How RNA and DNA viruses help us understand oncogenes and tumor suppressors

Jeff Engler
Department of Biochemistry and Molecular Genetics
Office Telephone: 934-4734
Email: engler@uab.edu


Characteristics of Cancer Cells

- Cancer cells undergo unregulated growth
- Cancer cells become immortal (active growth when they should be quiescent)
- Cancer cells have increased nutrient uptake
- Cancer cells in tissue culture become anchorage independent.
- The cell cycle in cancer cells becomes active
  - Growth signaling pathways activated (oncogenes – RNA tumor viruses)
  - Pathways to prevent cell proliferation are disrupted (tumor suppressors – DNA tumor viruses)

Cancers develop in many steps due to many mutation events

Many different pathways can lead to tumor formation

Figure 29-41: Molecular Biology of the Cell, 6th Edition
Some types of cancer associated with tumor viruses

• Leukemias (derived from lymphoid cells)
• Carcinomas (derived from epithelial or endothelial cells)
• Sarcomas (derived from connective tissue cells)

Definitions:

Oncogene: “Gain of function”
An altered gene whose product can act in a dominant fashion to help make a cell cancerous. Usually, an oncogene is a mutant form of a normal gene (a “proto-oncogene”) involved in the control of cell growth or division.

Tumor Suppressor gene: “Loss of function”
A gene whose normal activity prevents formation of a cancer. Loss of this function by mutation enhance the likelihood that a cell can become cancerous (a normal process to maintain control of cell division is lost).

Oncogenes can increase tumor susceptibility in transgenic mice

Three lines of transgenic mice which over-express oncogenes in mammary or salivary glands.

Figure 23-36: Molecular Biology of the Cell, 4th Edition.
How do oncogenes and tumor suppressors work?

There are many pathways affected by oncogenes and tumor suppressor proteins

General overview of DNA and RNA transforming viruses

1. Role in natural oncogenesis
   - Rous sarcoma virus forms solid tumors in chickens

2. Potential for involvement in human tumors
   a) DNA Viruses
      i. HPV - cervical cancer
      ii. Herpes (Epstein Barr Virus (EBV)) - mononucleosis
         • Burkitts lymphoma, nasopharyngeal carcinoma
         • Immunologic defect allows cancer to occur
      iii. Hepatitis B Virus - Hepatocellular carcinoma
   b) RNA Viruses
      i. HTLV-1 - T-cell leukemia (Japan)
         • 1% of those people infected will develop cancers
      ii. Hepatitis C Virus - Hepatocellular carcinoma
Retrovirus life cycle requires integration into the chromosome.

Genomic Organization of Simple Retroviruses

Chronology of understanding oncogenes

1. ts mutants of RSV - single gene responsible
2. spontaneous loss of transforming ability and genetic information
3. generation of src specific probes - identification of similar sequences in uninfected cells
4. each acute transforming retrovirus possesses a unique oncogene
Oncogene-encoding viruses

Replication defective acute transforming viruses – need helper virus to grow
Mixtures of wild type and transforming viruses in culture or in infected animals

Chronology of understanding oncogenes

1. ts mutants of RSV - single gene responsible
2. spontaneous loss of transforming ability and genetic information
3. generation of src specific probes
4. each acute transforming retrovirus possesses a unique oncogene
5. each retroviral oncogene has a cellular proto-oncogene counterpart

Proto-oncogenes encode components of the growth factor signal transduction pathway

Oncogenes can activate these signaling pathways in one of 4 ways:
- Oncogenic growth factors
- Mutated adaptor proteins
- Mutated cell-surface receptors
- Mutated transcription factors
Four classes of oncogenes

Class One: oncogenes that mimic growth factors to induce cell proliferation
Rare – only two have been identified

Sis:
• from simian sarcoma virus – a secreted protein that mimics PDGF
• from PI-FeSV – a cat sarcoma virus

Proto-oncogenes mainly encode components of growth factor signal transduction pathways
Components shown in yellow are known proto-oncogenes

Class Two: Mutated Receptors
Oncogenes that result from mutations of cell-surface receptors, usually resulting in an overactive or constitutive protein-tyrosine kinase (PTK).
Examples:
- *fms* – from McDonough feline sarcoma virus – CSF-1 receptor
- *kit* – from HZ4 feline sarcoma virus – SCL receptor
- *erbB* – from avian erythroblastosis virus – epidermal growth factor (EGF) receptor
- *ros* – from UR2 avian sarcoma virus – related to insulin receptor
- *sea* – from S13 avian sarcoma virus – related to human growth factor (HGF) receptor
Genetic Changes convert a Proto-oncogene into an Oncogene

![Genetic Changes convert a Proto-oncogene into an Oncogene](image)

**neu**
Mutation (Val to Gln)

![Genetic Changes convert a Proto-oncogene into an Oncogene](image)

**erbB**
Delete extracellular domain

Class Three: Intracellular transducers
4 types of oncogene transducers

- **Protein-tyrosine kinases**
  - add a phosphate to specific tyrosine amino acids
- **Protein-serine/threonine kinases**
  - add a phosphate to specific serine or threonine amino acids
- **G-protein (Ras) proteins**
  - Trimeric GTPases that bind GTP to become active as signal transducers
- **Phospholipase C (PKC)**
  - Activated by certain G-proteins to trigger inositol phospholipid signaling pathway

Proto-oncogenes mainly encode components of growth factor signal transduction pathways

![Proto-oncogenes mainly encode components of growth factor signal transduction pathways](image)

Mutated signal transducer molecules send incorrect “on” signals

Mutated transcription factors turn on genes at inappropriate times
Class Four: transcription factor oncogenes

Examples:
- Jun
- Fos
- Myc (many examples in chicken, cat leukemia viruses)
- Myb (chicken myeloblastosis virus)
- Rel (NF-κB family - turkey reticuloendotheliosis virus)
- Maf (interact with Fos and Jun – chicken sarcoma)
- erbA (thyroid hormone receptor – from chicken erythroblastosis virus)

The same oncogenes can be found in more than one virus isolate

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Oncogenesis by virus insertion

Most retroviruses cannot transform cells.
Non acute (chronic) transforming viruses don’t carry oncogenes – can’t transform cells in culture.
Non acute transforming viruses are still capable of replication.
Non acute transforming viruses can cause tumors in animals but over a 1-2 year time frame.
These viruses can transform cells by insertional mutagenesis:
1. Avian leukemia virus – insert near myc
2. Mouse mammary tumor virus - int-1, int-2
How do tumors (and viruses) overproduce oncogene proteins?

- Deletion or point mutation in coding sequence
- Gene amplification
- Chromosome rearrangement

Hyperactive protein
Protein overproduced

Retrovirus life cycle requires integration into the chromosome

Fig. 1 from Trends in Mol. Medicine 2:43-45 (2003)

Insertional activation of proto-oncogenes

- Viral enhancer activation
- Viral promoter insertion
- Post-transcriptional deregulation
- Insertional inactivation or gene truncation

Viral enhancer acts on a nearby gene (dominant)
Viral promoter transcribes a nearby oncogene (dominant)
Altered transcription, processing, or stability (dominant)
Inactivate a gene (recessive mutation)

Fig. 2 from Trends in Mol. Medicine 2:43-45 (2003)
Some DNA tumor viruses block tumor suppressor pathways

Examples of DNA tumor viruses
- Human Adenoviruses – all serotypes transform cells in vitro, but only a few can cause tumors in rats
- Human Papillomaviruses (HPV) – high risk types associated with cervical cancer
- Papovaviruses
  - Simian Virus 40 (SV40)
  - JC virus
  - BK virus

Many DNA tumor viruses encode proteins that bind to and sequester Rb

Retinoblastoma
- A rare form of ocular tumor
- Occurs in childhood
- Tumors develop from neural precursor cells in the immature retina
- About one child in 20,000 is afflicted.
- Two forms of the disease:
  - hereditary (multiple tumors affecting both eyes) - germline mutation in one copy of gene predisposes individual to retinoblastoma
  - non-hereditary (single tumor in one eye)
- deletion in chromosome 13 (recessive mutation)
Retinoblastoma gene (Rb)

- The Rb gene encodes an anti-proliferation protein (tumor suppressor).
- Mutations in gene implicated in breast and small cell lung cancers.
- Regulates transcription of genes involved in growth control through transcription factor E2F
- Introduction of cloned Rb gene into retinoblastoma and osteosarcoma cells suppresses neoplastic phenotype.
- Evidence that transforming proteins of DNA viruses bind to and inactivate RB protein

How do oncogenes and tumor suppressors work?

Rb shuts off cell proliferation by binding to E2F (a transcription factor)
Viral proteins sequester tumor suppressors to promote cell proliferation

RNA tumor viruses carrying the *myc* oncogene can also disrupt Rb indirectly

- The *myc* oncogene is a transcription factor that can regulate many genes involved in cell proliferation.
- The *myc* oncogene has been identified in retroviruses from both chickens and cats and can cause lymphomas or other carcinomas.
- The *myc* oncogene disrupts the Rb binding to E2F by overexpressing proteins that control Rb phosphorylation (an indirect effect).

Myc acts as a transcription factor to activate cell proliferation genes

A dominant mutation (an oncogene)
Summary

- RNA and DNA tumor viruses have helped define oncogenes and tumor suppressors.
- RNA tumor viruses generally exert their effects through growth signaling pathways, turning them on in the absence of growth stimuli.
  - “add gasoline to the system”
- DNA tumor viruses generally act by sequestering proteins that control cell proliferation (Rb, p53), to shift the cells into S phase.
  - “release the brakes”