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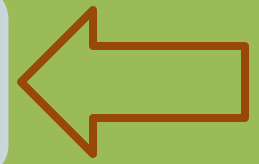
Cancer Risk from Radiation Exposure: The Role of Genetic Susceptibility

Radiation Epidemiology & Dosimetry Course

Levels of Variation

Whole Organism	• Assays such as $LD_{50/30}$
Clinical radiosensitivity	• Consequence of radiotherapy • e.g. skin erythema, lung fibrosis
Susceptibility to Radiation Carcinogenesis	• Risk differences in populations • Epidemiology studies
Tissue radiosensitivity	• By specific tissues/organs • Epidemiology/clinical studies
Cellular radiosensitivity	• e.g. cell killing, chromosomal damage, DNA damage



Adapted from Human Radiosensitivity: Report of AGIR, 2013

Genetic Susceptibility to Radiation

- Rare syndromes with extreme radiosensitivity
- Ataxia-telangiectasia (AT)
  - Rare childhood neurodegenerative disease
  - Caused by mutations in *ATM* gene

- Cultured fibroblasts from patients three times as sensitive to radiation (Taylor et al., 1975)

How Does this Affect the General Population?

Figure 1a Increasing Incidence with Radiation Dose

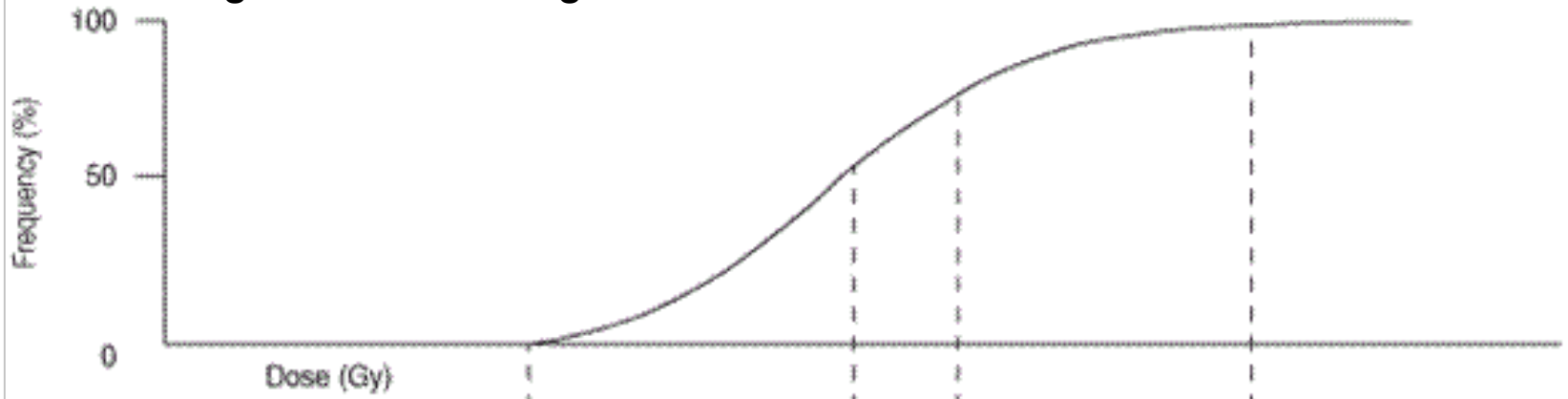
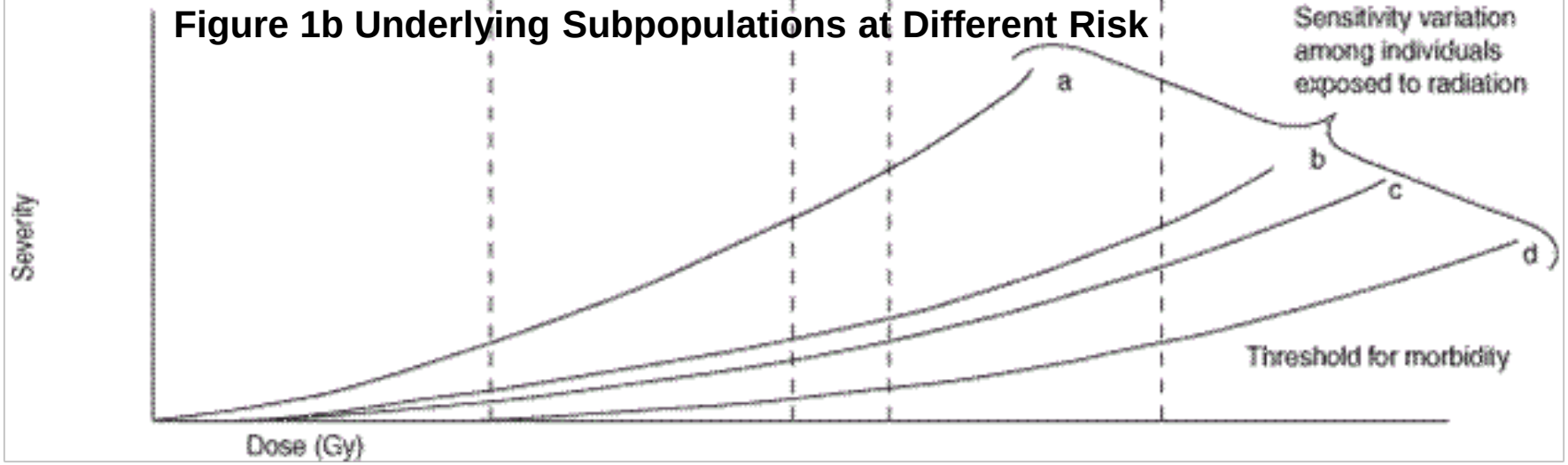


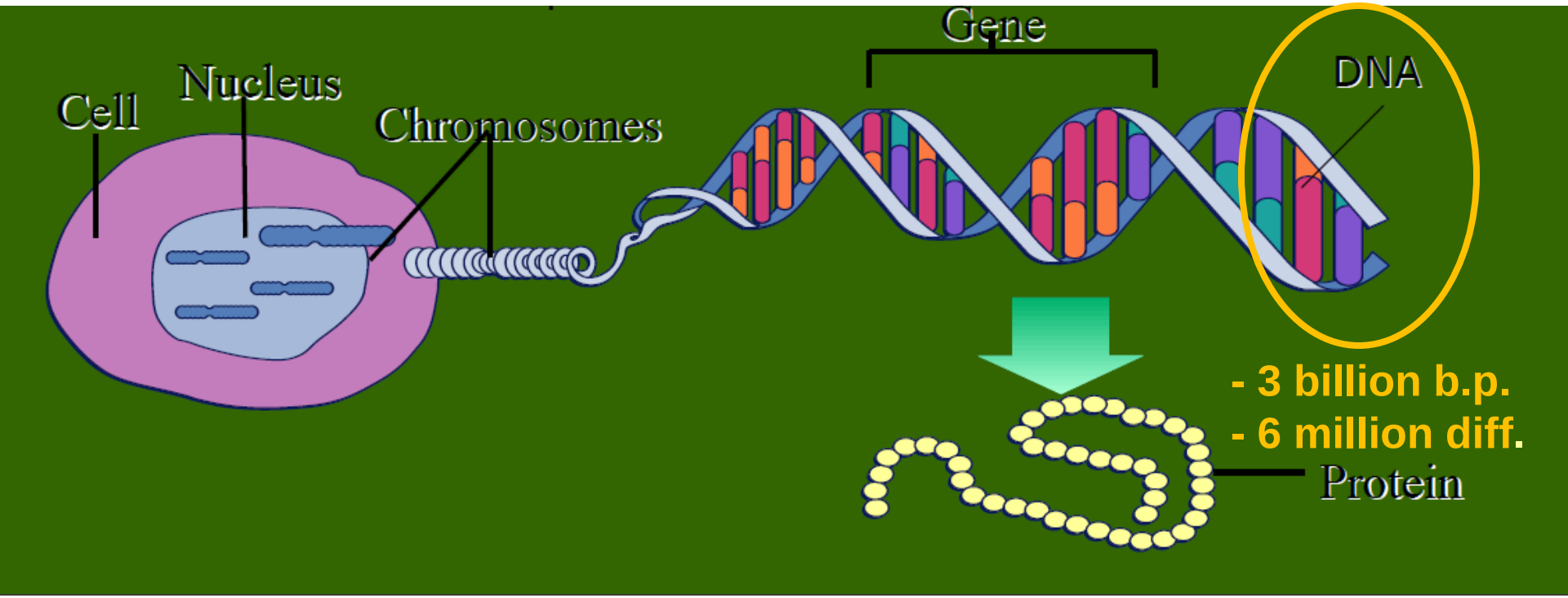
Figure 1b Underlying Subpopulations at Different Risk



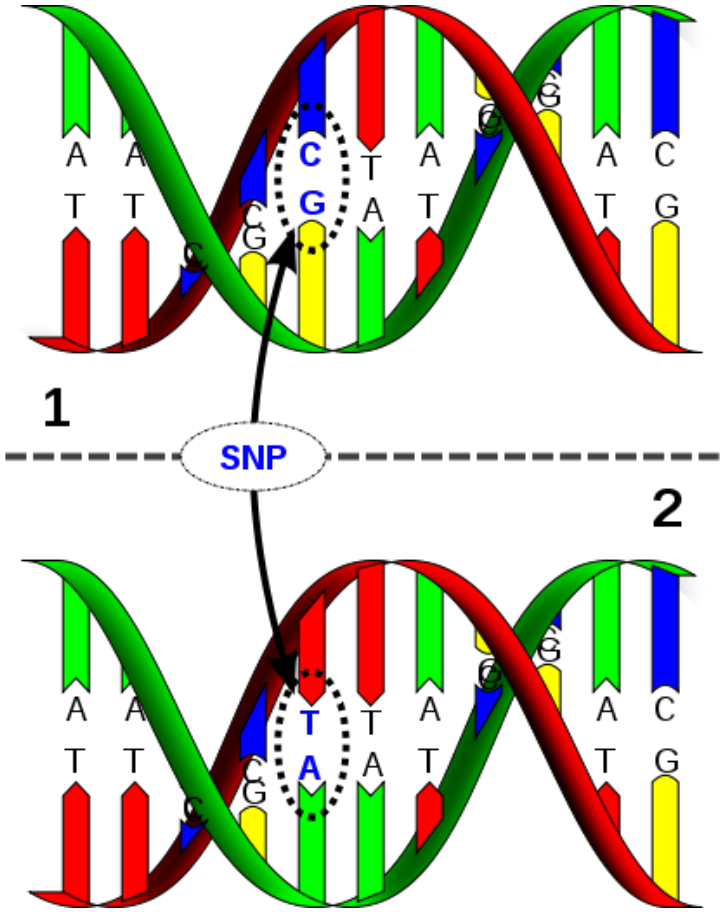
Adapted from Hendry et al, 2006

Types of Genetic Variation

- Chromosomes, genes, RNA, DNA



Single Nucleotide Polymorphisms (SNPs)

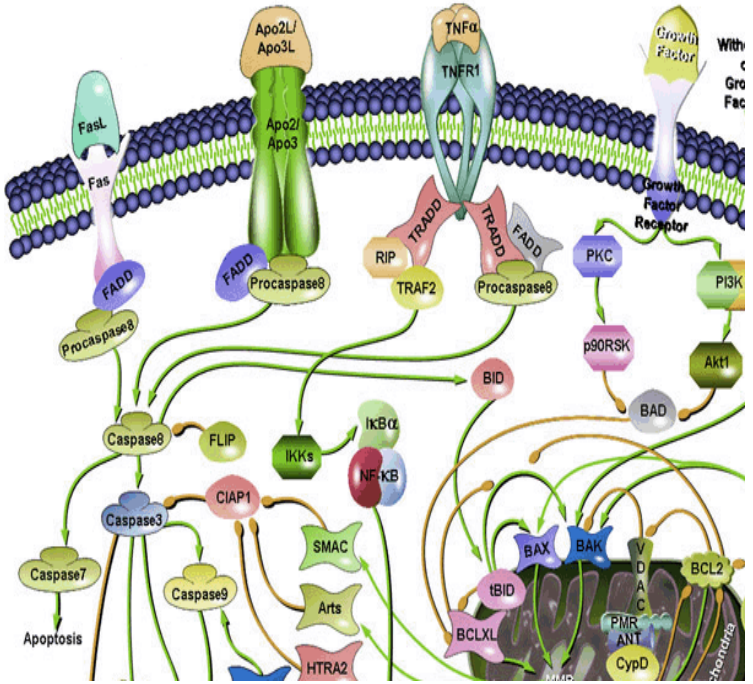


- Most common genetic variation
- Each individual has two alleles
  - CC (common referent)
  - CT (heterozygote)
  - TT (homozygous variant)
- Much of the variation appears meaningless
- Some variation increases risk of cancer

Approaches to Study Genetic Variation - 1

1. Candidate gene approach

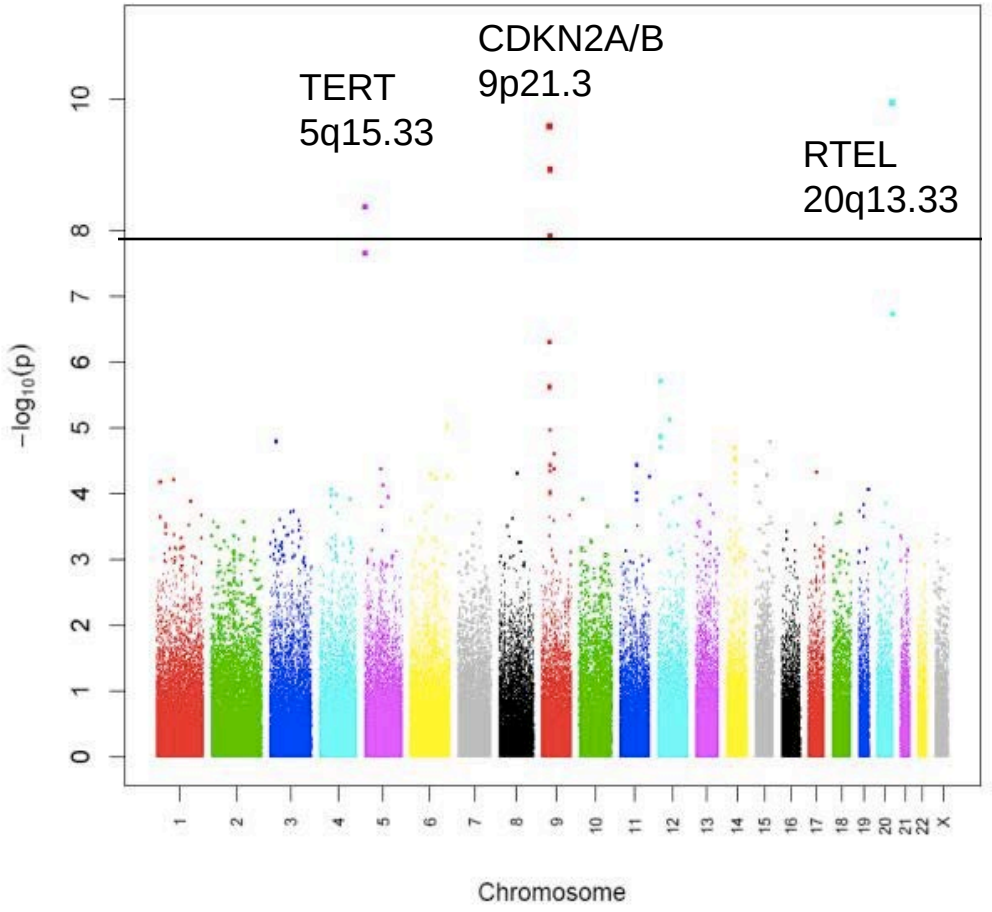
2. Pathways of Interest
   - DNA repair
   - Cell-cycle control
   - Apoptosis
   - Immune-related
   - Oxidative Response



Approaches to Study Genetic Variation - 2

Agnostic: GWAS, exome/genome-wide sequencing

- Agnostic – no assumptions about underlying biology
- GWAS approach has identified 475+ risk loci in germline DNA for cancers
- Typically 600,000 to 5 million markers across genome

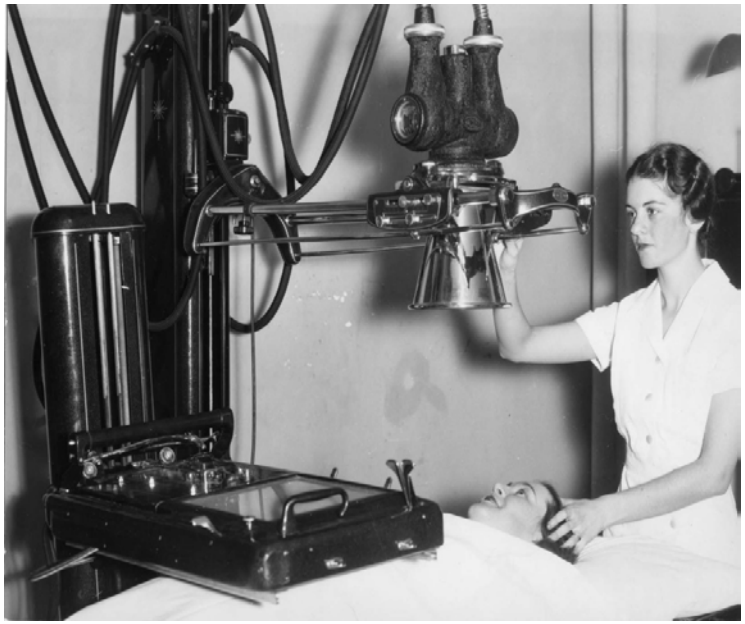


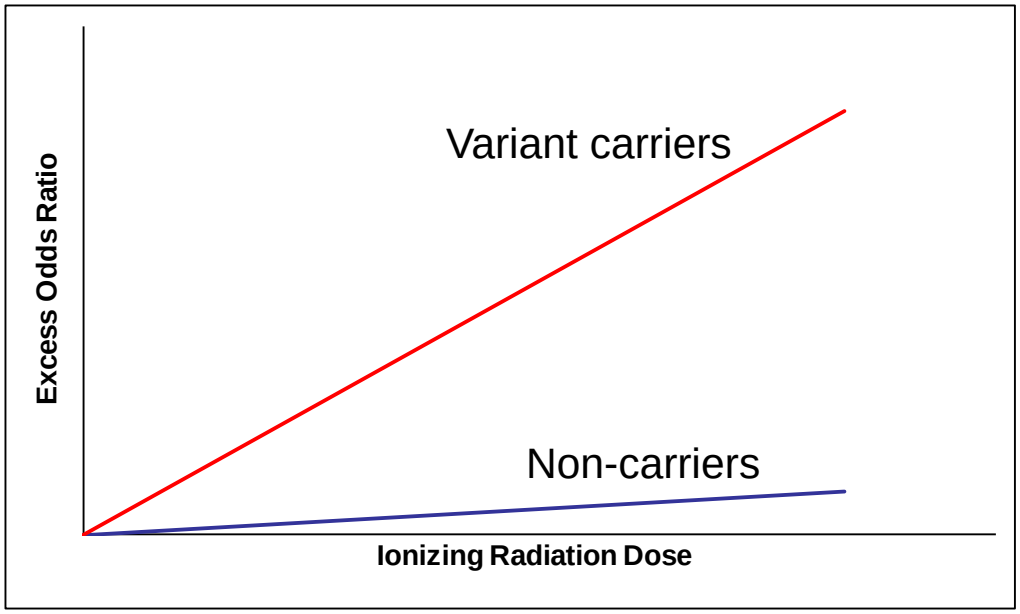
Examples from Radiation

Genetic Susceptibility to
Radiation-related Breast Cancer

Candidate Gene Approach: Occupational Radiation

- Nested case-control study within USRT cohort
  - 858 breast cancer, 1083 cancer-free controls
- Radiation dose
  - occupational, personal medical
- Blood samples for DNA





Occupational Radiation, USRT Breast Cancer SNP-radiation interactions:

Pathway (SNPs, genes examined)	Gene	SNP effect	Radiation Interaction	Replication
DNA repair (61 SNPs, 21 genes)	*PRKDC*	✓	N	
	BRCA2	✓	N	
Apoptosis and proliferation (16 SNPs, 8 genes)	*IL1A*	✓	✓	?
	CASP8	✓	N	
Oxidative stress and inflammation (28 SNPs, 16 genes)	*PTGS2*	✓	N	
	IL1B	✓	N	
	IL4	✓	N	
GWAS Identified (38 SNPs, 35 genes)	*MRPS30*	✓	✓	?

Sigurdson et al, 2007; Rajaraman et al, 2008; Bhatti et al, 2008, 2010; Schonfeld et al, 2010

Summary: Candidate Gene approach

- Focus on genes thought to be involved in radiation toxicity
- A few signals, but not consistent
- Limited knowledge of underlying biology; ability to query genome

DNA Repair Damage	**Radiation Fibrogenesis**	**Oxidative Stress**	**Endothelial Cell Damage**
ATM,NBN, BRCA1,2 H2AFX, RB1, XRCC1,4,5,6, PRKDC, LIG4, Cyclins, CDKs, CDK inhibitors	*TP53, BCL2, CASP3, TNF, ILIA, IL6, TGFB1,2,3 SMADs*	*SOD1,2,3*	*FGF2 VEGF*

Alternate Approach: Rare Mutations in Breast Cancer Susceptibility Genes

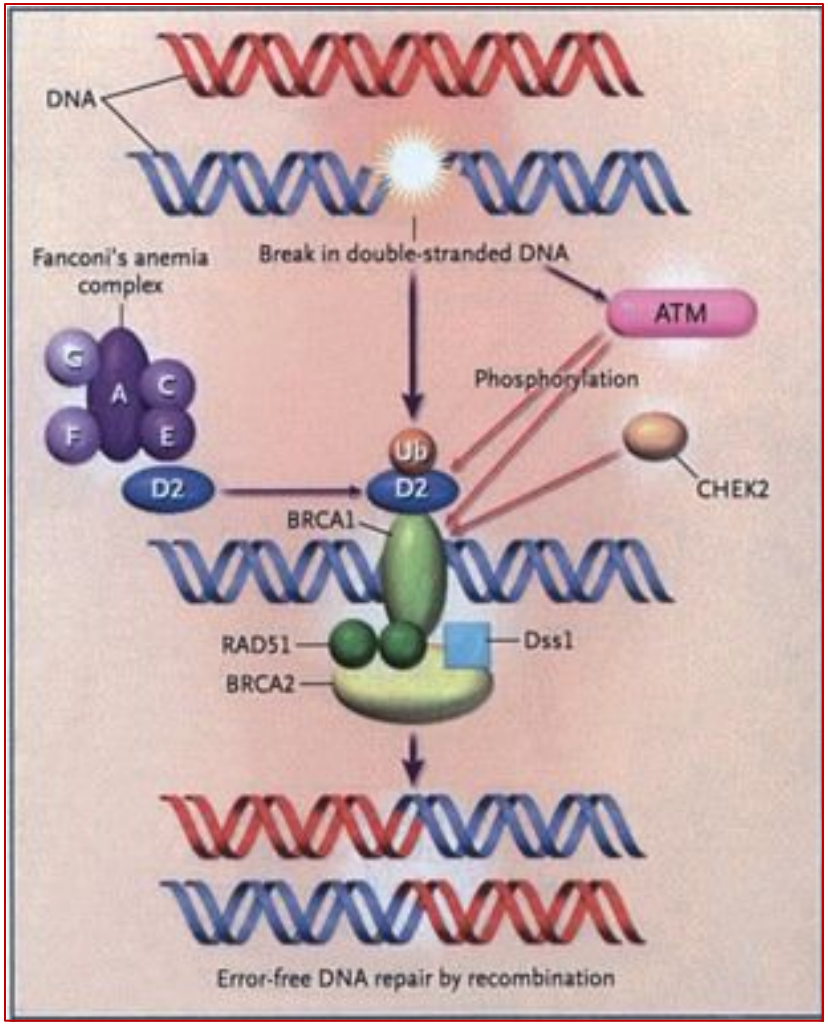


Figure source: Venkitaraman, NEJM 2003

- *BRCA1,BRCA2, ATM, RAD51*
- Repair of double-strand DNA breaks by homologous recombination
- Inactivation in these genes predisposes to breast (and other) cancers

Diagnostic Radiation (X-rays) and Breast Cancer

Self Report
- misclassification
- bias

Population	Exposure		Citation
• 1,601 *BRCA1/2* carriers cohort • UK, Canada, Netherlands, France	Chest x-rays - ever/never; <20yr - no. x-rays by age	ref never x-ray Higher with younger age, more reported x-rays	Andrieu et al, JCO, 2006
• 138 *BRCA1* breast cancer; 158 non-mutation breast cancer • Poland	Chest x-rays - ever - <30	OR=1.7 (0.9, 3.0) OR=1.8 (1.2, 2.9)* ref non-carrier	Gronwald et al, Br Ca Res Treat, 2008
• 379 *BRCA1*, 6 non-mutation • France	0yr, ge	HR = 4.29 (2.1,8.8)* No variation by age at exposure, number x-rays	Lecarpentier et al, Br Ca Res Treat, 2011
• 454 *BRCA1*, 273 *BRCA2* carriers <50yrs • US, Canada, Aus/NZ	Chest x-rays, ever/never	OR=1.16 (0.64-2.11) for *BRCA1* OR=1.22 (0.62-2.42) for *BRCA2*	John et al, CEBP, 2013

Overlapping populations

Confounding by indication?

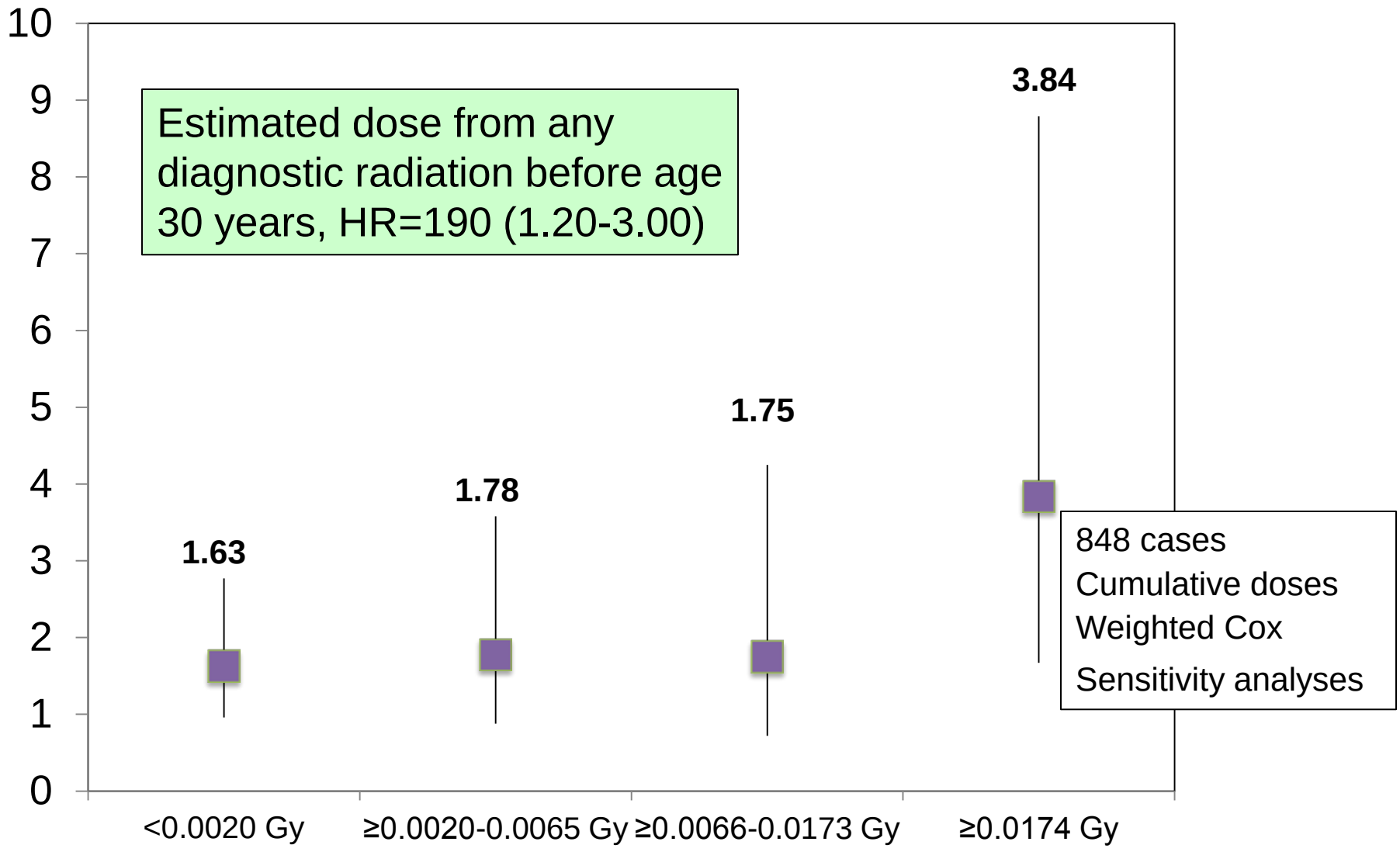
Diagnostic Radiation (Mammograms) and Breast Cancer

Population	Exposure	Odds Ratio (95% Confidence Interval)	Citation
1,600 *BRCA* breast cancer cases, 1,600 non-cancer controls	Age at 1st Mammography >1yr before dx	OR=1.03 (0.85, 1.25)	Narod et al, Lancet Oncol, 2006
162 *BRCA* carriers; 34 cases	No. of mammograms >1yr before enrollment	OR=0.94 (0.88, 1.00)	Goldfrank et al, 2006
2,346 *BRCA 1/2* carriers; 238 cases	Any prior graphy; first gram age <30 yrs	HR=0.79 (0.53, 1.19); HR 0.90 (0.35, 2.34)	Giannakes et al, Breast Ca Res Treat 2014

Limited Power

- Mammograms more likely to be accurately reported
- Inconsistent results

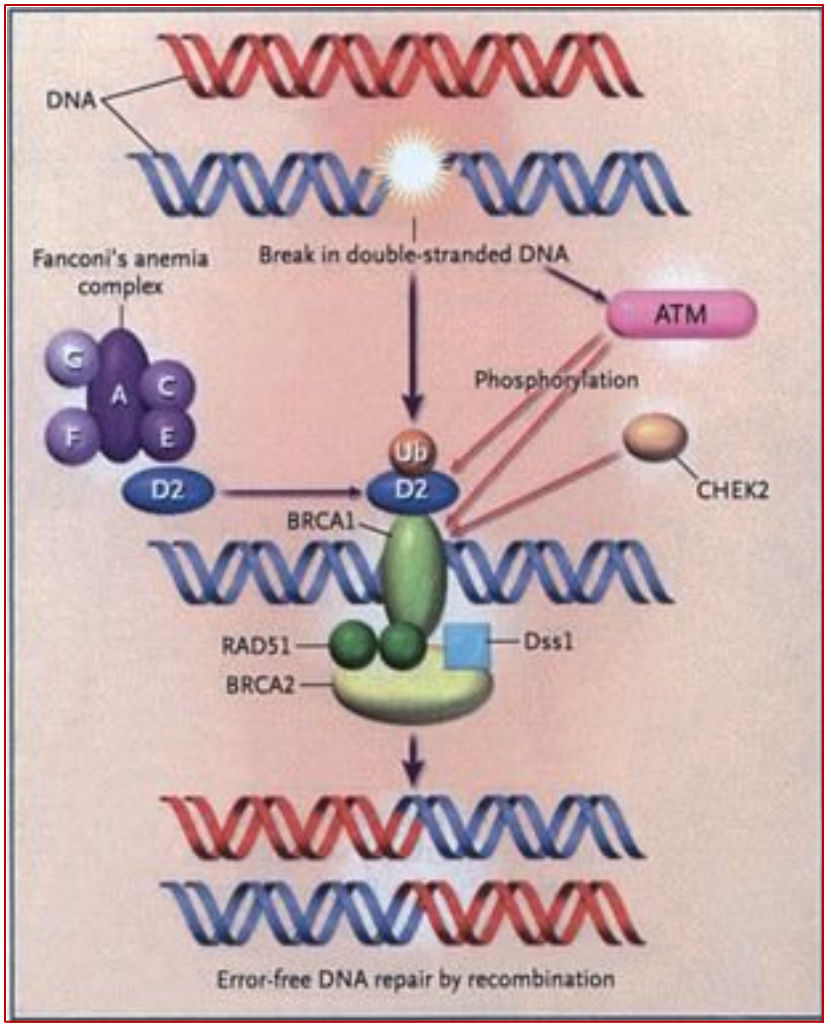
Risk of Breast Cancer in 1,993 *BRCA 1/2* carriers



Pipje et al, BMJ 2012

Therapeutic Radiation and Breast Cancer - 1

- 247 contralateral breast cancer (CBC) cases
- 51 pathogenic germline mutations in *BRCA1, BRCA2, CHEK2, ATM* in 247 CBC patients
- Radiotherapy-related risk for DDRP germline mutation carriers
  - OR=2.2 (1.03, 4.62) overall
  - OR=2.51 (1.03, 6.10) for CBC five or more years after RT



Broeks et al, Breast Can Res, 2007

Therapeutic Radiation and Breast Cancer - 2

WECARE Nested Case-Control Study

- 708 contralateral breast cancer cases; 1,397 controls
- No increase in risk with radiation dose in carriers

Gene	Radiation	Cases/Cntrls	OR (95% CI)
BRCA1/2 carrier	--	96/62	4.5 (3.0 - 6.8)
No mutation	≥ 1 Gy	118/406	1.2 (1.0 - 1.6)
BRCA1/2 Carrier	≥ 1 Gy	21/26	1.0 (0.4 - 2.8)

Bernstein JB et al., EJC, 2013

ATM and risk of contralateral breast cancer

	Radiation Gy	Case/Cntrl	OR (95% CI)	OR* (95% CI)	ERR/Gy
Wild type		271/480			
Any rare *ATM* variant	Adj	148/264	1.1 (0.8-1.4)		
Missense	Adj	75/129	1.2 (0.8-1.7)		
Missense	0	26/30	0.6 (0.3-1.1)	1.0 (ref)	
	0.01-0.99	21/45	1.7 (0.9-3.1)	2.7 (1.2-6.4)	
	≥ 1.0	21/38	2.0 (1.1-3.9)	3.3 (1.4-8.0)	1.3 (0.1-3.9)
Deleterious Missense	0	14/14	0.6 (0.2-1.3)	1.0 (ref)	
	0.01-0.99	12/17	2.8 (1.2-6.5)	5.3 (1.6-17.3)	
	≥ 1.0	11/15	3.3 (1.4-8.0)	5.8 (1.8-19.0)	2.6 (0.0-10.6)

Bernstein et al, JNCI 2010

GWAS Studies of Radiation-related Cancer

1. WECARE study
   - CBC after primary breast cancer (*J Bernstein)*
   - Omni-1Quad

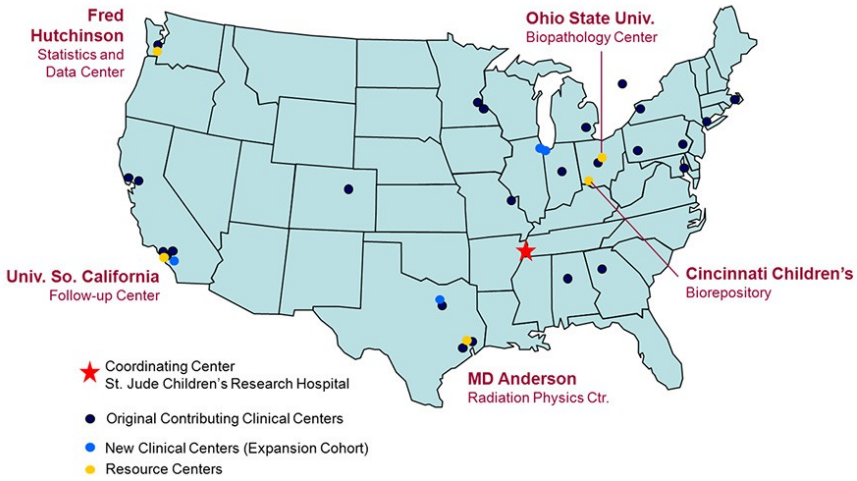
2. Childhood Cancer Survivor Study
   - Secondary malignancies after Hodgkin's Lymphoma Survivors *(K Onel)*
   - All secondary malignancies after eight most common childhood cancers *(L Morton)*

Childhood Cancer Survivor Study

CCSS
An NCI-Funded Resource

1970 1980 1990 2000 2010

- 14,359 childhood cancer survivors
  - dx 1970-1986, age <21 yrs
  - ≥5 yr survival post-dx
- Detailed treatment data
  - RT + Chemo (49%)
  - Radiotherapy (11%)
  - Chemotherapy (22%)
- ~ 45% with biospecimens

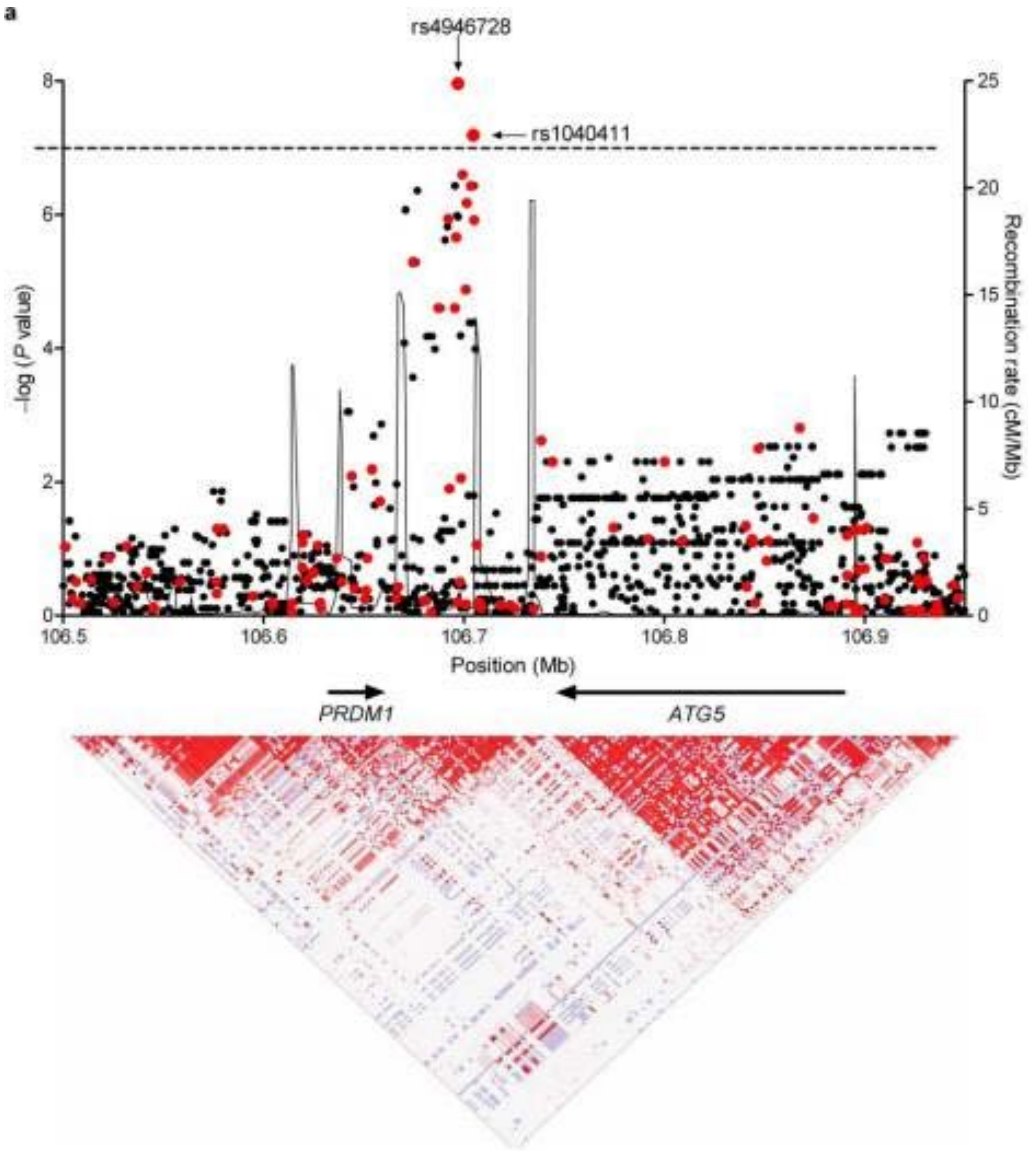


Slide courtesy of Lindsay Morton

SMNs in Hodgkin's Lymphoma Survivors

- HL dx before age 18yrs, treated with RT
- Genotyped on Affymetrix 6.0
- Discovery set
  - 100 SMN cases, 89 SMN-free controls
  - rs4946728, *PRDM1*
- Independent replication
  - 62 SMN, 71 controls

  (CCSS, MSK, USC)
- Greater number of risk haplotypes associated with lower *PRDM1* mRNA expression

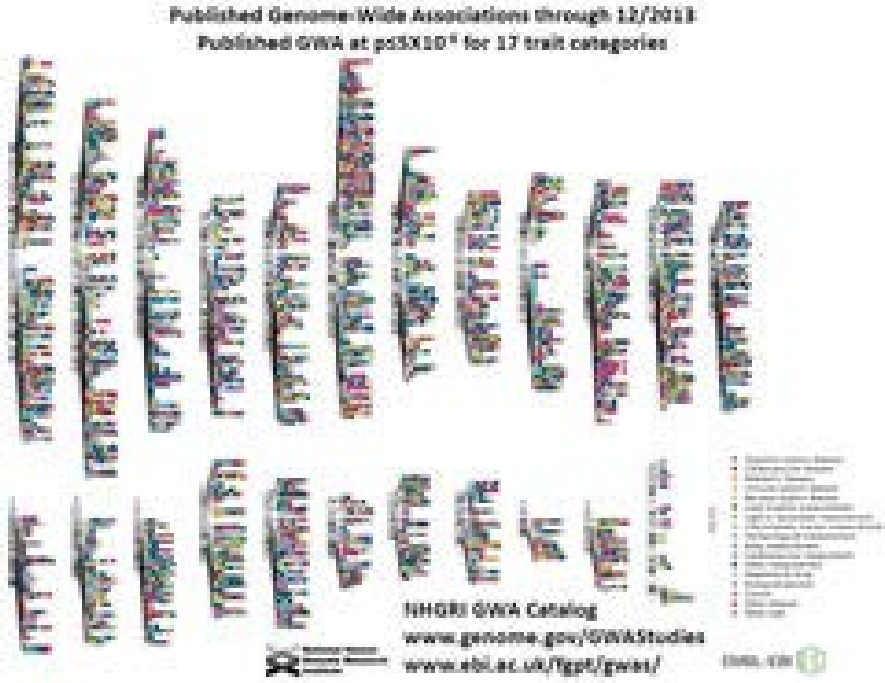


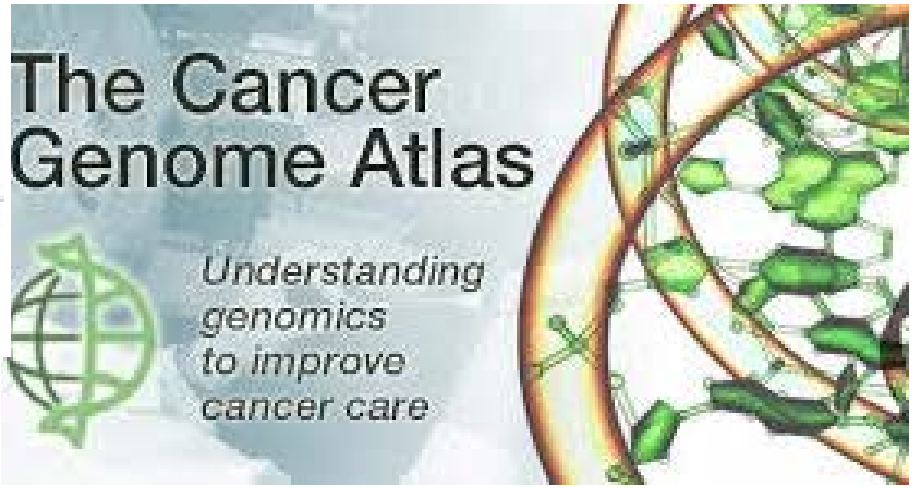
Best et al, Nature Medicine, 2011

Childhood Cancer Survivor Study GWAS

- 5,739 survivors
  - >1,500 subsequent malignancies in 877 persons
- Illumina Omni5M+E SNP chip (>4 million loci)
- Identify genetic variants associated with:
  1) Second cancers after childhood cancer
  2) First primary childhood cancer
  3) Other late adverse effects after childhood cancer

Parallel Investigations: Germline vs. Tissue

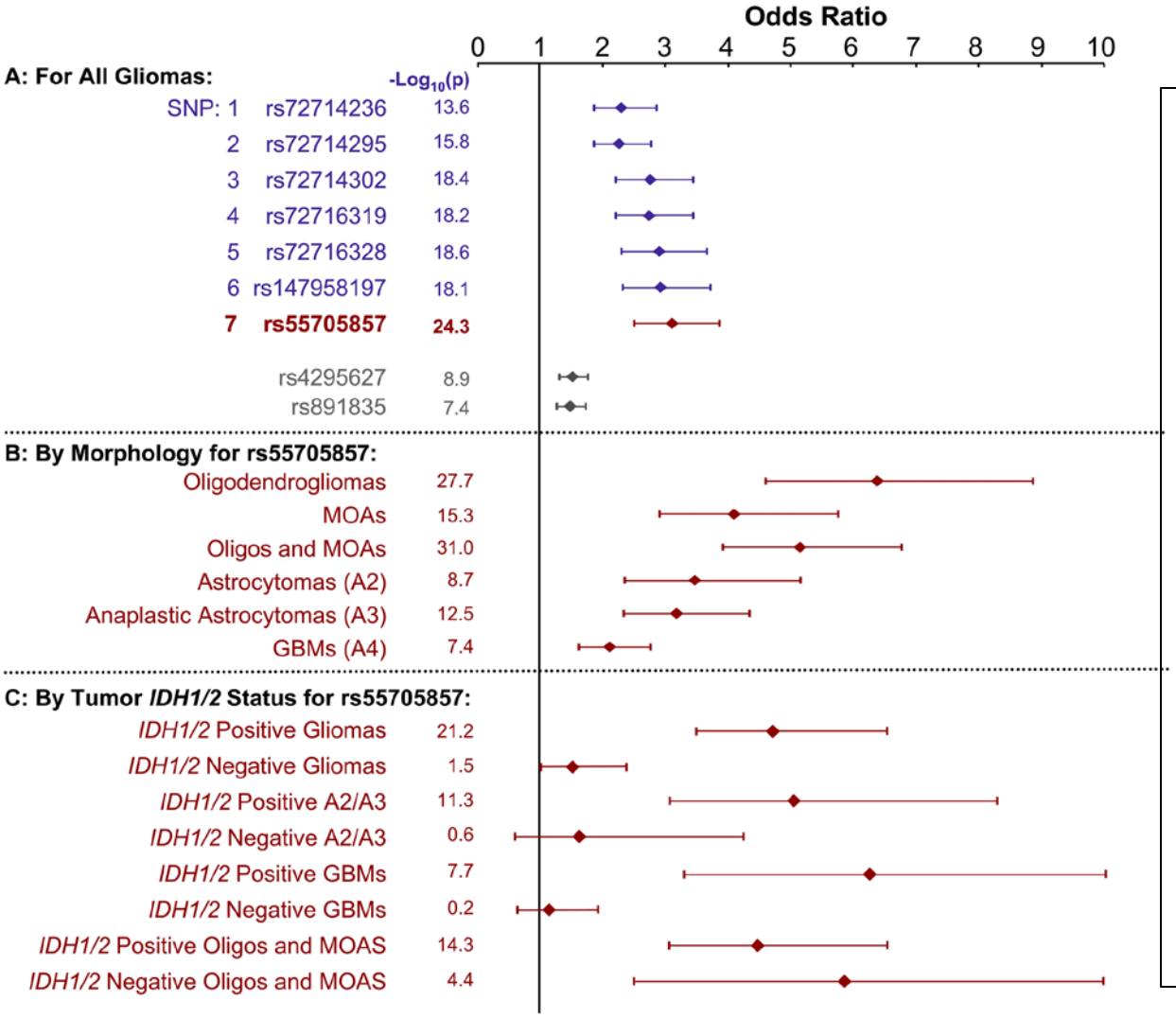




Tumor Sequencing: Molecular Classification

- The Cancer Genome Atlas (2005); International Cancer Genome Consortium (2008)
- Within tumors (>30 types)
  - Gliomablastoma: four distinct subtypes (and specific gene mutations) with different survival and response to treatment
  - Breast cancer: confirmed four main subtypes, with distinct mutations in each. Similarity of basal-like breast cancers and serous ovarian tumors
- Across tumors
  - Common tumor-driving mutations (*BAP1, FBXW7, TP53*) correlated with poor survival across several cancer types

Integrating Germline and Tumor Tissue (example from Glioma)



2008: Tumor

- *IDH1/2* (Isocitrate dehydrogenase)

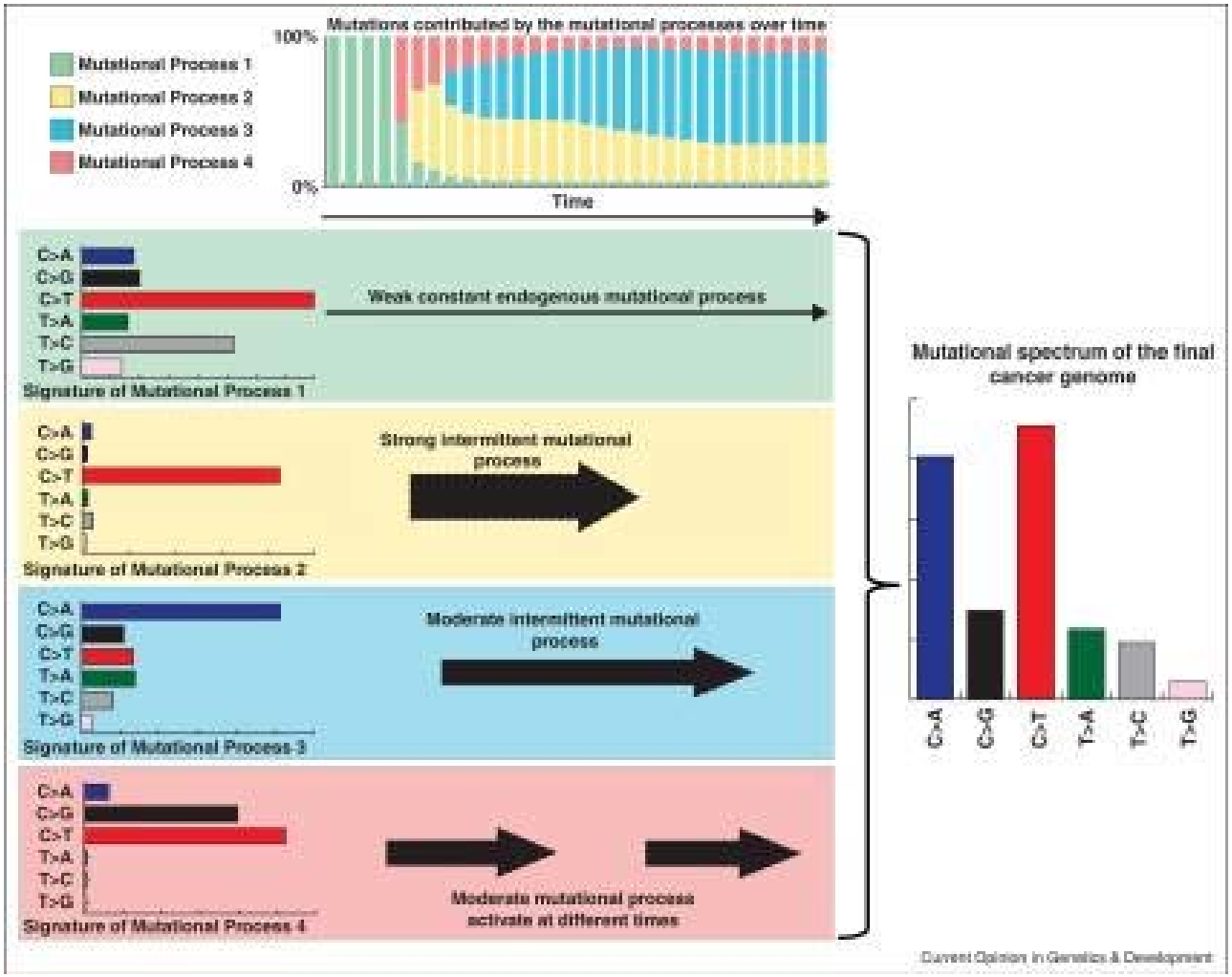
2009: Germline

- 8q24.21, rs429562 *CCDC26*

2012: Jenkins et al, 2012

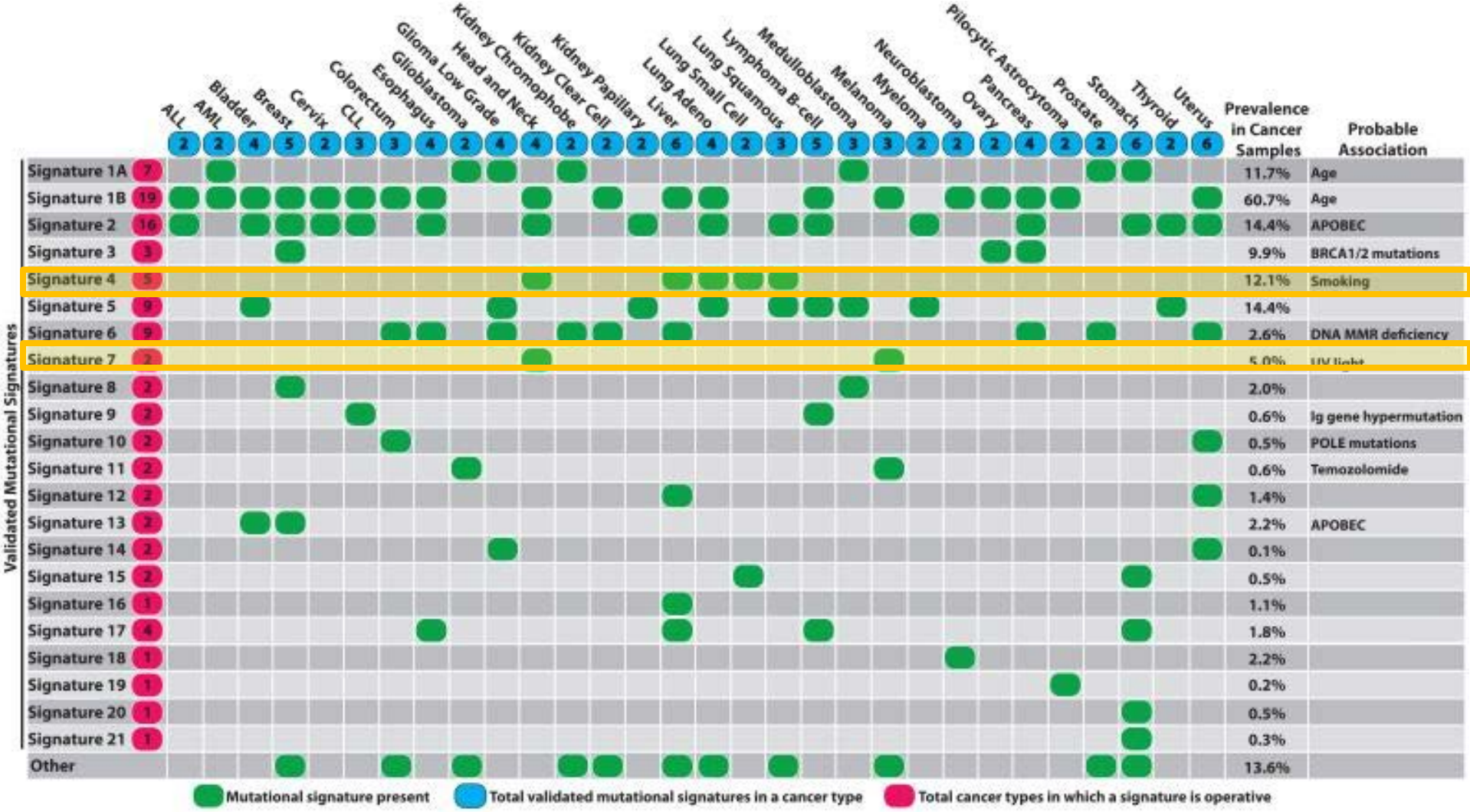
- Used fine mapping to narrow down region
- *IDH* status more predictive of risk associated with locus than histology

The Search for “Mutational Signatures”



Alexandrov, Curr Opin Gen Dev, 2014

Sanger Institute, 7,042 tumor tissue samples



22 validated signatures

Challenges: Genetic Susceptibility to Radiation

- Setting
  - High versus low dose radiation
- Outcome
  - Cancer
  - Intermediate outcomes
- Analytical challenges
  - Power
  - Volume of data, methods need to be developed
- Replication

But, exciting times ahead…

- Continue to explore genetic susceptibility
  - Germline + tumor
  - Whole genome sequencing of special populations
  - Integration of platforms: methylation, RNA, proteins
- Interaction with other risk factors
  - Age, sex, smoking
  - BMI, infection, co-morbidities

Questions and Answers

U.S. Department of Health and Human Services
National Institutes of Health | National Cancer Institute
www.dceg.cancer.gov/RadEpiCourse
1-800-4-CANCER
Produced May 2015