



**Mouse Retroviruses and Solving Mysteries
Surrounding Human Chronic Myelogenous Leukemia**

**Naomi Rosenberg, PhD
Tufts University School of Medicine
Boston, MA**

Overview of Today's Presentation

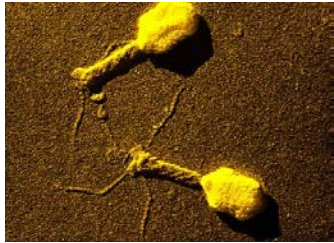
- Why Viruses are Important (& Really Neat)
- Retroviruses Basics
- Abelson Virus & Lessons Learned

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Viruses are Common

They are everywhere & there are a lot of them



Bacterial virus biomass exceeds that of all elephants on the planet!



Viruses infect all living things

They enter through our lungs, our digestive tract, eyes, micro-breaks in epithelium

Our bodies harbor many types of viruses

human & animal viruses

bacterial viruses

plant viruses

Viruses are Important Causes of Disease



They have caused important human diseases for centuries



Animals & plants can also be devastated



GOT A VIRUS?

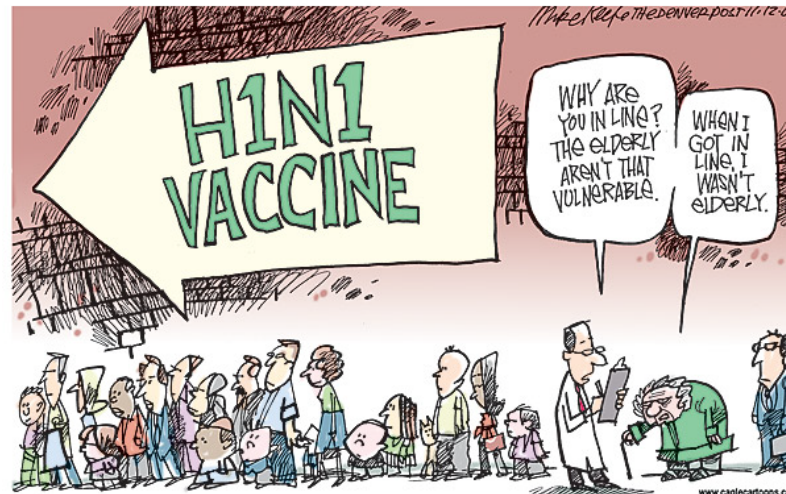


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Controlling Virus Diseases is Difficult

Solutions tend to be

- rare
- costly
- sometimes ineffective



Viruses Continue to be Important in Driving Molecular Biology Discovery

Study of viruses helped us discover

- mRNA
- Splicing
- Enhancers and Repressors
- Mechanisms of DNA Replication
- Mechanisms of Translational Control
- miRNA
- Mechanisms of Macromolecular Assembly

Viruses can be used to change and manipulate biology

- Virus vectors speed molecular discovery
- Gene therapy

Fundamental Properties of Viruses

Viruses are intracellular obligate parasites

Viruses carry their genetic information as nucleic acid

- Either DNA or RNA – not both

Viruses orchestrate synthesis of viral components using host cell machinery

Progeny viruses form by self-assembly of newly synthesized components inside cells

Progeny viruses disseminate to new cells to continue replication

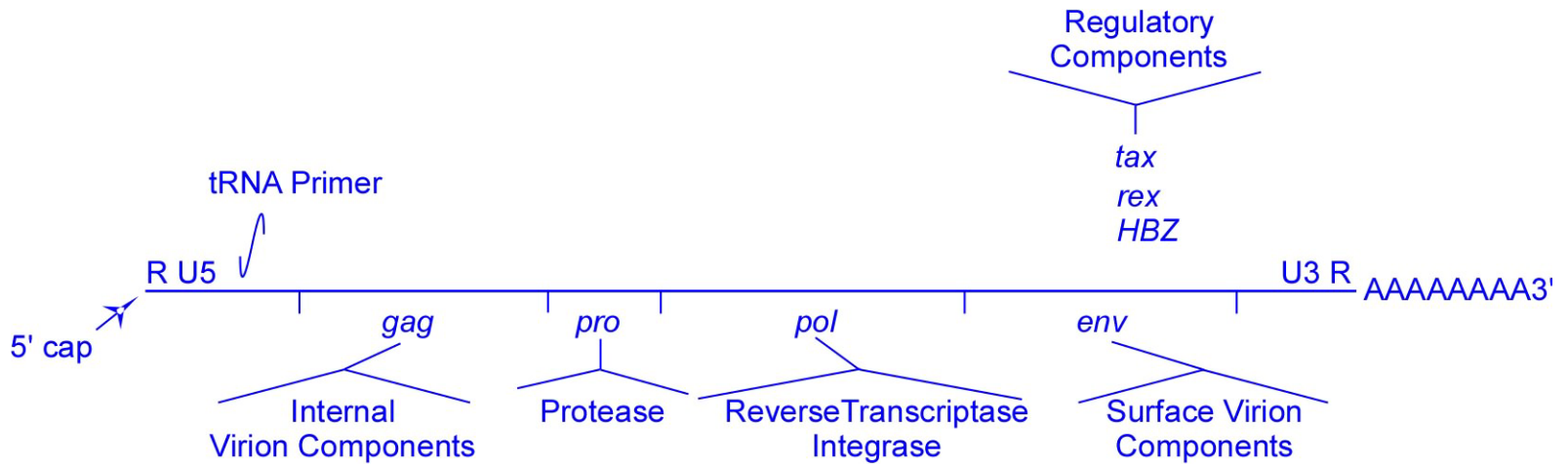
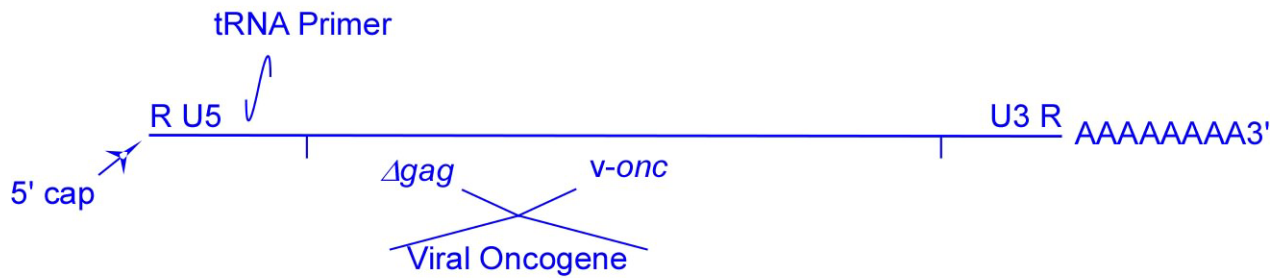
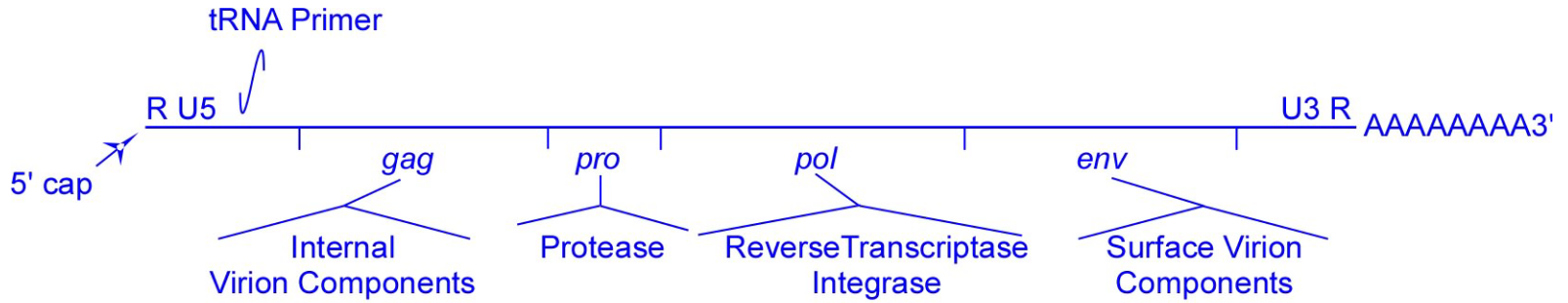
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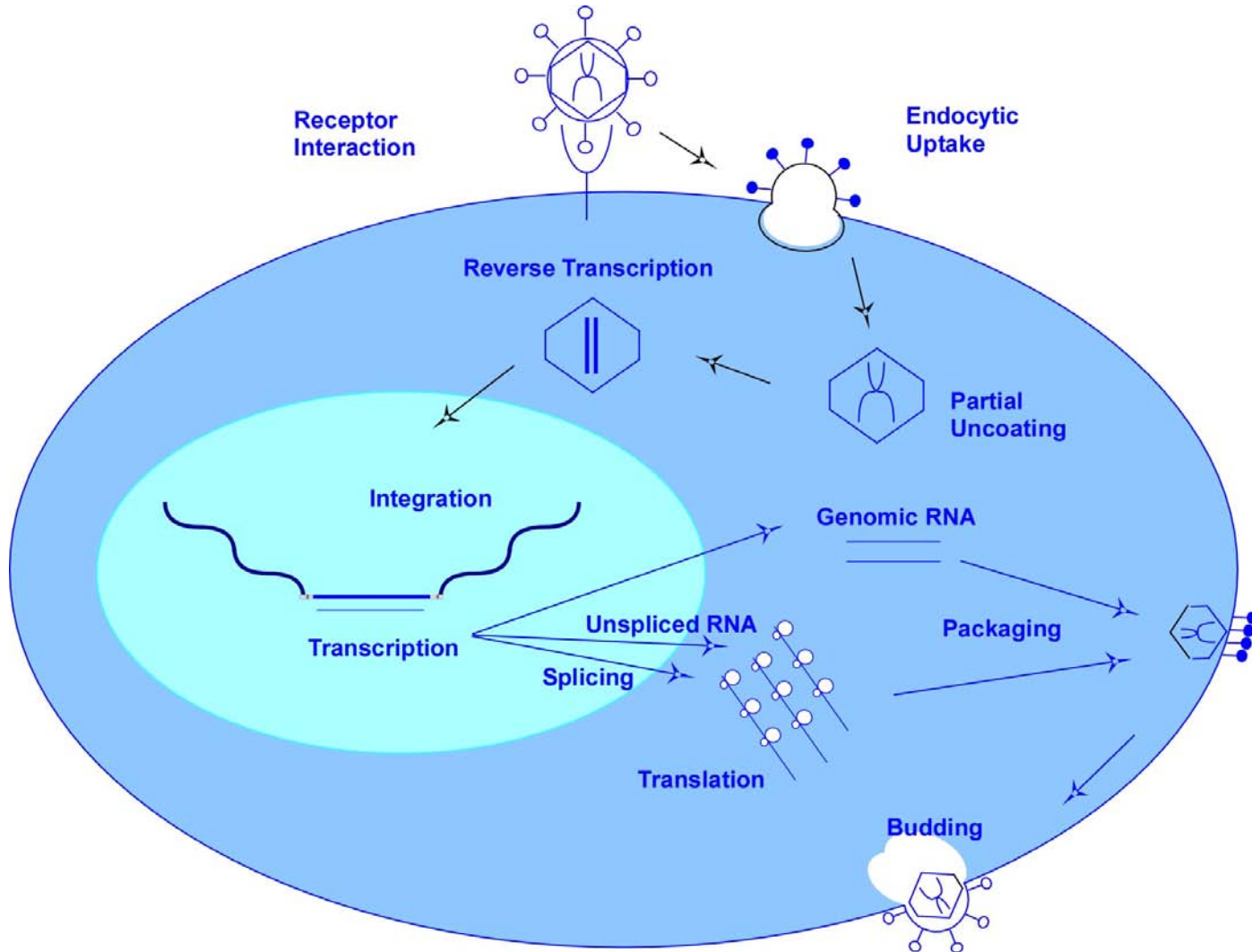
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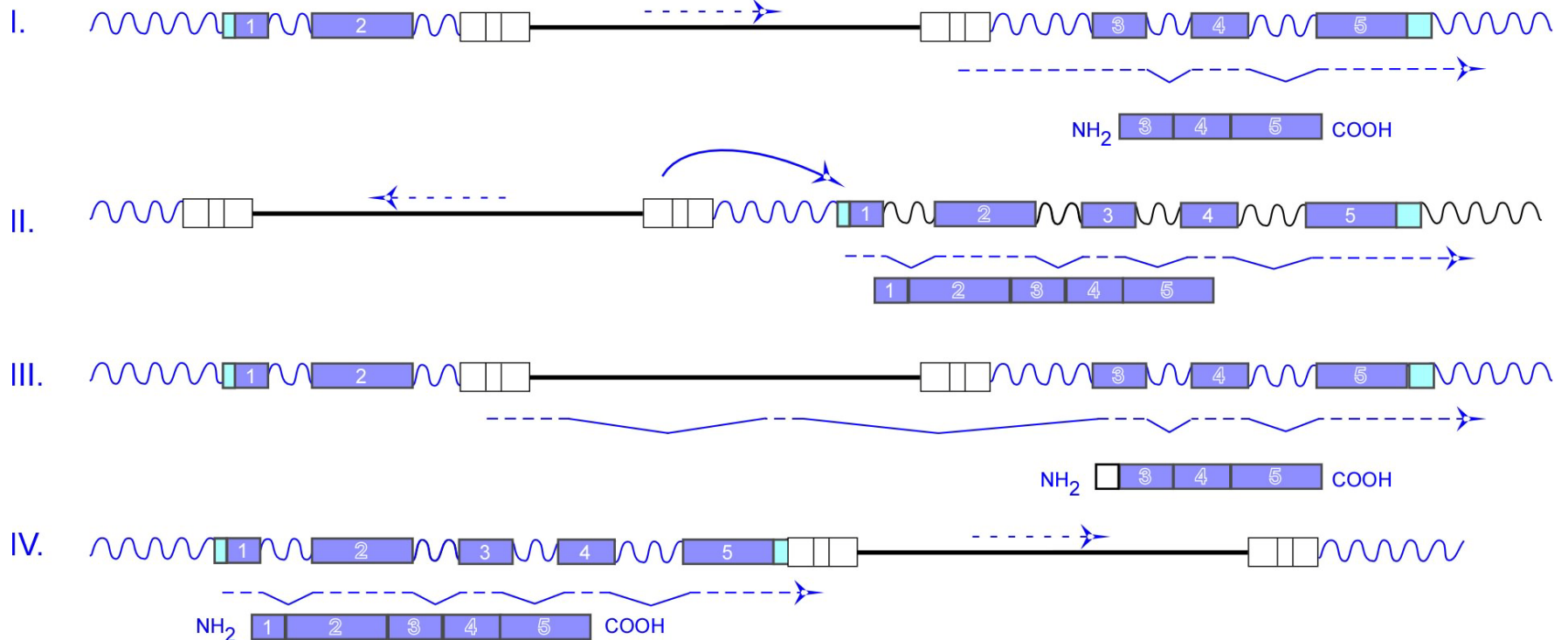
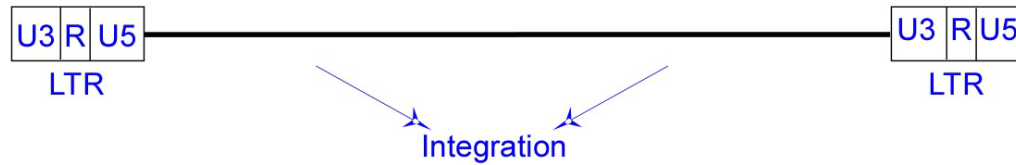
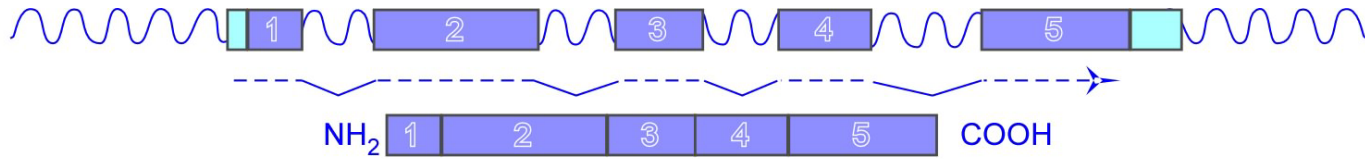
Genome Structure of Oncogenic Retroviruses



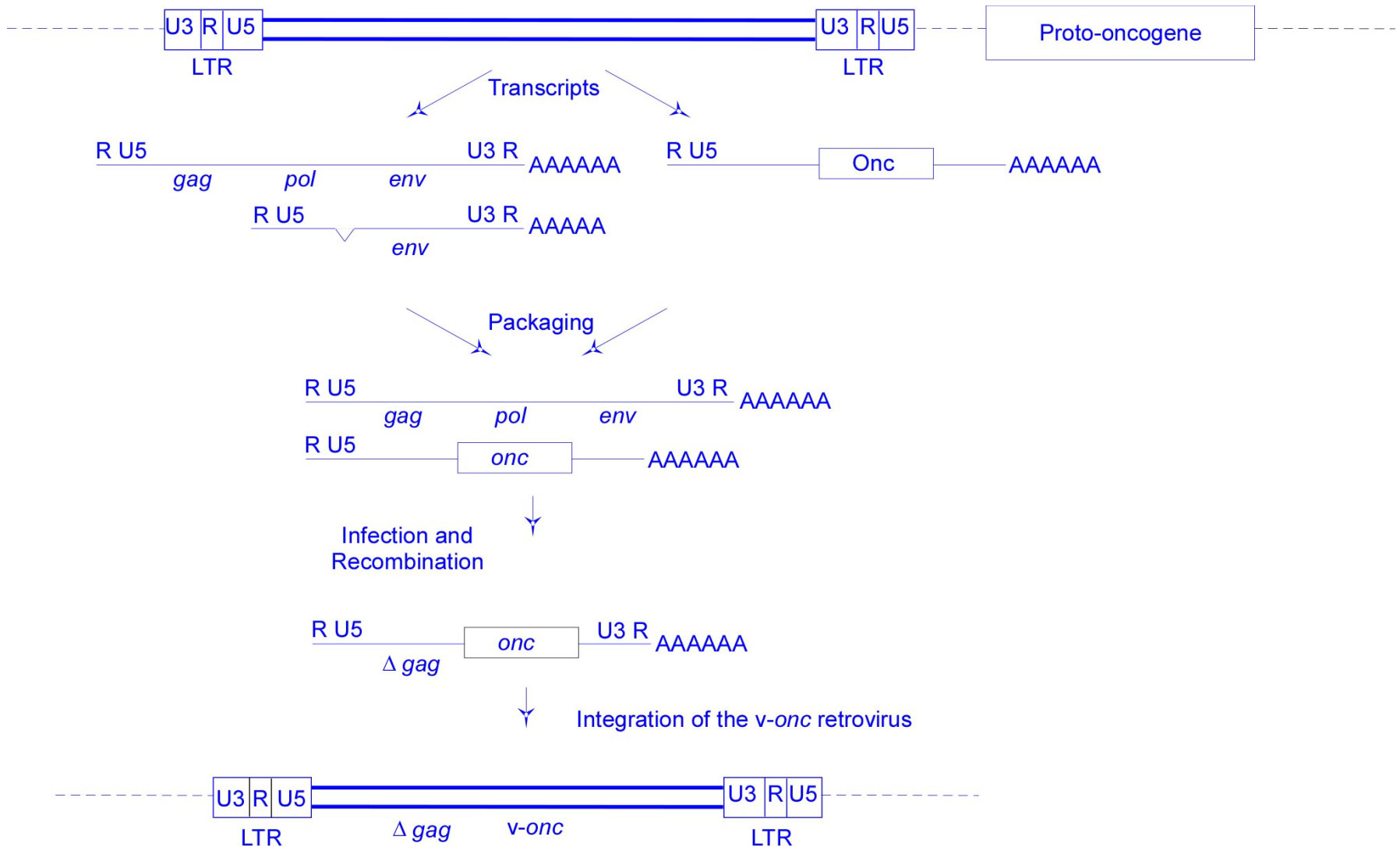
The Life Cycle Involves Reverse Transcription and Integration into the Host Genome



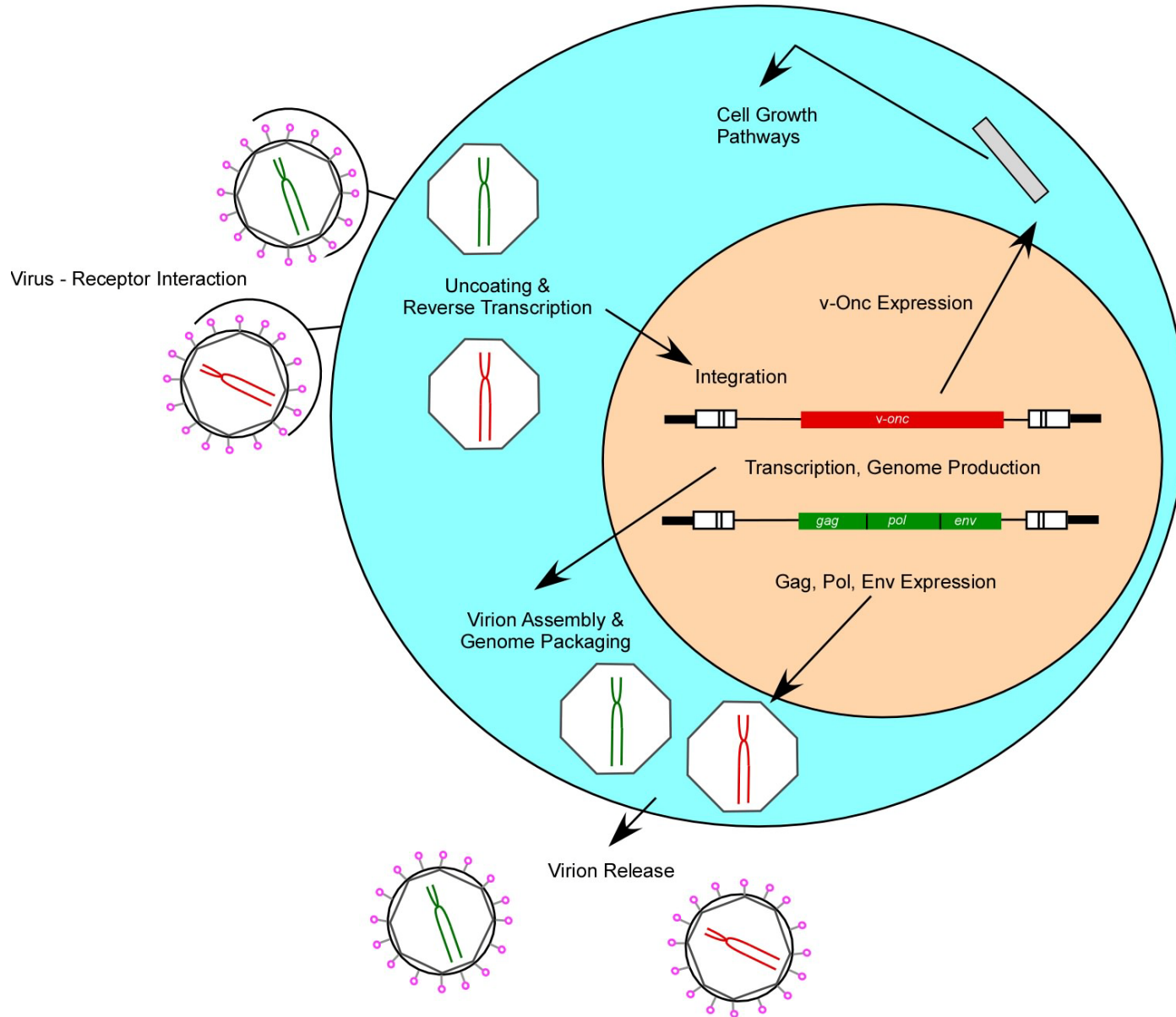
Integration is an Insertional Mutagenesis Event and Can Lead to Cancer Development



Integration Can Lead to Oncogene Capture



“Helper” Viruses Enable Replication of Most v-onc Gene Containing Retroviruses

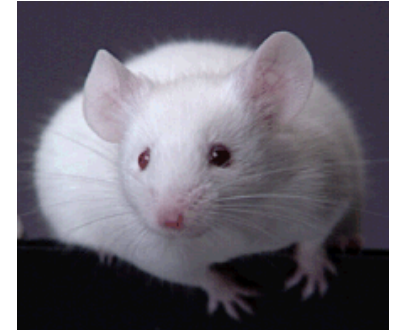


Where Do v-*onc* Gene Containing Retroviruses Come From?



Approximately 100 independent oncogenic isolates have been identified in animals.

In a natural setting, v-*onc* gene containing retroviruses are not transmitted horizontally.



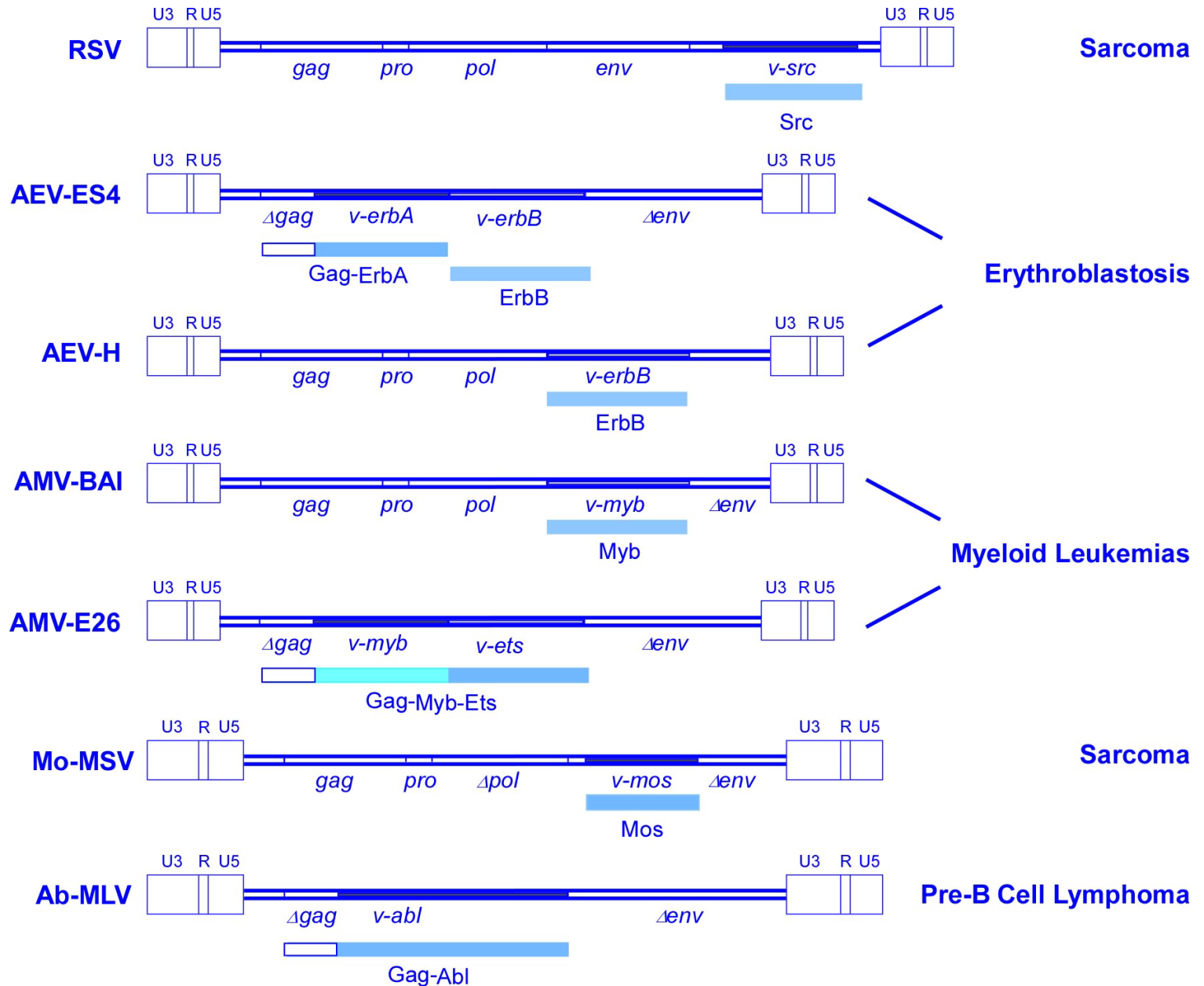
Study of these viruses has established key principles of oncogenesis.



Key signaling pathways identified by studying these viruses function in human tumors.



Oncogenes Influence the Type of Disease

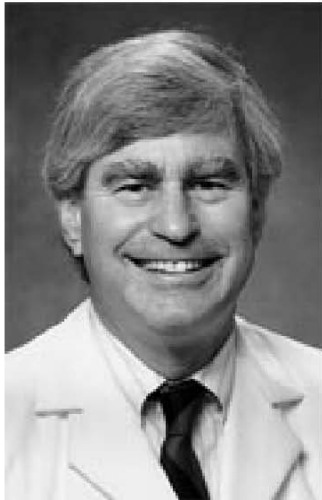


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Abelson Virus – Another Novel Leukemia Virus

What Have We Learned?



1969

Herbert Abelson, MD, discovers Abelson virus



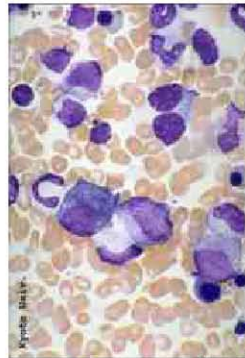
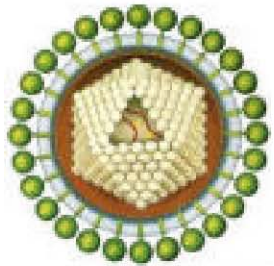
All infected mice develop leukemia quickly

Abelson virus is required for the leukemia

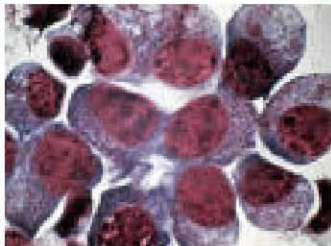
The cancer has a distinct symptoms

In Vitro Transformation with a Mouse Leukemia Virus

1973



Mouse Bone Marrow



Infected Leukemia Cells



Leukemic Mouse

Abelson Projects

Transformation of hematopoietic cells \pm Ab virus

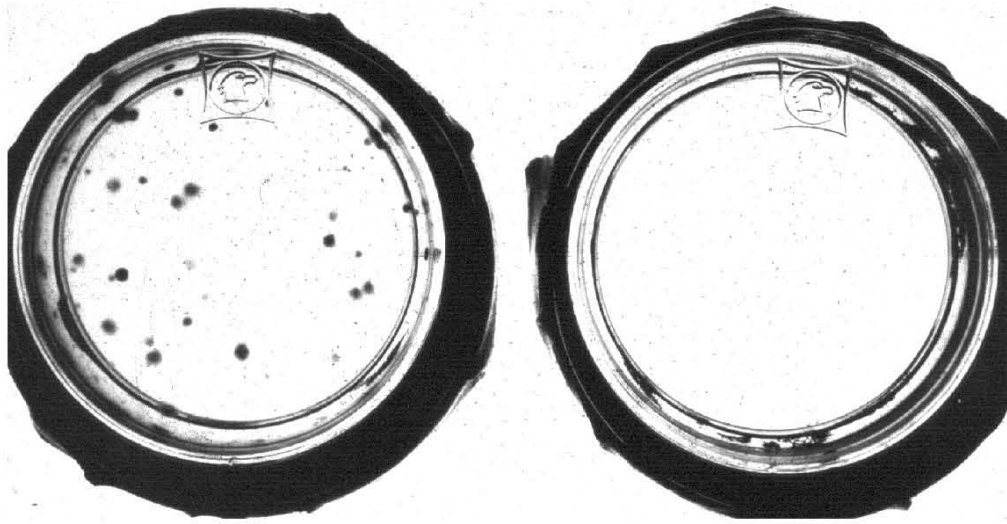
A. Tissue sources

1. bone marrow
 - a. skull
2. lymph node
3. spleen
4. fetal liver

B. Technical Aspects

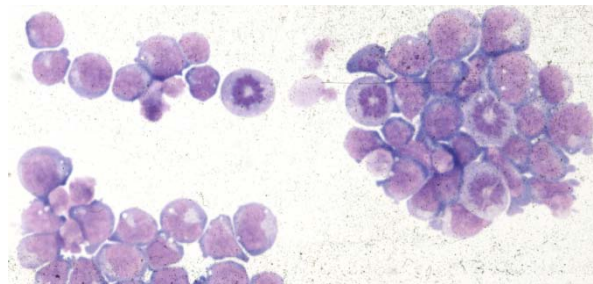
1. removal
2. mode of culture
 - a. outgrowth or maintenance
 - b. method of passage
 - c. suspensions
3. Recognition of cell types
 - a. morphology
 - b. staining for abs.
 - c. animal inoculation.

Transformation Works!



Infected

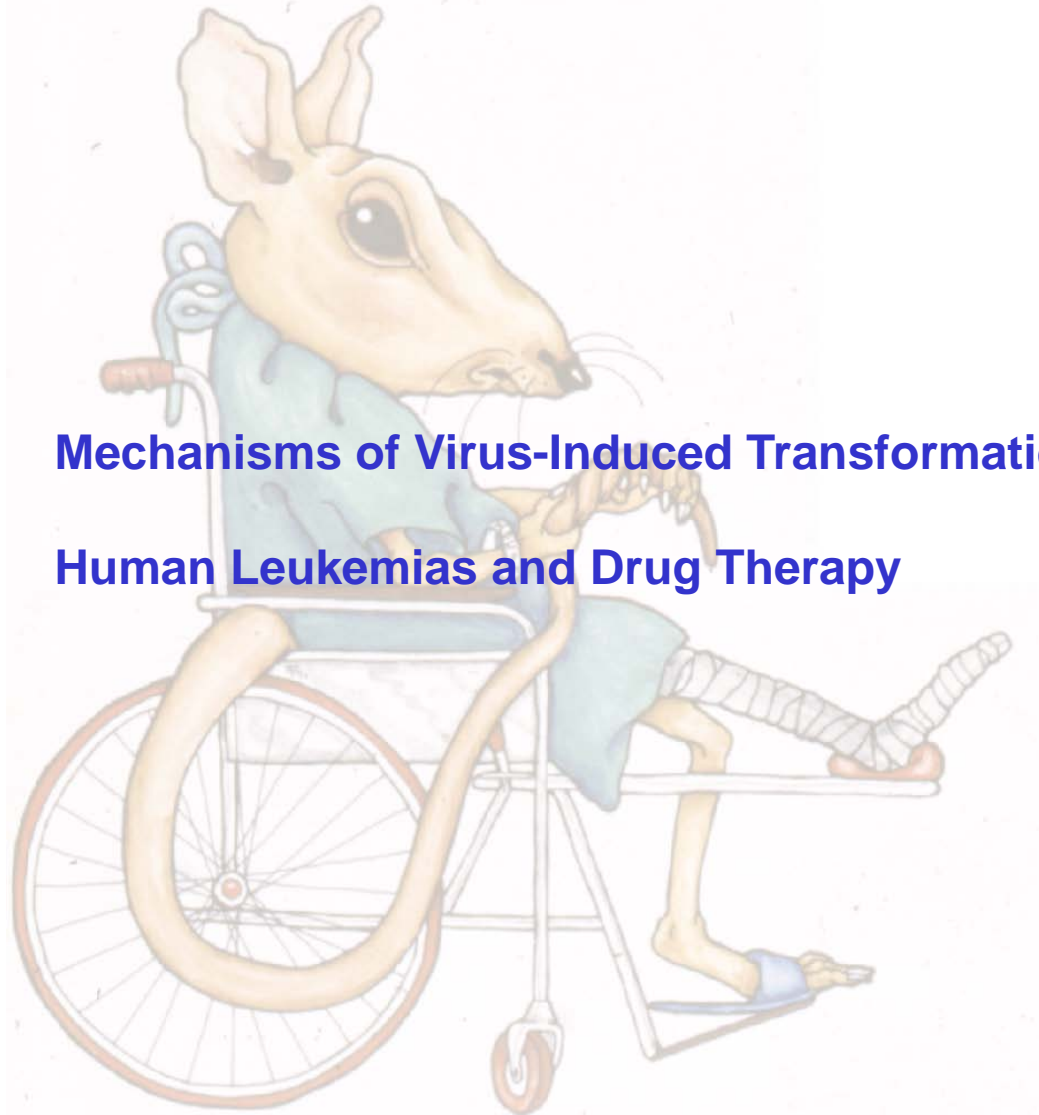
Uninfected



How Did Work with This Virus Advance Research on Human Cancer?

Mechanisms of Virus-Induced Transformation

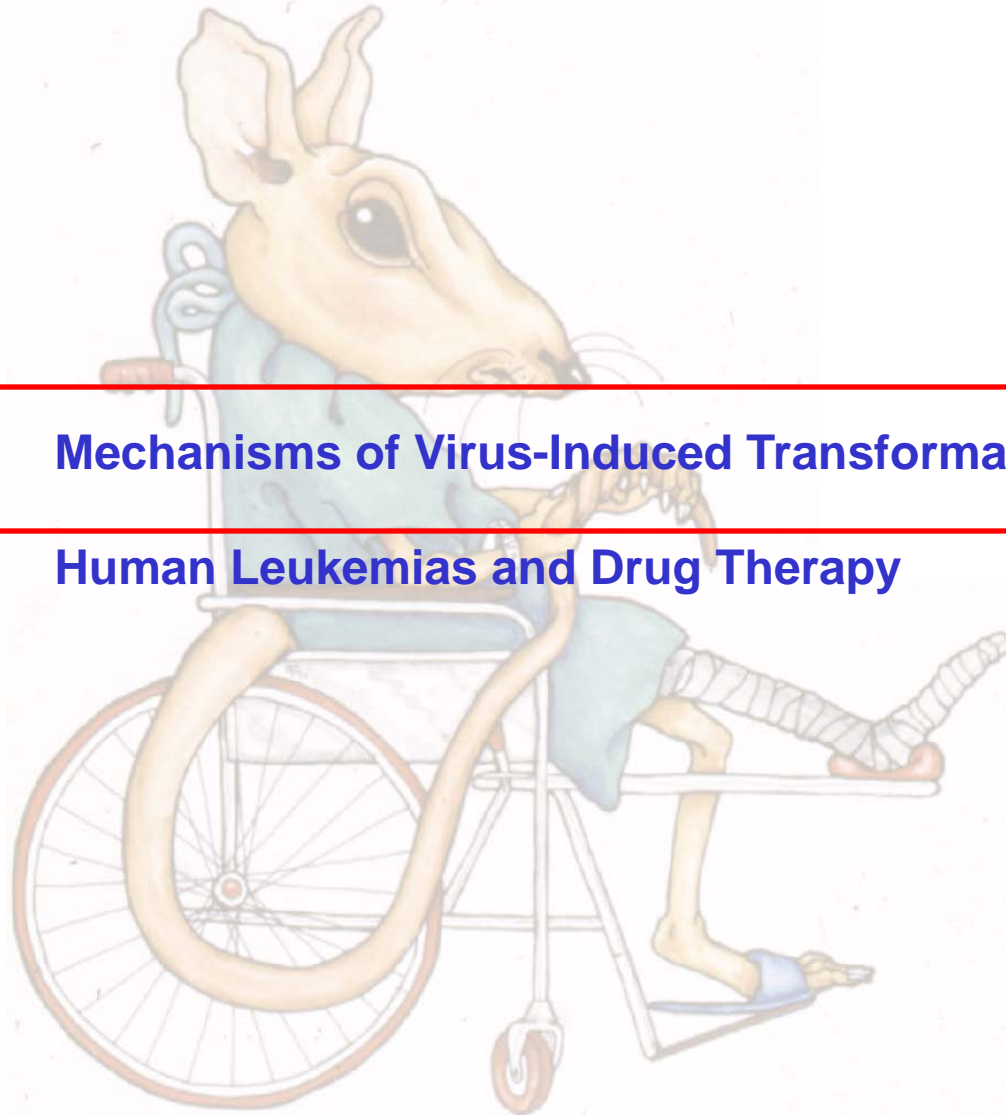
Human Leukemias and Drug Therapy



How Did Work with This Virus Advance Research on Human Cancer?

Mechanisms of Virus-Induced Transformation

Human Leukemias and Drug Therapy



The v-Abl Protein is Required for Transformation

Owen Witte, MD



David Baltimore, PhD



Naomi Rosenberg, PhD

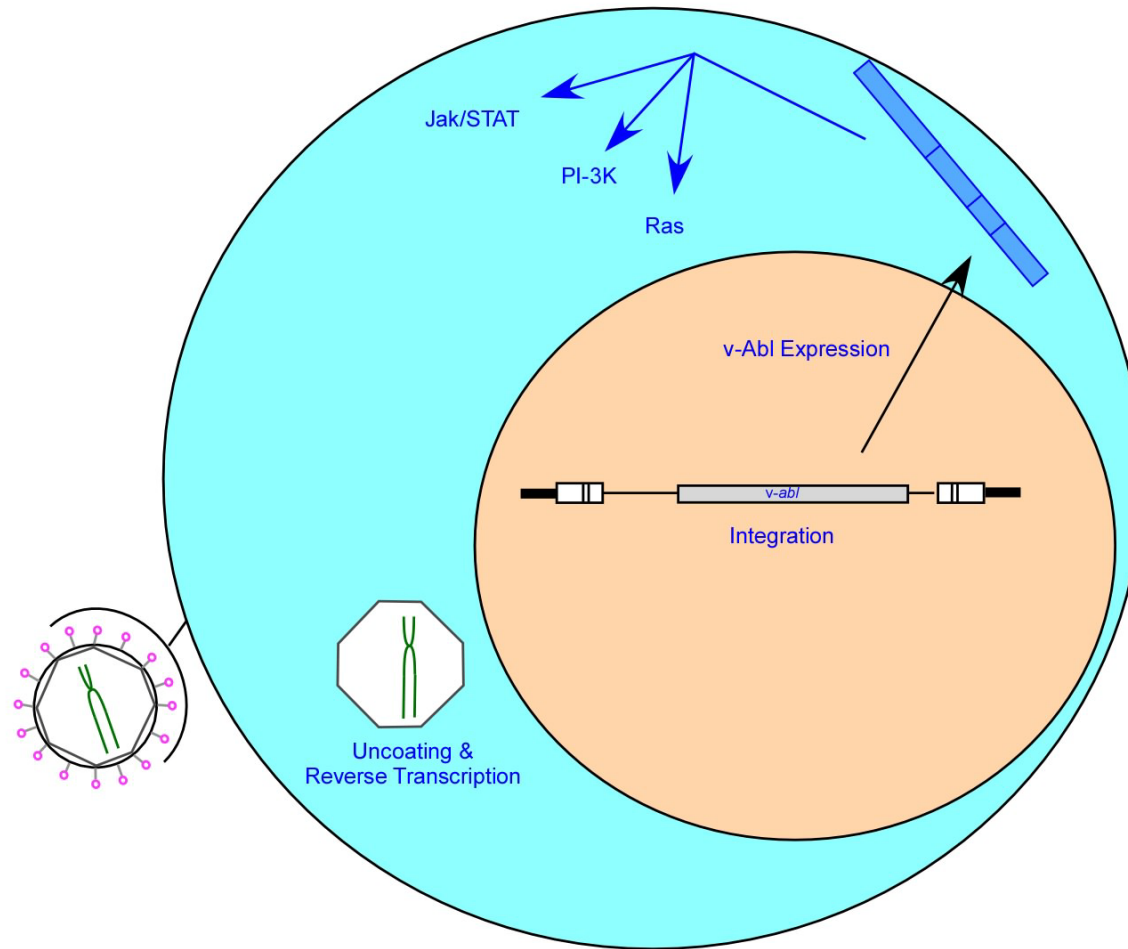
v-Abl Protein



v-Abl is a Tyrosine Kinase

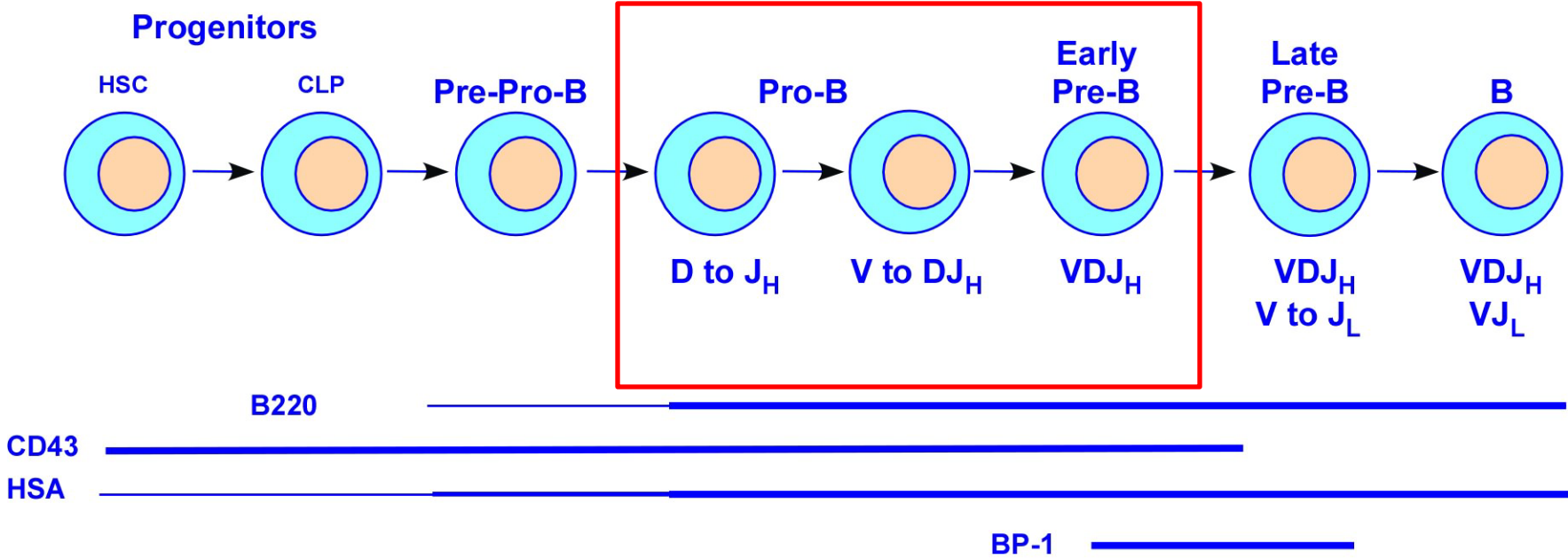
Mimics Normal Signals that Make Cells Grow and Survive

Receptor Binding and Cell Division are Required for v-Abl Expression



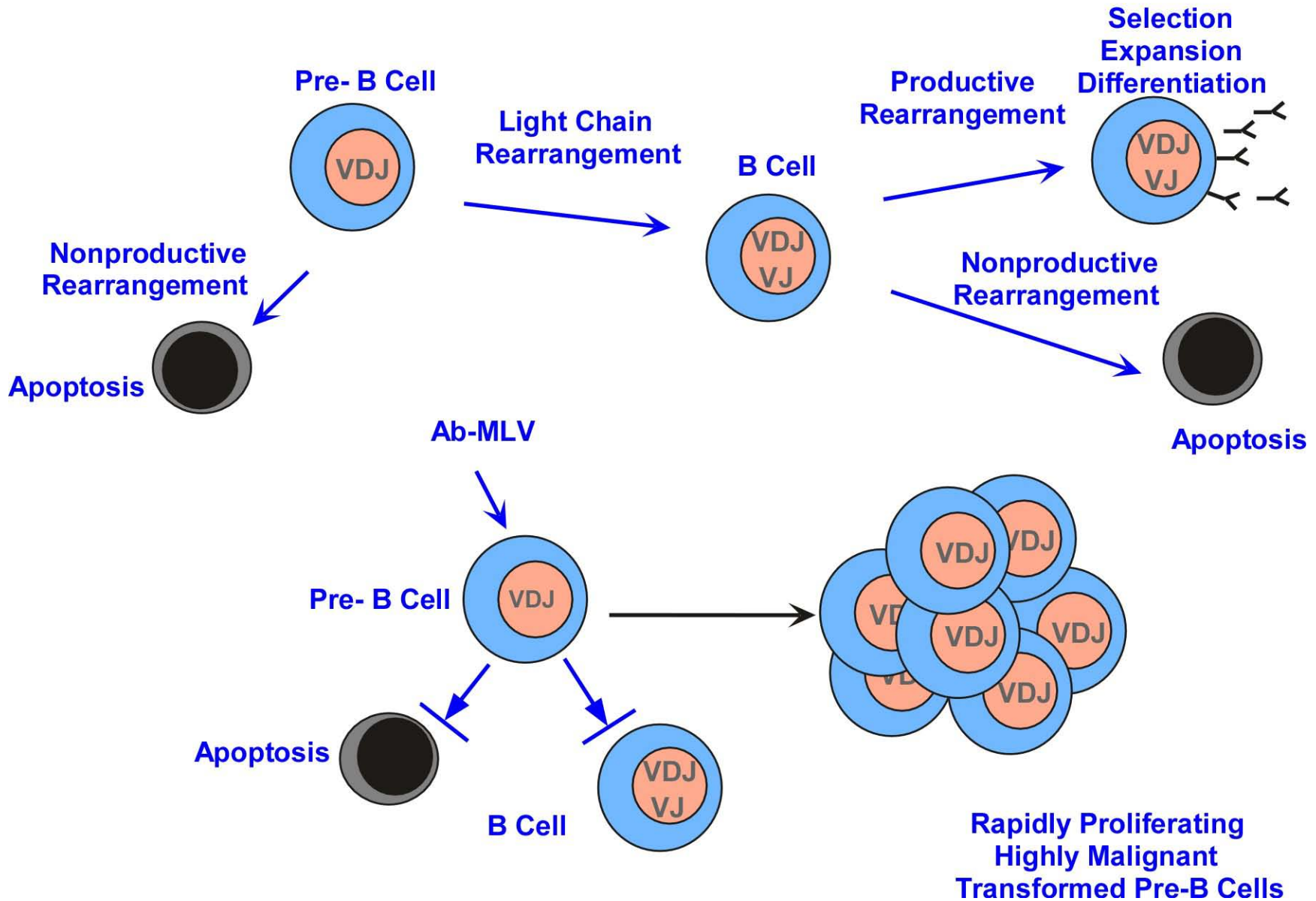
Many Cells Can Be Infected

Transformed Lymphoid Cells Resemble Pre-B Cells

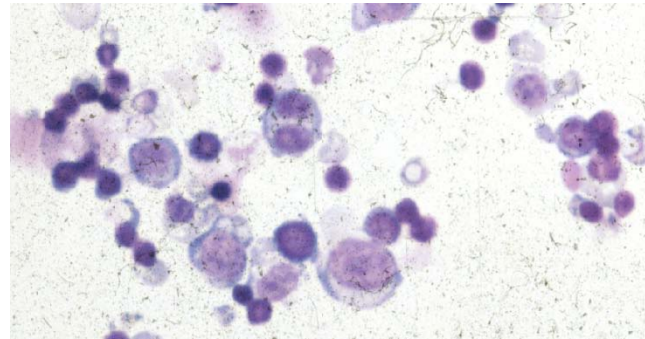
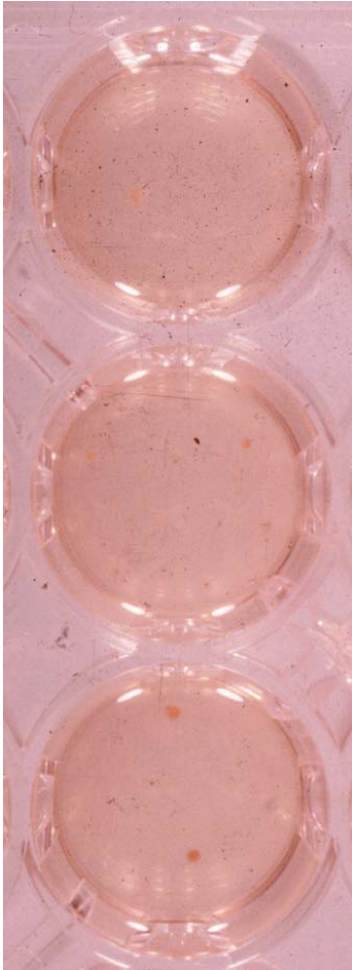


Transformant	% B220 +	% HSA +	% CD43 +	% BP-1 +
1	98	99	96	72
2	76	99	98	95
3	99	99	92	99
4	97	91	92	99

Abelson Virus Infects Pre-B Cells and Blocks Differentiation



Ab-MLV Stimulates Erythroid and Lymphoid Cells

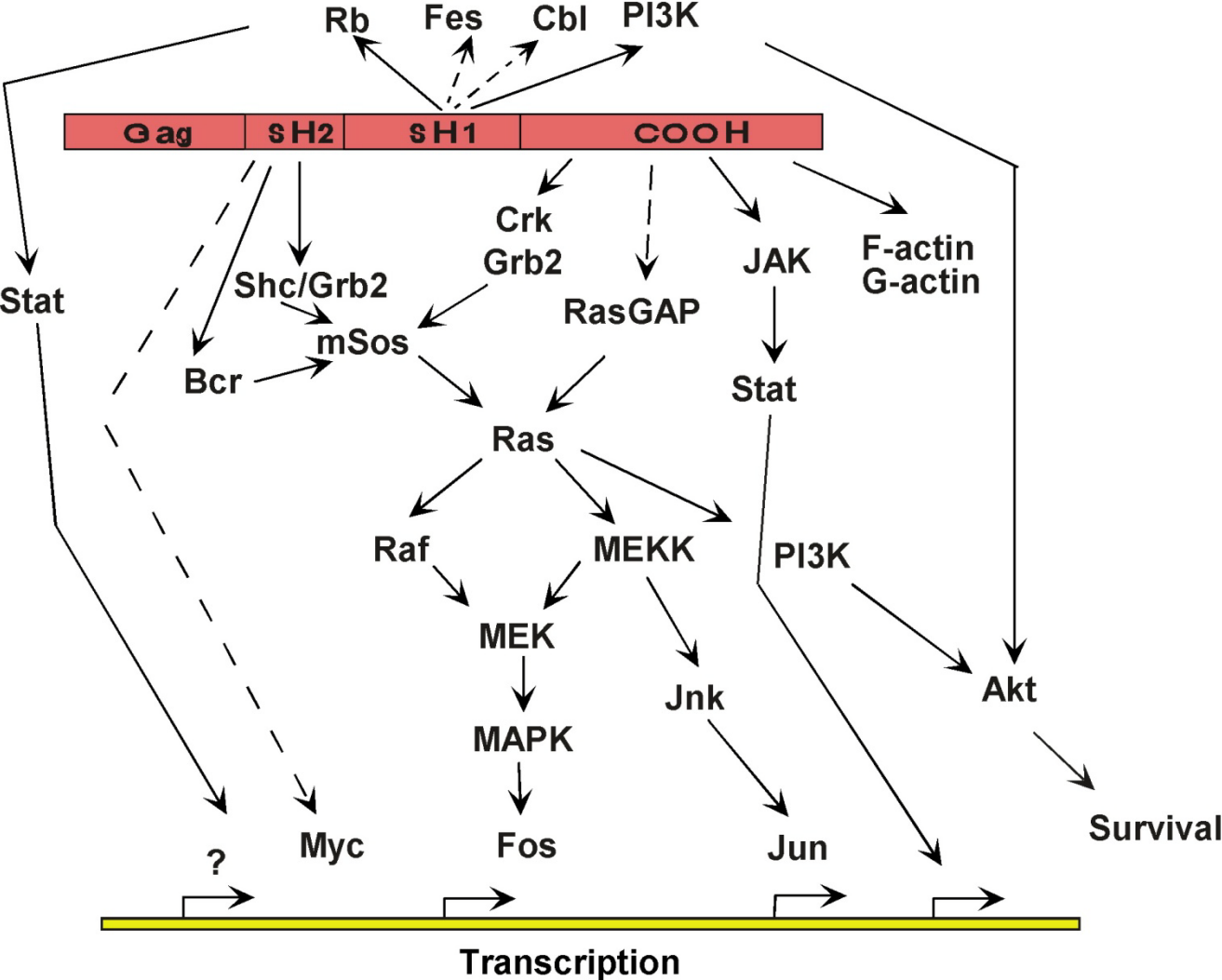


Fetal erythroid cells are stimulated to grow but don't transform

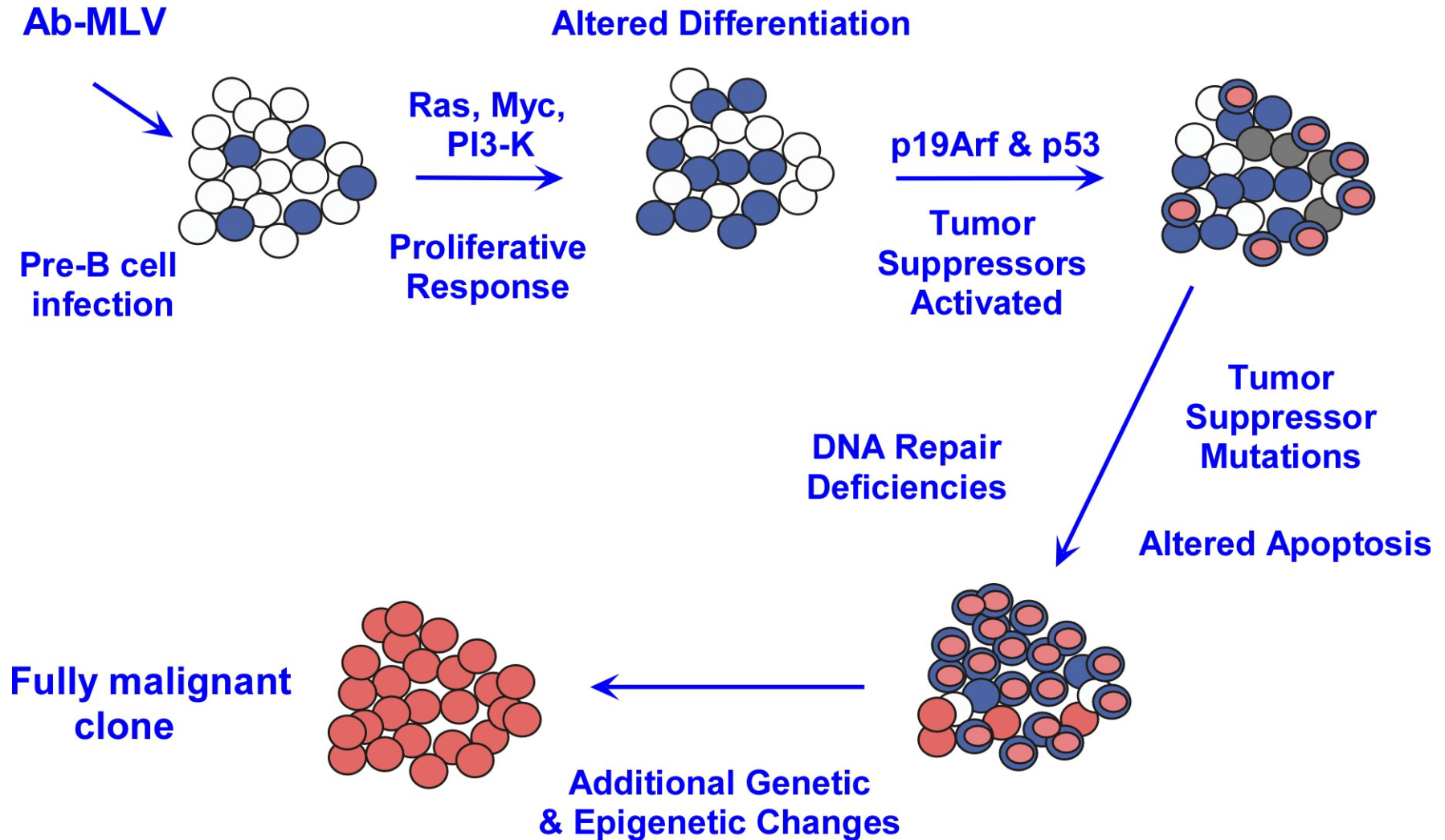
Lymphoid cells transform

Outcome reflects the context in which the *v-*onc gene is expressed**

The Answer Must Lie in Signaling Pathways But Which Ones are Key?



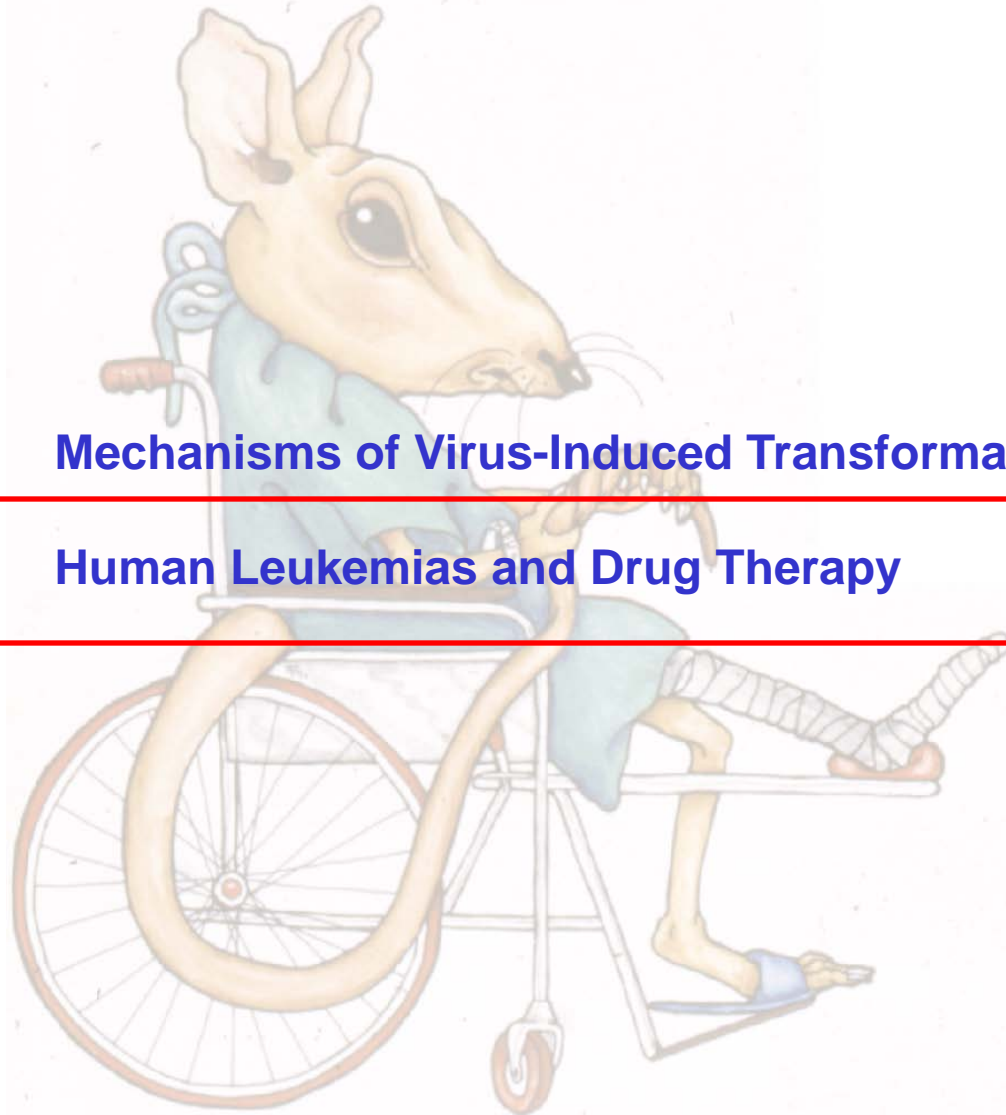
What Cellular Factors Cooperate with the *abl* Oncogene to Induce Malignant Disease?



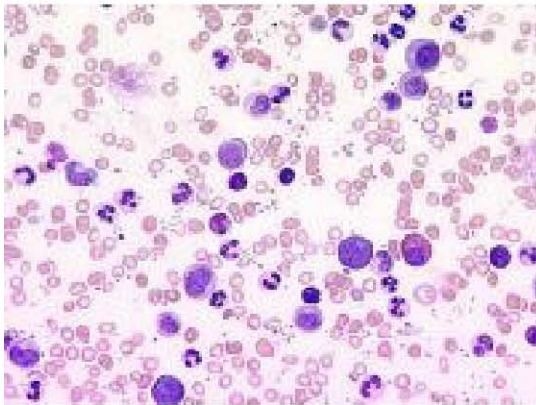
How Did Work with This Virus Advance Research on Human Cancer?

Mechanisms of Virus-Induced Transformation

Human Leukemias and Drug Therapy



Chronic Myelogenous Leukemia



**Leukemia Cells in the Blood
of a CML Patient**

20% of all Leukemias

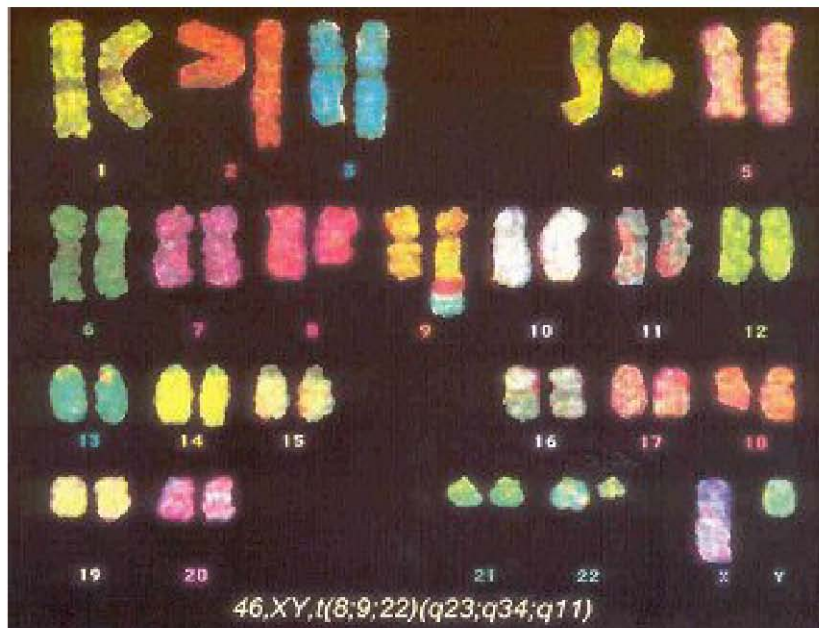
1-1.5 Cases per 100,000 People

Chronic Phase of 3 - 4 Years

Blast Phase of 3 - 6 Months

Disease Arises in Hematopoietic Stem Cells

CML Cells Have a Unique Chromosome Abnormality



Peter Nowell, MD

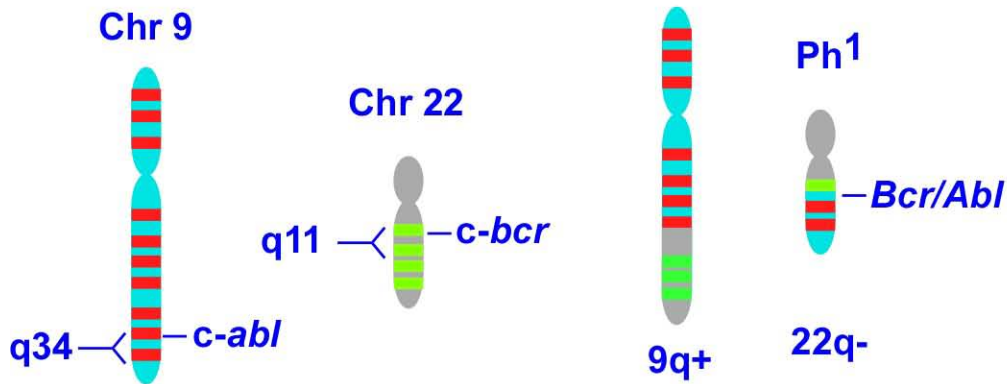


Janet Rowley, MD



David Hungerford, MD

Abl is Linked to CML



P210 Bcr/Abl



Owen Witte, MD



John Groffen, PhD

ABL is Associated with Other Leukemias



P210 BCR/ABL

CML ~ 100%



P190 BCR/ABL

B-ALL - 25-30%



P230 BCR/ABL

CML, Myeloid Leukemias, T-ALL - ~1%



ETV6/ABL

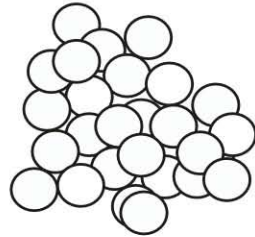
Various (ALL, CML, MPD) - <25 cases

Other translocations occur rarely
All active the tyrosine kinase activity of ABL

P210 BCR-ABL Expression Causes CML in Mice

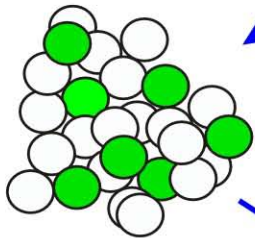


Infect in Culture

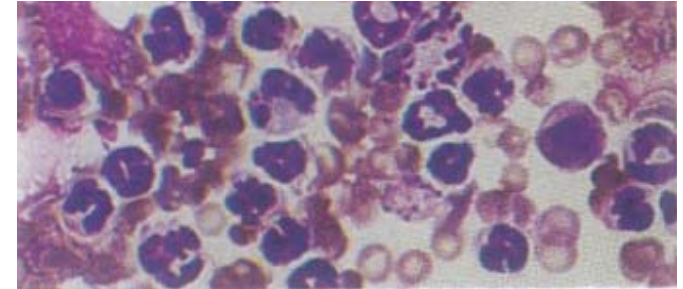


Stem Cell Enriched Bone Marrow Cells

Expand in Cytokine-rich Cocktail



Transplant Irradiated Mice



CML-like Disease Develops from BCR/ABL positive cells

Understanding Basic Mechanisms Leads to New Drug Development

Alex Matter, MD

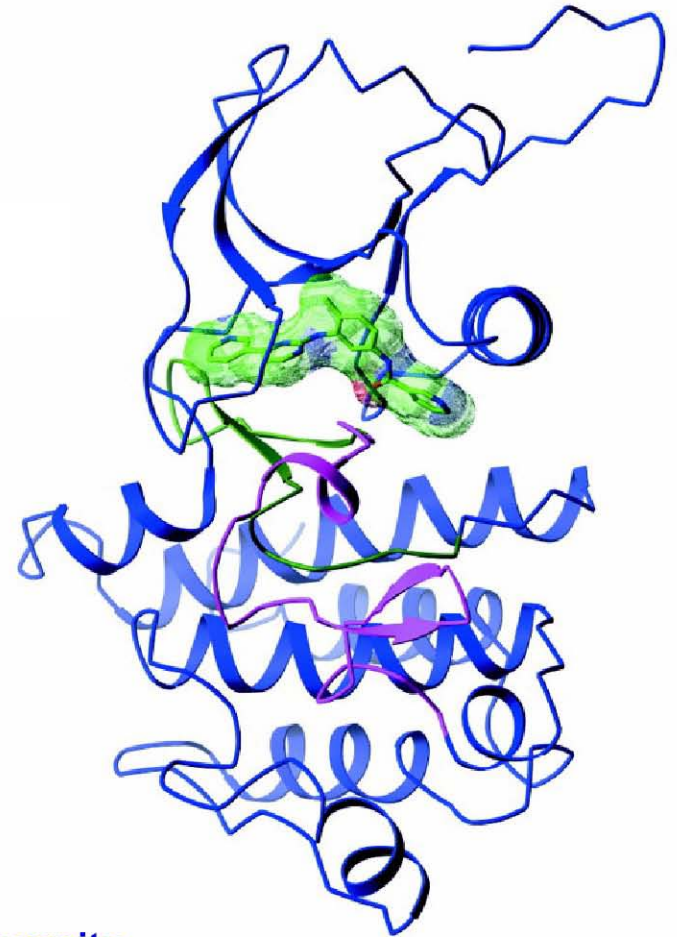


Brian Druker, MD

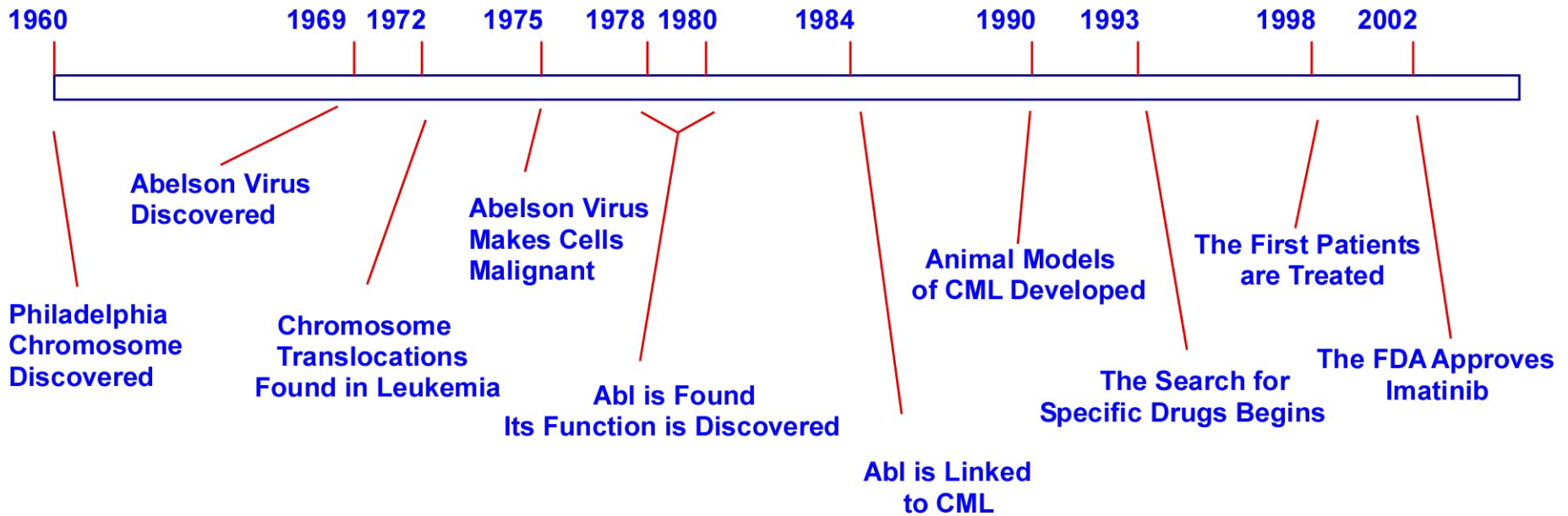


Nicholas Lydon, PhD

50% NCI
10% Novartis
10% Oregon Health Sciences University
30% Leukemia and Lymphoma Society



The Steps to Developing Imatinib



Implications and Challenges Following Approval Of Imatinib

**Has lengthened the chronic phase of CML but <10%
achieve molecular remission for > 2 years;**

**Basis for next generation inhibitors (Nilotinib,
Dasatinib, others);**

Patients develop resistance by multiple mechanisms;

Not effective for all BCR/ABL-related diseases

Paved the way for other targeted cancer therapeutics



The Abl Family of Oncogenes

Ab-MLV	Mouse	P120 v-Abl P160 v-Abl	Pre-B Cell Lymphoma
HZ2-FeSV	Cat	P110 v-Abl	Fibrosarcoma
BCR/ABL	Man	P210 Bcr/Abl P190 Bcr/Abl	Chronic Myelogenous Leukemia Acute Lymphocytic Leukemia
c-ABL	<i>C. elegans</i> to Man	P150 c-Abl	Neurological and Lymphoid Development Cell Cycle; Stress Responses

How Did We Arrive at These Insights?

Gerald Waneck
Leslie Schiff
Leslie Serunian
Bob Huebner
Dominic Picarella
Alan Engelman
Lalita Ramakrishnan
Michelle Kelliher
Christopher Barry
Yunn-Yi Chen
Li-chun Wang
Kelly Thome
Glen Raffel
Laura Baldwin
Dan Liu
Arash Radfar
Stephanie Nelson
David Warren
Li Gong



Jenia Jenab-Wolcott
Celine Mainville
Zohar Sachs
Anu Raghavan
Linda Baughn
Erica Marchlik
Mirja Gunthart
Caleb Lee
Chae-ryun Yi
Rebekah Zimmerman
Brendan Stuart

Judy Runnells
Doug Weckstein
Michel Starobinksi
Kalindi Parmar
Indira Unnikrishnan
Shawn Fessler