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# Cancer Biology – How a cell responds to DNA Damage

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## EDUCATIONAL GOALS

- **How proteins can transmit signals to each other.**
- **The definition of a tumor suppressor gene and an oncogene.**
- **How networks of proteins can function together in proliferation and damage control systems.**
- **Why disruption of these systems contributes to cancer formation.**
- **How these disrupted systems can be targeted for cancer therapy.**

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## Outline

- Organization and systems within cells
- Cancer results from disruption of key systems
- Damage response system
- Exploiting disrupted systems for cancer therapy

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## Cells are complex machines

- Efficient energy use
- Adapt to environment
- Self-repair
- Cell division / proliferation

Pictures of pentium 4, race car and cell electron micrograph

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## Cellular Building Blocks

- DNA - master blueprint
- RNA - working copies of blueprint
- Proteins - physical structure
  - structural proteins
  - enzymatic proteins
  - signaling proteins
- Lipids - key component of membranes

DNA is duplicated by a process of 'replication'.
RNA is created by 'transcription'.
Proteins are created by 'translation'.

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## Cells composed of organelles

- nucleus
- cytoskeleton
- mitochondria
- endoplasmic reticulum
- golgi apparatus

Electron micrograph of cell organelles

The nucleus contains genetic material. Mitochondria generate energy. Endoplasmic reticulum are involved in protein production and the golgi apparatus in processing protiens. There are several other organelles that are not listed.

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## Cells have biochemical systems

- Energy production
- DNA replication
- RNA transcription
- Protein translation
- Cytoskeleton
- Signaling systems
- Damage response

Diagram of Kreb's cycle

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## Hallmarks of cancer

- Uncontrolled cell proliferation
- Unlimited replication
- Sustained angiogenesis
- Evasion of apoptosis
- Invasion

Photo of lung cancer

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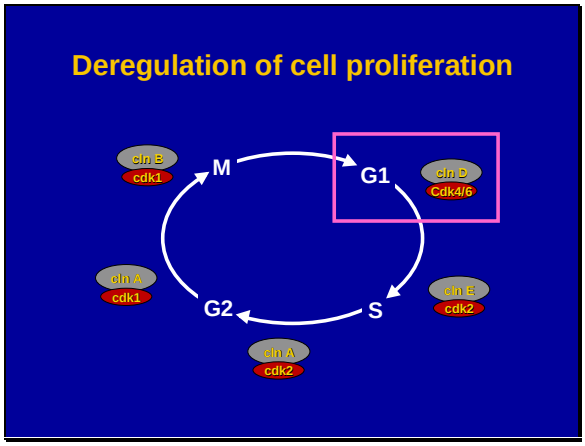


The cell cycle describes the phases that a cell passes through in order to divide. There are 4 phases:
G1 = Gap 1 - primarily involved in cell growth in preparation for division.
S = synthesis of DNA - the entire genome must be duplicated during this time.
G2 = Gap 2 - pause before mitosis
M = mitosis - segregation of duplicated DNA into two separate daughter cells and division of the cell contents.

At the core of the cell cycle, proteins called kinases drive progression into the next phase. These kinases are tightly regulated.

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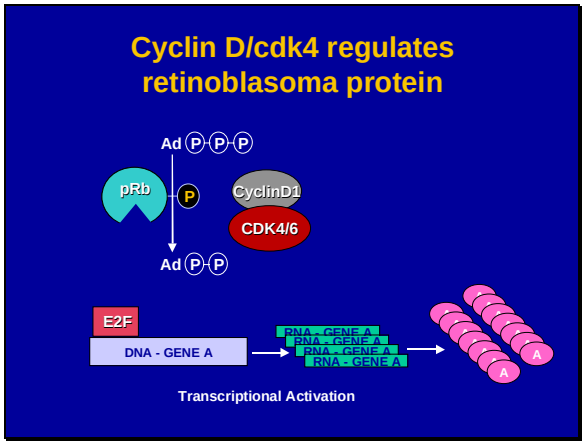


CyclinD1/CDK4 is a kinase complex that catalyzes the addition of phosphate to the retinoblastoma protein (Rb). When Rb is not phosphorylated, it binds to the E2F protein and prevents it from working. After phosphorylation, E2F is released and can then bind to DNA and drive the production of select RNAs that code for specific proteins needed for progression into S-phase.

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## Tumor suppressors and oncogenes

- Tumor suppressor
  - protein product of gene inhibits the development of cancer.
  - Need to lose both copies of the gene to have an effect.
- Oncogene
  - Deregulation or mutation of protein / gene promotes the development of cancer.
  - Need to alter only one copy of gene to have an effect.

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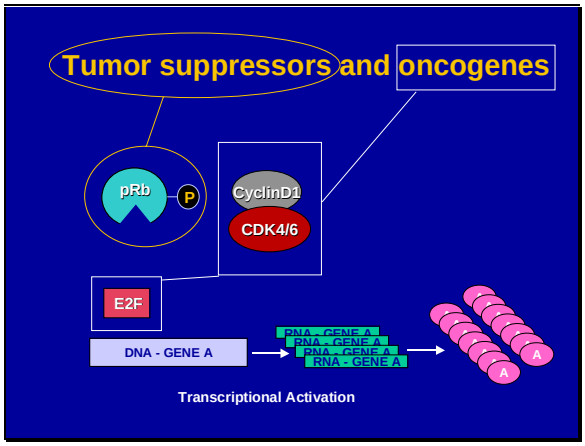


Retinoblastoma is a tumor suppressor
Cyclin D, Cdk4, and E2F are oncogenes

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## Summary

- Cell cycle is a biochemical system that drives proliferation.
- Protein function regulated by:
  - complex formation
  - phosphorylation
- Proteins regulate RNA transcription.
- A tumor suppressor prevents tumor formation.
- An oncogene promotes tumor formation

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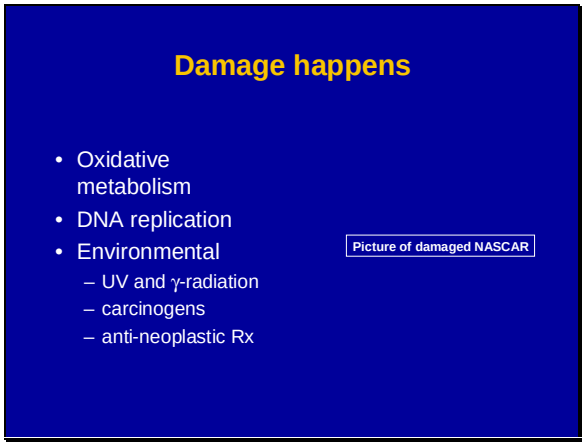


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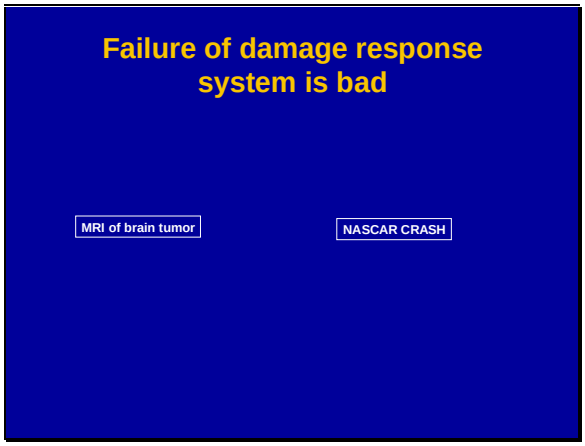


The DNA damage response system is critical for the prevention of cancer and is often disrupted as one of the first steps in developing cancer.

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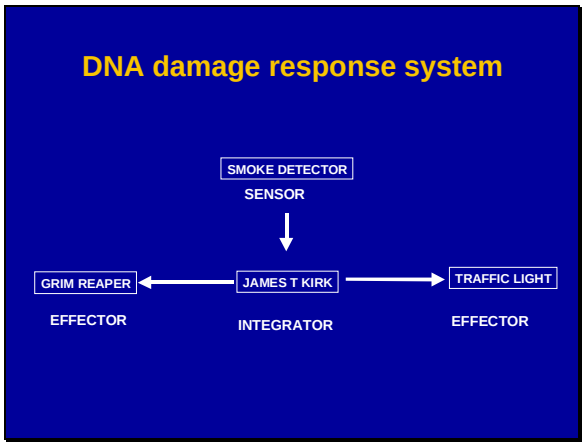


The DNA damage response system is composed of 3 classes of proteins. Sensors primary role is to recognize damage and recruit help from integrators and effectors. Integrators recognize damage or signals from sensors and 'decide' what the most effective response would be. They can signal to one or more effectors depending on the context of the damage. Effectors execute the commands from the integrators.

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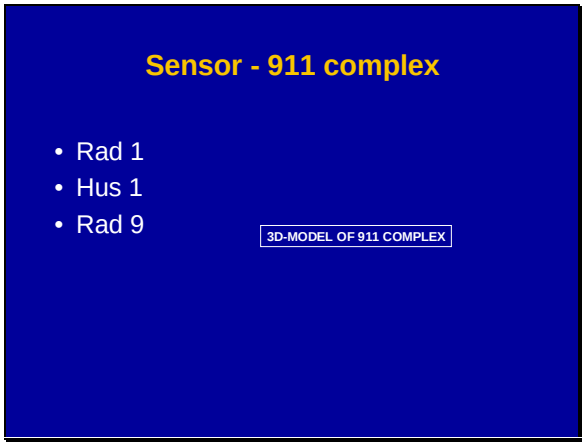


These three proteins bind together and localize to sites of DNA damage after radiation.

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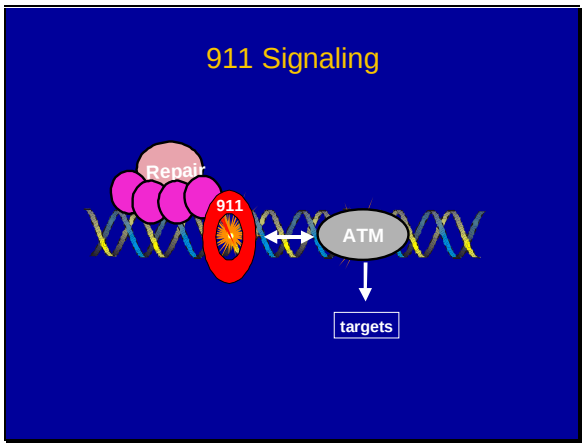


The 911 complex recruits repair complexes to sites of damage and may interact with integrators like ATM or related proteins.

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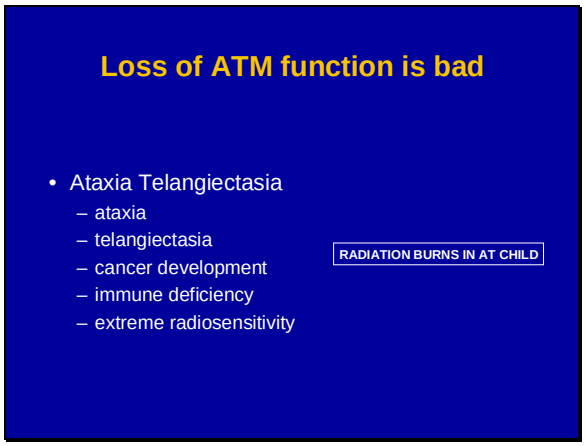


Lack of ATM function is an inherited disease known as ataxia telangiectasia. It is present from birth and is usually diagnosed when children begin to walk because of ataxia (drunken walk). These children are extremely radiosensitive.

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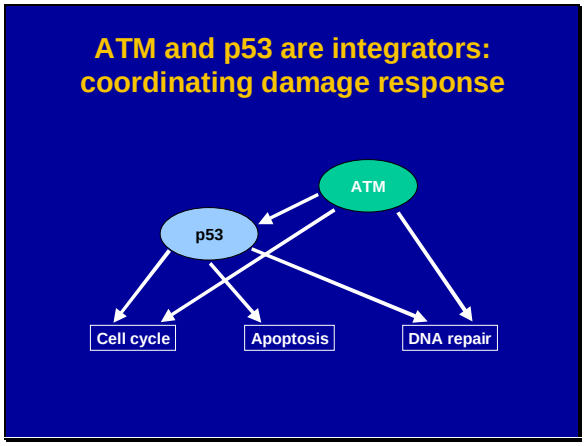


ATM functions as an integrator and signals to the cell cycle and DNA repair machinery. It also signals to another integrator p53. P53 is one of the most commonly mutated tumor suppressors. P53 also controls apoptosis, which is the cellular equivalent of suicide.

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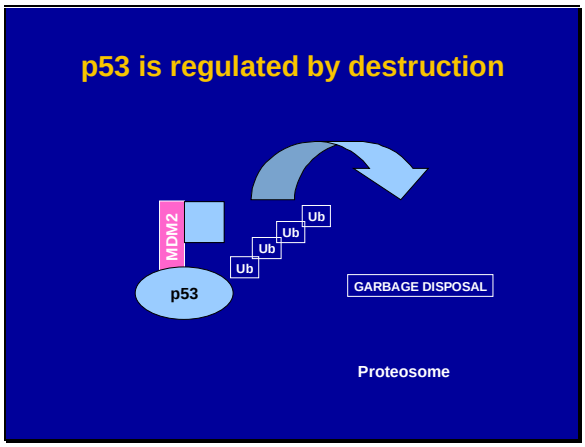


P53 is normally at very low levels in the cell. This is achieved by the protein MDM2, which places a ubiquitin (Ub) tag on proteins. This tag targets p53 for the proteosome, which is the cellular equivalent to a garbage disposal. Thus, p53 is found at low levels in the cell because it is continually destroyed.

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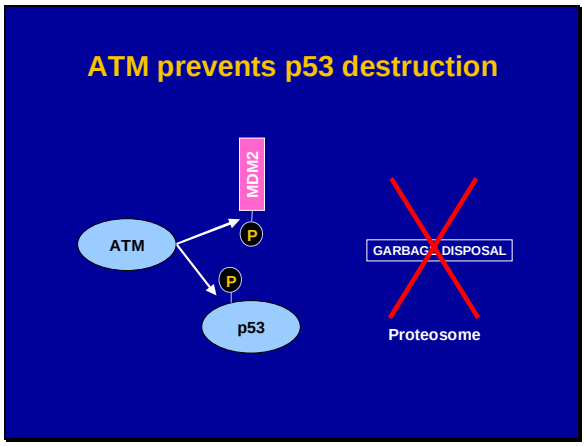


Follwing damage, ATM phosphorylates both MDM2 and p53. This prevents MDM2 from associating with p53 and prevents p53 from being destroyed.

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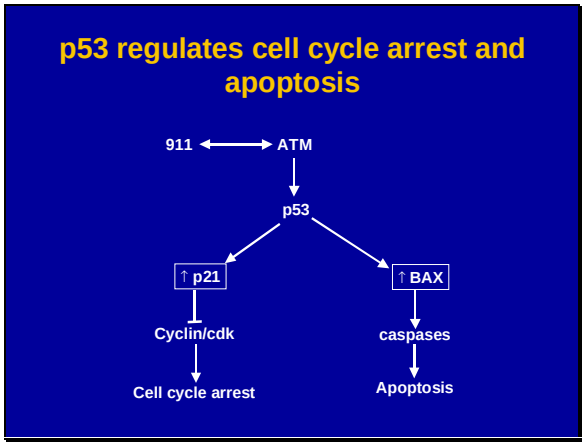


Following damage, p53 accumulates in the cell and can drive either cell cycle arrest or apoptosis (suicide) depending on cell type and extent of damage.

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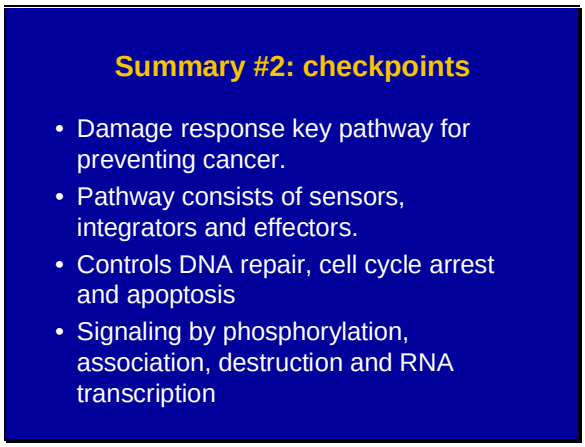


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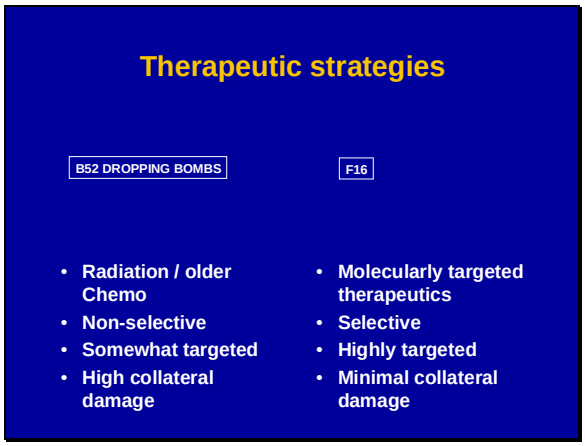

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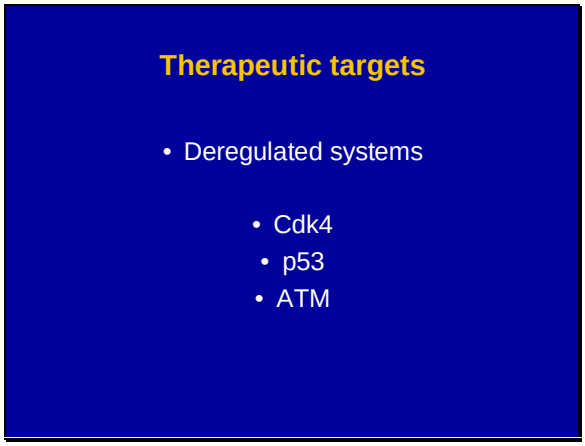


Novel therapeutic targets are aimed at systems in cancer cells that are deregulated.

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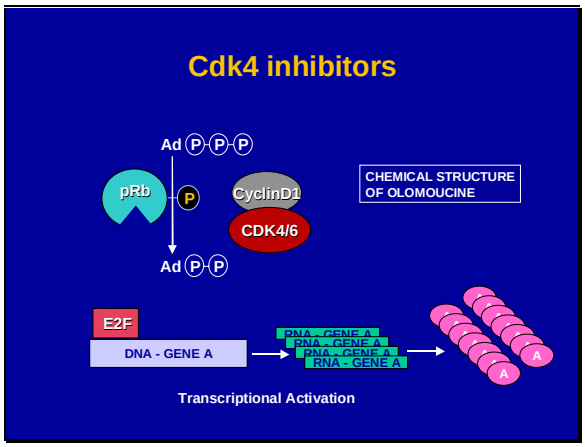


cyclinD/cdk4 are often amplified in tumors. Therefore, a drug that inhibits cdk4 function may have more profound effects on cancer cells than normal cells.

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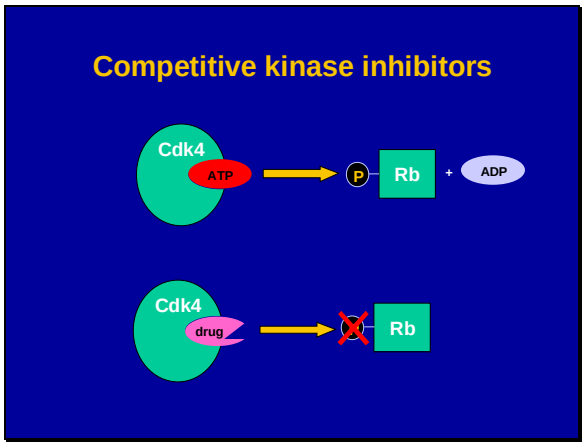


Cdk4 is a kinase that uses ATP as a phosphate source. Cdk4 has a pocket where ATP must bind for the protein to be active. Drugs can be identified that bind in the same pocket and prevent ATP from binding. This would inhibit Cdk4 activity.

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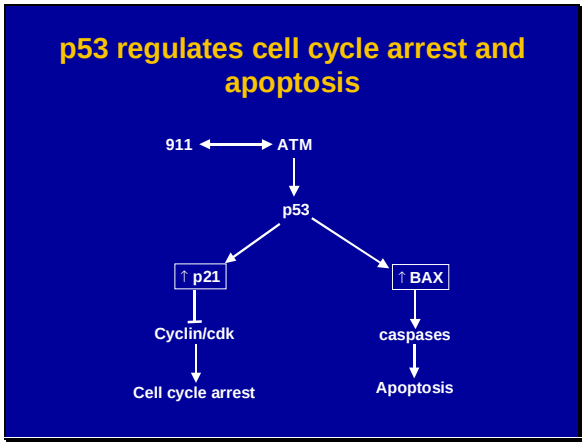


P53 is commonly mutated and so strategies that either restore p53 function or kill cells that lack p53 function could be useful for treating cancers.

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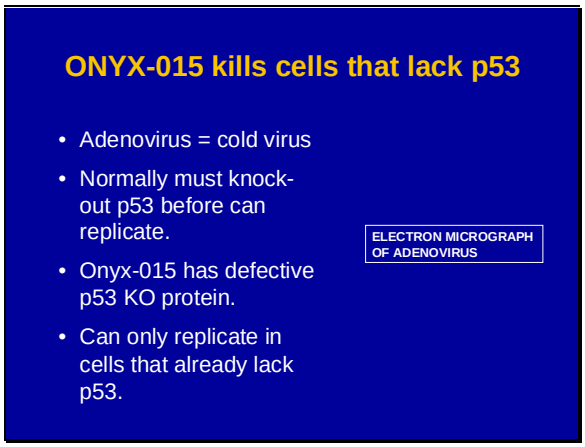


Cold viruses normally must knockout p53 function before they can take-over and ultimately kill a cell. By genetic engineering, the protein responsible for knocking out p53 function has been modified in the ONYX-015 virus so it can only replicate and kill cells that already lack p53 function. Therefore, normal cells will not be affected, while tumor cells without p53 function will be killed.

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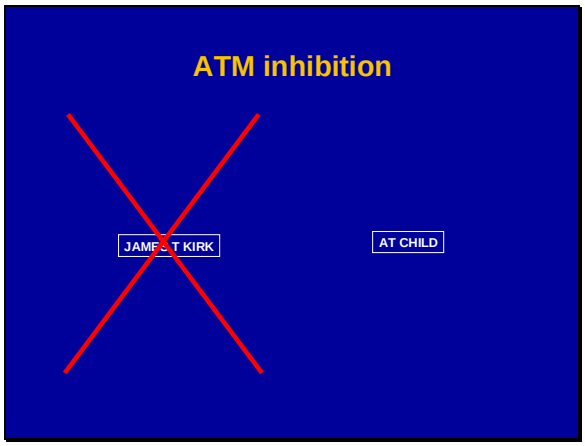


AT kids are very radiosensitive because they lack normal ATM function. This suggests that a drug that inhibits ATM activity might sensitize tumors to radiation. However, it would be important to only sensitize tumor cells and not normal tissues.

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## Caffeine inhibits ATM

- High levels of caffeine inhibit ATM activity.
- Caffeine selectively sensitizes cells to radiation that have lost p53 function.
- Selective ATM inhibitors are being identified.

CAN OF COKE

Caffeine is a prototype of an ATM inhibitor. Interestingly, caffeine only radio-sensitizes tumor cells that have lost p53 function.

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## Summary #3: Targets

- Selectively target deregulated systems.
- Small molecules disrupt protein function.
- Viral therapy can destroy cells.
- Combine targeted therapies with with radiation or chemotherapy.

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## GOALS

- Protein signaling
  - add/remove phosphate
  - bind to other proteins
  - destruction
  - drive RNA production
- Tumor suppressor
  - suppresses cancer - both copies lost
- Oncogene
  - promotes cancer - one copy activated

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## GOALS (2)

- DNA damage system
  - sensor
  - integrator
  - effector
- Disruption of damage systems
  - prevents cells from repairing damage leads to cancer
- Novel therapeutics
  - exploit differences between normal and tumor systems.