Oligo-A sites

The Achilles heel in the HIV genome

HIV has the ability to integrate as a DNA molecule within the host cell's chromatin. Dr John N Anderson, Full Professor at Purdue University, and his team showed that the HIV genome in the host-integrated protein-coding strand possesses highly conserved structural features that have the potential to be exploited as pharmacological targets in the design of specific anti-HIV drugs.

The Human Immunodeficiency Virus (HIV) is able to generate a large amount of genetic variation, posing a major obstacle to the treatment of AIDS and to the development of an HIV vaccine. Although substantial progress has been made in the development of anti-HIV drugs – which has culminated in the use of antiviral drug therapy – the emergence of multi-drug-resistant variants of HIV remains a concern for the treatment of the disease in the future. From a drug design point of view, it is important to identify unusual features of the virus so that they might be used as a basis for treatments that can selectively target a viral feature that is conserved in the viral population and is critical for viral survival.

THE PECULIAR FEATURES OF HIV

HIV has the ability to integrate as a DNA molecule within the host cell's chromatin.

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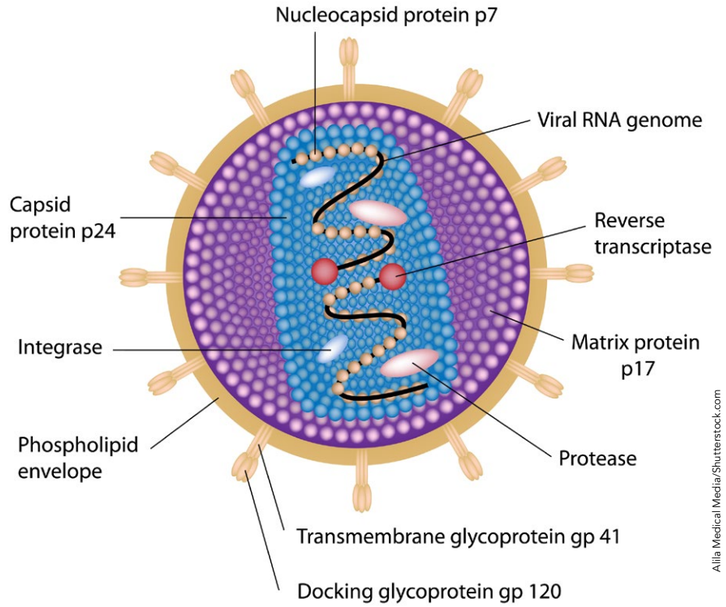
One of the two strands of the integrated HIV genome acts as a protein coding strand. For those unfamiliar with genetic mechanisms, the protein coding strands of genes serve as templates for the production of messenger RNAs (mRNA) during a process known as transcription. The mRNAs then travel to the cytoplasm where they associate with ribosomes. The informational content of the mRNAs is then 'read' to dictate the order or sequence of amino acids in specific proteins. The informational content in DNA and RNA is in the sequence or order of nucleotides. There are four main types of nucleotides in DNA, which are known by their familiar symbols of A, C, T, and G.

THE NUCLEOTIDE 'BIAS' AND THE GENETIC DIVERSITY IN HIV

Dr John Anderson and his team at Purdue University showed the A-rich and C-deficient regions typical of the nucleotide bias of HIV are the major factor in determining the composition of retroviral proteins. This helped them understand why proteins of HIV are rich in lysine and glutamate and other amino acids encoded by A-rich and AG-rich codons, while at the same time they are depleted in proline, which would be encoded by C-rich codons.

In order to further understand the genetic bias of the HIV genome, Dr Anderson and his collaborators compared the HIV genome to the gene databases of other organisms, identifying a total of 101 genes in bacterial and invertebrate species that indicate strong similarities with the HIV genomic composition. The researchers speculated that the similarities between the HIV proteins and the antigens from other pathogens may indicate a common pathogenic strategy for the promotion of immune dysregulation, which is a common feature of HIV infections and infections caused by many other pathogens.



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Structure of HIV.

In order to integrate with the host genome, the HIV virus uses a process known as reverse transcription, where complementary DNA (cDNA) is generated from an RNA template. The process is made possible by the action of the reverse transcriptase enzyme. It is generally assumed that the low fidelity of the HIV reverse transcriptase promotes the genetic variation of HIV.

Dr Anderson and his team proposed that this nucleotide bias is another driving force behind the genetic variation of the virus. The substitution of G–A transitions over T–C transitions has minimal deleterious effects on protein function and, consequently, produces large numbers of viable variants in the population. Genes that encode antigens for many bacterial, protozoan, and metazoan pathogens are also characterised by the unusual nucleotide bias, which is reflected in the composition of the encoded proteins, making it likely that diverse pathogens employ similar mechanisms for the generation of genetic variation.

OLIGO-A SITES IMPACT DNA CURVATURE AND THE WIDTH OF THE MINOR GROOVE

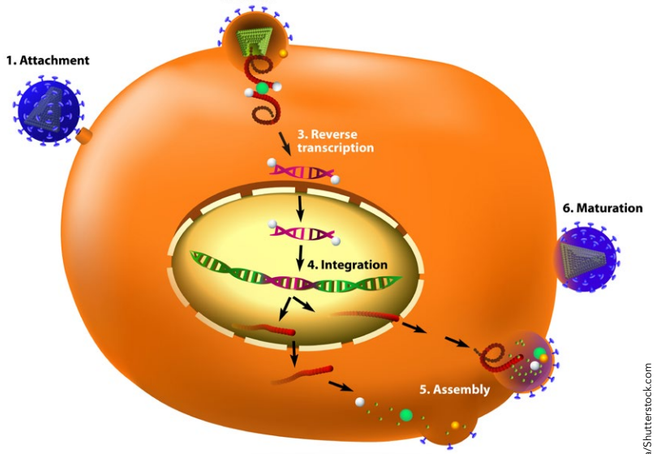
The HIV cDNA contains specific regions of four to six nucleotide base pairs, known as oligo-A sites, enriched in A because of the A genome bias. Oligo-A sites dictate the DNA structure, since a single site can produce a deflection in the DNA helical axis from a straight line by as much as 36 degrees. When such sites are spaced ten nucleotide base pairs apart, the deflections sum to produce DNA curvature and the formation of circular DNA structures. These structures may reflect organised chromatin of the HIV when it is integrated into the host genome.

Another important structural feature associated with oligo-A sites is the narrowness of the DNA minor groove. In double-stranded DNA, the backbones of the two strands are closer together on one side of the helix than on the other. The regions where the backbones are far apart are known as major grooves and those where they are close together are known as minor grooves. Dr Anderson and his colleagues propose that narrow minor grooves could act as important pharmacological targets for minor groove binding drugs (MGBDs).

THE THERAPEUTIC POTENTIAL OF MINOR GROOVE BINDING DRUGS

Since drugs that bind in the DNA minor groove disrupt chromatin on sequences that contain multiple closely spaced oligo-A tracts, which are prevalent in HIV DNA, Dr Anderson and his collaborators recently published a study where they

HIV has the ability to integrate as a DNA molecule within the host cell's chromatin.



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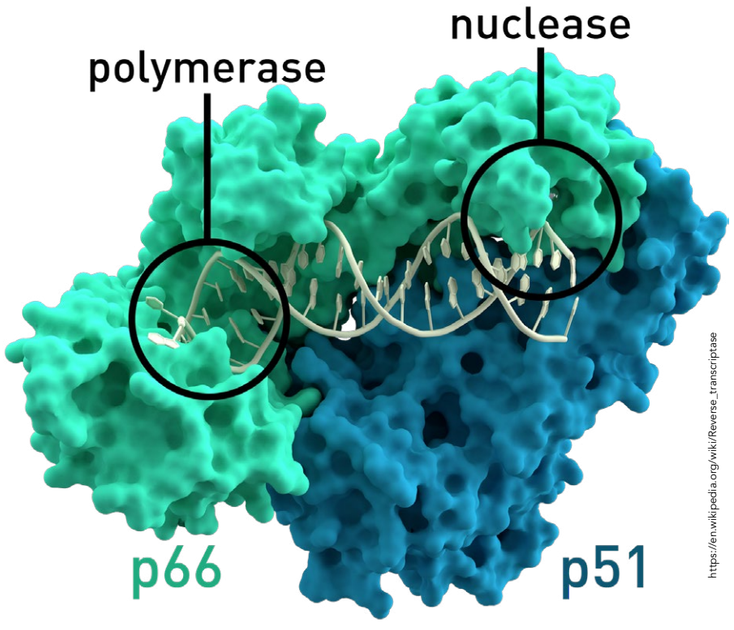
HIV replication cycle. The virus uses a process known as reverse transcription to generate complementary DNA, which integrates with the host genome.



https://en.wikipedia.org/wiki/Reverse_transcriptase

Crystallographic structure of HIV-1 reverse transcriptase.

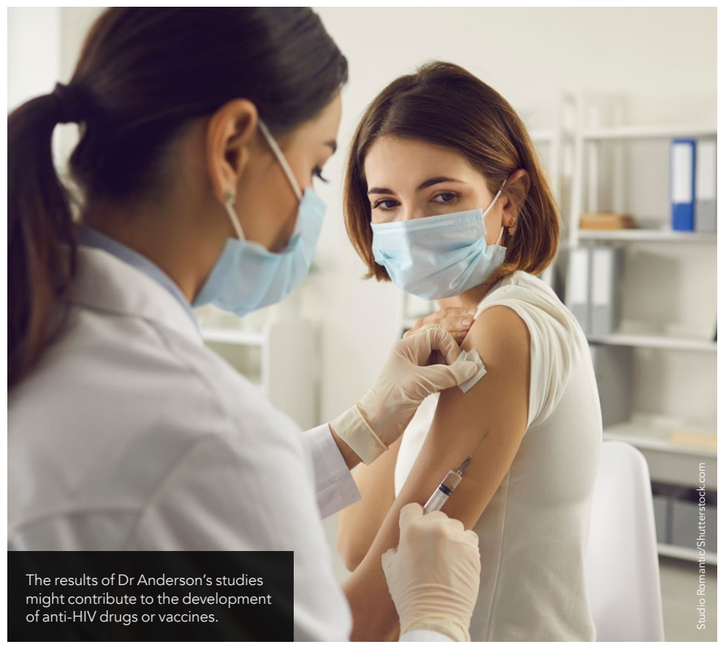
proposed that these drugs exert an inhibitory effect on the assembly of HIV chromatin. To test their hypothesis, Dr Anderson and his team incubated HIV cDNA both in the presence and in the absence of several MGBDs. The MGBDs inhibited the assembly of chromatin on HIV cDNA, in vitro and in yeast cells, in a manner that was proportional to the presence of oligo-A sites. These studies demonstrated that the narrow DNA minor grooves represent a possible 'Achilles heel' in the HIV genome and that MGBDs might serve as a novel class of anti-HIV agents that act by preferentially disrupting chromatin assembly. The laboratory group also demonstrated that MGBDs inhibited the assembly of chromatin on selected membrane antigen genes from other pathogens that displayed the A-bias. These drugs have been shown by others to reduce the pathogenicity of these organisms. Whether this effect is involved in the anti-pathogenic action of the drugs is a topic for future investigation.

Although the study did not show that MGBDs block viral replication, the researchers did not rule this out as a desirable consequence of the MGBDs' mechanism of action. The team now hopes to involve a wider network of collaborators to undertake studies using HIV-infected cells to further test the anti-HIV potential of MGDBs, particularly as treatments of multidrug-resistant variants of HIV.

MOTIVATION TO STUDY HIV

The studies described by the Anderson group with HIV began with an analysis of the structure of a highly curved segment of DNA that the group had isolated from a bird. To determine if there were similar segments in other organisms, the group searched for sequence similarities in large sequence databases from other organisms. The most similar sequences uncovered by these searches were from HIV and related viruses. Close inspection of these viral sequences revealed that the matching sequence elements were multiple closely spaced oligo-A sites that were seen in the original bird sequence. These findings led to an extensive characterisation of the nucleotide and protein sequence patterns of all known retroviruses, and the studies are described in the publication by Bronson and Anderson (1994).

The narrow DNA minor grooves represent a possible 'Achilles heel' in the HIV genome.



The results of Dr Anderson's studies might contribute to the development of anti-HIV drugs or vaccines.

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Behind the Research

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Research Objectives

Dr John N Anderson researches the structural features of the HIV genome, with a view to helping advance the design of anti-HIV drugs.

Detail

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Bio

John Anderson was born in Pittsburgh, Pennsylvania, and received his BS degree in biology at Slippery Rock College in Pennsylvania. He obtained his Doctor of Philosophy (PhD) from Purdue University where he studied estrogen receptors with James H Clark. He was then an ACS postdoctoral fellow with Robert T.Schimke at Stanford University where his work centered on gene isolation. He has been a full professor at Purdue University since 1986.

Dr Anderson has a long-standing research interest in the role of chromatin structure in the control of gene expression. Additional research topics include DNA structure, genomics, and retroviral evolution. He teaches a graduate-level course in endocrinology and courses in molecular biology for undergraduate students at Purdue. His most recent work has focused on the effects of DNA methylation in the control of nucleosome positioning and stability. He has published more than 80 peer-reviewed research publications and several books for teaching college-level biology.

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Collaborators

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References

Balzarini, J, Camarasa, M, Pérez-Pérez, M, San-Félix, A, Velázquez, S, Perno, C, De Clercq, E, Anderson, J, Karlsson, A, (2001). Exploitation of the low fidelity of human immunodeficiency virus type 1 (HIV-1) reverse transcriptase and the nucleotide composition bias in the HIV-1 genome to alter the drug resistance development of HIV. *Journal of virology,* 75(13), 5772–5777. Available at: doi.org/10.1128/JVI.75.13.5772-5777.2001

Collings, C, Little, D, Schafer, S, Anderson, J, (2019). HIV chromatin is a preferred target for drugs that bind in the DNA minor groove. *PLOS ONE,* 14(12), p e0216515. Available at: doi.org/10.1371/journal.pone.0216515

Fitzgerald, D, Bronson, E, Anderson, J, (1996). Compositional similarities between the human immunodeficiency virus and surface antigens of pathogens. *AIDS research and human retroviruses,* 12(2), 99–106. Available at: doi.org/10.1089/aid.1996.12.99

Albert, F, Bronson, E, Fitzgerald, D, Anderson, J, (1995). Circular structures in retroviral and cellular genomes. *The Journal of biological chemistry,* 270(40), 23570–23581. Available at: doi.org/10.1074/jbc.270.40.23570

Bronson, E, Anderson, J, (1994). Nucleotide composition as a driving force in the evolution of retroviruses. *Journal of molecular evolution,* 38(5), 506–532. Available at: //doi.org/10.1007/BF00178851

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