The characteristics of retroviruses

Retroviruses have various characteristics that make them unique as gene delivery vehicles. Their life cycle includes an integrated state in the DNA of the host chromosome.

Retroviruses are the only animal viruses that integrate into the host cell's genome during the normal growth cycle. They use an integrase that acts in a site-specific manner to join the ends of the viral cDNA to target sequences in host cell DNA. The linear ds cDNA made in the cytoplasm is transported to the nucleus where it is also found as circles and as integrated DNA. Two forms of circular DNA are generally found: one having a single Long Terminal Repeat (LTR) and one having two LTRs. It is now thought that the original integrated proviruses were linear molecules with two LTRs.

The retroviral promoter can direct high-level, efficient expression of genes encoded within the viral capsid of its genome using chromatin.

The retroviral genomes can accommodate changes to its configuration.

Retroviruses offer gene therapy researchers aid for delivering genes to target cells at high efficiency that allows for long-term, stable expression of introduced genetic elements

The retroviral life cycle begins in the nucleus of an infected cell.

At the beginning of the life cycle the retroviral genome is a DNA element integrated into and covalently attached to the DNA of the host cell.

Full-length genomic mRNA is made starting at the beginning of the repeat at the 5′ LTR (Long Terminal Repeat).

The free particle can infect new cells by binding to a cell surface receptor. The specificity of the virus-cell interaction is determined most commonly by the envelope proteins of the retrovirus. Infection leads to injection of the virus nucleoprotein core (consisting of many gag-derived proteins, full-length genomic RNA, and the reverse transcriptase protein).

Once inside the cell, the nucleoprotein complex accesses intracellular DNA nucleotide triphosphate pools, where the reverse transcriptase protein initiates and creation of a double-stranded DNA copy of the genome of the virus is prepared for integration into the host cell chromosome. When reverse transcription is completed, the viral enzyme integrase looks for an appropriate storage place for the DNA, which the integrase clips the host DNA to and binds the double-stranded DNA into the host DNA.

The virus is the able to initiate a new round of replication again.

3 major proteins encoded in a retroviral genome

- Gag is a polyprotein and is an acronym for Group Antigens (ag).
- Pol is the reverse transcriptase.
- Env is the envelope protein.

The group antigens form the viral core structure and are the major proteins which comprise the nucleoprotein core particles.

Reverse transcriptase is the essential enzyme that carries out the reverse transcription process that take the RNA genome to a double-stranded DNA preintegrate form. General transcription and proteins are encoded from spliced mRNA of retroviruses.

Transcription proceeds through the genome and mRNA is polyadenylated and processed using signals in transcribed regions from the 3′ LTR at the end of the transcribed R (repeat). The full-length message can be spliced to lead to production of envelope proteins (or other proteins depending upon retroviral class). Unspliced full-length mRNA can give rise to gag-pol proteins. Gag and Pol are made as either Gag protein or a Gag-Pol precursor.

Translated proteins assemble a retroviral particle at the cell surface. Full-length genomic unspliced mRNA is bound by gag-derived proteins and incorporated into the budding particle.

Virion structures – In retroviruses particle shapes can be divided into distinct categories:

A-type particles are immature intracellular forms derived from endogenous retrovirus-like elements and the immature form of MMTV.

B-type particles correspond to the extracellular form of MMTV and are characterised by prominent surface protein “ spikes” and a dense asentric nucleocapsid.

C-type particles form at the surface of the cell at the site of budding. Lentiviruses bud like C type particles but have a distinctive blunted cone shaped core.

D-type particles are the MMPV related viruses of sub-human primates, and differ from B-type particles by a lack of surface spikes.

The gag (group specific antigen) gene encodes the viral matrix, capsid and nucleoproteins

The protease encodes a product that cleaves the gag polyprotein precursor. It can be encoded as part of Gag or a Gag-Pro-Pol polyprotein

The major read-through product is derived from the pol gene which encodes the reverse transcriptase and an integrase which is involved in provirus integration.

The envelope gene encodes the surface glycoprotein (SU) – transmembrane (TM) polyprotein.

Viral entry

Retroviruses enter by at least two different manners, dependent upon the retroviral subclass. The viral envelope is critical in each case for recognising appropriate surface receptors to initiate viral fusion to the host target cells.

The RNA genome in the free retrovirus is arranged as a diploid genome with identical sequences. The mRNA associates with a tRNA primer (pro, trp, or lys) that is bound by complementary base pairing to 18 base pairs to the U5 region.

The integrated form (proviral) of all retroviruses contain transcription regulatory sequences primarily in Long Terminal Repeats (LTR). LTR sequences are derived from sequences unique to the 5′ end of viral RNA (U5), from sequences unique to the 3′ end of viral RNA (U3), and from sequences repeated at both ends of the viral RNA. The integrated provirus is

larger than the viral genome but its complexity is the same because of duplication of U3 and U5 during synthesis.

Replication of retroviruses is sensitive to the transcription inhibitors Actinomycin D, alpha-amanitin nucleoside and analogues like 5-bromodioxyuridine and cytosine arabinoside. 5 bromodioxyuridine and cytosine arabinoside are thought to inhibit DNA replication.