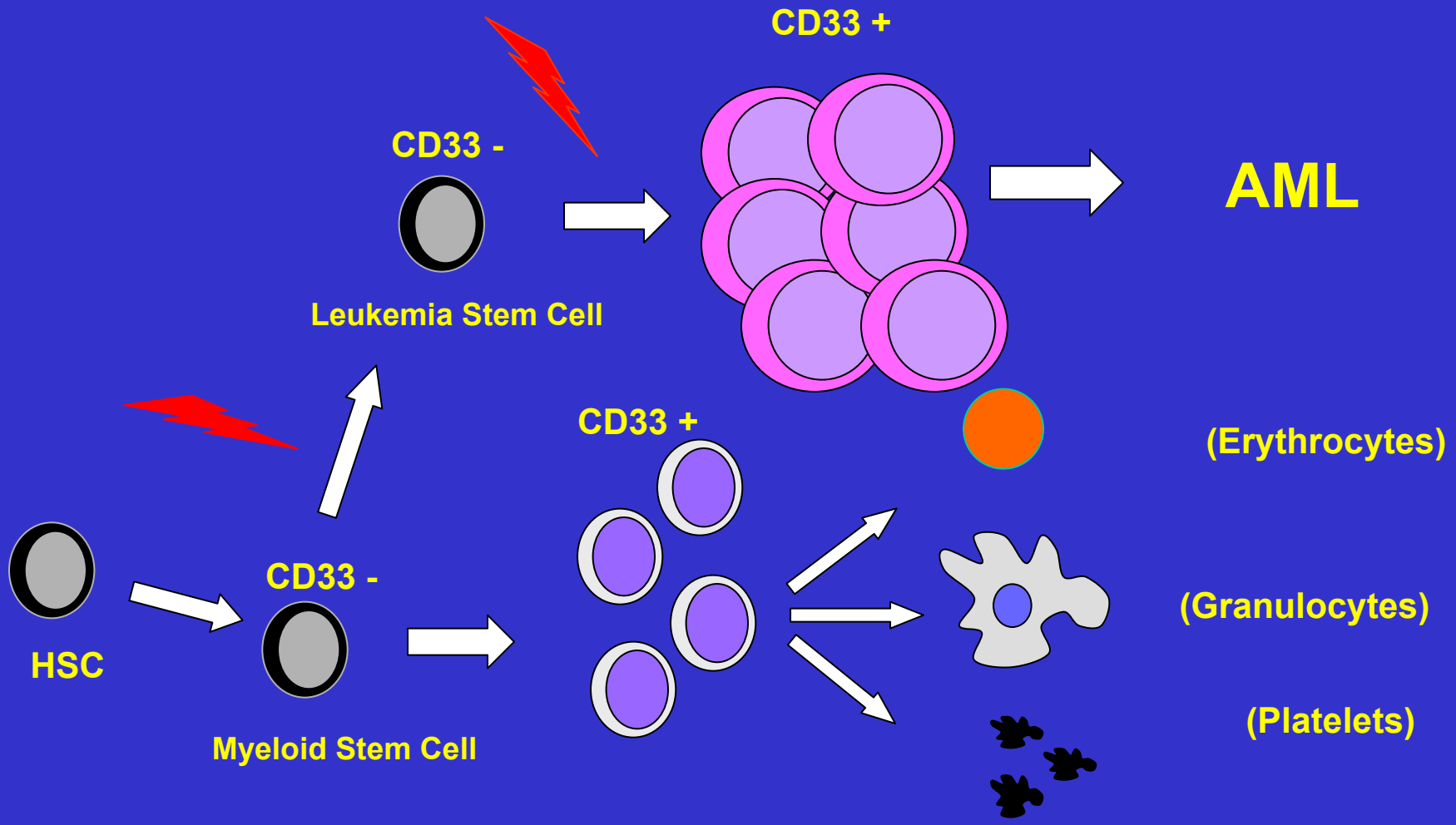
Hierarchy Model of Hematopoiesis and Leukemogenesis



PATHOPHYSIOLOGY OF ACUTE LEUKEMIA: DETERMINANTS OF CLINICAL PRESENTATION

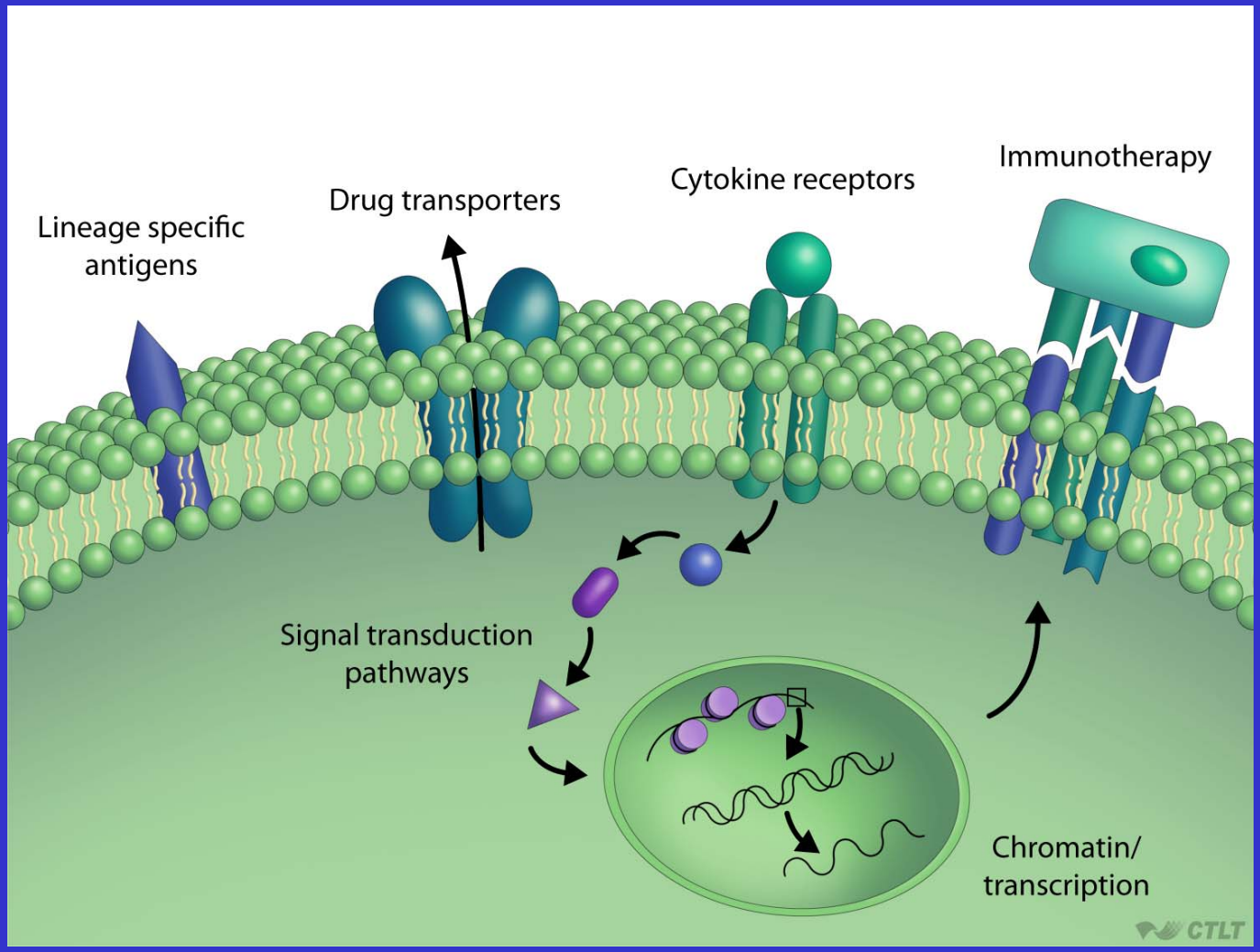
Factor	Clinical Consequences	Correction
Bone Marrow Failure	Anemia Bleeding Infection	Restore Normal Hematopoiesis
Leukemia Phenotype	Leukostasis Endothelial Damage DIC Extramedullary Tissue Infiltration Tumor Lysis Syndrome	Cytoreduction

LEUKEMIA BIOLOGY: A SIMPLISTIC VIEW

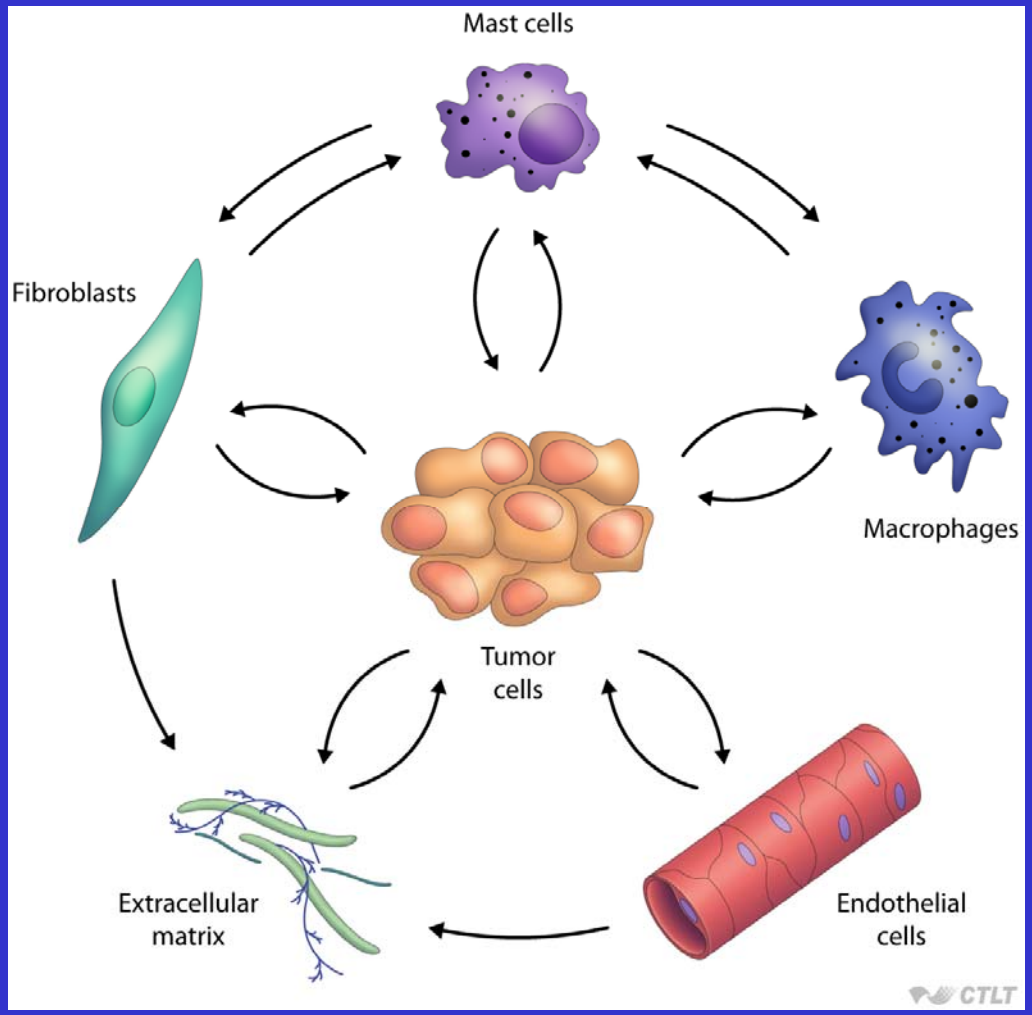
TWO MAJOR DEFECTS:

- **INABILITY TO DIFFERENTIATE**
- **INABILITY TO DIE IN RESPONSE TO STRESS**

Biological Advances:
Understanding Cellular Mechanisms → Cancer



Tumor and Host Microenvironment: Growth Factors, Motility Factors, Angiogenic Factors



AML demographics 2006

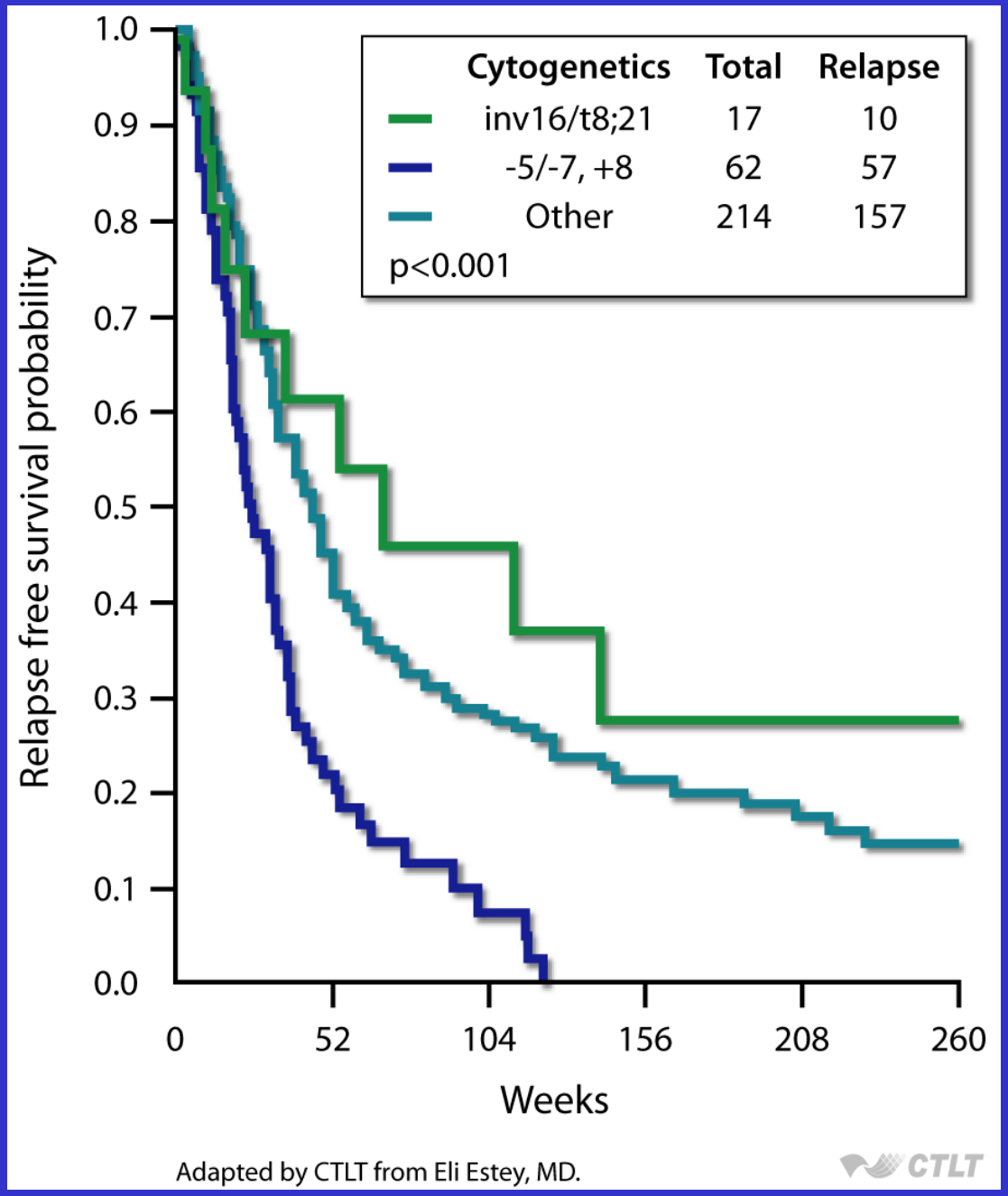
- New cases = 12,000; deaths = 9,000
- Median age = 68 yrs
- Incidence = 3.8 per 100,000
  - <65 yrs = 2.1 per 100,000
  - *≥ 65 yrs = 18 per 100,000*
- Chance of developing AML
  - For a 50 yr old = 1 in 50,000
  - *For a 70 yr old = 1 in 5,000*

AML: Much work to do

Age	CR	DFS	Early Death	OS (median)
<60	70%	45%	10%	30% (24 mo)
>60	45%	20%	25%	10% (10 mo)

Based on CALGB, MRC trials in which adults of all ages were eligible

Slide kindly provided by Dr. Richard Stone


Cytogenetics Total Relapse
inv16/t8;21 17 10
-5/-7, +8 62 57
Other 214 157
p<0.001
Relapse free survival probability
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
0
52
104
156
208
260
Weeks
Adapted by CTLT from Eli Estey, MD.
CTLT

Why is AML in the older adult so difficult to treat?

Disease Biology

- DNA Damage/Toxin Exposure Increases with Age
- Detoxifying Enzyme Activity Decreases with Age
- Immune Surveillance Decreases with Age

Host Biology

- Intolerance for Cytotoxic Chemotherapy

DNA REPAIR PATHWAYS

IDENTIFYING CRITICAL GENES IN THE PROCESS OF LEUKEMOGENESIS

Repairing DNA Damage: Lessons Learned from Familial Cancer Syndromes

Selected Familial Leukemia-Prone Syndromes: Lesions in Genes Responsible for Repairing DNA Damage

- Fanconi Anemia (Repair of interstrand DNA crosslinks)
- Xeroderma Pigmentosum (Nucleotide excision repair)
- Bloom Syndrome (RecQ Helicase)
- Ataxia Telangiectasia
- Li-Fraumeni (p53)

Polymorphisms in DNA Repair Genes May Determine Risk, Response, and Toxicity

- XPD Lys751Gln polymorphism → increased risk of developing AML with -5q and/or -7q
- ERCC1 8092 C>A polymorphism → increased risk for childhood ALL in males
- XPD Gln751/Asp312G haplotype → increased CR for adults with AML >age 55
- ERCC1 polymorphisms → lung and metabolic tox
- XRCC3 241Met polymorphism → decreased risk for liver toxicity

Fanconi Anemia Proteins: A "Master" Switchboard for Repairing of DNA Damage

All 11 FA proteins cooperate in a pathway to:

- **Recognize and repair DNA damage due to DNA crosslinking agents (alkylators, platins, mitomycin C)**
- **Coordinate with other DNA repair proteins (ATM, ATR, Bloom, BRCAs!!) to form complexes that localize to and repair damaged sites**

FA Genes as Targets for Inactivation in AML (Kennedy and D'Andrea, JCO, 2006)

- Inherited biallelic inactivating mutations → high incidence (~ 30%) of developing cancer by age 40, especially AML with adverse cytogenetics (-7, abnormalities of chromosomes 1 q and 3q)
- Heterozygous Carriers → high incidence of breast/ovarian, pancreatic (esp. FANCD1/BRCA2), low incidence of AML (FANCA missense mutations)
- Somatic inactivation of FANC genes in AML:
  - Deletions/mutations: FANCA, FANCD1, FANCG
  - Methylation: FANCA, FANCC, FANCF, FANCG

GENES AND ENVIRONMENT:
A LEUKEMOGENIC COMBINATION

AML: A Prototype Environmental-Occupational Malignancy

Etiologic Factors	% AML	Genetic Abnormalities
Env/Occ Exposure Benzene, Petroleum, Organic solvents, Arsenical Pesticides, Radiation	~30-35%	-5/5q, -7/7q, 3q26 t(8;21), +8, +21
Therapeutic Agents	~20%	
DNA Damaging Agents		-5/5q, -7/7q, 3q26 AML-1 mutation (21q22)
Topo-II Directed Drugs		Translocations (11q23)

Benzene: The Classical Environmental Hematotoxin

- 1930's: 1st association between benzene and bone marrow disorders
- 1950's: Direct link between chronic exposure and acute leukemia demonstrated in long-term study from Turkey when benzene-based adhesives were first used (Askoy 1972)
- 1970's: Large-scale NCI cohort study in China beginning 1972 → dose-dependent incidence of MDS/AML <10 ppm → RR 3.2, >25ppm → RR 7.1 (Hayes 1997)

Selected Mechanisms of Benzene Hematotoxicity

- Intermediary benzene metabolites → direct DNA damage → activate p53 and, in turn, p21 → cell cycle arrest (Yoon, Exp Hematol 2001)
- Hydroquinone causes selective deletion of chromosome 7 (by FISH) in CD34+ marrow cells (Stillman, Exp Hematol, 2000)
- Low level airborne exposure to benzene (gas station attendants, police) → altered methylation of p15 (↑) and MAGE ↓) (Bokati, Ca REs 2007)

NAD(PH): QUINONE OXIDOREDUCTASE (NQO1)

- NQO1 gene located on chromosome 16q23
- Induced by antioxidants → protects against oxidative stress
- Inactivated by ^{609}C → T polymorphism

Intersection of Benzene and $^{609}C \rightarrow T$ NQ01 Polymorphism

- NQO1 detoxifies marrow-genotoxic *benzene* metabolites (phenols, benzoquinones) to less toxic hydroxyquinone metabolites
- Inactivating $^{609}C \rightarrow T$ associated with >7-fold increased RR for benzene poisoning (Rothman,Ca Res, 1997)
- Low exposure (<1ppm) → decrease in WBC, platelets, colony-forming progenitors; most toxicity in $^{609}C \rightarrow T$ NQO1 and/or fully active MPO (converts benzene to toxic quinones) (Lan, Science, 2004)

More Evidence Tying $^{609}C \rightarrow T$ to Increased Risk of Leukemia

- ↑ frequency low or null NQO1 activity in all acute leukemias: for *de novo* leukemias, highest frequency seen in *de novo* AML with *inv(16)* (Smith, Blood 2001)
- ↑ prevalence in AMLs with -5/-7 and T-AML, with greatest prevalence in homozygotes (Larson, Blood 1999)
- ↑ risk 11q23 (*MLL* gene)-rearranged infant ALL (Smith, Blood 2002) or infant ALL without *MLL* rearrangements (Lanciotti, Leukemia 2005)

GLUTATHIONE S-TRANSFERASES (GSTs)

- Family of isoenzymes (α,μ,ρ,Θ) that detoxifies anthracyclines and epipodophyllotoxins (VP16): induced by oxidative stress (DNA and lipid damage by reactive oxygen species)
- Null polymorphisms are common for each subfamily
- Epidemiologic relationships between "null" μ or Θ genotypes and increased risk for developing CLL and MDS – relationships with genesis of treatment-related AML and *de novo* childhood ALL not so clear

GST Polymorphisms May Relate to Chemotherapy Drug Metabolism and Clinical Outcome

- Homozygous deletions in GSTM1 and/or T1 → drug resistance and short overall survival in AML (Voso, Blood 2002) but not in childhood ALL (Davies, Blood 2002)
- High blast glutathione levels → ↑ risk of relapse in childhood ALL (Kearns, Blood 2001)
- Null GST-T1 genotype → increased early death (toxicity) in AML (Naoe, Leukemia 2002)
- SWOG multiple AML therapies → no clear impact of GST polymorphisms on toxicity, early mortality achievement or duration of remission (Weiss, Leukemia 2006)

Treatment-Related AML After Treatment for Breast Cancer

- 1970's: Case reports of women < age 60 who received chemo but no XRT → AML (Davis, Cancer 1972)
- 1980's: AML RR post-breast cancer therapy: 2.4 local XRT alone, 10 alkylators alone; 17.4 alkylators + XRT (Curtis, NEJM 1992)
- 1990's: ↑AML risk with DNA intercalators Epirubicin (+ alkylators: Pederson-Bjerrgaard, JCO 1992; Praga, JCO 2005) and Mitoxantrone (+ XRT: Carli, Leukemia 2000; Chaplain JCO, 2000) – risks are dose-dependent
- 1990's (NSABP trials): AML risk relates to CY dose (in CY/Adr combo), XRT, and use of G-CSF

BREAST CANCER AND AML: A "LOCAL" PERSPECTIVE

- Among 230 women ages 18-86, *33 (14.3%)* have a history of breast cancer and an additional *47 (20.3%)* have positive family history (mainly breast/ovarian, also heme and/or multiple ca's)
- Of the 33 women with AML and Hx breast cancer:
  - 19 (58%) < 60 at breast ca Dx (med 51, range 36-76)
  - 20 (61%) did not have chemotherapy:
    - 8 → no chemo or XRT, 12 → XRT only
    - 4 → chemo only, 9 → chemo + XRT

Therapy-Related AML: Could We Predict Who Is At Risk?

Development of T-AML appears to be increased in patients with:

- Defective expression of mismatch repair proteins → microsatellite instability (Ben-Yehuda, Blood 1996; Zhu, Blood 1999; Rund, Leukemia 2005)
- Variant polymorphisms in HLX1 (homeobox gene) and Rad51 (DNA repair) (Kawad, Blood 2006)

In contrast, the CYP3A4 1*B genotype may confer protective effect against T-AML (Rund, Leukemia 2005)

CLINICAL TRIALS: THE ROAD TO MEDICAL ADVANCES

Clinical trials are important because:

- At the present time, we don't have a reasonable "standard of care" because we are not curing nearly enough people
- Clinical trials with new ideas and new drugs the offer a chance to do better
  - What we have learned from clinical trials in years past is the basis for what we do today (and we are better than we were 2-3 decades ago!)
- What we learn from today's trials will help us to develop more effective approaches tomorrow

TYPES OF CLINICAL TRIALS

- Risk Assessment → Prevention
- Early Detection and Diagnosis
- Treatment
  - Primary Disease
  - "Secondary Prevention" (Adjuvant Therapy, Minimal Residual Disease/Maintenance)
  - Recurrent/Refractory/Metastatic Disease
- Quality of Life
  - Quantity vs. Quality – how can we measure that?
  - What is the cost to the person and to society?

"Secondary" Leukemia Research Agenda: Molecular Epidemiology

Define pathways/processes leading to loss of cell cycle regulation, inappropriate cell survival and net genomic instability

- Genetic Susceptibility (Host response to Toxins and DNA Damage)
  - DNA Repair Genes and Pathways
  - Pharmacogenomics (Intracellular toxin metabolism)
- "Epigenetic" Factors
  - DNA damage→ altered gene expression through methylation and/or histone deacetylation

"Secondary" Leukemia Research Agenda: Clinical Epidemiology

- Define populations at risk for leukemogenesis: A *folie a deux* between genetics and environment
  - Longitudinal studies to quantitate dose-risk relationships for leukemogenic agents
  - Serial monitoring of "at risk" populations to identify "leukemia intitiation"

"Secondary" Leukemias: Clinical Interventions

- Prevention for patients at high risk
  - Screening for predisposition → intervene before cumulative genetic damage ensues: Vaccines????
  - Avoid specific agents in patients with susceptibility to certain types of DNA damage
- Innovative treatment approaches
  - Epigenetic modulation
  - Overcome pro-survival pathways
  - Inhibit DNA repair pathways

CHALLENGES TO TRANSLATIONAL CLINICAL RESEARCH : Common Misconceptions

Patient samples grow on trees

Man is related to any/all:

1. Mouse 2. Cell line 3. Nude mouse 4. Agar colony

Isolated cells are just like cells in their natural habitat

Patients, like inbred mice, are all the same

Insurance companies really have the patient's interest at heart

IRBs protect all of us from the bad, mad doctors who just want to "spearmint" on those poor, unsuspecting patients

The FDA makes thoughtful decisions about drug safety and efficacy

Why should clinical research cost anything? After all, the patients are already there

A MAJOR CHALLENGE FOR ALL AREAS OF MEDICINE

How do we get the best medicine *to EVERYONE, not just those who can pay or be paid for??

***What is the best medicine? In the setting of inadequate cure rates, it is a clinical trial that offers the possibility of doing better!**