



BHARATHIDASAN UNIVERSITY

Tiruchirappalli- 620024, Tamil Nadu, India

Programme: M.Sc., Biomedical Science (5 Year Integrated Program)

Course Title : Medical Genetics

Course Code : BM59C19

Unit-IV

Genomic Imprinting

Dr. K. PREMKUMAR

Professor

Department of Biomedical Science

Genomic Imprinting

- Refers to the genetic phenomenon of parent-of-origin gene expression.
- Does not obey Mendel's laws.
- Monoallelic expression of genes based on the sex of the transmitting parent.
- Genomic imprinting a reversible form of gene inactivation and explains the difference between paternal and maternal genomes.
- Genes subjected to genomic imprinting are called “imprinted genes” and make up <1% of genes.

Evolution of Genomic Imprinting

- Imprinting evolved nearly 125 million years ago.
- Current evidences show that Protherian mammals lacked imprinted genes, while the Metatherians and Eutherians show gene imprints; this means that Imprinting evolved in a common ancestor to the Therian mammals.
- **Evidences:**
- Identification of 125 year old marsupial-like mammalian fossil.
- Molecular analyses of amino acid sets of BRCA 1 proteins indicate that Meta – Eu-therians split occurred in 182-190 million years ago.
- **Theories on the origin of Imprinting:**
- Two hypothesis have been proposed for the origin of imprinting.
 - Parental conflict Hypothesis
 - Foreign DNA Silencing Hypothesis

Imprinted Genes

- Genes in which one copy of the allele is epigenetically marked / imprinted in either the egg / sperm.
- 156 novel candidate imprinted genes are identified.
- Humans possess less no. of genes than mice.
- Parental imprints are established during gametogenesis and further maintained in epigenetic state.

Features:

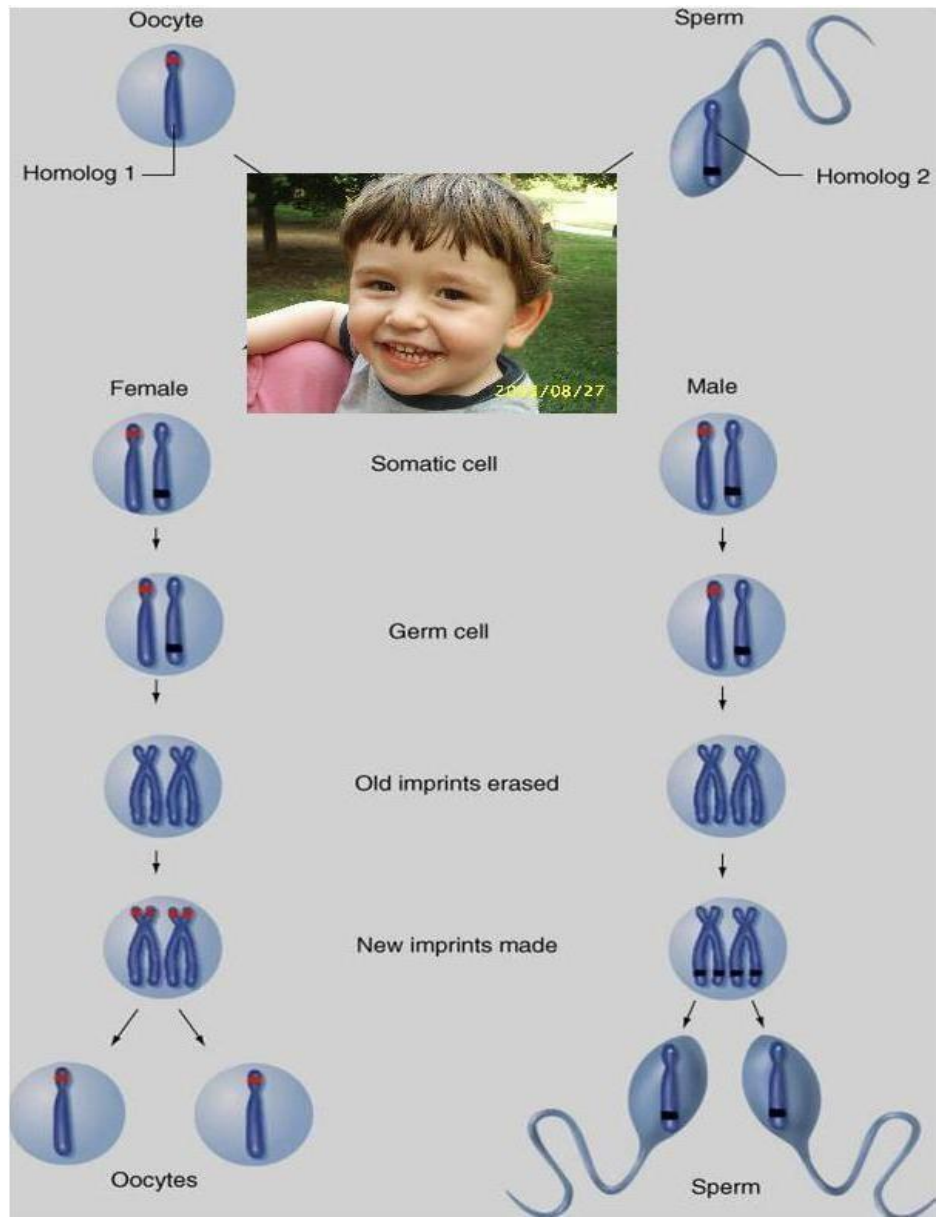
- ✓ Functionally haploid.
- ✓ Cluster in large regions of the genome. Each cluster is controlled by a cis-acting imprinting centre (IC).
- ✓ Code for proteins, untranslated RNA transcripts.

Mechanism of Imprinting

- Multi-step, dynamic process.
- Involves epigenetic modifications that are erased and re-programmed during gametogenesis.
- DNA methylation and histone modifications : play important roles in establishment of imprint.

Steps:

- a) Chromosome is marked as to its parental origin.
- b) Stable maintenance of the mark as cells divide and differentiate.
- c) Recognition of mark by transcriptional machinery, mono-allelic expression.
- d) Erasure and resetting of mark in the germ cells.

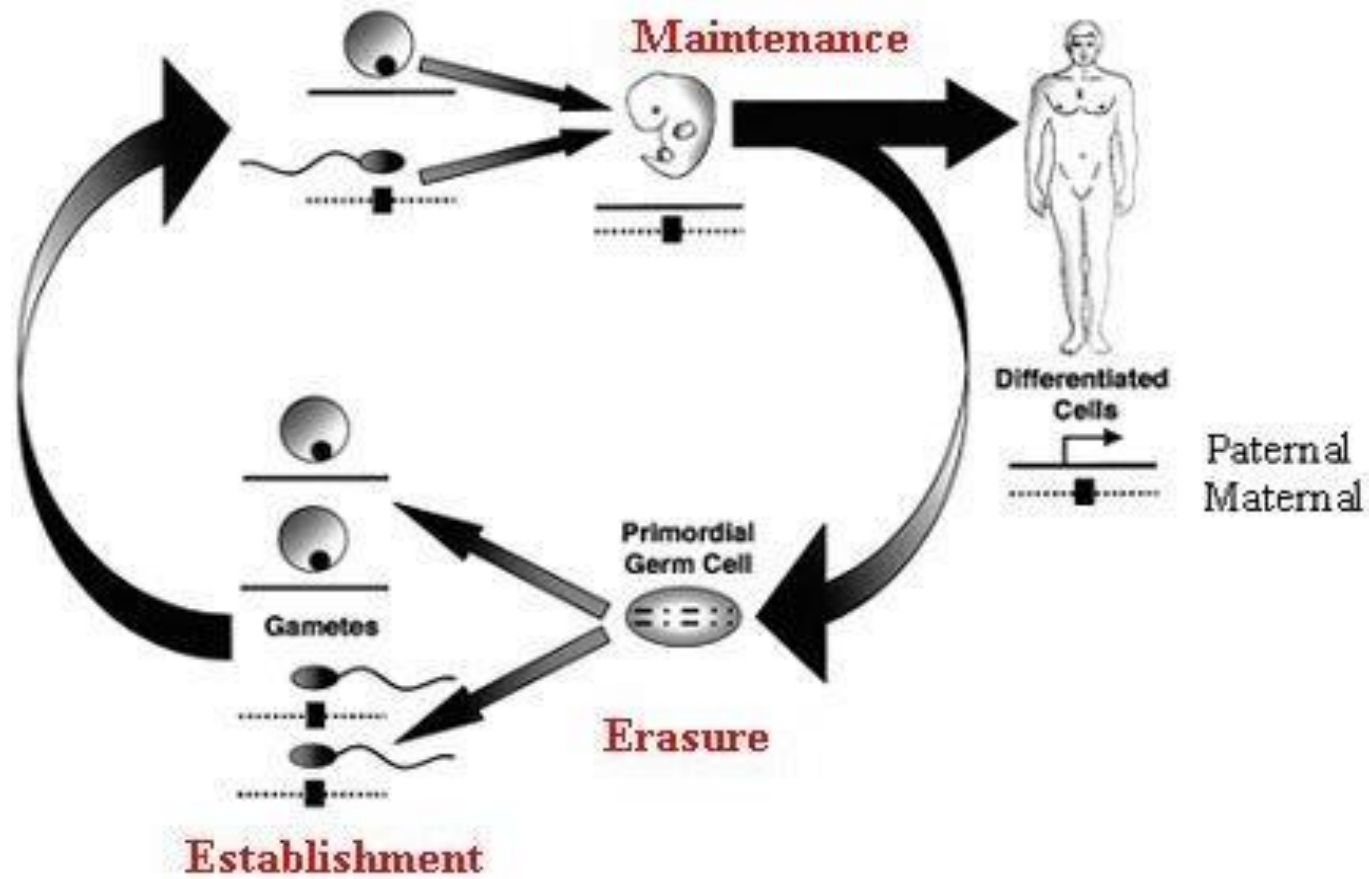


DNA Methylation and Genomic Imprinting

- DNA methylation plays an important role in chromatin structure and gene expression.
- Has been reported as the optimal method for genomic imprinting.
- The copy of the gene to be inactivated is coated with methyl group.
- Involvement of DNA Methyl Transferase.
- Normal level of methylation required for the maintenance of differential expression.

REGULATION OF GENOMIC IMPRINTING:

- Epigenetic regulation of imprinting.
- Other regulatory elements include:
 - Non-coding RNAs
 - Differentially methylated regions
- Control imprinted genes in clusters.



Significance of Genomic Imprinting

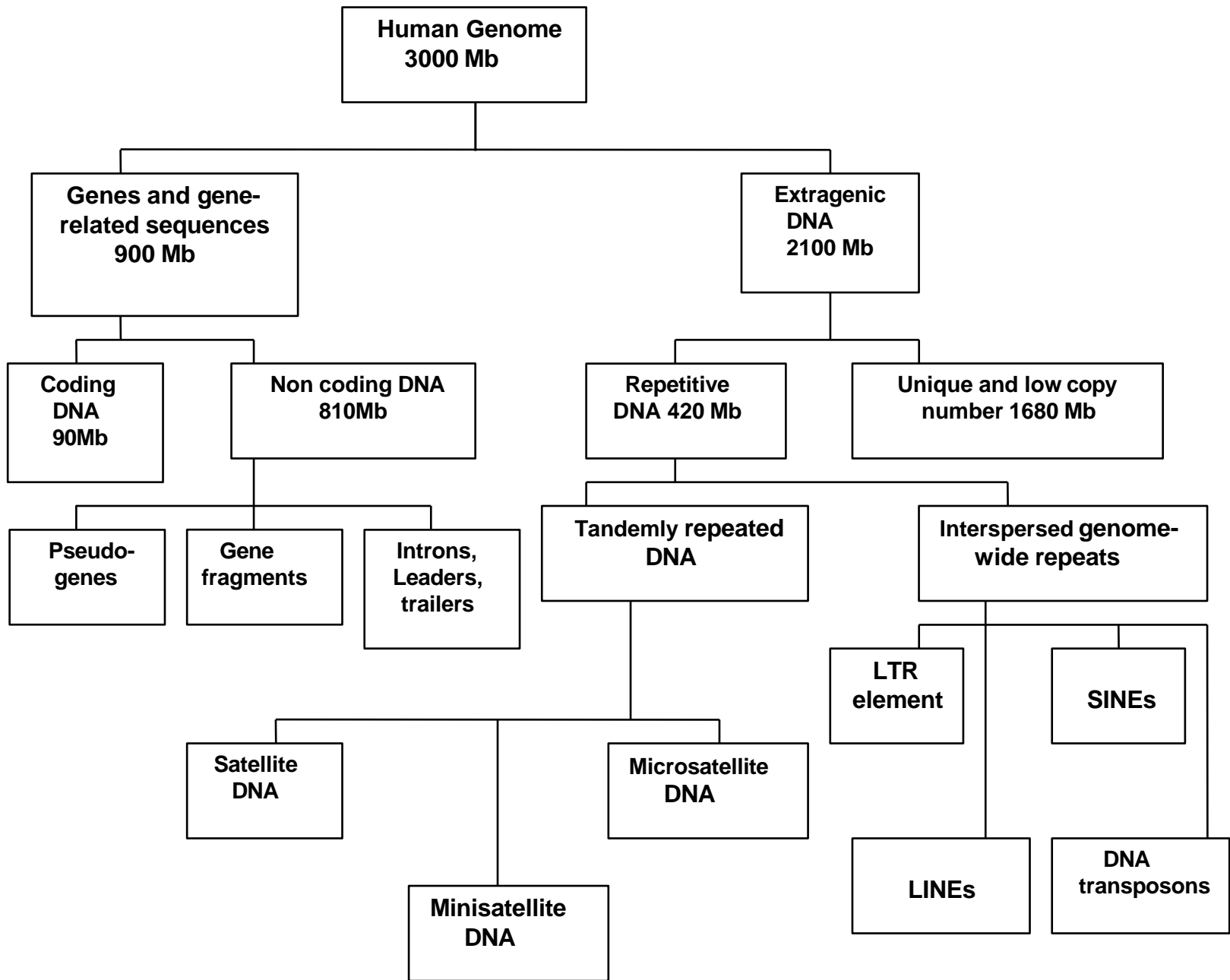
- Play major role in embryonic growth and development; post natal development; cell cycle proliferation and cognitive functions.
- Paternally expressed genes favour cell proliferation to increase growth of placenta.
- Can cause problems in cloning.
- Unbalanced gene expression insufficient to support healthy phenotype.
- Trnscription of wild type alleles can be silenced.
- Uniparental disomy occurrence.
- Aberrant imprinting causes clinical diseases.

Repetitive Sequences

- Certain base sequences are repeated many times and make up nearly 10% of the mammalian genome; such are termed as repetitive DNA.
- Main Features:
 - Short sequences in large clusters.
 - Rapid rate of renaturation.

Types:

- **Tandem repeats:** repeats lie adjacent to each other.
- **Interspersed repeat:** repeats are scattered through the genome.



Tandemly Repeated DNA

- Common feature in eukaryotes only.
- Made up of successive identical or related repeat units.
- Vary in length and also in length of the repeating unit.
- Arisen by expansion of progenitor sequence by Replication Slippage or DNA Recombination process.
- Also termed as satellite DNA as they form satellite bands (human genome – 3 satellite bands)
- Satellite bands – atypical density and GC content.
- Types:

Satellite DNA

Minisatellites and Microsatellites.

Tandemly Repeated DNA

Satellite DNA:

- Consists of large repetitive sequences made up of large repeat units. (Seq length - >100 kb; L.O.R - <5 - >200 bp)
- Arises due to unequal crossing over.
- This DNA is not transcribed or translated.
- Density is different due to methylation patterns.
- Location is determined by In-Situ Hybridisation.
- Found in the heterochromatic regions at or near centromere.
- Eg: Alphoid satellite DNA – Human satellite DNA with primary unit 171 bp.

Tandemly Repeated DNA

Minisatellites:

- Shorter tandem repeats (repeat length – 20 kb; L.O.R – upto 25 bp).
- Highly polymorphic in nature.
- Also called VNTRs – variable no. of tandem repeats.
- Variation in no. of repeats is due to recombination-like events.
- Enriched in sub telomeric regions of the chromosomes
- Eg : telomeres with TTAGGG repeating units.

Microsatellites:

- Shortest tandem repeats (repeat length – <10 kb; L.O.R – <= 4bp).
- Very common in the genome; highly polymorphic.
- Variation in repeat no. is a result of slippage occurring during replication.

Tandemly Repeated DNA

Significance:

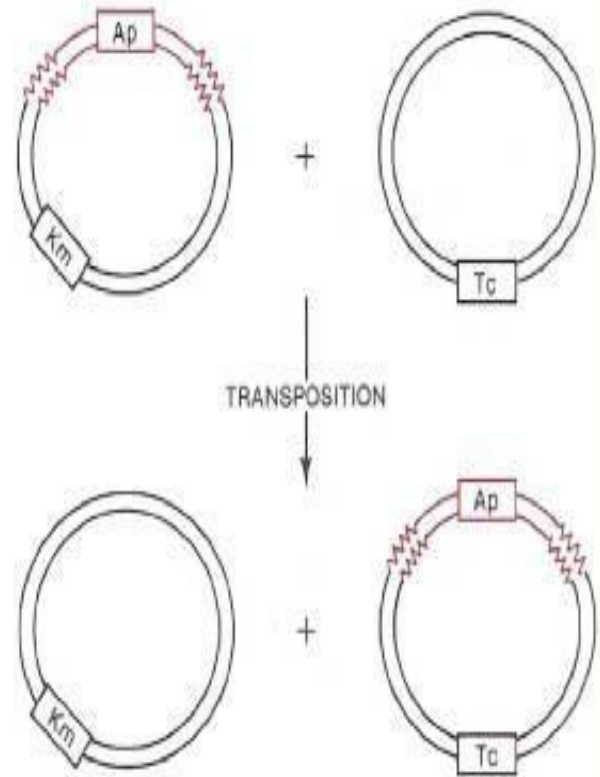
- Satellite DNA probably form the functional core of centromeres. As they are important for the kinetochore assembly during cell division.
- Minisatellites are hypothesized to have regulatory functions. Eg: VNTR in insulin promoter.
- Microsatellites are highly variable such that they serve as best tools for genetic profiling.
- Length of microsatellites – trinucleotide expansion diseases. Eg: Huntington's Chorea, Myotonic Dystrophy.

Interspersed Repeats

- Arises by the mechanism of transposition which results in a copy of repeat unit occurring in the genome at a distant position from original sequence.
- Generally referred to as transposons.
- Transposition is the mechanism by which transposons are inserted in a new position.
- Salient feature: no requirement of sequence similarity between the initial region containing transposons and final location.

Transposons:

- Called as “mobile genetic elements”, “mobile DNA” or as “jumping genes”.
- Discovered by Barbara McClintock .
- Based on the transposition mechanism, they are classified into class I (retrotransposons) and class II (DNA transposons) transposons.



Transposons

Retrotransposons:

- Most important transposable elements in the human genome making up nearly 45% of it.
- Transposition is brought about by copy-and-paste mechanism.
- Copy no. is duplicated.
- Requires an RNA intermediate.

Retrotransposons

Autonomous

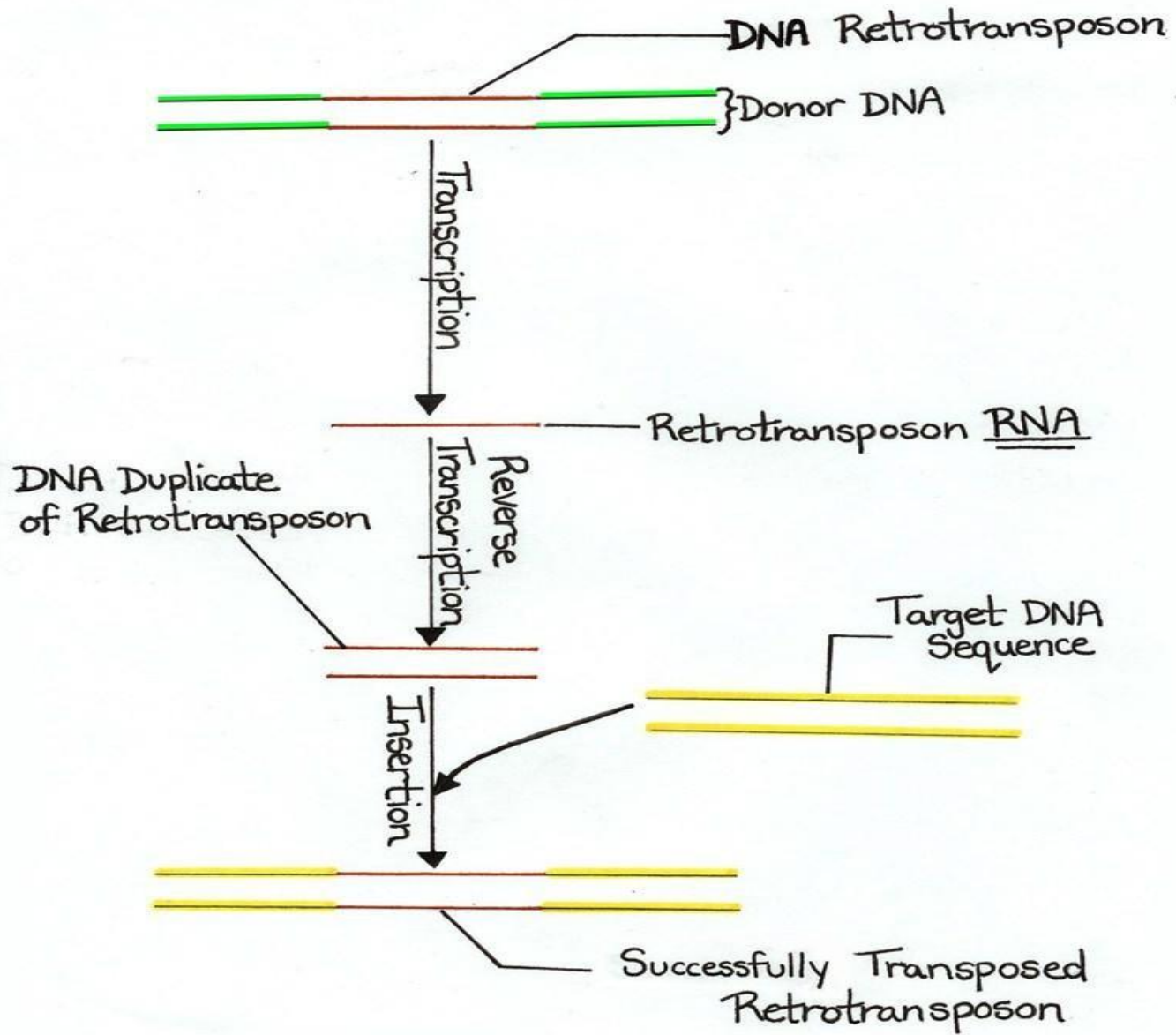
Code proteins for transposition

Eg: LINEs

Non- Autonomous

Does not code proteins

Eg: SINEs



Retrotransposons

LTR (Long Terminal Repeats) Elements:

- ✓ Retroviruses.
- ✓ Endogenous viruses.

Non – LTR elements:

- ✓ LINEs – Long Interspersed Nuclear Elements.
(make up 21% of the human genome)

Non-Autonomous retrotransposons:

- ✓ SINEs – Short Interspersed Nuclear Elements.
(formed mostly by Alu Elements)

LINES

Long interspersed elements

5,000 bps genes reverse
transcribed RNA
No introns promoter

SINES

Short interspersed elements

500 bps genes reverse
transcribed RNA
No introns promoter

Alu sequences
11% of the total DNA

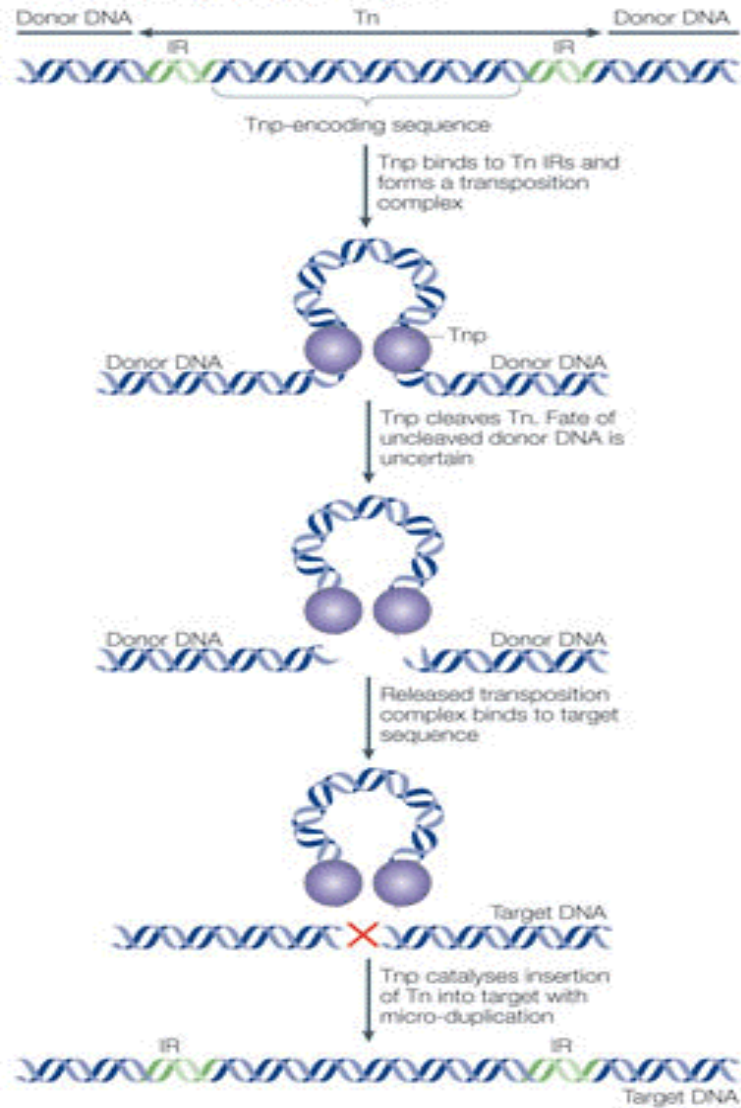
Transposons

DNA transposons:

- Generally regarded as inactive in human genome.
- Transposition requires no RNA intermediate.
- Transposition is brought about by cut-and-paste mechanism or replicative transposition.
- Copy number remains stable.

- Mechanism:
 - 1) The transposase enzyme binds to the ends of the element.
 - 2) Transposase then cuts the transposon out and ligates the resulting free chromosomal DNA ends.
 - 3) The free complex transposon-transposase makes a staggered cut at the target site producing sticky ends, and ligates the transposons into the target site.

a Cut-and-paste DNA transposition



Transposons

Significance:

- LTR and LINE elements can change gene expression.
- Retrotransposons lead to deletions and mutations.
- Transposons are speculated to have some real physiological functions, as their expression is upregulated during stress responses.
- Transposons are the widely used tools for mutagenesis in *D.melanogaster*.

DNA methylation & Transposons

- Methylation of transposons evolved roughly 500 million years ago.
- Timothy Bestor and his colleagues of Columbia University discovered that 90% of all methylated DNA sequences within the mammalian genome consist of transposable elements.
- Methylation had evolved as a very successful method of shutting down unnecessary or destructive effects of transposons.
- Methylation seen in human genome but not in Drosophila.
- There are 50% transposons in human genome but only 0.2% of mutation while there is only 10-20% transposons in Drosophila but nearly 85% mutation occurrence.