACAATGAGACACCAGGGATCAGATATCAGTACAATGTGCTTCCACAGGGATGG
AAAGGATCACCGGCAATATTCCAGGCTAGCATGACAAAAATATTAGAACCCTTTAGATCAAAAAAT
CCAGAAATAGTTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAG
CATAGAATAAAAGTAGAGGAGTTGAGAGATCATCTATTGAAGTGGGGATTTACTACACCAGACAAA
AAGCATCAAAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE A:SUS

019 GCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTCTAGAAATGGAGAAGGAAGGAAA
AATTTCAAAAATTGGGCCTGAAAATCCATACAACACTCCAGTGTTTGCTATAAAGAAAAAAGATAG
CACTAAATGGAGAAAATTAGTAGATTTTAGAGAACTCAATAAGAGAACTCAAGACTTCTGGGAAGT
TCAGTTAGGAATACCACATCCAGCAGGATTAAAAAAGAAAAAATCAGTAACAGTATTAGATGTGGG
GGACGCATATTTTTCCGTTCCCTTAGATAAAGAATTTAGAAAATATACTGCATTCACCATACCTAG
TATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTCCCACAGGGATGGAAAGGATC
ACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAGCCCTTTAGAAAACAAAATCCAGAAAT
GGTTATCTATCAATACGTGGATGATTTGCTTGTAGGATCTGACTTAGAAATAGGGCAGCATAGAGC
AAAAATAGAGGAGTTAAGAGAACATCTATTGAAATGGGGATTTACCACACCAGATAAAAAACATCA
AAAAGAACCTCCATTTCTTTGGATGGGTT

SUBTYPE D:RES NRTI,NNRTI

020 AACAATGGCCATTGACAGAGGAAAAAATAAAAGCATTAACAGAAATCTGTACAGAAATGGAAAAGG
AAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCAATAAAGAAAA
AAGATAGCACTAAGTGGAGAAAATTAGTAGACTTCAGAGAGCTCAATAAAAGAACACAAGACTTTT
GGGAAGTTCAGTTAGGAATACCGCATCCAGCGGGCCTAAAAAAGAAAAAATCAGTAACAGTGCTAG
ATGTGGGGGATGCATATTTTTCAGTTCCTTTAGATAAAGAGTTCAGAAAATATACTGCATTCACCA
TACCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGA
AAGGATCACCGGCAATATTCCAGAATAGCATGCTAAAAATTTTAGAGCCCTTTAGATCAAAGAATC
CAGAAATAATTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGAGCAGC

ATAGATCAAAAGTAGAAGAGTTGAGAGCTCATCTATTGAGATGGGGACTAACTACACCAGACAAAA
AGCATCAGAAAGAACCTCCATTCCTTTGGATGGGWTATGARCTA

SUBTYPE A:SUS

021 ACAATGGCCATTGACAGAAGAAAAAATAAAAGCACTAACAGAAATTTGTATGGAAATGGAAAAGGA
AGGAAAAATTTCAAGAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCCATAAAGAAAAA
AGACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGAGAACTTAATAAGAGAACTCAAGACTTCTG
GGAGGTTCAACTAGGAATACCACATCCTGCAGGGCTAAAAAARAAAAAATCAGTAACAGTACTGGA
TGTGGGTGATGCATATTTTTCAGTTCCATTGTATGAAGACTTTAGAAAATATACCGCATTCACCAT
ACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTTCCACAAGGATGGAA
AGGATCACCAGCAATATTCCAAAGTAGTATGACAAAAATCCTAGAACCTTTTAGAAGAAAAAATCC
AGAAATAGTTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGGCAGCA
TAGAATAAAAATAGAGGAATTAAGGGAACACCTATTGAAGTGGGGATTTACCACACCAGACAAAAA
GCATCAGAAAGAACCCCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE D:SUS

022 ACAATGGCCATTGACAGAAGAAAAAATAAAAGCACTAACAGAAATTTGTATGGAAATGGAAAAGGA
AGGAAAAATTTCAAGAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCCATAAAGAAAAA
AGACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGAGAACTTAATAAGAGAACTCAAGACTTCTG
GGAGGTTCAACTAGGAATACCACATCCTGCAGGGCTAAAAAAGAAAAAATCAGTAACAGTACTGGA
TGTGGGTGATGCATATTTTTCAGTTCCCTTGTATGAAGACTTTAGAAAATATACCGCATTCACCAT
ACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTTCCACAAGGATGGAA
AGGATCACCAGCAATATTCCAAAGTAGTATGACAAAAATCCTAGAACCTTTTAGAAGAAAAAATCC
AGAAATAGTTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGGCAGCA
TAGAATAAAAATAGAGGAATTAAGGGAACACCTATTGAAGTGGGGATTTACCACACCAGACAAAAA
GCATCAGAAAGAACCCCCATTTCTTTGGATGGGTTATGAAC

SUBTYPE D:SUS

023 TTAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCACTAACAGACATTTGTAATGAAATGGAAA
AGGAAGGGAAAATTTCAAAGATTGGGCCTGAAAATCCATACAATACCCCAATATTTGCCATAAAGA
AAAAGGACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGAGAGCTTAATAAGAGAACTCAAGACT
TCTGGGAAGTTCAACTAGGAATACCACATCCTGCAGGGCTAAAGAAGAAAAAATCAGTAACAGTAC
TGGATGTGGGTGATGCATATTTTTCAGTTCCCTTGGATGAAGACTTTAGAAAATATACTGCATTCA
CCATACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTGCCACAAGGAT
GGAAAGGATCACCGGCAATATTTCAAAGTAGCATGACAAAAATCTTAGAGCCTTTTAGAAAACAAA
ATCCAGAAATGGTTATCTATCAATATATGGATGATTTGTATGTAGSATCTGACTTAGAAATAGGGC
AGCATAGAATAAAAATAGAGGAATTAAGGGAACACCTATTGAAGTGGGGATTTACCACACCAGACA
AAAAGCATCAGAAAGAACCCCCATTTCTTTGGATGGGTTATGAACT

SUBTYPE D:RES NRTI

024 AACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGAAAAGG
AGGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCGATCAAGAAAA
AAGATAGCACTAAATGGAGGAAATTAGTAGACTTCAGAGAGCTCAATAAAAGAACACAAGATTTTT
GGGAAGTKCAATTAGGGATACCACATCCAGCRGGCCTAAAAAAGAAAAAATCAGTAACAGTACTAG
ATGTGGGGGACGCATATTTTTCAGTCCCCTTAGATAAAGACTTTAGAAAATATACTGCATTCACCA
TACCTAGTACAAACAATGAGACACCAGGAATCAGATATCAATACAATGTGCTTCCACAGGGATGGA
AAGGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCTTTTAGATTAAAGAATC
CAGAAATAATTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAGC
ATAGAACAAAAATAGAGGAGTTAAGAGCCCATCTATTGAGCTGGGGGTTTACTACACCAGACAAAA
AGCATCAGAAAGAACCTCCATTCCTTTGGATGGGWTATGA

SUBTYPE A:SUS

025

AAACAATGGCCATTGACAGAAGAAAAAATAAGAGCATTAMCAGAAATTTGTACAGAAATGGAAAAG
GAAGGAAAAATTTCRAAAATTGGGCCAGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAA
AAAGACAGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAGCTTAATAAAAGAACTCAAGATTTT
TGGGAAGTTCAATTAGGAATACCGCACCCAGCGGGCCTAAAAAAGAAYAAATCAGTAACAGTACTA
GATGTGGGGGACGCATATTTTTCRGTTCCCTTAGATGAAAGYTTTAGAAAGTATACTGCGTTCACC
ATACCTAGTACAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCCACAGGGATGG
AAAGGYTCACCATCAATATTCCAGAGTAGCATGACAAAAATCTTAGARCCCTTTAGAGCAAAAAAT
CCAGAAATAATTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAG
CATAGAACAAAARTAGAAGARTTGAGAGCTCATCTATTGAGCTGGGGATTTACTACCCCAGACAAA
AARCATCAGAAAGAACCGCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE A:RES NRTI

026

AAACAATGGCCATTGACAGAAGAAAAAATAAGAGCATTAACAGAAATTTGTACAGAAATGGAAAAG
GAAGGAAAAATTTCGAAAATTGGGCCAGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAA
AAAGACAGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAGCTTAATAAAAGAACTCAAGATTTT
TGGGAAGTTCAATTAGGAATACCGCACCCAGCGGGCCTAAAAAAGAATAAATCAGTAACAGTACTA
GATGTGGGGGACGCATATTTTTCGGTTCCCTTAGATGAAAGTTTTAGAAAGTATACTGCGTTCACC
ATACCTAGTACAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCCACAGGGATGG
AAAGGCTCACCATCAATATTCCAGAGTAGCATGACAAAAATCTTAGAACCCTTTAGAGCAAAAAAT
CCAGAAATAATTATCTATCAATACGTGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAG
CATAGAACAAAAATAGAAGAATTGAGAGCTCATCTATTGAGCTGGGGATTTACTACCCCAGACAAA
AAGCATCAGAAAGAACCGCCATTTCTTTGGATGGGTTATGAACTAA

SUBTYPE A:RES NRTI,NNRTI

027

AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAGATGGAAAAG
GAAGGGAAAATTTCAAAAATTGGACCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAA
AAAGATAGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAGAGAACTCAGGACTTC
TGGGAAGTTCAATTAGGAATACCACACCCAGCAGGTTTAAAAAAGAAGAAATCGGTAACAGTACTA
GATGTGGGGGATGCATATTTTTCAGTTCCTTTAGATGAAAGCTTTAGAAAGTATACTGCATTCACC
ATACCTAGTACAAACAATGAGACACCAGGAGTCAGGTATCAATATAATGTGCTTCCACAGGGATGG
AAAGGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTATAGATCAAAAAAT
CCAGAAATAATTATTTATCAATACATGGATGATTTGTATGTAGCATCTGATTTAGAAATAGGACAA
CATAGAGCAAAAATAGAGGAGCTGAGAGCTCATCTATTAAGTTGGGGGTTTACTACACCAGACAAA
AAGCATCAGAAAGAACCCCCATTTCTTTGGATGG

CRF01_AE:RES NNRTI

028

TGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAGATGGAAAAGGAAGGG
AAAATTTCAAAAATTGGACCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAAAAAGAT
AGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAGAGAACTCAGGACTTCTGGGAA
GTTCAATTAGGAATACCACACCCAGCAGGTTTAAAAAAGAAGAAATCGGTAACAGTACTAGATGTG
GGGGATGCATATTTTTCAGTTCCTTTAGATGAAAGCTTTAGAAAGTATACTGCATTCACCATACCT
AGTACAAACAATCAGACACCAGGAGTCAGGTATCAATATAATGTGCTTCCACAGGGATGGAAAGGA
TCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTATAGATCAAAAAATCCAGAA
ATAATTATTTATCAATACGTGGATGATTTGTATGTAGCATCTGATTTAGAAATAGGACAACATAGA
GCAAAAATAGAGGAGCTGAGAGCTCATCTATTAAGTTGGGGGTTTACTACACCAGACAAAAAGCAT
CARAAAGAACCCCCATTTCTTTGGATGGGTTATGAACT

CRF01_AE :RES NRTI,NNRTI

029

AACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTAAWGARATGGAAAAGG
AAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATATAATACTCCAATATTTGCMATAAAGAAAA
AAGACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGAGAACTWAATAARAGAACTCAAGACTTYT
GGGAARTTCAATTAGGAATACCRCATCCTGCAGGGCTAAAAAAYAAAAARTCAGTAACAGTACTRG
ATGTGGGAGATGCATATTTTTCAGTTCCCTTAYRTGAAGATTTTAGAAAGTATACTGCATTTACCA
TACCTAGTRYAAAYAATGAGACACCAGGGRTTAGATATCAGTACAATGTGCTTCCACAGGGATGGA
AAGGRTCACCAGCAATATTYCAAAGTAGCATGACAAAAATCTTAGAACCTTTTAGAAAACAAAATC
CAGARGTGGTTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGKMARC
ATAGAACAAAAATAGAGGAATTAAGGGAACAYCTATTAARGTGGGGRTTTACCACACCAGACAAAA
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SUBTYPE D:RES NRTI

030

AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTGAAGAGATGGAAAAG
GAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAGTGTTTGCTATAAAGAAA
AAGGATAGCACTAAATGGAGAAAATTAGTAGATTTTAGAGAGCTCAATAAAAGAACACAGGACTTC
TGGGAAGTTCAATTAGGAATACCCCATCCTGCAGGTTTAAAAAAGAAAAAATCAGTAACAGTACTA
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ATACCTAGTATAAACAATGAGACACCAGGAACCAGGTATCAGTACAATGTGCTTCCACAAGGATGG
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CCAGGAATAATTATTTATCAATACGTGGATGACTTGTATGTAGCATCTGATTTAGAAATAGGGCAG
CATAGAACAAAAGTAGAAGAATTGAGAGCTCATCTATTGAGTTGGGGATTTACTACACCAGACAAA
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SUBTYPE A:RES NRTI,NNRTI

031

GTTAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAMATGGAA
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TTTTGGGAAGTCCAATTAGGGATACCACACCCAGCAGGGTTGAAAAAGAAAAAATCAGTGACAGTA
TTGGATGTGGGGGATGCATATTTTTCAGTTCCTTTAGATGAAGACTTCAGAAAATATACTGCATTT
ACAATACCTAGTATAAACAATGAAACACCAGGAATTAGATATCAGTATAATGTGCTTCCACAGGGA
TGGAAAGGATCACCAGCAATATTCCAAAGCAGCATGACAAAAATTTTAGAGCCCTTTAGGGCGCAA
AACCCAGAAATAGWTATCTATCAATACATGGATGACTTGTATGTAGGATCTGACTTAGAAATAGGG
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SUBTYPE B:RES NRTI

032

CAATGGCCATTGACAGAAGAAAAAATAAAAGCACTAACAGAAATTTGTAAAGAAATGGAAAAGGAA
GGAAAAATTTCAAGAATTGGGCCTGAGAATCCATACAATACTCCAATATTTGCCATAAGAAAGAAA
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GAAGTTCAATTAGGGATACCACACCCAGCAGGGTTAAAAAAGAAAAAATCAGTGACAGTACTAGAT
GTGGGAGATGCATATTTTTCAGTTCCTTTAGATGAAGGCTTCAGAAAATATACTGCATTCACCATA
CCTAGTAKRAAYAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTTCCGCAAGGATGGAAA
GGATCACCGGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAACCCTTTAGGAAACAAAATCCA
GAAATGGTTATCTRTCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGGCAACAT
AGAATAAAAATAGGAGAGTTAAGGGAACACCTATTGAAGTGGGGATTTACTACACCAGACAAAAAG
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SUBTYPE D:RES NRTI,NNRTI

033

AAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCAATAAAGAAAAAAGA
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AGTTCAATTAGGGATACCGCATCCAGCGGGCCTAAAAAAGAAAAAATCAGTAACAGTACTAGATGT
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SUBTYPE A:SUS

034 AACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGATATGGAAAAGG
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CAGAAATAGTTATTTATCAGTACATGGATGATTTATATGTAGGATCTGACTTAGAAATAGGGCAGC
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SUBTYPE D:SUS

035 GATGGAAAAGGAAGGAAAGATTTCAAAAATAGGGCCTGAAAATCCATACAATACTCCAGTATTTGC
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TGCATTCACAATACCTAGTATAAACAATGAAACACCAGGGATTAGATATCAATACAATGTGCTTCC
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ATCAAAAAATCCAGAAATAATTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGA
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SUBTYPE B:SUS

036 AAAGGAAGGAAAAATTTCAAGAATTGGGCCTGAAAATCCATACAACACTCCATTATTTGCTATAAA
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CTTCTGGGAAGTTCAATTAGGAATACCGCATCCAGCAGGTTTAAAAAAGAAAAAATCAGTAACAGT
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CACCATACCTAGTACAAACAATGAGACACCAGGAGTCAGGTATCAGTACAATGTGCTCCCACAAGG
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SUBTYPE A:SUS

037 AACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGAAAAGG
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ATGTGGGAGATGCATATTTTTCAGTTCCTTTAGATGAAAGTTTTAGAAAATACACTGCATTCACCA
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ATAGAACAAAAATAGAAGAGTTAAGAGCTCATCTACTGAGCTGGGGATTTACTACACCAGACAAAA
AACATCAGAAAGAACCTCCATTCCTTTGGATGGGTTATGAAC

CRF01_AE :RES NNRTI

038 AACAATGGCCATTGACAGAAGAGAAAATAAAAGCATTAACAGAAATTTGTGGAGAAATGGAAAAGG
AAGGAAAAATTTCAAAGATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCAATAAAGAAAA
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GGGAGGTTCAATTAGGAATACCACATCCAGCAGGCCTAAAAAAGAAGAAATCAGTAACAGTACTGG
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CAGACATAGTGATCTATCAATACATGGATGACTTGTATGTAGGATCTGATCTAGAAATAGGGCAGC
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AGCATCAGAAAGAACCCCCATTCCTTTGGATGGGTTATGAACTA

SUBTYPE A:SUS

039 AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGATATGGAAAAG
GAAGGAAAGATTTCAAAAATTGGGCCTGAAAATCCATATAATACTCCAATATTTGCAATAAAGAAA
AAAGATAGTACCAAATGGAGAAAATTGGTAGATTTCAGAGAGCTCAACAAAAGAACACAAGACTTT
TGGGAAGTTCAATTAGGAATACCGCATCCAGCGGGCTTAAAAAAGAAAAAATCAGTAACAGTACTA
GATGTGGGGGATGCATATTTTTCAGTTCCTTTAGATGAAAACTTTAGAAAATACACCGCGTTCACC
ATACCGAGTATAAACAATGAGACACCAGGAGTCAGATATCAGTACAATGTGCTTCCACAGGGATGG
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CCAGAAATAATTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAG
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SUBTYPE A:SUS

040 CAATGGCCATTGACAGAAGAGAAAATAAAAGCTTTAATAGAAATTTGTACAGAAATGGAAAAGGAA
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GAAGTCCAATTAGGGATACCTCACCCCGCGGGTCTAAAGAAGAAAAAATCAGTAACAGTACTAGAT
GTGGGGGATGCATATTTCTCAGTTCCCTTAGATGAAAACTTTAGAAAGTATACAGCATTCACTATA
CCTAGTGTAAATAATGAGACACCAGGGATTAGATACCAGTACAATGTGCTGCCTCAGGGATGGAAA
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GAAATGGTAATTTGCCAATATATGGATGATTTATATGTAGGATCTGATTTAGAAATAGGGCAGCAT
AGAGCAAAAATAGAAGAATTAAGAAAACATCTATTGAATTGGGGATTTACCACACCAGATAAAAAA
TATCAGAAAGAACCCCCATTCCTTTGGATGG

SUBTYPE G:RES NRTI

041 CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTATAGAGATGGAAAAGGAA
GGAAAAATTTCAAAAATTGGGCCTGAGAATCCATACAATACTCCAATATTTGCTATAAAGAAAAAA
GACAGCACTAAATGGAGGAAACTAGTAGATTTTAGAGAGCTCAATAAAAGAACACAAGACTTCTGG
GAAGTTCAATTAGGGATACCGCATCCAGCGGGACTAAAAAAGAAAAAATCAGTAACAGTACTGGAT
GTGGGGGACGCATATTTTTCAGTTCCTTTACATAAAGACTTTAGAAAATATACTGCATTCACCATA
CCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGAAA
GGATCACCGGCAATTTTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAATAATCCA
GAAATAGTTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAACTAGGGCAGCAT
AGAGCAAAAATAGAAGAGTTGAGGGCGCATTTATTGAGCTGGGGATTAACTACCCCAGACAAAAAG

CATCAGAAAGAGCCGCCATTTCTTTGGATGG

SUBTYPE A:SUS

042 ACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTGACAGAAATTTGTACAGAGATGGAAAAGGA
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GGAAGTTCAATTAGGGATACCGCATCCAGCGGGCCTAAAAAAGAAGAAATCAGTAACAGTACTAGA
TGTGGGGGACGCATATTTTTCAGTTCCTTTAGATGTAGACTTTAGAAAGTATACTGCGTTCACCAT
ACCTAGTACAAACAATGAGACACCAGGAATAAGGTATCAGTACAATGTGCTTCCACAGGGATGGAA
AGGATCACCGGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAAAATCC
AGAAATAATTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGTCAGCA
TAGAGCAAAAGTAGAGGAGTTGAGAGCTCATCTATTGAGTTGGGGGTTTACTACACCAGATAAAAA
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SUBTYPE A:SUS

043 AACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGAAAAGG
AAGGAAAAATTTCAAAAATTGGGCCTGAGAATCCATACAATACCCCAATATTTGCTATAAAGAAAA
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ATGTGGGTGATGCATATTTTTCAGTTCCCTTAGATGAAGACTTTAGAAAATATACCGCATTCACTA
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ATAGAACAAAAATAGAGGAATTAAGGGGACACCTATTGAAGTGGGGATTCACCACACCAGACAAAA
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SUBTYPE D:SUS

044 AGTTAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACGGAAATGGA
AAAGGAGGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCGATAAA
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CTTTTGGGAAGTTCAATTAGGGATACCGCATCCAGCGGGCCTAAAAAAGAAAAAATCAGTAACAGT
ACTGGATGTGGGGGACGCATACTTTTCAGTTCCTTTACATAAGGACTTTAGAAAGTATACTGCGTT
CACCATACCTAGTACCAACAATGAGACACCAGGAATCAGATATCAGTACAATGTACTTCCACAGGG
ATGGAAAGGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAA
AAATCCAGAAATAATCATCTATCAATACATGGATGATTTGTATGTAGGATCTGATTTAGAAATAGG
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SUBTYPE A:SUS

045 CAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTTTAGAGATGGAAAAGGAGGGAAAGATTTCAA
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GGATACCGCATCCAGCGGGCCTAAAAAAGAAGAAATCAGTAACAGTACTAGATGTGGGGGATGCAT
ATTTTTCAGTTCCTTTACATGAAGACTTTAGAAAGTATACTGCATTCACCATACCTAGCACAAACA
ATGAGACACCAGGAATCAGATATCAGTAYAATGTGCTTCCACAAGGATGGAAAGGATCACCAGCAA
TATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGGTTAAAAAATCCAGAAATAATTATCT
ATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAGATAGGGCAGCATAGRACAAAAATAG
AAGAGTTGAGGGCTCATCTATTGAGCTGGGGATTTACTACACCAGACAAAAAGCATCAGAAAGAAC
CTCCATTCCTTTGGATGGGTTATGAACTA

CRF01_AE:SUS

046

CAATGGCCATTGACAGAGGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGAAAAGGAA
GGAAAAATTTCAAGAATTGGGCCTGAAAATCCATATAATACTCCAATATTTGCCATAAAGAAAAAA
GACAGTACTAAATGGAGGAAATTAGTGGATTTCAGAGAGCTCAATAAAAGAACTCAAGATTTTTGG
GAAGTTCAATTAGGAATACCGCATCCAGCGGGCTTAAAAAAGAAAAAATCAGTAACAGTACTGGAT
GTGGGGGACGCATATTTTTCAGTTCCCTTAGATGAAGGCTTTAGGAAGTATACGGCGTTCACCATA
CCTAGTACAAACAATGAGACACCAGGGATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGAAA
GGGTCTCCAGCAATATTCCAGAGTAGTATGACAAAAATCTTAGAGCCCTTTAGGCTAAAAAATCCA
GAAGTAACTATCTATCAATACATGGATGACTTATATGTAGGGTCTGATTTAGAAATAGGGCAGCAT
AGAACAAAAGTAGAGGAGTTGAGAGATCATCTATTGAGCTGGGGATTAACTACACCAGACAAAAAG
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SUBTYPE A:RES NNRTI

047

CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTATAGATATGGAAAAGGAA
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GAAGTTCAATTAGGAATACCACACCCTGCAGGGCTAAAAAAGAAAAAATCAGTAACAGTACTGGAT
GTGGGTGATGCATATTTTTCAGTTCCCCTATATGAAGATTTTAGAAAATATACTGCATTCACCATA
CCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGAAA
GGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTATAGATTAAAAAATCCA
GAAATAATTATCTATCAATACATGGATGACTTGTATGTGGGATCTGATTTAGAAATAGGGCAGCAT
AGAACAAAAATAGAAGAGTTGAGAGCTCATCTATTGAGCTGGGGATTTACCACACCAGACAAAAAG
CATCAAAAAGAACCTCCATTTCTTTGGATGG

SUBTYPE D:SUS

048

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ACAAGACTTTTGGGAAGTTCAATTAGGAATACCACATCCAGCAGGCCTGAAGAAGAAAAAATCAGT
AACAGTACTAGATGTGGGGGATGCATATTTTTCAGTTCCTCTAGATGAAAGCTTTAGAAAGTATAC
TGCATTCACCATACCTAGTAGAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCC
ACAAGGATGGAAAGGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGATCCCTTTAG
ATCAAAAAATCCAGAAATAGTTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGA
AATAGAGCAGCACAGAACAAAAATAGAAGAATTAAGAGCTCATCTATTGAGCTGGGGATTCACTAC
ACCAGACAAAAAGCATCAGAAAGAACCTCCATTCCTTTGGATGGG

CRF01_AE:SUS

049

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AATAAAGAAAAAAGATAGCACTAAGTGGAGGAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAAC
ACAAGACTTTTGGGAAGTCCAATTAGGAATACCGCATCCAGCGGGCCTAAAAAARAAAAAATCAGT
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AGCATTCACCATACCTAGTAYAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCC
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CRF01_AE :SUS

050

AAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCCATAAAGAAAAAAGA
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GATCCAATTAGGAATACCCCATCCCGCAGGTTTAAAAAAGAAYAAATCAGTCACAGTACTAGATGT
GGGGGATGCATATTTTTCAGTCCCCTTAGATAAAGATTTTAGAAAATATACAGCATTCACTATACC
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CATAGTGATCTACCAATATATGGATGATTTATATGTAGGATCWGACYTAGAAATAGGGCARCATAG
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TCAGAAAGAACCTCCATTMCAATGGATGGGATATGAGCTCCATCCTGACAAATGGACGGTACAGCC
TATACAGCTGCCAGAAAAAGAAAGCTGGACTGTCAATGATATACAAAAGTTAGTGGGAAAACTAAA
TTGGGCAAGTCAGATTTATGCA

SUBTYPE G:RES NNRTI

051 CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTCAAGAGATGGAAAAGGAA
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GAAGTTCAATTAGGAATACCGCATCCTGCAGGTTTAAAGAAGAAAAAAGCAGTAACAGTACTGGAT
GTGGGGGATGCATATTTTTCAGTGCCTTTAGATGAAAACTTTAGAAAGTATACTGCATTCACCATA
CCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGAAG
GGATCACCAGCAATATTTCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGAGCACAAAATCCA
GGAATAATTATCTATCAATACATGGATGACTTATATGTAGGATCTGATTTAGAAATAGGGCAACAT
AGAGCAAAAGTGGAGGAGTTGAGAGCTCATCTATTACAATGGGGATTTACTACACCAGATAAAAAA
CATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAAC

SUBTYPE A:SUS

052 ATCCATACAATRBBSSAVKRKYDGCTWTAWWGAAWAAAGACAGCACTAAAYGGAGAADATTAGTAS
AYYTCAGAGAACTTAATAAAAGAACACCAGACTTTTGGGAAGTTCAATTAGGGATACCGCATCCAG
CGGGCCTAGAAAAGAAAAAATCAGTAACAGTATTGGATGTGGGGGACGCATATTTTTCAGTGCCTT
TAGATGAAAACTTTAGAAAATATACTGCATTCACCATACCTAGTACAAACAATGCGACACCAGGAG
TCAGGTATCAGTACAATGTACTTCCACAGGGATGGAAAGGATCCCCAGCAATATTCCAGAGTAGCA
TGACAAAAATCTTAGAGCCCTTCAGATCTAAAAATCCAGACATAATTATCTATCAATACGTGGATG
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ATTTATTGAGTTGGGGATTDDCTACACCAGACAAAAAGCATCAGAAAGAAC

SUBTYPE A:RES NNRTI/NRTI

053 GTCCAAAAGTTAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAG
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CTATAAAGAAAAAAGACAGTACCAAGTGGAGAAAATTAGTAGATTTCAGGGAACTTAATAAGAGAA
CTCAAGATTTCTGGGAAGTTCAATTAGGAATACCACACCCGGCAGGGCTAAAAAARAAAAAATCAG
TAACAGTACTGGATGTGGGTGATGCCTATTTTTCAGTTCCCTTATGTGAAGAGTTTARAAAATATA
CTGCATTTACCATACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTTC
CACAGGGATGGAAAGGATCACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAACCCTTTA
GAGAACAAAATCCAGAAATAGTTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAG
AAATAGGGCAGCATAGAGCAAAAATAGAGGAACTAAGAGAACATCTATTGAGGTGGGGATTTACCA
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SUBTYPE D:SUS

054 AAAAAGTTAAAACAATGGCCATTGACAGAAGAAAAAATAAAGGCATTGATAGAAATTTGTACAGAG
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CARGACTTTTGGGAAGTTCAATTAGGRATACCACACCCAGCAGGGTTAAAARAGAAAAAATCAGTG
ACAGTACTAGATGTGGGGGATGCRTATTTTTCAGTWCCTTTAGATGAAAGCTTCAGGAAATATACT

GCATTCACCATACCRAGTRTAAACAATGAGACACCAGGAATCAGRTATCAGTACAATGTGCTTCCA
CAAGGATGGAAAGGATCACCRGCAATATTCCAAGCTAGYATGACAAAAATYCTGGAACCTTTTAGG
AAACAAAATCCAGAAATGATTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAA
ATAGGGCAACATAGAGCAAAAATAGAGRAATTAAGGGRACACCTGTTRAAGTGGGGGTTTACTACA
CCAGACAAAAAGCATCAGAAAGAACCTCCATTYCTTTGGATGGGGTTATGAAM

SUBTYPE D:SUS

055 TTTTCCAAAAGTTAAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTC
AAGAGATGGAAAAGGAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAGTAT
TTGCTATAAAGAAAAAAGATAGCACAAAATGGAGAAAATTAGTAGATTTTAGAGAACTTAATAAAA
GAACTCAGGATTTTTGGGAAGTTCAATTAGGAATACCGCATCCTGCAGGTTTAAAGAAGAAAAAAG
CAGTAACAGTACTGGATGTGGGGGATGCATATTTTTCAGTGCCTTTAGATGAAAACTTTAGAAAGT
ATACTGCATTCACCATACCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGC
TTCCACAGGGATGGAAGGGATCACCAGCAATATTTCAGAGTAGCATGACAAAAATCTTAGAGCCCT
TTAGAGCACAAAATCCAGGAATAATTATCTATCAATACATGGATGACTTATATGTAGGATCTGATT
TAGAAATAGGGCAACATAGAGCAAAAGTGGAGGAGTTGAGAGCTCATCTATTACAATGGGGATTTA
CTACACCAGATAAAAAACATCAGAAAGAACCTCCATTTCTTTGGATGGGGTTATGAAC

SUBTYPE A:SUS

056 ATATTATGMHATWTRMAKAARMAAAGAWAGCACTAAAKGGARRAAATTAGTAGATTTCAGAGAGCT
CAACAAAAGAACACAAGACTTTTGGGAAGTTCAGTTAGGGATACCGCATCCAGGGGGCCTAAAAAA
GAAGAAATCAGTAACAGTACTGGATGTGGGVGATGCATATTTTTCAGTTCCCTTAGATGAAAGCTT
TAGAAAATATMCTGCATTCACCATACCTAGTACAAACAATGAGAGACCAGGAATAAGGTATCAGTA
CAATGTGCTTCCACAGGGATGGAAAGGATCACCAGCAATCTTCCAGAGTAGTATGACAAAAATCTT
AGAGCCCTTTAGACTAWAAWWTYYWKAAAWAATTATCTGTCAATACGTGGATGACTTGTATGTAGG
ATCTGATTTAGAAATAGGGCAGCATAGAGCAAAAATDGCAGAATTAAGAGCTCATCTATGRAGCTG
GGGATTHHAYACACCAGACAAAAAGCATCAARDDWRAACCCC

SUBTYPE A:RES NNRTI/NRTI

057 CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAGATGGAAAAGGAA
GGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAGTATTTGCTATAAAGAAAAAA
GATAGCACAAAATGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAACACAAGACTTTTGG
GAAGTTCAATTAGGAATACCGCATCCAGCAGGCCTAAAAAAGAAAAGATCAGTAACAGTGCTAGAT
GTGGGAGATGCATATTTTTCAGTTCCTTTACATAAAGATTTTAGAAAGTATACTGCATTCACCATA
CCTAGTACAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCCACAGGGATGGAAA
GGATCACCAGCAATATTCCAGTATAGCATGACAAAAATCTTAGAGCCCTTTAGATTAAAAAATCCA
GAAATAGTTATCTATCAATACATGGATGACTTGTATGTGGGATCTGATTTAGAAATAGGGCAGCAT
AGAACAAAAATAGAAGAATTAAGAGCTCATCTATTGAGCTGGGGATTTACTACACCAGACAAAAAG
CATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAAC

SUBTYPE A:SUS

058 WWTAWADAGAMAAGATGGCACTACATGKAGGAAATTAGTAGATTTCAGAGAACTCAATAAAAGAAC
ACAAGACTTTTGGGAAGTTCAGTTGGGAATACCACATCCAGGAGGCCTAGAAAAGAAAAAATCART
AACAGTACTAGATGTGGGGGATGCATATTTTTCAGTTCCTTTGCATGAAGACTTTAGAAAATATAC
TGCATTCACCATACCTAGTATAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCC
ACAGGGATGGAAAGGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAG
AGCAAAGAATCCAGAGATGACTATTTGTCAATACGTGGATGACTTGTATGTATCATCTGATTTAGA
AATAGAGCAGCATAGAGCAAAAATDGDDGAGTTGAGAGCTCATCTATTGAACTGGGGATTTACYAC
CCCAGACAAAAAGCATCAGADAGAA

SUBTYPE A:RES NNRTI/NRTI

059

AAAAAATAAAAGCATTAACAGAAATTTGTATGGAAATGGAGAAAGAAGGAAAAATTTCAAAAATTG
GGCCTGAAAATCCATACAATACTCCAATATTTGCCATAAAGAAAAAGGACAGTACAAAATGGAGAA
AATTGGTAGATTTCAGAGAACTTAACAAGAGAACGCAAGATTTCTGGGAAGTTCAATTAGGAATAC
CGCATCCTGCAGGGCTAAAAAARAARAAATCAGTAACAGTACTGGATGTGGGTGATGCATATTTTT
CAGTTCCCTTATATGAAGATTTTAGGAAGTATACTGCATTCMCCATACCCAGTATAAACAATGAGA
CMCCAGGAATTAGATATCAGTACAATGTACTTCCACAGGGATGGAAAGGATCACCGGCAATATTCC
AAAGTAGTATGACAAAAATCTTARAACCCTTTAGGAAGAAAAATCCAGAAATGGTCATCTATCAAT
ACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGACAGCATAGAACAAAAATAGAAGAAT
TAAGGGAACATTTATTGAGGTGGGGATTTACCACACCAGACAAAAAACATCAGAAAGAACCTCCAT
TTCTTTGGATGGGTTA

SUBTYPE B:SUS

060

AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGCAATTTGTGACGAAATGGAAAAA
GAAGGAAAGATTACAAAAATTGGGCCTGAAAATCCATATAACACTCCAGTATTTGCTATAAAAAAG
AAGGACAGTACAAAATGGAGAAAATTAGTAGATTTCAGGGAACTCAATAAAAGAACTCAAGACTTT
TGGGAAGTTCAATTAGGAATACCGCACCCGGCAGGGTTAAAAAAGAAAAAATCAGTGACAGTACTG
GATGKGGGGGATGCATATTTTTCAGTACCTTTAGATAAAGACTTCAGGAAATATACTGCATTCACC
ATACCTAGTATAAACAATGAAACACCGGGAATTAGATATCAATATAATGTGCTTCCACAAGGATGG
AAAGGATCACCAGCAATATTCCAGAGTAGCATGACAAGAATCTTAGAGCCTTTTAGAGCAAAAAAC
CCAGAAATGGTTATCTATCAATATATGGATGACTTATATGTAGGATCTAATTTAGAAATGATGCAA
CATAGAGCAAAAATAGAGGAGTTAAGAGAACACCTATTGAGATGGGGATTTACCACACCAGACAAG
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SUBTYPE C:SUS

061

CAATGGCCATTGACAGAAGAAAAAATAAAAGCACTAACAGAAATTTGTATAGACATGGAAAAGGAA
GGAAAAATTTCAAGAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAAAAA
GACAGTACCAAGTGGCGAAAATTAGTAGATTTCAGAGAACTTAATAAGAGAACTCAAGATTTCKGG
GAAGTTCAACTAGGAATACCACATCCTGCAGGGCTAAAAAAGAAAAAATCAGTTACAATACTGGAT
GTGGGTGATGCATATTTTTCAGTTCCCTTGGATAAAGAATTTAGAAAATACACTGCATTCACCATA
CCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTATAATGTGCTTCCACAAGGGTGGAAA
GGATCACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAGCCCTTTAGGAAACAAAATCCA
GAAATAGTTATCTRTCAATACATGGATGACTTGTATGTAGGGTCTGACTTAGAAATAGGGCAGCAT
CGAGCAAAAATAGAACAGTTGAGAGCTCATCTATTGAGATGGGGATTTAMTACACCAGACAAGAAG
CATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTMAA

SUBTYPE D:RES NNRTI

062

TGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAGATGGAAAAGGAAGGG
AAAATTTCAAAAATTGGACCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAAAAAGAT
AGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAGAGAACTCAGGACTTCTGGGAA
GTTCAATTAGGAATACCACACCCAGCAGGTTTAAAAAAGAAGAAATCGGTAACAGTACTAGATGTG
GGGGATGCATATTTTTCAGTTCCTTTAGATGAAAGCTTTAGAAAGTATACTGCATTCACCATACCT
AGTACAAACAATGAGACACCAGGAGTCAGGTATCAATATAATGTGCTTCCACAGGGATGGAAAGGA
TCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTATAGATCAAAAAATCCAGAA
ATAATTATTTATCAATACATGGATGATTTGTATGTAGCATCTGATTTAGAAATAGGACAACATAGA
GCAAAAATAGAGGAGCTGAGAGCTCATCTATTAAGTTGGGGGTTTACTACACCAGACAAAAAGCAT
CAGAAAGAACCCCCATTTCTTTGGATGGGTTATGAAC

SUBTYPE A:RES NNRTI

063

AACAATGGCCATTGACAGAAGAAAAAATAAAGGCATTGATAGAAATTTGTACAGAGATGGAAAAGG
AAGGAAAAATTTCAAGAATTGGGCCTGAGAATCCATACAATACTCCAGTATTTGCCATAAAAAAGA
AAGACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGGGAACTCAATAAAAGAACCCAAGACTTTT
GGGAAGTTCAATTAGGGATACCACACCCAGCAGGGTTAAAAGAGAAAAAATCAGCGACAGTACTAG
ATGTGGGGGATGCGTATTTTTCAGTACCTTTAGATGAAAGCTTCAGGAAATATACTGCATTCACCA
TACCAAGTATAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCCACAAGGATGGA
AAGGATCACCAGCAATATTCCAAGCTAGCATGAAAAAAATTCTGGAACCTTTTAGGAAACAAAATC
CAGAAATGATTATCTATCAATACGTGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGGCAAC
ATAGAGCAAAAATAGAGGAATTAAGGGGACACCTGTTGAAGTGGGGGTTTACTACACCAGACAAAA
AGCATCAGAAAGAACCTCCATTCCTTTGGATGGGTTATGAAC

SUBTYPE D:RES NNRTI/NRTI

064

TCCAAAAGTTAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGA
CATGGAAAAGGAAGGAAAAATTTCAAAAATTGGGCCTGAAAACCCATACAATACTCCAGTATTTGC
TATAAAGAAAAAAGATAGCACTAAATGGAGAAAACTAGTAGATTTTAGAGAGCTCAATAAAAGAAC
TCAAGACTTCTGGGAGGTTCAATTAGGAATACCGCATCCCGCAGGTTTAAAAAAGAAGAAATCAGT
AACAGTACTAGATGTGGGGGACGCATATTTCTCAGTTCCTTTAGATGAAAATTTTAGAAAGTACAC
TGCATTCACCATACCTAGTATAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCC
ACAAGGATGGAAGGGATCACCAGCAATATTTCACAGTAGCATGACAAAAATCTTAGAGCCCTTTAG
ATCAAAAAATACAGAAATAATTATCTATCAATACATGGATGACCTGTATGTAGCATCTGATTTAGA
AATAGGGCAGCATAGAGCAAAAGTAGAGGAATTAAGAGCTCATCTATTGAGCTGGGGGCTTACTAC
ACCAGACAAAAAGCATCAGAAAGAACCTCCATTTCTTTGGATGGGTTA

SUBTYPE A:RES NNRTI

065

GTTAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTCTAGAAATGGAG
AAGGAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAACACTCCAGTGTTTGCTATAAAG
AAAAAAGATAGCACTAAATGGAGAAAATTAGTAGATTTTAGAGAACTCAATAAGAGAACTCAAGAC
TTCTGGGAAGTTCAGTTAGGAATACCACATCCAGCAGGATTAAAAAAGAAAAAATCAGTAACAGTA
TTAGATGTGGGGGACGCATATTTTTCCGTTCCCTTAGATGAAGAATTTAGAAAATATACTGCATTC
ACCATACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTCCCACAGGGA
TGGAAAGGATCACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAGCCCTTTAGAAAACAA
AATCCAGAAATGGTTATCTATCAATACGTGGATGATTTGCTTGTAGGATCTGACTTAGAAATAGGG
CAGCATAGAGCAAAAATAGAGGAGTTAAGAGAACATCTATTGAAATGGGGATTTACCACACCAGAT
AAAAAACATCAAAAAGAACCTCCATTTCTTTGGATGGGWTATGAACTA

SUBTYPE A:RES NNRTI/NRTI

066

ATGGCCATTGACAGAAGAAAAAATAARAGCATTAACAGAAATTTGTGCAGACATGGAAAAGGAAGG
AAAAATTTCAAAAATTGGGCCTGAAAACCCATACAATACTCCAGTATTTGCTATAAAGAAAAAAGA
TAGCACYAAATGGAGAAAACTAGTAGATTTTAGAGAGCTCAATAAAAGAACTCAAGACTTCTGGGA
AGTTCAATTAGGAATACCGCATCCCGCAGGKTTAAAAAAGAAGAAATCAGTAACAGTACTAGATGT
GGGGGACGCATATTTCTCAGTTCCTTTAGATGAAAATTTTAGAAAGTACACTGCATTCACCATACC
TAGTATAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGAAGGG
ATCACCAGCAATATTTCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAAAATACAGA
AATAATTATCTATCAATACATGGATGACCTGTATGTAGCATCTGATTTAGAAATAGGGCAGCATAG
AGAAAAAGTAGAGGAATTAAGAGCTCATCTATTGAGTTGGGGGCTTACTACACCAGACAAAAAGCA
TCAGAAAGAACCTCYATTTCTTTGGATGGGTTA

SUBTYPE A:RES NNRTI

067

AAACAATGGCCATTGACAGAAGAAAAAAWAAAAGCATTAACMGAGATTTGTACAGATATGGAAAAG
GAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCCATATTTGCAATAAAGAAA

AAAGATAGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAACTTAATAAAMGAACACAAGACTTT
TGGGAAGTTCAATTAGGAATACCGCATCCAGCGGGCCTAAAAAGAAAAAGATCAGTAACAGTACTA
GATGTGGGGGATGCATATTTTTCAGTACCCTTATATGAAGATTTTAGAAAGTATACTGCATTCACC
ATACCTAGTACAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGTTGCCGCAGGGATGG
AAGGGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAAAAT
CCAGAAATAATTATCTATCAATACATGGATGACTTATATGTAGGATCTGATTTAGAAATAGGGCAG
CATAGAGCAAAGATAGAAGAGCTAAGAGCTCATCTATTGAGCTGGGGATTTACTACACCAGACAAA
AAGCATCAAAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTAA

SUBTYPE A:SUS

068 AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTCTAGAAATGGAGAAG
GAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAACACTCCAGTGTTTGCTATAAAGAAA
AAAGATAGCACTAAATGGAGAAAATTAGTAGATTTTAGAGAACTCAATAAGAGAACTCAAGACTTC
TGGGAAGTTCAGTTAGGAATACCACATCCAGCAGGATTAAAAAARAAAAAATCAGTAACAGTATTA
GATGTGGGGGACGCATATTTTTCCGTTCCCTTAGATGAAGAATTTAGAAAATATACTGCATTCACC
ATACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTCCCACAGGGATGG
AAAGGATCACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAGCCCTTTAGAAAACAAAAT
CCAGAAATGGTTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGGCAG
CATAGAGCAAAAATAGAGGAGTTAAGAGAACATCTATTGAAATGGGGATTTACCACACCAGATAAA
AAACATCARAAAGAACCTCCATTTCTTTGGATGGGTTATGAAC

SUBTYPE A:SUS

069 CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTGCAGACATGGAAAAGGAA
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GAGGTTCAATTAGGAATACCGCATCCCGCAGGTTTAAAAAARAARAAATCAGTAACAGTACTAGAT
GTGGGGGACGCATATTTCTCAGTTCCTTTAGATGAAAATTTTARAAAGTACACTGCATTCACCATA
CCTAGTATAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGAAG
GGATCACCAGCAATATTTCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAAAATACA
GAAATAATTATCTATCAATACATGGATGACCTGTATGTAGCATCTGATTTAGAAATAGGGCAGCAT
AGAGCAAAAGTAGAGGAATTAAGAGCTCATCTATTGAGTTGGGGGCTTACTACACCAGACAAAAAG
CATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTACATCCTGATAAA

SUBTYPE A:RES NNRTI

070 CAATGGCCATTGACAGAAGAAAAAATAAAGGCATTGAYAGAAATTTGTACAGAGATGGAAAAGGAA
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GACAGTACWAAGTGGAGAAAATTAGTAGATTTCAGGGAACTCAATAAAAGRACCCARGACTTTTGG
GAAGTTCAATTAGGRATACCACACCCAGCAGGGTTAAAARAGAAAAAATCAGTGACAGTACTAGAT
GTGGGGGATGCRTATTTTTCAGTWCCTTTAGATGAAAGCTTCAGGAAATATACTGCATTCACCATA
CCRAGTRTAAACAATGAGACACCAGGAATCAGRTATCAGTACAATGTGCTTCCACAAGGATGGAAA
GGATCACCRGCAATATTCCAAGCTAGYATGACAAAAATYCTGGAACCTTTTAGGAAACAAAATCCA
GAAATGATTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGGGCAACAT
AGAGCAAAAATAGAGRAATTAAGGGRACACCTGTTRAAGTGGGGGTTTACTACACCAGACAAAAAG
CATCAGAAAGAACCTCCATTYCTTTGGATGGGTTATGAACTA

CRF10_CD:SUS

071 CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTCAAGAGATGGAAAAGGAA
GGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAGTATTTGCTATAAAGAAAAAA
GATAGCACAAAATGGAGAAAATTAGTAGATTTTAGAGAACTTAATAAAAGAACTCAGGATTTTTGG
GAAGTTCAATTAGGAATACCGCATCCTGCAGGTTTAAAGAARAAAAAAGCAGTAACAGTACTGGAT

GKGGGGGATGCATATTTTTCAGTGCCTTTAGATGAAAACTTTARAAAGTATACTGCATTCMCCATA
CCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGKGCTTCCACAGGGATGGAAG
GGATCMCCAGCAATWTTTCARAGTAGCATGACAAAAATYTTAGAGCCCTTTAGAGCACAAAATCCA
GGAATAATTATCTATCAATACATGGATGACTTATATGTAGGATCTGATTTARAAATAGGGCAACAT
ARAGCAAAAGTGGAGGAGTTGAGAGCTCATCTATTACAATGGGGATTTACTACACCARATAAAAAA
CATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACT

SUBTYPE A:SUS

072 CATTAHCAGATATTTGTACAGAAATGGAAAAGGAAGGAAAAATTTCAAGAATTGGGCCTGAAAATC
CATATAATACACCAATATTTGCCATAAAGAAAAAAGATAGTACTAAGTGGAGAAAATTAGTAGATT
TCAGAGAACTTAATAAGAGAACTCAAGACTTCTGGGAAGTTCAACTAGGRATACCACATCCTGCAG
GGCTAAAAAAGAAAAAATCAGTAACAGTACTGGATGTGGGTGATGCATATTTTTCAGTGCCCTTAY
RTGAAGACTTTAGAAAATATACTGCATTCACCATACCTAGTAKAAACAATGAAACACCAGGAATTA
GATATCAGTACAATGTGCTTCCACAAGGCTGGAAAGGATCACCGGCAATATTCCAAAGTAGCATGA
CAAAGATCTTAGAACCTTTTAGAAAACAAAACCCGGAAATGGTTATTTATCAATACATGGATGATT
TGTATGTAGGATCTGATTTAGAAATAGGGCAGCATAGAACRAAAATAGAAGAATTAAGGGAGCACC
TATTGAAGTGGGGCTTTACCACACCAGACAAAAAGCATCAGAAAGAACCTCCATTTCTTTGGATGG
GTTATG

SUBTYPE D:SUS

073 CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGAAAAGGAA
GGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCCATATTTGCAATAAAGAAAAAA
GATAGCACTAAGTGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAACACAAGACTTTTGG
GAGGTTCAATTAGGAATACCGCATCCAGCGGGCCTAAAAAARAAAAAATCAGTGACAGTACTAGAT
GTGGGGGATGCATACTTTTCAGTTCCCTTAGATGAAAATTTTARAAAGTATACTGCATTCACCATA
CCTAGCACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGGAAA
GGATCACCGGCAATATTCCAGAGTAGCATGACAAAAATTTTAGAGCCCTTTAGATTAAAAAATCCA
GAAATAATCATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAACTAGGGCAGCAT
AGAGCAAAAATAGAGGAGTTGAGAGCTCATCTATTGAGCTGGGGATTTACTACACCAGACAAAAAG
CATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATRAAC

SUBTYPE A:SUS

074 AAAGCATTAHCAGAAATTTGTACAGAGATGGAAAAGGAAGGAAAAATTTCAAAAATTGGGCCTGAA
AATCCATACAATACTCCAATATTTGCAATAAAGAAAAAAGATAGCACTAAATGGAGAAAATTAGTA
GATTTCAGAGAGCTCAATAAAAGAACACAAGACTTTTGGGAAGTTCAATTAGGAATACCGCATCCA
GCGGGTTTAAAAAAGAAAAAATCAGTAACAGTATTAGATGTGGGGGATGCATATTTCTCAGTTCCT
TTAGATGAAAGCTTTAGAAAGTATACTGCATTCACCATACCTAGTACAAACAATGAGACACCAGGG
ATCAGATATCAGTACAATGTGCTTCCACAGGGATGGAAAGGATCACCAGCAATATTCCAGAGTAGC
ATGACAAAAATCTTAGAGCCCTTTAGAGCAAAAAATCCAGAAATAATTATTTATCAATACATGGAT
GACTTATATGTAGGATCTGATTTAGAAATAGGGCAGCATAGAGCAAAAATAGAGGAGTTAAGAGCT
CATCTATTGAGRTGGGGATTTACTACACCAGACAAAAAGCATCAGAAAGAACCCCCATTCCTTTGG
ATGGGTTATGAA

SUBTYPE A:SUS

075 AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGCAAAGAGATGGAAAAG
GAAGGAAAAATTTCAAAAATTGGGCCTGACAATCCATACAATACTCCAGTATTTGCTATAAAGAAA
AAAGATAGCACTAAATGGAGAAAATTAGTAGATTTTAGAGAGCTCAATAAAAGAACTCAGGACTTC
TGGGAAGTTCAATTGGGGATACCACATCCCGCAGGCTTAAAAAAGAAAAAATCAGTAACAGTATTA
GATGTAGGGGACGCATATTTTTCAGTCCCTTTAGATGAAAACTTTAGAAAGTATACTGCATTCACC
ATACCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGG

AAAGGATCACCAGCAATATTCCAGAGTAGCATGATAAAAATCTTAGAGCCCTTTAGATCAAAAAAT
CCAGAAATAATTATATATCAATACATGGATGACTTATATGTAGGATCTGATTTAGAAATAGGGCAG
CATAGAACAAAAGTAGAGGAGTTGAGAGCTCATCTATTGAATTGGGGATTTACTACACCAGACAAA
AAGCATCAGAAAGAACCCCCATTTCTTTGGATGGGATATGA

SUBTYPE A:SUS

076 TAACAGAAATTTGTACAGATATGGAAAAGGAAGGAAAAATTTCAAGAATTGGGCCTGAAAATCCAT
ACAATACTCCARTATTTGCCATAAAGAAAAAAGACAGTACTAAGTGGAGAAAATTASTAGATTTTA
GAGAACTTAATAAAAGAACTCAAGACTTCTGGGAAGTTCAACTAGGAATACCACATCCTGCAGGGC
TAAAAAARAAAAAATCAGTAACAGTACTGGACGTGGGTGATGCATWTTTTTCAGTTCCCTTARATG
AAAACTTTARAAAATATACCGCATTCACCATACCCAGTATAAATAATGAGACACCAGGAATTAGAT
ACCAGTACAATGTGCTTCCACAAGGATGGAAAGGATCACCAGCAATATTCCAAAGCAGCATGACAA
AAATCCTAGAACCTTTTAGGAAACAAAATCCAGAAATRGTTATCTATCAATACATGGATGATTTGT
ATGTAGGATCTGACTTARAAATAGGGCAGCATAGAACAAAGATAGAGGAACTAAGGGAACACTTAT
TGAAGTGGGGGTTTACCACACCARACAAAAAGCATCAGAAAGAACCTCCATTTCTTTGGATGGGTT
ATGAACTA

SUBTYPE D:RES NRTI

077 AAACAATGGCCATTGACAGAAGAAAAAATAAAGGCATTAACAGAAATTTGTACAGAAATGGAAAAG
GAAGGAAAAATCTCAAGAATTGGGCCTGAAAATCCATACAATACTCCAGTATTTGCTATAAAGAAA
AAAGACAGCACCAAATGGAGGAAATTAGTAGATTTCAGAGAACTCAATAAAAGAACTCAGGATTTC
TGGGAAGTTCAATTAGGAATACCGCATCCAGCAGGTTTAAAAAARAAAAAATCAGTAACAGTACTA
GATGTGGGGGACGCATATTTTTCAGTGCCTTTAGATGAAAACTTTAGAAAGTATACTGCATTCACC
ATACCTAGTACAAACAATGAGACACCAGGAATCAGATATCAGTATAATGTGCTTCCACAGGGATGG
AAAGGATCACCAGCAATATTCCAAAGCAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAAAAT
CCAGAAATAATTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAG
CATAGAGAGAAAATAGAGGAGTTAAGGGCTCATCTATTGAGCTGGGGGTTTACTACACCAGACAAA
AAGCATCAGAAGGAACCTCCATTTCTTTGGATGGGTTATGAACTA

CRF01_AE:SUS

078 CATTAACAGAAATTTGTACAGAAATGGAAAAGGAAGGAAAAATTTCAAAAATTGGGCCTGAAAATC
CATACAATACTCCAATATTCGCGATAAAGAAAAAAGACAGCACTAAATGGAGAAAATTAGTAGATT
TCAGAGAGCTCAATAAAAGAACTCAAGACTTTTGGGAAGTTCAATTAGGAATACCGCATCCAGCGG
GCTTAAAAAAGAAAAAATCAGTAACAGTACTAGATGTGGGGGACGCATATTTTTCAGTTCCCTTAG
ATGAAAGCTTTAGAAAATATACTGCATTCACCATACCTAGTACAAACAATGAGACACCAGGGATTA
GATATCAGTACAATGTGCTTCCACAGGGATGGAAAGGATCACCAGCAATATTCCAGAGTAGCATGA
CAAAAATCTTAGAGCCCTTCAGAGCAAAAAATCCAGAAATAATTATCTATCAATACATGGATGACT
TGTATGTAGGATCTGATTTAGAAATAGGACCGCATAGGGCAAAAATAGAAGAATTGAGAGCTCATC
TATTGAGCTGGGGATTAACTACACCAGACAAGAAGCATCAGAAAGAACCTCCATTCCTTTGGATGG
GTTATGAAC

SUBTYPE A:SUS

079 AATTTGTAAAGAGATGGAGAAGGAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATAC
TCCAGTATTTGCTATAAAGAAAAAAGATAGCACTAAATGGAGAAAATTAGTAGATTTTAGAGAGCT
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GAAAAAATCAGTAACAGTACTAGATGTGGGAGACGCATATTTTTCAGTTCCTTTAGATGAAAACTT
TAGAAAGTATACAGCATTCACCATACCTAGTATAAACAATGAGACACCAGGAATCAGGTATCAGTA
CAATGTGCTTCCACAGGGATGGAAAGGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTT
AGAGCCCTTCAGATCACAAAATCCAGAAATAATTATCTATCAATACATGGATGACTTGTATGTAGG
ATCTGATTTAGAAATAGGGCAGCATAGAGCAAAAGTAGAGGAGTTGAGAAGTCATCTATTGAAGTG

GGGATTTACCACACCAGACAAAAAGCATCAGAAAGAACCCCCATTTCTTTGGATGGGTT

SUBTYPE A:SUS

080 CATTAACAGAAATTTGTGCAGATATGGAAAAGGAAGGAAAGATTTCAAAAATTGGGCCTGAGAATC
CATACAATACTCCAATATTTGCTATAAAGAAAAAAGACAGCACTAAATGGAGAAAATTAGTAGATT
TCAGAGAACTTAATAAAAGAACTCAAGACTTTTGGGAAGTTCAATTAGGAATACCGCATCCAGCGG
GGTTAAAAAAGAAAAAATCAGTAACAGTACTAGATGTAGGGGACGCATATTTTTCAGTTCCCCTAG
ATGAAAGCTTTAGAAAGTATACAGCATTCACAATACCTAGTACAAATAATGAGACACCAGGAATCA
GATATCAGTACAATGTGCTCCCACAGGGATGGAAAGGATCACCAGCAATATTCCAGAGCAGCATGA
CAAAAATCTTAGAGCCCTTTAGATCAAAAAATCCAGAAATAATTATCTATCAATACATGGATGACT
TATATGTGGGATCTGATTTAGAAATAGGGCAGCATAGAGCAAAAATAGAAGAGTTAAGAGCTCATC
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GTT

SUBTYPE A:SUS

081 AATTTGTAAAGAGATGGAGAAGGAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATAC
TCCAGTATTTGCTATAAAGAAAAAAGATAGCACTAAATGGAGAAAATTAGTAGATTTTAGAGAGCT
CAATAAAAGAACTCAGGACTTCTGGGAAGTTCAATTAGGAATACCACATCCCGCAGGTTTAAAAAA
GAAAAAATCAGTAACAGTACTAGATGTGGGAGACGCATATTTTTCAGTTCCTTTAGATGAAAACTT
TAGAAAGTATACAGCATTCACCATACCTAGTATAAACAATGAGACACCAGGAATCAGGTATCAGTA
CAATGTGCTTCCACAGGGATGGAAAGGATCACCAGCAATATTCCAGAGTAGCATGACAAAAATCTT
AGAGCCCTTCAGATCACAAAATCCAGAAATAATTATCTATCAATACATGGATGACTTGTATGTAGG
ATCTGATTTAGAAATAGGGCAGCATAGAGCAAAAGTAGAGGAGTTGAGAAGTCATCTATTGAAGTG
GGGATTTACCACACCAGACAAAAAGCATCAGAAAGAACCCCCATTTCTTTGGATGGGTT

SUBTYPE A:SUS

082 GTTAAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGA
AAAGGAAGGAAAAATTTCAAAAATTGGGCCTGAGAATCCATACAATACCCCAATATTTGCTATAAA
GAAAAAAGACAGTACT:AAGTGGAGAAAATTAGTGGATTTTAGAGAACTTAATAAGAGAACTCAAG
ATTTCTGGGAAGTTCAATTAGGAATACCACATCCTGCAGGATTAAAAAAGAAAAATTCAGTAACAG
TACTGGATGTGGGTGATGCATATTTTTCAGTTCCCTTAGATGAAGACTTTAGAAAATATACCGCAT
TCACTATACCTAGTATAAATAACGAGACACCAGGAGTTAGATATCAGTACAACGTGCTTCCACAAG
GATGGAAAGGGTCACCATCAATATTTCAAAGTAGCATGACAAAGATCTTAGAACCTTTTAGAAAAC
AAAATCCAGAAATAGTTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAG
GGCAGCATAGAACAAAAATAGAGGAATTAAGGGGACACCTATTGAAGTGGGGATTCACCACACCAG
ACAAAAAGCATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTACACCCTGA

SUBTYPE D:SUS

083 TGGCCATTGACAGAAGAAAAAATAAAAGCATTAACCAGCAATTTGTGAAGATATGGAGARGGAAGG
AAAAATTACAAGAGTTGGGCCTGAGAATCCATATAACACTCCAATATTTGCCATAAAAAAGAAAGA
TAGTACTAAGTGGAGAAAATTAGTAGACTTTAGGGAACTCAATAAAAGAACTCAAGACTTTTGGGA
AGTCCAATTAGGGATACCACACCCAGCAGGGTTAAAAAAGAAAAAATCAGTGACAGTACTGGATGT
GGGGGATGCATATTTYTCAGTTCCTTTAGATGAGAGYTTTAGRAAATATACTGCATTCACCATACC
TAGTACAAATAATGAAACACCAGGAATTAGATATCAATACAATGTRCTTCCACAGGGATGGAAAGG
ATCACCAGCGATATTTCAGAGTAGTATGACAAAAATCTTAGAGCCCTTTAGGGCACAAAACCCAGA
AATAGTTATCTATCAATATATGGATGACTTGTATGTAGGATCTGACTTAGAAATAGGGCAACATAG
GGCAAAAATAGAGGAGTTAAGAGAACATCTATTAAGGTGGGGATTTACCACACCAGACAAAAAGCA
TCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTABATCCTGA

SUBTYPE C:SUS

084

CCAAAAGTTAAACAATGGCCATTGACAGAAGAAAAGATAAAAGCATTAACAGAAATTTGTAAAGAA
ATGGAAGCTGAAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCT
ATAAAGAAAAAAGATAGCACTAAGTGGAGAAAATTGGTAGACTTTAGAGAGCTCAATAAAAGAACT
CAGGACTTCTGGGAAGTTCAATTAGGAATACCGCATCCCGCGGGGTTAAAAAAGAAAAAATCAGTA
ACAGTACTAGATGTGGGGGATGCATATTTTTCAGTTCCTTTACATGAGAGCTTTAGAAAATATACT
GCATTCACCATACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTGCCA
CAAGGATGGAAAGGGTCACCGGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCATTTAGA
GCAAAAAATCCAGAAATGRTTATCTATCAATACRTGGATGACCTGTATGTAGGATCTGATTTAGAA
ATAGGGCAGCATAGAGAAAAAATAGAACAATTAAGAGCTCACTTATTGAAATGGGGATTTACTACA
CCAGACAAAAAGCATCAGAAAGAACCTCCATTCCTTTGGATGGGWTATGARCTA

SUBTYPE A:RES NRTI

085

AACAATGGCCATTGACAGAAGAAAAAATAAAAGCACTAACAGAAATTTGTACAGAAATGGAAAAGG
AAGGAAAAATCTCAAGAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCCATAAAGAAAA
AAGACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGRGAACTTAATAARAGGACTCAAGACTTCT
GGGAAGTTCAACTAGGAATACCACATCCWGCAGGGTTAAAAAAGAAAAAATCAGTAACAGTACTGG
ATGTGGGKGATGCATATTTTTCAGTTCCCTTATATGAAGACTTTAGAAAGTATACTGCATTCACCA
TACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAATACAATGTGCTTCCACAAGGATGGA
AAGGATCACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAACCTTTTAGAMRACAAAATC
CAGAAATAGTTRTCTATCAATACATGGATGATTTGTAYGTAGGATCTGATTTAGACATAGGGCAGC
ACAGAAYAAAAATAGAGGAATTAAGAGAACACCTCTTGAAGTGGGGATTTACCACGCCAGATAAAA
AGCATCAGAAAGAACCCCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE D:SUS

086

ARAAAAWAAAAAGCATTAACAGAAATTTGTGCAGATATGGAAAAGGARGGAAAAATTTCAAGAATT
GGGCCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAAAAAGACAGTACYAAATGGAGR
AAATTAGTRGATTTCAGRGAACTYAATAARAGAACTCAAGATTTYTGGGARGTYCAATTAGGAATA
CCACATCCTGCRGGGYTRAAAAAGAAAAAATCAGTAACAGTAYTGGATGTRGGKGATGCATATTTT
TCAGTTCCCTTATATGAARAYTTYAGAAAATATACTGCATTCACCATACCTAGTATAAACAATGAG
ACACCAGGARTYAGRTATCAATACAATGTGCTWCCACAGGGATGGAAAGGATCACCAGCAATATTC
CAGAGTAGCATGACAAAAATCTTGGAGCCCTTTAGAAAACAAAAYCCAGAMATARTTATCTATCAA
TACATGGATGATTTGTATGTAGGATCTGAYTTRGAAATAGGGCAGCATAGAACAAAAATAGATGAA
CTAAGAGAACATCTATTGAAGTGGGGATTTACCACACCAGATAAAAAACATCAGAAAGAACCTCCA
TTTCTTTGGATGGGTTATGAACTACATCCTGA

SUBTYPE D:SUS

087

ACAATGGCCATTGACAGAAGAGAAAATAAAAGCATTAACAGCAATTTGTGAAGATATGGAAAAGGA
AGGAAAAATTTCAAGAATTGGGCCTGAAAATCCATATAACACTCCAGTATTTGCCATAAAAAAGAA
GGACAGTACTAAGTGGAGAAAATTAGTAGATTTCAGGGAGCTCAATAAAAGAACTCAAGACTTTTG
GGAAGTTCAATTAGGGATACCACACCCAGCAGGGTTAAAGAAGAAAAAATCAGTGACAGTATTGGA
TGTGGGGGATGCATATTTCTCAGTACCTTTAGATGAAAACTTCAGGAAGTAYACAGCATTCACCAT
ACCTAGTATAAACAATGAAACACCAGGAATTAGATATCAATATAATGTGCTTCCACAGGGATGGAA
AGGATCACCATCAATATTTCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGGATAAAAAACCC
AGACATAGTTATCTATCAATATATGGATGATTTGTATGTAGGATCTGATTTAGAAATAGGGCAACA
TAGAGCAAAAATAGAGGAGTTAAGGGATCATCTATTGAAGTGGGGATTTACTACACCAGACAAGAA
ACATCAGAAAGAACCTCCATTTCTTTGGATGGGTTTA

SUBTYPE C:SUS

088

CAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGAAAAGGAA
GGAAAAATTTCAAAAATTGGSCCTGAAAAYCCATACAATACTCCAATATTTGCTATAAAGAAAAAA

GAYAGYACTAAATGGAGGAAATTAGTGGACTTCAGAGAACTCAATAAAAGAACTCAAGAYTTTTGG
GAAGTTCARTTAGGAATACCACAYCCAGCGGGCTTAAAAAAGAAAAAATCAGTAACAGTACTAGAT
GTGGGGGAYGCATATTTTTCAGTTCCCTTAGATGAAARCTTTAGAAAGTATACTGCATTCACCATA
CCTAGTAYAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTTCCGCAGGGATGGAAR
GGATCACCRGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAGAACCCA
GAAATAATTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTGGAAATAGGGCAACAT
AGAACAAAAGTAGAAGAGTTGAGAGATCATCTATTGAGCTGGGGATTTACTACACCAGACAAAAAG
CATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE A:SUS

089 ACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAGTAGAAATTTGTACAGAAATGGAAAAAGA
AGGAAAAATTACAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAAAA
GGACAGCACTAAATGGAGGAAACTAGTAGATTTCAGAGAGCTCAATAAAAGGACACAAGACTTTTG
GGAAGTTCAATTAGGGATACCGCATCCAGCGGGCCTAAAAAAGAAAAAATCAGTAACAGTACTGGA
TGTGGGGGATGCATATTTTTCAGTCCCTTTAGATGAAAGCTTTAGGAAATATACTGCGTTCACCAT
ACCTAGTACAAACAATGAGGCACCAGGAATTAGATATCAATACAATGTGCTTCCACAAGGATGGAA
AGGATCACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAACCTTTTAGAAAGCAAAATCC
AGAAATAGTTATCTATCAATACATGGATGACTTGTATGTCGCATCTGACTTAGAAATAGGGCAACA
TAGAACAAAGATAGAAGAATTAAGGGAACACCTATTGAAATGGGGATTTACCACACCAGACAAAAA
GCATCAGAAAGAACCTCCATTKCTKTGGATGGGTTATGAACTA

SUBTYPE D:RES NNRTI

090 TAAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTATGGAAATGGAAAA
GGAGGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCAATAAGGAA
AAAAGACAGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAACACAAGACTT
TTGGGAGGTTCAATTAGGAATACCACATCCAGCTGGCCTAAAAAARAAAAAATCAGTAACAGTACT
AGATGTGGGGGACGCATATTTTTCAGTTCCTTTACATGAAGATTTTAGGAAGTATACTGCGTTCAC
CATACCTAGTACAAACAATGAGACACCAGGAATCAGATATCAGTACAATGTGCTCCCACAGGGATG
GAAAGGATCACCGTCAATATTCCAGGCTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAAAA
TCCAGAGCTAGTTATKTATCAGTACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCA
GCACAGAGCAAAAATAGAAGAGCTGAGAGCTCATCTATTGAGCTGGGGATTTACTACACCAGACAA
AAAGCATCAGAAAGAACCTCCATTCCTCTGGATGGGATATGAGCTA

SUBTYPE A:SUS

091 AACAATGGCCATTGACAGAAGAAAAAATAAAAGCACTAACAGAAATTTGTATAGACATGGAAAAGG
AAGGAAAAATTTCAAGAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAAA
AAGACAGTACCAAGTGGCGAAAATTAGTAGATTTCAGAGAACTTAATAAGAGAACTCAAGATTTCK
GGGAAGTTCAACTAGGAATACCACATCCTGCAGGGCTAAAAAAGAAAAAATCAGTTACAATACTGG
ATGTGGGTGATGCATATTTTTCAGTTCCCTTGGATAAAGAATTTAGAAAATACACTGCATTCACCA
TACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTATAATGTGCTTCCACAAGGGTGGA
AAGGATCACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAGCCCTTTAGGAAACAAAATC
CAGAAATAGTTATCTRTCAATACATGGATGACTTGTATGTAGGGTCTGACTTAGAAATAGGGCAGC
ATCGAGCAAAAATAGAACAGTTGAGAGCTCATCTATTGAGATGGGGATTTAMTACACCAGACAAGA
AGCATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE D:RES NNRTI/NRTI

092 GTTAAACAATGGCCATTGACAGCAGAAAAAATAAAAGCATTAACAGAAATTTGTTTAGAAATGGAA
AAGGAAGGAAAAATTTCTAAAATTGGGCCTGAAAATCCATACAATACTCCAGTATTTGCAATAAAG
AAAAAAGATAGCACTAAATGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAACACAAGAC
TTTTGGGAAGTTCAATTAGGAATACCGCATCCAGCGGGMCTAAAAAAGAAAAAATCAGTAACAGTA
CTAGATGTGGGGGATGCATATTTTTCTGTCCCCTTAGATGAAGAATTTAGAAAGTATACTGCATTC
MCCATACCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAATACAATGTGCTTCCACAGGGA
TGGAAAGGATCACCGGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAG
AATCCAGAGATAATTATCTATCAATACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGG
CAACATAGAGCAAAAATAGAAGAGTTGAGAGCTCATCTATTGAGCTGGGGATTTACTACACCAGAC
AAAAAGCATCAGAAAGAACCTCCATTCCTTTGGATGGGGTATGAACTA

SUBTYPE A:SUS

093 CAATGGCCATTGACAGAAGAAAAAATAAAAGCACTAACAGAAATTTGTATGGAAATGGAAAAGGAA
GGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAACACTCCAGTATTTGCTATAAAGAAAAAA
GACAGCACTAAGTGGAGAAAATTAGTAGATTTCAGAGAGCTCAATAAAAGAACTCAAGACTTTTGG
GAAGTTCAGTTAGGAATACCACACCCAGCAGGGTTAAAAAAGAAAAAGTCAGTGACAGTATTGGAT
GTGGGGGATGCATATTTTTCAGTTCCTTTAGATGAAGGATTCAGGAAATATACTGCATTCACCATA
CCTAGTACAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTTCCACAAGGATGGAAA
GGATCACCGGCAATATTCCAAAGTAGCATGACAAGAATCTTAGAACCTTTTAGAAAACAAAATCCA
GAAATAGTTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAGAGCAACAT
AGAGCAAAAATAGAGGAATTAAGGGAACACCTATTGAAGTGGGGGTTTACCACACCAGATAAAAAG
CATCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE B:SUS

094 AAACAATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTACAGAAATGGAGAAG
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AAGGACAGCACTAAGTGGAGAAAATTAGTAGATTTTAGAGAGCTCAATAAAAGAACTCAAGATTTT
TGGGAAGTCCAATTAGGAATACCGCATCCAGCGGGCCTAAAAAAGAAAAAATCAGTAACAGTACTG
GATGTGGGGGATGCATATTTCTCAGTTCCTTTAGATGAAAGCTTTAGAAAGTATACTGCGTTCACC
ATACCTAGTACAAACAATGAGACACCAGGAATCAGGTATCAGTACAATGTGCTTCCACAGGGATGG
AAGGGATCACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAGCCTTTTAGATCAAAAAAT
CCAGAAATAATTATCTATCAGTACATGGATGACTTATATGTAGGATCTGATTTAGAAATAGGGCAG
CATAGAGAAAAAATAGAGGAGTTGAGAGCTCATCTATTGAGCTGGGGACTTACTACCCCGGACAAA
AAGCATCAGAAAGAACCGCCATTTCTTTGGATGGGTTATRAACTA

SUBTYPE A:SUS

095 ATGGCCATTGACAGAAGAAAAAATAAAAGCATTAACAGAAATTTGTAAAGAGATGGAAAAGGAAGG
AAAAATTTCAAAAATCGGGCCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAAAAAGA
TAGCACTAAATGGAGAAAATTAGTAGATTTTAGAGAGCTCAATAAAAGAACTCAGGACTTCTGGGA
AGTTCAATTAGGAATACCACATCCAGCAGGTTTAAAAAAGAAAAAATCAGTAACAGTACTAGATGT
GGGGGACGCATATTTTTCAGTTCCTTTAGATGAAAACTTTAGAAAGTATACTGCGTTCACCATACC
TAGTATAAACAATGAGACACCAGGAATCAGGTATCAATACAATGTGCTCCCGCAGGGATGGAAAGG
ATCACCAGCAATATTCCAAAGTAGCATGACAAAAATCTTAGAGCCCTTTAGAGCAAAAAATCCAGA
AATAATTATCTATCAATATATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGACAGCATAG
ARCAAAAGTAGAGGAATTGAGAGCTCATCTATTGAGCTGGGGATTTACTACACCAGACAAAAAGCA
TCAGAAAGAACCTCCATTTCTTTGGATGGGTTATGAACTA

SUBTYPE A:SUS

096

AACAATGGCCATTGACAGAAGAAAAGATAAAAGCATTAACAGAAATTTGTAAAGAAATGGAAGCTG
AAGGAAAAATTTCAAAAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCTATAAAGAAAA
AAGATAGCACTAAGTGGAGAAAATTGGTAGACTTTAGAGAGCTCAATAAAAGAACTCAGGACTTCT
GGGAAGTTCAATTAGGAATACCGCATCCCGCGGGGTTAAAAAAGAAAAAATCAGTAACAGTACTAG
ATGTGGGGGATGCATATTTTTCAGTTCCTTTACATGAGAGCTTTAGAAAATATACTGCATTCACCA
TACCTAGTATAAACAATGAGACACCAGGAATTAGATATCAGTACAATGTGCTGCCACAAGGATGGA
AAGGGTCACCGGCAATATTCCAGAGTAGCATGACAAAAATCTTAGAGCCATTYAGAGCAAAAAATY
CAGAAATGRTTATCTATCAATACRTGGATGACCTGTATGTAGGATCTGATTTAGAAATAGGGCAGC
ATAGAGAAAAAATAGAACAATTAAGAGCTCACTTATTGAAATGGGGATTTACTACACCAGACAAAA
AGCATCAGAAAGAACCTCCATTCCTTTGGATGGGTTATGAACTA

SUBTYPE A:RES NRTI

097

AAAAAATAAAAGCATTAACTGAAATTTGTACAGAAATGGAAAAGGAAGGAAAAATTTCAAAAATTG
GGCCTGAAAATCCATACAATACTCCAATATTTGCAATAAGGAAAAAAGATAGTACTAAATGGAGGA
AATTAGTGGATTTCAGAGAGCTCAATAAAAGAACACAAGATTTTTGGGAAGTTCAATTAGGGATAC
CACATCCAGCGGGCCTAAAGAAGAAYAAATCAGTAACAGTACTAGATGTGGGGGATGCATATTTTT
CAGTTCCTTTACATGAAGACTTTAGAAAATATACTGCGTTCACCATACCTAGTACAAACAATGAGA
CACCAGGAATCAGATATCAGTACAATGTGCTACCACAGGGATGGAAAGGATCACCAGCAATATTCC
AGAGTAGCATGACAAAAATCTTAGAGCCCTTTAGATCAAAAAATCCAGAAATAAGCATCTATCAAT
ACATGGATGACTTGTATGTAGGATCTGATTTAGAAATAGGGCAACATAGAGCAAAAATAGAGGAGT
TAAGAGCTCATCTATTGAGCTGGGGGTTTACTACACCAGACAAAAAGCATCAGAAAGAACCTCCAT
TCCTTTGGATGGGTTATGAACTACATCCTGACAAGTGGACAGTCCAGCCTATAAAGCTGCCA

SUBTYPE A:RES NNRTI

098

AAGAAAAAATAAAAGCATTAACAGATATTTGTACAGAAATGGAAAAGGAAGGAAAAATTTCAAGAA
TTGGGCCTGAAAATCCATATAATACACCAATATTTGCCATAAAGAAAAAAGATAGTACTAAGTGGA
GAAAATTAGTAGATTTCAGAGAACTTAATAAGAGAACTCAAGACTTCTGGGAAGTTCAACTAGGAA
TACCACATCCTGCAGGGCTAAAAAAGAAAAAATCAGTAACAGTACTGGATGTGGGTGATGCATATT
TTTCAGTGCCCTTATGTGAAGACTTTAGAAAATATACTGCATTCACCATACCTAGTATAAACAATG
AAACACCAGGAATTAGATATCAGTACAATGTGCTTCCACAAGGCTGGAAAGGATCACCGGCAATAT
TCCAAAGTAGCATGACAAAGATCTTAGAACCTTTTAGAAAACAAAACCCGGAAATGGTTATTTATC
AATACATGGATGATTTGTATGTAGGATCTGATTTAGAAATAGGGCAGCATAGAACGAAAATAGAAG
AATTAAGGGAGCACCTATTGAAGTGGGGCTTTACCACACCAGACAAAAAGCATCAGAAAGAACCTC
CATTTCTTTGGATGGGTTATGAACTAYAAYCCTG

SUBTYPE D:SUS

099

CAATGGCCATTGACAGAAGAAAAAAWWAAAAGCACTAAACAGAAATTTGTACAGATATGGAAAAGG
AAGGAAAAATTTCAAGAATTGGGCCTGAAAATCCATACAATACTCCAATATTTGCCATAAAGAAAA
AAGACAGTACTAAGTGGAGAAAATTAGTAGATTTTAGAGAACTTAATAAAAGAACTCAAGACTTCT
GGGAAGTTCAACTAGGAATACCACATCCTGCAGGGCTAAAAAAGAAAAAATCAGTAACAGTACTGG
ACGTGGGTGATGCATATTTTTCAGTTCCCTTAGATGAAAACTTTAGAAAATATACCGCATTCACCA
TACCCAGTATAAATAATGAGACACCAGGAATTAGATACCAGTACAATGTGCTTCCACAAGGATGGA
AAGGATCACCAGCAATATTCCAAAGCAGCATGACAAAAATCCTAGAACCTTTTAGGAAAMMAAAAT
CCAGAAATGGTTATCTATCAATACATGGATGATTTGTATGTAGGATCTGACTTAGAAATAAGGGCA
GCATAGAAACAAAGATAGAGGAACTAARGGAAMCMCTTATTGAAGTGGGGGGTTTACCACACCAGA
CAAAAAGCATCARAAAGAACCTCCATTTCTTTGRAKGGRKTAWKAAAMTAA

SUBTYPE D:SUS

100

AAAWAAAAAGCATTAACAGAAATATGTAGGGAGATGGAAGAAGAAGGAAAAATTACAAAAATTGGG
CCTGAAAATCCATATAACACTCCAGTATTTGCYATAAAAAAGAAGGACAGTACTAAGTGGAGAAAA

TTAGTAGACTTCAGGGAACTCAATAAAAGAACTCAAGACTTTTGGGAAGTTCAATTAGGAATACCA
CACCCAGCAGGATTAAAAAGGAACAAATCAATAACGGTACTGGATGTGGGAGATGCATATTTTTCA
GTTCCTTTAGATGAAGATTTCAGRAAATACACTGCATTCACCATACCTAGTATAAACAATGAAACA
CCAGGAGTTAGATACCAATATAATGTGCTTCCACAAGGATGGAAAGGATCACCAGCAATATTCCAG
AGTAGTATGACAAGAATCTTAGAGCCCTTTAGAGCAAGAAACCCAGAGATAGTTATCTATCAATAT
ATGGATGACTTGTATGTAGGATCTGATTTAGAACTAAAGCAACATAGAGCAAAAATAGAAGAGTTA
AGAGAACACCTATTGAAATGGGGATTTACCACACCAGACAAGAAACATCAGAAAGAACCCCCATTT
CTTTGGATGGGTTATGAACTACATCCTGA

SUBTYPE A:RES NNRTI

Appendix 1.3: *ARVPredictor* Test Performance Results Table

SeqID	Stanford HIV Database						*ARVPredictor*					
	HIV Subtype	Susceptibility	RT Mutation Output				HIV Subtype	Susceptibility	RT Mutation Output			
			NNRTI	NRTI		Other Mutations			NNRTI	NRTI		Other Mutations
				TAMs	Non-TAMs					TAMs	Non-TAMs	
1	A	Susceptible	**V179T**	None	None	E28EA, V35T, E40D, V60I, K122E, D123G, F214L	A	Susceptible	**V179T**	None	None	SAME
2	A	Susceptible	**V179T**	None	None	V35T, T39M, V60I, D121H, K122E, D123G, R125RK, R 211S	A	Susceptible	**V179T**	None	None	SAME
3	A	Susceptible	None	None	None	V35T, T39M, V60I, D121H, K122E, D123G, I135T, A15 8S, T211S	A	Susceptible	None	None	None	SAME
4	A	Susceptible	None	None	None	V35T, V60I, K122E, D123S, I135T, K173S, Q174K, D17 7E, T200A, Q207A, R211S	A	Susceptible	None	None	None	SAME
5	B	Susceptible	None	None	None	E40D, I47L, K49R, V60I, D 121C, K122E, D123E, R125 RK, K173E, D177E, T200A, Q207E	B	Susceptible	None	None	None	SAME
6	A	**NNRTI**	**K103KN**	None	None	K32KN, V35T, K43E, V60I, K122E, D123G, I135T, K17 3I, Q174K, V179I, T200A, Q207A, R211S	A	**NNRTI**	**K103KN**	None	None	SAME
7	D	Susceptible	None	None	None	V35T, E40D, K49R, V60I, D121Y, K122E, Q174R, D1 77E, I178V, T200I, Q207E, R211K	D	Susceptible	None	None	None	SAME
8	B	Susceptible	None	None	None	V35I, K49R, K102KE, K122 E, D123S, S162A, D177E, I 178M, V179I, T200A, E204 K, Q207E, R211K	B	Susceptible	None	None	None	SAME

9	B	**NNRTI, NRTI**	**V106VA, F227FL, M230I**	None	**K65KR**	V35I, K49R, K102KE, K122E, D123S, I135IV, S162A, D177E, I178M, V179I, T200A, E204EK, Q207E, R211K	C+D	**NNRTI, NRTI**	**V106VA, F227FL**	None	**K65KR**	**M2301**
10	C + D	**NNRTI, NRTI**	**V106A**	None	**M184V**	V35I, K49R, K102E, K122E, D123S, S162A, T165K, D177E, I178M, V179I, T200A, Q207G, R211K	C+D	**NNRTI, NRTI**	**V106A**	None	**M184V**	SAME
11	C	**NNRTI, NRTI**	**K103N**	None	**M184V**	V35T, E36A, T39E, S48T, K122E, D123N, I135IT, T139TK, K173A, D177E, I178M, T200A, Q207N, R211K	C	**NNRTI, NRTI**	**K103N**	None	**M184V**	SAME
12	C	**NNRTI, NRTI**	**K103N**	None	**M184V**	L34I, V35T, E36A, T39E, S48T, D86DE, K122E, D123N, T139K, E169EK, K173A, D177E, I178M, T200A, Q207N, R211K	C	**NNRTI, NRTI**	**K103N**	None	**M184V**	SAME
13	B	Susceptible	None	None	None	V35T, T39M, V60I, D121Y, K122E, D123DN, R125RK, Q174K, D177E, I178M, Q207E	B	Susceptible	None	None	None	SAME
14	D	**NNRTI, NRTI**	**A98G, Y181C, H221HY**	None	**M184V**	V35T, E40D, K43R, K49R, V60I, D121Y, K122E, D123E, I135T, D177E, I178M, I195L, T200I, E203EK, E204K, Q207E, R211K	D	**NNRTI, NRTI**	**A98G, Y181C**	None	**M184V**	**H221HY**
15	A	Susceptible	None	None	None	V35T, V60I, D121H, K122E, D123S, K173S, Q174K, D177E, T200A, Q207AT, R211S	A	Susceptible	None	None	None	SAME
16	A	Susceptible	None	None	None	V35T, T39A, E40D, V60I, D121H, K122E, I135T, K173S, Q174K, D177Q, V179I, T200A, Q207A, R211S	A	Susceptible	None	None	None	SAME
17	A	Susceptible	None	None	None	V35T, T39N, V60I, K122E, D123S, S162A, K173S, Q174K, D177E, T200I, I202V, Q207D, R211K	A	Susceptible	None	None	None	SAME
18	A	Susceptible	None	None	None	V35T, T39N, V60I, K122E, D123S, S162A, K173S, Q174K, D177E, T200I, I202V, Q	A	Susceptible	None	None	None	SAME

						207D, R211K						
19	A2	**NNRTI, NRTI**	**Y188L**	None	**M184V**	V35T, T39L, D123E, D177E, I178M, T200A, Q207E, R211K	A2	**NNRTI, NRTI**	**Y188L**	None	**M184V**	SAME
20	A	Susceptible	None	None	None	V35T, V60I, D123E, I135T, S162N, T165L, K173S, Q174K, D177E, V179I, G196E, T200S, I202V, Q207A, F214L	A	Susceptible	None	None	None	SAME
21	D	Susceptible	None	None	None	V35T, T39M, K49R, V60I, D121Y, K122E, K173R, Q174K, D177E, T200I, Q207E, R211K	D	Susceptible	None	None	None	SAME
22	D	Susceptible	None	None	None	V35T, T39M, K49R, V60I, D121Y, K122E, K173R, Q174K, D177E, T200I, Q207E, R211K	D	Susceptible	None	None	None	SAME
23	D	**NNRTI**	**G190GA**	None	None	V35T, E36D, T39N, V60I, K122E, D177E, I178M, T200I, Q207E, R211K	D	**NNRTI**	**G190GA**	None	None	SAME
24	A	Susceptible	None	None	None	V35T, V60I, I135T, K173L, Q174K, D177E, V179I, Q207A, R211S	A	Susceptible	None	None	None	SAME
25	A	**NNRTI**	**K103N**	None	None	K32R, V35PT, V60I, K122E, D123S, I135T, A158S, K173A, Q174K, D177E, V179I, I202IV, Q207A, R211S	A	**NNRTI**	**K103N**	None	None	SAME
26	A	**NNRTI, NRTI**	**K103N**	None	**M184V**	K32R, V35T, V60I, K122E, D123S, I135T, A158S, K173A, Q174K, D177E, V179I, Q207A, R211S	A	**NNRTI, NRTI**	**K103N**	None	**M184V**	SAME
27	A	**NNRTI**	**G190A**	None	None	V35T, V60I, K122E, D123S, I135T, I142V, F171Y, K173S, Q174K, D177E, V179I, T200A, Q207A, R211S	A	**NNRTI**	**G190A**	None	None	SAME

28	A	**NNRTI, NRTI**	**E138Q, G 190A**	None	**M184V**	V35T, V60I, K122E, D123S, I135T, I142V, F171Y, K173 S, Q174K, D177E, V179I, T 200A, Q207A, R211S	A	**NNRTI, NRTI**	**E138Q, G190A**	None	**M184V**	SAME
29	D	Susceptible	None	None	None	V35T, T39KN, V60I, V90VI , K102N, D121CHRY, K122 E, I135IATV, I142IV, D177 E, I178V, Q197QK, Q207E, R211RK	D	Susceptible	None	None	None	SAME
30	A	**NNRTI, NRTI**	**G190A**	None	**M184V**	V35T, T39E, I142T, T165A, K173S, D177G, V179I, I20 2V, Q207A, R211S, P226PR	A	**NNRTI, NRTI**	**G190A**	None	**M184V**	SAME
31	B	**NNRTI**	**V179VD**	None	None	V35T, E40ED, V60I, K122E , K173A, D177E, T200A, Q 207E, R211RK	B	**NNRTI**	**V179VD**	None	None	SAME
32	D	**NNRTI, NRTI**	**Y181YC**	None	**V75VM**	V35T, T39K, K49R, V60I, K64R, K122E, D123G, I135 IMR, D177E, I178M, T200I, E203G, Q207E, R211K	C+D	**NNRTI, NRTI**	**Y181YC**	None	**V75VM**	SAME
33	A	Susceptible	None	None	None	V60I, K122E, D123N, K173 T, Q174K, D177E, V179VI, T200A, Q207A, R211S	A	Susceptible	None	None	None	SAME
34	B	Susceptible	None	None	None	V35T, E40D, K49R, V60I, Q174K, D177E, Q207E	B	Susceptible	None	None	None	SAME
35	A	Susceptible	None	None	None	K122E, D123N, K173S, Q17 4K, D177E, V179I, I202V, Q207A	A	Susceptible	None	None	None	SAME
36	A	Susceptible	None	None	None	K49R, V60L, D121H, K122 E, R125RK, I135T, I142V, K173T, Q174K, D177E, Q2 07A, F214L	A	Susceptible	None	None	None	SAME
37	A	**NNRTI**	**Y181YFI N**	None	None	V35T, V60I, V90VI, K122E, D123S, I135T, I142V, K173 S, Q174K, D177E, V179I, Q 207A, R211S	A	**NNRTI**	**Y181YF IN**	None	None	SAME
38	A	Susceptible	None	None	None	V35T, T39G, V60I, I135T, K173S, Q174K, Q207A, R2 11N	A	Susceptible	None	None	None	SAME
39	A	Susceptible	None	None	None	V35T, E40D, V60I, K122E, D123N, I142V, K173S, Q17 4K, D177E, V179I, Q207A,	A	Susceptible	None	None	None	SAME

						R211S						
40	G	**NNRTI**	**Y181C, H 221Y**	None	None	V35I, V60I, K122E, D123N, I135V, K173R, Q174E, D177E, I178M, T200A, Q207K, R211N	G	**NNRTI**	**Y181C**	None	None	**H221Y**
41	A	Susceptible	None	None	None	V35T, T39I, V60I, D121H, I135T, K173S, Q174N, D177E, I195L, T200A, Q207A, R211S, F214L	A	Susceptible	None	None	None	SAME
42	A	Susceptible	None	None	None	V35T, K49R, V60I, K122V, I135T, K173S, Q174K, D177E, V179I, T200A, I202V, Q207A, R211S	A	Susceptible	None	None	None	SAME
43	D	Susceptible	None	None	None	V35T, V60I, K104N, K122E, I142V, A158S, D177E, Q207G, R211K	D	Susceptible	None	None	None	SAME
44	A	Susceptible	None	None	None	V35T, V60I, D121H, I135T, K173S, Q174K, D177E, V179I, T200A, Q207A, R211S	A	Susceptible	None	None	None	SAME
45	A	Susceptible	None	None	None	V35T, T39L, V60I, D121H, K122E, I135T, K173L, Q174K, D177E, V179I, Q207A, R211S	A	Susceptible	None	None	None	SAME
46	A	Susceptible	**V179T**	None	None	V35T, K49R, V60I, K122E, D123G, I135T, K173L, Q174K, D177E, I178V, I202V, Q207D, R211S, F214L	A	Susceptible	None	None	None	SAME
47	D	Susceptible	None	None	None	V35T, T39I, E40D, K49R, V60I, D121Y, K122E, I135T, F171Y, K173L, Q174K, D177E, V179I, Q207A, R211S	D	Susceptible	None	None	None	SAME
48	A	Susceptible	None	None	None	V35T, V60I, K122E, D123S, I135R, E169D, K173S, Q174K, D177E, G196E, Q207A, R211S	A	Susceptible	None	None	None	SAME
49	A	Susceptible	None	None	None	V60I, D121Y, K122E, I135IT, S162H, K173L, Q174K, D177E, V179I, Q207A, R21	A	Susceptible	None	None	None	SAME

50	CRF02_AG	**NNRTI**	**K103N, F227FL**	None	None	V60I, V90I, I135A, I142V, S162A, K173KT, Q174E, T200A, Q207E, L228Q, V245Q, D250E	CRF02_AG	**NNRTI**	**K103N, F227FL**	None	None	SAME
51	A	Susceptible	None	None	None	V35T, T39Q, S105A, K122E, D123N, I135T, K173A, D177G, V179I, T200A, I202V, Q207A, R211Q	A	Susceptible	None	None	None	SAME
52	A	**NNRTI, NRTI**	**K101E, E138A, G190A**	**F77FL, T215TAS**	M184V	Q85P, K122E, D123N, I135T, I142V, K173S, Q174K, V179I, T200A, Q207A, R211S, F214FL	A	**NNRTI, NRTI**	**K101E, E138A, G190A**	**F77L**	M184V	T215TAS
53	D	Susceptible	None	None	None	V35T, E40D, I47L, K49R, V60I, D121C, K122E, D123E, R125RK, K173E, D177E, T200A, Q207E	D	Susceptible	None	None	None	SAME
54	C+D	Susceptible	None	None	None	V35I, K49R, K102KE, K122E, D123S, I135IV, S162A, D177E, I178M, V179I, T200A, E204EK, Q207EG, R211K	C+D	Susceptible	None	None	None	SAME
55	A	Susceptible	None	None	None	V35T, T39Q, S105A, K122E, D123N, I135T, K173A, D177G, V179I, T200A, I202V, Q207A, R211Q	A	Susceptible	None	None	None	SAME
56	A	**NNRTI, NRTI**	**A98G, Y181C**	**L210*W, T215HNY**	**M184V,**	K122E, D123S, T128TP, I135T, T139R, K173L, Q174*K, N175NFIY, P176PFLS, D177*E, I178IK, V179I, T200A, I202IM, E203A, Q207A, R211S	A	**NNRTI, NRTI**	**A98G, Y181C**	**L210W**	None	**T215HNY**
57	A	Susceptible	None	None	None	V35T, K104R, D121H, I135T, S162Y, K173L, Q174K, D177E, Q207A, R211S	A	Susceptible	None	None	None	SAME
58	A	**NNRTI, NRTI**	**A98G, K101E, V106VI, V179T, Y181C, G190S**	None	**M184V**	D121H, K122E, K173A, Q174K, D177E, I178M, G196E, T200A, I202IM, E203EDGV, Q207A, R211N	A	**NNRTI, NRTI**	**A98G, K101E, V106VI, V179T, Y181C**	None	**M184V**	SAME

59	B	Susceptible	None	None	None	V35T, T39M, V60I, D121Y, K122E, T131TP, E169EK, Q174K, D177E, I178M, Q207E	B	Susceptible	None	None	None	SAME
60	C	Susceptible	None	None	None	V35T, E36A, T39D, S48T, V111VG, K166R, K173A, Q174K, D177E, I178M, D192N, I195M, G196M, T200A, Q207E	C	Susceptible	None	None	None	SAME
61	D	**NNRTI, NRTI**	**V108I, Y181YC**	**T215TN**	None	K32E, V35T, T39L, V60I, D121Y, K122E, D177E, R206RK, Q207E, R211K	D	**NNRTI**	**V108I, Y181YC**	None	None	**T215TN**
62	A	**NNRTI**	**G190A**	None	None	V35T, V60I, K122E, D123S, I135T, I142V, F171Y, K173S, Q174K, D177E, V179I, T200A, Q207A, R211S	A	**NNRTI**	**G190A**	None	None	SAME
63	C + D	**NNRTI, NRTI**	**V106A**	None	**M184V**	V35I, K49R, K102E, K122E, D123S, S162A, T165K, D177E, I178M, V179I, T200A, Q207G, R211K	C + D	**NNRTI, NRTI**	**V106A**	None	**M184V**	SAME
64	A	**NNRTI**	**G190A**	None	None	V35T, E40D, K122E, D123N, Q161H, K173S, Q174K, P176T, D177E, V179I, T200A, I202V, Q207A, R211S, F214L	A	**NNRTI**	**G190A**	None	None	SAME
65	A2	**NNRTI, NRTI**	**Y188L**	None	**M184V**	V35T, T39L, K122E, D123E, D177E, I178M, T200A, Q207E, R211K	A2	**NNRTI, NRTI**	**Y188L**	None	**M184V**	SAME
66	A	**NNRTI**	**G190A**	None	None	K32KR, V35T, T39A, E40D, K122E, D123N, K173S, Q174K, P176T, D177E, V179I, T200E, I202V, Q207A, R211S, F214L, P226PL	A	**NNRTI**	**G190A**	None	None	SAME
67	A	Susceptible	None	None	None	I31IK, V35T, E40D, V60I, K102R, K104R, D121Y, K122E, I135T, K173S, Q174K, D177E, V179I, T200A, Q207A, R211S	A	Susceptible	None	None	None	SAME
68	A	Susceptible	None	None	None	V35T, T39L, K122E, D123E, D177E, I178M, T200A, Q207E, R211K	A	Susceptible	None	None	None	SAME

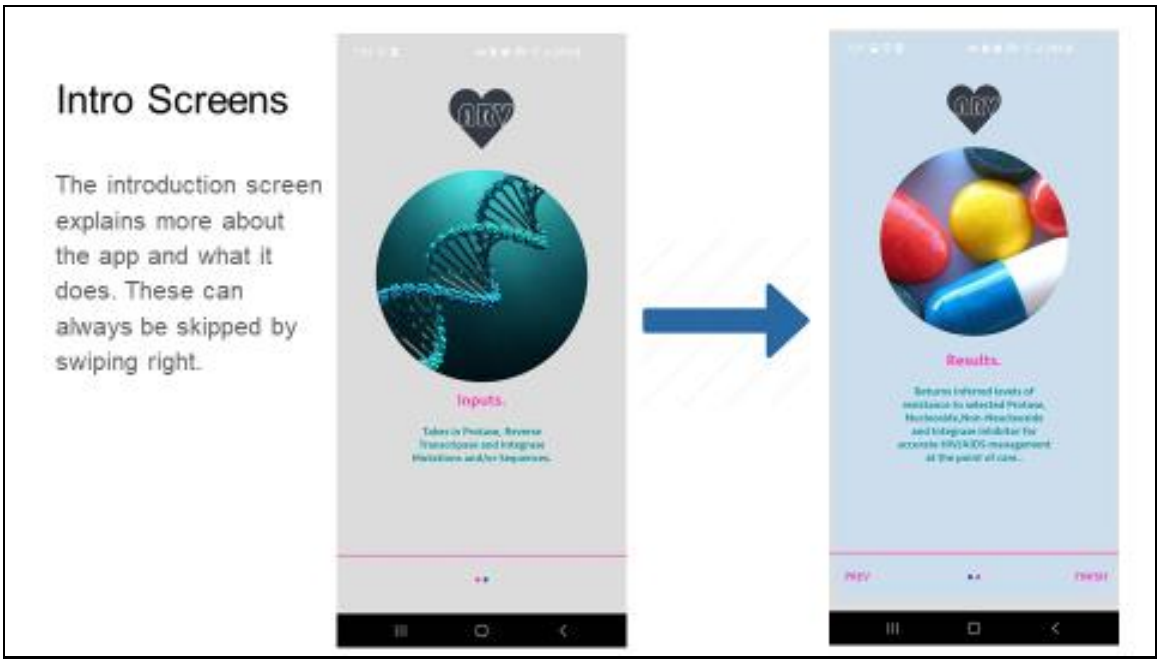
69	A	**NNRTI**	**G190A**	None	None	V35T, T39A, E40D, K122E, D123N, R125RK, K173S, Q174K, P176T, D177E, V179I, T200A, I202V, Q207A, R211S, F214L	A	**NNRTI**	**G190A**	None	None	SAME
70	C + D	Susceptible	None	None	None	V35IT, K49R, I50IV, P55PS, V60VI, K102KE, K122E, D123S, I135IV, S162A, D177E, I178M, V179I, T200A, E204EK, Q207EG, R211K	C + D	Susceptible	None	None	None	SAME
71	A	Susceptible	None	None	None	V35T, T39Q, S105A, V111VG, K122E, D123N, R125RK, T131TP, I135T, V148VG, K173A, D177G, V179I, E194EK, R199RK, T200A, I202V, Q207A, R211Q, D218DN	A	Susceptible	None	None	None	SAME
72	D	Susceptible	None	None	None	V35PST, E36D, K49R, V60I, D121CHRY, K122E, I135IR, D177E, I178M, Q207E, R211K	D	Susceptible	None	None	None	SAME
73	A	Susceptible	None	None	None	V35T, V60I, K122E, D123N, R125RK, I135T, K173L, Q174K, D177E, V179I, I195L, T200A, Q207A, R211S	A	Susceptible	None	None	None	SAME
74	A	Susceptible	None	None	None	V35PST, V60I, K122E, D123S, I135T, K173A, Q174K, D177E, V179I, T200A, Q207A	A	Susceptible	None	None	None	SAME
75	A	Susceptible	None	None	None	V35T, T39K, E53D, K122E, D123N, I135T, T165I, K173S, Q174K, D177E, V179I, I202V, Q207A, R211N	A	Susceptible	None	None	None	SAME
76	D	**NRTI**	None	None	**Y115YF**	V35T, E40D, K49R, V60VI, V75VL, D121DN, K122E, D123N, R125RK, D177E, I178IM, E194EK, Q207E, R211K, D218DN	D	**NRTI**	None	None	**Y115YF**	SAME
77	A	Susceptible	None	None	None	V35T, K49R, K122E, D123N, I135T, K173S, Q174K, D177E, V179I, T200E, Q207	A	Susceptible	None	None	None	SAME

						A, R211S						
78	A	Susceptible	None	None	None	V35T, V60I, K122E, D123S, I135T, K173A, Q174K, D177E, V179I, Q197P, T200A, Q207A, R211S, F214L	A	Susceptible	None	None	None	SAME
79	A	Susceptible	None	None	None	T39K, K122E, D123N, K173S, D177E, V179I, T200A, I202V, Q207S, R211K	A	Susceptible	None	None	None	SAME
80	A	Susceptible	None	None	None	V35T, T39A, E40D, V60I, K122E, D123S, I135T, K173S, Q174K, D177E, V179I, T200A, Q207A, F214L	A	Susceptible	None	None	None	SAME
81	A	Susceptible	None	None	None	T39K, K122E, D123N, K173S, D177E, V179I, T200A, I202V, Q207S, R211K	A	Susceptible	None	None	None	SAME
82	D	Susceptible	None	None	None	V35T, V60I, K104N, K122E, I142V, A158S, D177E, Q207G, R211K	D	Susceptible	None	None	None	SAME
83	C	Susceptible	None	None	None	V35T, E36S, T39E, E40D, K43KR, S48T, K49R, I50V, V60I, K122E, D123S, I135T, K173A, D177E, T200A, Q207E	C	Susceptible	None	None	None	SAME
84	A	**NRTI**	None	None	**M184V**	V35T, T39K, K43A, V60I, D121H, K122E, D123S, K173A, Q174K, D177E, I178M, V179VI, T200E, E204Q, Q207A, R211K	A	**NRTI**	None	None	**M184V**	SAME
85	D	Susceptible	None	None	None	V35T, K49R, V60I, D121Y, K122E, K173KQR, D177E, I180IV, E194D, T200TI, Q207E, R211K	D	Susceptible	None	None	None	SAME
86	B	Susceptible	None	None	None	V35T, T39A, E40D, K49R, V60I, D121Y, K122E, D123DN, I142IV, D177DE, V179VI, E203D, Q207E, R211K	B	Susceptible	None	None	None	SAME
87	C	Susceptible	None	None	None	V35T, E36A, T39E, E40D, K49R, K122E, D123N, A158S, K173I, Q174K, T200A,	C	Susceptible	None	None	None	SAME

						Q207D, R211K						
88	A	Susceptible	None	None	None	V35T, V60I, K122E, D123NS, I135IT, K173S, Q174K, D177E, V179I, I202V, Q207D, R211S	A	Susceptible	None	None	None	SAME
89	B	**NNRTI**	**G190A, F227FL**	None	None	S48T, V60I, K122E, D123S, I135T, T139A, D177E, Q207E, R211K	B	**NNRTI**	**G190A**	None	None	F227FL
90	A	Susceptible	None	None	None	V35T, T39M, V60I, K64R, D121H, K122E, I135T, A158S, S162A, K173S, Q174K, D177E, I178L, I180IM, T200A, Q207A, R211S	A	Susceptible	None	None	None	SAME
91	D	**NNRTI, NRTI**	**V108I, Y181YC**	**T215TN**	None	V35T, T39I, E40D, K49R, V60I, W88WG, D123E, D177E, T200A, E204Q, Q207A	D	**NNRTI, NRTI**	**V108I, Y181YC**	None	None	**T215TN**
92	A	Susceptible	None	None	None	E28A, V35T, T39L, K122E, D123E, T131TP, I135T, K173S, Q174K, D177E, V179I, T200A, Q207A, R211S	A	Susceptible	None	None	None	SAME
93	D	Susceptible	None	None	None	V35T, T39M, K122E, D123G, I135T, K166R, D177E, G196E, T200A, Q207E, R211K	D	Susceptible	None	None	None	SAME
94	A	Susceptible	None	None	None	V35T, V60I, K122E, D123S, I135T, K173S, Q174K, D177E, V179I, T200E, Q207A, R211S, F214L	A	Susceptible	None	None	None	SAME
95	A	Susceptible	None	None	None	V35T, T39K, V60I, K122E, D123N, K173A, Q174K, D177E, V179I, T200TA, I202V, Q207A, R211S	A	Susceptible	None	None	None	SAME
96	A	**NRTI**	None	None	**M184MV**	V35T, T39K, K43A, V60I, D121H, K122E, D123S, K173A, Q174K, P176PS, D177E, I178M, V179VI, T200E, E204Q, Q207A, R211K	A	**NRTI**	None	None	**M184MV**	SAME
97	A	**NNRTI**	**K103N**	None	None	V35T, V60I, K64R, D121H, K122E, I135T, K173S, Q174	A	**NNRTI**	**K103N**	None	None	SAME

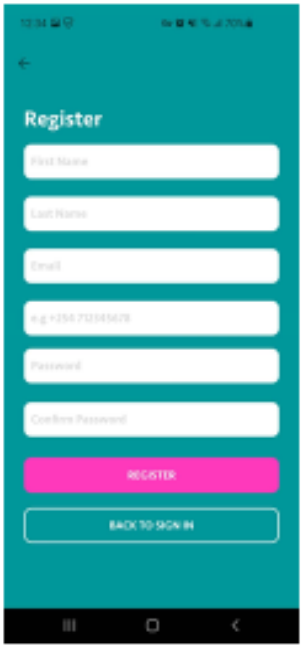
						K, D177E, V179S, T200A, Q207A, R211S, V245K						
98	D	Susceptible	None	None	None	V35T, E36D, K49R, V60I, D121C, K122E, D177E, I178M, Q207E	D	Susceptible	None	None	None	SAME
99	D	Susceptible	None	None	None	I31IKN, A33S, L34T, V35T, E40D, K49R, V60I, K122E, D123N, Q174QK, D177E, I178M, R206RK, Q207E, H208HP, R211K	D	Susceptible	None	None	None	SAME
100	C	**NNRTI**	**K103N**, **V106I**	None	None	V35T, T39R, K43E, S48T, K102R, K122E, I142V, K166R, K173A, Q174R, D177E, I195L, G196K, T200A, Q207E, R211K	C	**NNRTI**	**K103N**, **V106I**	None	None	SAME

Appendix 1.4: *ARVPredictor:* User Guide



Login Page

On this page, you the user is to provide the phone number they registered with and their password before clicking the Login button. However if the user is not registered, they should click the register button for registration page.

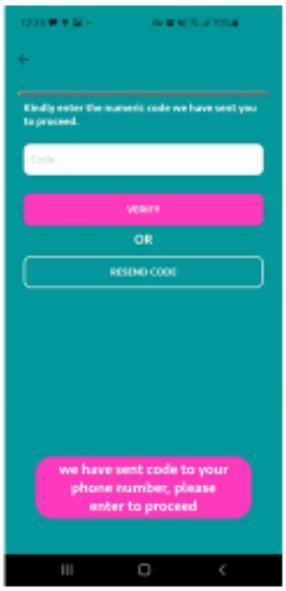


Registration Page

On this page., the user provides they first and second names, theri email,phone number and password. They then click register to initiate the registration process.

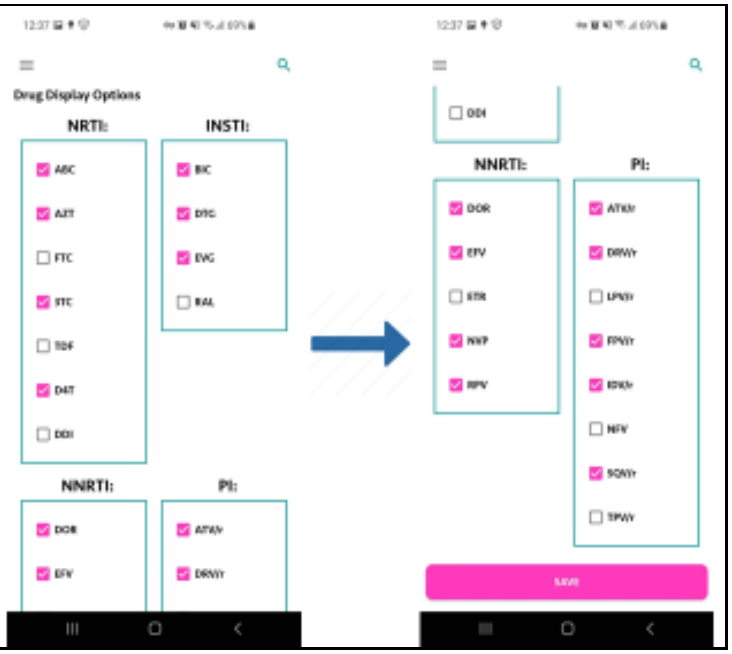
Phone Number Verification

In order to be sure about the phone number provided, we send a verification code through SMS. this code should be provided on this page for verification purposes.



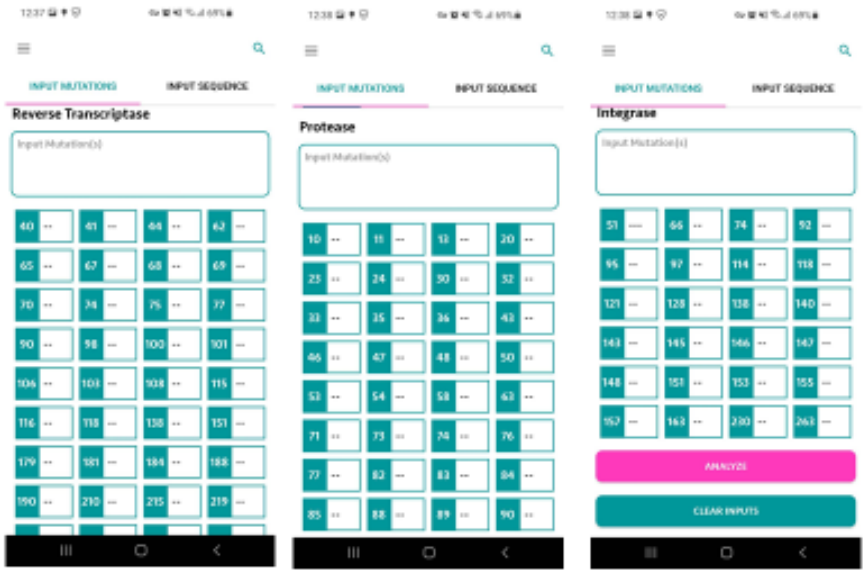
Drug Selection Page

On this page, the user selects the drug varieties that they want to be part of the results during mutation or sequence analysis. It is advisable to select all the available drugs.



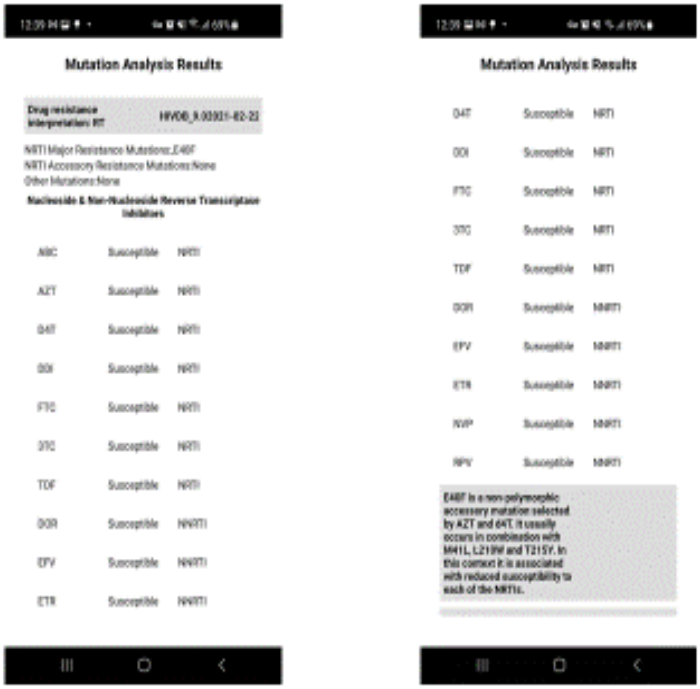
Mutation Selection Page

On this screen, the user is able to select the various mutations to analyze. There is the analyze button to initiate analysis.



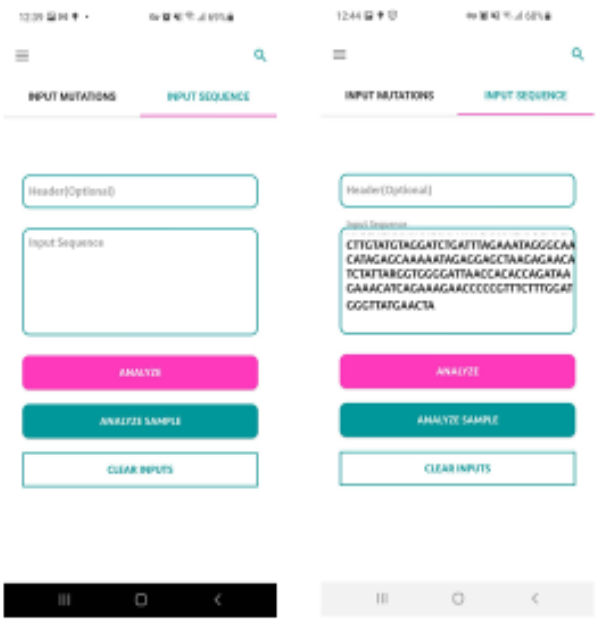
Mutation Analysis Result Page

On this screen, the user is set to view the results from analysis of particular mutation sets. These are very comprehensive results and are produced depending on the number of mutations provided.



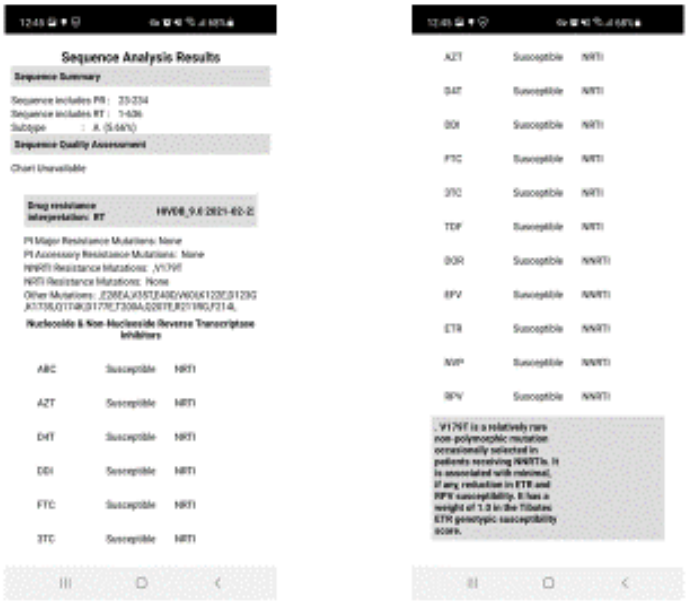
Sequence Analysis Page

This screen expects the sequence string from the user and a header parameter. To ensure no interference, the string should be copied from a separate file and pasted on the provided text area. The headers define extra parameters needed for the analysis.



Sequence Analysis Result Page

On this screen, the user will be able to view the results of their sequence analysis. The results are as well comprehensive and are displayed according to the number of sequences selected.



Appendix 1.5: Informed consent form

Title of the Research Study: An Android–Based Mobile App (*ARVPredictor)* for the detection of HIV Drug –Resistance Mutations and Treatment at the Point of Care: Development Study.

Investigator(s) –	Beatrice A. Ongadi	-	Corresponding Investigator
	Prof. George Obiero		
	Dr. Raphael Lihana		
	Dr. John Kiiru		
	Dr. Musa Ng'ayo		

You are being requested to take part in this research study which aims at evaluating a mobile android application (*ARVPredictor*). The application is currently under development and will be used for the detection of HIV Drug–Resistance Mutations and Treatment at the Point of Care.

The final tool forms part of the requirements for award of the Degree of Doctor of Philosophy in Bioinformatics for **Beatrice Ongadi** enlisted here in as the Corresponding Investigator.

The Summarized information below tells you important things you should think about before deciding to participate. Please ask questions about any of the information before you decide whether to contribute though the following contacts: betongadi@gmail.com or bongadi@students.uonbi.ac.ke. Tel: +254 728 324 324

KEY INFORMATION FOR YOU TO CONSIDER

- **Voluntary Consent**. You are being asked to volunteer for this research study. It is up to you whether you choose to participate or not. There are no penalties and you will not lose anything if you decide not to joint or if after you join, you decide to quit.
- **Purpose.** We are doing this research to evaluate a mobile android application (*ARVPredictor*). The application is currently under development and will be used for the detection of HIV Drug–Resistance Mutations and Treatment at the Point of Care.
- **Duration.** Your part of the study will last 6 months.
- **Procedures and Activities.** You will be requested to test the usability of the app based on your professional qualification and or experience. If you have any questions you will be free to call or send email for clarity.
- **Benefits**. There will be no immediate and direct benefits to participants in this study. Participants and others may benefit from the future interventions that will be informed by information learned from this study.
- **Alternatives.** Participation is voluntary and the only alternative is to voluntarily choose to not participate in the study.
- **Confidentiality:** No information containing the identity of any study participants will be released or published without their consent. All personal information collected will be accessed only by the principal investigator; and they will be stored in a password secured laptop. During analysis the identifying information will be ripped off and unique identifiers assigned.

(REQUIRED FOR DATA ANALYSIS)
I agree to take part in the study voluntarily and for my information to be used for research or shared with other researchers without identifiers.

SIGNATURE OF PARTICIPANT

Printed Name of Participant:_____

Signature of Participant:_____

Date:_____

(REQUIRED FOR FUTURE USE)

I authorize the storage of data collected as a part of this study for use in future research studies without identifiers.

SIGNATURE OF PARTICIPANT

Printed Name of Participant:_____

Signature of Participant:_____

Date:_____

SIGNATURE OF THE CORRESPONDING INVESTIGATOR

Signature:_____

Date: _____

Appendix 1.6: Requirement Gathering and Testing Questionnaire

Participant's Name (Optional): _____ **Date**: _____

Profession (Tick as appropriate): Virologist___ ICT Expert___Application Developer___Other (Specify):_____

Title of the Research Study: An Android–Based Mobile App *(ARVPredictor)* for the detection of HIV Drug –Resistance Mutations and Treatment at the Point of Care: Development Study.

Section A: ICT Experts/Web Developers/ Software Developers		
1.	Are you familiar with Android applications and how they work?	☐ Yes ☐ No ☐ Further Comments:_____
2.	Did you manage to download and install *ARVPredictor?*	☐ Yes ☐ No
3.	**Acceptance testing:** (Do you think the application works as intended?)	☐ Yes ☐ No ☐ Not Sure ☐ Further Comments:_____
4.	**Integration testing:** (Are the software components operating together)	☐ Yes ☐ No ☐ Not Sure ☐ Further Comments:_____
5.	**Unit testing:** (Are all the application units functioning independently as expected)	☐ Yes ☐ No ☐ Not Sure ☐ Further Comments:_____
6.	**Functional testing:** (Kindly verify each function through Black box testing --- Are all function working as expected).	☐ Yes ☐ No ☐ Not Sure ☐ Further Comments:_____
7.	**Performance testing:** (How can you gauge performance in relation to different workloads)	☐ Satisfactory ☐ Not Satisfactory ☐ Needs Improvement ☐ Further Comments:_____
8.	**Regression testing:** (Are all new features affecting the functionality of the application)	☐ Yes ☐ No ☐ Not Sure

		☐ Further Comments:_____
9.	**Stress testing:** (Kindly stress test the application in your own way and give a feedback)	☐ Kind of test done_____ ☐ Comments_____
10.	Please give general comments on how to improve the application	☐ _____ _____ _____ _____ ☐
Section B: Virologist/HIV Care givers/Pharmacist		
1.	Do you directly deal with HIV patients?	☐ Yes ☐ No ☐ Not Sure ☐ Rarely ☐ Further Comments:_____
2.	Do you directly handle ARVs?	☐ Yes ☐ No ☐ Not Sure ☐ Rarely ☐ Further Comments:_____
3.	Can you explain how different ARVs work?	☐ Yes ☐ No ☐ Not Sure ☐ Further Comments:_____
4.	Do you directly sequence HIV data?	☐ Yes ☐ Never ☐ Not Sure ☐ Rarely ☐ Further Comments:_____
5.	Did you have a problem accessing and downloading *ARVPredictor?*	☐ Yes ☐ No ☐ Not Sure ☐ Didn't get it ☐ Further Comments:_____
6.	Did you have a problem using *ARVPredictor?*	☐ Yes ☐ No ☐ Not Sure ☐ Didn't get it ☐ Further Comments:_____
7.	Kindly use the application in our own way	☐ Relevance_____

	and comment on the following areas: Relevance, Usability Speed, availability.	☐ Usability_____ ☐ Speed _____ ☐ Availability_____ ☐ Further Comments:_____ _____ _____
8.	Please give general comments on how to improve the application	☐ _____ _____ _____ _____ ☐ ___ ☐