• Overview
  – What is cancer?
  – What sort of genes contribute to cancer phenotypes?

• Genetic approaches to the study of cancer: Is cancer a genetically tractable system?
  – Naturally existing mutations
  – *In vitro* and *in vivo* systems

• Application of genetics research to cancer care (translational research)
  – Clinical cancer genetics
  – Targeted therapies
Cancer Is a Genetic Disease

- Cancers arise as a consequence of the accumulation of alterations in genes that control cell proliferation, differentiation and death
  - some of the genetic changes in tumor cells are unselected (hitchhiking)

- Mutational targets frequently involve key genes and pathways that play critical roles in normal development
• Mutation is consequence of natural processes and in many regards is **not** preventable

• The hypermutable state of the cancer cell affords many opportunities for adaptive evolution and “clinically relevant” disease can be seen as having achieved a high degree of fitness (the “winning” genome)

• Tumor behavior (phenotype) is determined by a complex interplay between the cancer cell genome, the host genome and environment
Cancer is an Oligogenic Multifactorial Trait

• Long lived cells (with stem cell like features) acquire multiple mutations (alterations in DNA make-up) and suffer from numerous environmental insults

• Selected mutations lead to
  - uncontrolled cellular proliferation
  - invasion of neighboring tissues
  - ability to spread or metastasize
Acquired Capabilities of Cancer

Self-sufficiency in growth signals

Evading apoptosis

Insensitivity to anti-growth signals

Sustained angiogenesis

Tissue invasion & metastasis

Limitless replicative potential

The Hallmarks of Cancer

Douglas Hanahan* and Robert A. Weinberg†

*Department of Biochemistry and Biophysics and Hormone Research Institute
University of California at San Francisco
San Francisco, California 94143

†Whitehead Institute for Biomedical Research and Department of Biology
Massachusetts Institute of Technology
Cambridge, Massachusetts 02142
Six Key Defects... Many Ways to Get to the Same End

<table>
<thead>
<tr>
<th>Component</th>
<th>Acquired Capability</th>
<th>Example of Mechanism</th>
</tr>
</thead>
<tbody>
<tr>
<td>🔄</td>
<td>Self-sufficiency in growth signals</td>
<td>Activate H-Ras oncogene</td>
</tr>
<tr>
<td>⌚</td>
<td>Insensitivity to anti-growth signals</td>
<td>Lose retinoblastoma suppressor</td>
</tr>
<tr>
<td>⚽️</td>
<td>Evading apoptosis</td>
<td>Produce IGF survival factors</td>
</tr>
<tr>
<td>💫</td>
<td>Limitless replicative potential</td>
<td>Turn on telomerase</td>
</tr>
<tr>
<td>🌈</td>
<td>Sustained angiogenesis</td>
<td>Produce VEGF inducer</td>
</tr>
<tr>
<td>🍼</td>
<td>Tissue invasion &amp; metastasis</td>
<td>Inactivate E-cadherin</td>
</tr>
</tbody>
</table>

B

Diagram showing a color-coded pathway leading to Cancer.
Emerging Hallmarks and Enabling Characteristics

Emerging Hallmarks

- Deregulating cellular energetics
- Avoiding immune destruction
- Genome instability and mutation
- Tumor-promoting Inflammation

Enabling Characteristics

Hallmarks of Cancer: The Next Generation

Douglas Hanahan¹,²,³ and Robert A. Weinberg³,⁴

646 Cell 144, March 4, 2011 ©2011 Elsevier Inc.
• Cancer phenotype reflect the interplay of *inherited* and *acquired* genetic defects AND environmental insults (*NOTE: both impact the epigenetic state*)

• Important environmental factors include
  
  - age
  - diet
  - lifestyle
  - gender

• Some cancer are *inherited*: a germline defect leads to tumor formation

• Most cancers are *sporadic*: acquired somatic defects
Genetics and Cancers

- 5% inherited mutations
- 15% familial clustering
- 80% sporadic acquired mutations
Few lines of investigation have taught us more about cancer than the study of inherited tumor susceptibility syndromes. Initially, the mutations responsible for these diseases were thought to promote malignancy in a straightforward manner, through inactivation of "tumor suppressor" genes, which directly modulate cell birth or cell death.

not so simple….oncogenes, DNA repair genes, and genes that do not easily fit into a single class

Kinzler and Vogelstein, Science 1998
next-generation sequencing has applications that are immediately relevant to the medical field. In cancer genetics, for example, specific cancer alleles can now be detected in tissues through ultra-deep sequencing of genomic DNA... not so simple....too much data and too few phenotypes...don’t forget there will be opportunity to assess the interaction between inherited and acquired abnormalities

Cancer Causing Mutations Involve Genes That Play Key Roles in Regulating Cell Growth and Maintaining Genetic Integrity

- **tumor suppressor genes**
  - negative growth regulators that inhibit cell growth
- **oncogenes**
  - positive growth regulators that promote cell division
- **DNA repair genes**
  - maintain integrity of DNA
• **tumor suppressor genes**
  *loss of function (null, hypomorph, dominant negative)*
  *typically a cellular recessive defect in tumor cell*

• **oncogenes**
  *gain of function (hypermorph, neomorph)*
  *typically a dominant defect in tumor cell*

• **DNA repair genes**
  *loss of function (null, hypomorph, dominant negative)*
  *typically a cellular recessive defect in tumor cell*
Caretaker genes Inactivation of these genes contributes directly to the neoplastic growth of the tumor; thus, they normally function as gatekeepers" to prevent runaway growth.

Gatekeeper genes Inactivation of a caretaker gene results in a greatly increased mutation rate and is equivalent to a constant exposure to mutagens. It is not surprising that such defects should lead to cancer, but restoration of caretaker function to a cancer cell will not affect its growth. As these indirectly acting genes are never required for neoplasia, most nonhereditary (sporadic) tumors will evolve without them.

Landscapers genes...altered terrain for epithelial cell growth and can be thought of as a "landscaper" defect.

Kinzler and Vogelstein, Science 1998
Is Cancer Genetically Tractable?

Human Populations
(people and the tumors they develop)

Naturally existing mutations

Translation to patient care

in vitro and in vivo models
mouse cell line
Naturally Existing Mutations: Acquired Abnormalities

• Recurrent somatic mutations or mutations in the same pathway can point to key cancer “drivers”

• Genes important in normal development are frequent mutational targets in human cancers
Genetic Classification of Cancer

• Sporadic: acquired (somatic) mutations or epimutations

• Familial: modest increase in risk likely due to genetic and environmental factors

• Hereditary: early, rate-limiting genetic event (mutation) is inherited conferring dramatic increase in risk. Subsequent acquired (somatic defects) are usually necessary for disease development
Naturally Existing Mutations: 
*Inherited Cancer Susceptibility*

- Highly informative in that tumors arising in genetically susceptible individuals begin with the same ‘initiating’ event (more genetic homogeneity)
- Genes important in inherited cancer susceptibility are frequently altered in sporadic tumors and/or point to pathways that are abnormal in tumors
Characteristics of Hereditary Cancer

• Multiple affected individuals within a family
• Early age of onset
• Bilaterality in paired organs
• Other cancers suggestive of a cancer syndrome
Hereditary Cancer Syndromes

• Most often dominant inheritance
  – tumors can be the result of a cellular recessive defect or dominant mutation
  – inherited recessive cancers exist but are less frequently observed

• Variable expressivity

• Variable penetrance
Defective DNA Mismatch Repair & Inherited Cancer Susceptibility

• Inherited DNA mismatch repair loss of function defect lead to cancer susceptibility. The syndromes are variably referred to as HNPCC (hereditary nonpolyposis colorectal cancer) or Lynch syndrome
  – colon, endometrial, urinary, ovarian and other tumors
• DNA tumor phenotype is a consistent feature of tumors in patients with inherited mutations in DNA mismatch repair
  – MSH2, MLH1, PMS2 and MSH6
DNA Mismatch Repair (DMMR)

Recognition of base-base mispairs and single base insertion-deletion mispairs

Recognition of insertion-deletion mispairs (2-3 base pairs)
MSH2 Mutation and Lynch Syndrome

41 Brain ca
29 Colon ca
46 Endo ca

42 Endo ca

69 Renal ca
57 Jejunum
59 Colon ca

50 Pan ca

46 Liver met

92 Ov ca

53 Colon ca
51 Ov ca

52 Endo ca

45 Endo ca
43 Colon ca

36 Endo ca
MSH6 Mutation and Lynch Syndrome

Family 1524
Dominant Inheritance, Cellular Recessive Cancer Defect

Germline (blood) DNA

Tumors

Family 1524
insert
TCAAAAGGGGACATAGAAAA

158 bp (mutant)
139 bp (wild-type)

1524 endometrial cancer
1524:09 pituitary adenoma
DNA Mismatch Repair Defects In Human Cancers

- manifests as an easily recognizable tumor phenotype referred to as microsatellite instability (MSI)
- causally associated with tumor formation confers a ‘mutator’ phenotype (Loeb, 1991)
- common molecular defect in specific cancer types (colon ~15%, endometrial ~28%)
Microsatellite Instability (MSI) Phenotype

- uncorrected strand slippage mutations revealed by PCR analysis of patient tumor and normal DNA
- five reference markers (di- and mononucleotide repeats, Boland et al. 1998)

MSI-H: novel tumor bands with 2 or more markers
MSH-L: novel tumor bands with 1 marker
MSS: no novel tumor bands

\{ Defective \} \{ Normal \}
Microsatellite Instability (MSI) Phenotype

- Uncorrected strand slippage mutations revealed by PCR analysis of patient tumor and normal DNA
- Similar DNA phenotype seen in model organisms
DNA Mismatch Repair

- mutS proteins recognize damage; mutL proteins are central to lesion repair
  - loss of function results in a 100-1000X fold increase in mutation rate

- loss of function abolishes and S-phase apoptotic signal
DNA Mismatch Repair

- mutS proteins recognize damage; mutL proteins are central to lesion repair
- loss of function results a 100-1000X fold increase in mutation rate
DMMR Mutation & Mutator State

Normal

Atypical Hyperplasia

Cancer

Decades

DMMR Mutation

Months or years
DMMR Mutation & Apoptotic Signaling

- Exonucleolytic degradation, replication arrest
- DNA-damage bypass
- ATR/ATRIP/RPA
- MutSα

Cell-cycle arrest and apoptosis

DNA-damage signalling

Jiricny, Nature Reviews Molecular Cell Biology 2006
• Genes important in inherited cancer susceptibility are frequently altered in sporadic tumors and/or point to pathways that are abnormal in tumors.

624 consecutive endometrial cancer cases
(median age 64.6 years, range 26-92)

- MSI-H: 180 (28.8%)
- MSI-L: 20 (3.2%)
- MSS: 424 (67.9%)

Defective DMMR
Microsatellite instability (MIN)

Aneuploidy
Chromosomal instability (CIN)

Most often in sporadic cancers
Epigenetic Change Associated with Loss of MMR in MSI-positive Endometrial Cancers

- Mutation in MMR genes is uncommon

- Methylation of the MLH1 promoter (CpG island) seen in the majority of MSI-positive endometrial cancers and correlates with lack of MLH1 gene expression in sporadic tumors and tumor cell lines

Epigenetic Silencing

Unmethylated Promoter

MLH1

Transcription factor

Hypermethylated Promoter/hypoacetylated

Protein

MLH1

Condensation of promoter chromatin

Hypermethylated Promoter (Condensed)

Transcription factor

Silencing of gene

Image source: http://www.npaci.edu/features/00/Mar/protein.jpg
MLH1 Methylation Correlates with Lack of Expression

MLH1 IHC

MLH1 unmethylated  MLH1 methylated
Is Cancer Genetically Tractable?

Human Populations
(people and the tumors they develop)

Translation to patient care

in vitro and in vivo models
mouse cell line
Mouse Models for Cancer (DMMR)

- Genetically engineered mice with null alleles get tumors....sort of
- Combining genetic defects results in more cancers and ‘predicted’ phenotypes...sort of
- Useful but not entirely satisfactory

*Yeast and bacterial models provide important basic insights*
Cancer Cell Lines for DMMR

- Very helpful for complementation and functional studies
- Somewhat informative for identification of mutations and other abnormalities secondary to loss of MMR
- Useful but not entirely satisfactory…
Is Cancer Genetically Tractable?

Human Populations
(people and the tumors they develop)

Translation to patient care

\textit{in vitro} and \textit{in vivo} models
mouse cell line
• Overview
  – What is cancer?
  – What sort of genes contribute to cancer phenotypes?

• Genetic approaches to the study of cancer: is cancer a genetically tractable system?
  – Naturally existing mutations
  – In vitro and in vivo systems

• Application of genetics research to cancer care (translational research)
  – Clinical cancer genetics
  – Targeted therapies
Clinical Cancer Genetics Research

- MSH2 and MHL1 mutations strongly associated with colon cancer susceptibility (mid ’90s)
- Germline MSH6 mutations are “common -- ~2%” in endometrial cancer patients unselected for family or medical history. MSH6 confers high risk for gynecologic cancers and a much lower risk for colon cancer
  - *MSH6 mutation testing is now standard of care for women suspected to have Lynch syndrome and extended to at risk family members (mid ‘00s)*
- Risk-appropriate cancer surveillance (primarily colon)
- Best approaches: 2009 --- based on family studies
Naturally Existing Mutations and Clinical Phenotypes

Risks of Lynch Syndrome Cancers for *MSH6* Mutation Carriers


- 26% risk for endometrial cancer by age 70
  
  *(compared with 44% for MSH2 and MLH1 cancers)*

- 10% risk for colon cancer by age 70
  
  *(compared with 33% for MSH2 and MLH1 cancers)*

  …and risk for both increases to age 80

Baglietto et al. JNCI 2010
### Table 5. Management for at-risk members of Lynch syndrome families with *MSH6* mutation

<table>
<thead>
<tr>
<th>Management</th>
<th>Recently published recommendations for Lynch syndrome as a whole</th>
<th>Levels of certainty* regarding net benefit for Lynch syndrome as a whole</th>
<th>Recommendations for <em>MSH6</em> mutation carriers by authors of this article</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Cancer screening options</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Colonoscopy</td>
<td>Every 1–2 y beginning at age 20–25 y (age 30 y in <em>MSH6</em> families) or 10 y younger than the youngest age at diagnosis in the family, whichever comes first (33); every 1–2 y starting at age 20–25 y for men and age 30 y for women (34); every 1–2 y starting at ages 20–25 y (3)</td>
<td>High†</td>
<td>Every 1–2 y beginning at age 30 y‡</td>
</tr>
<tr>
<td>Endometrial sampling</td>
<td>Every year beginning at age 30–35 y (33); every 1–2 y starting between ages of 30 and 35 y (34)</td>
<td>Moderate (when combined with transvaginal ultrasound) (35)</td>
<td>Every year beginning at age 30–35 y‡</td>
</tr>
<tr>
<td>Transvaginal ultrasound for endometrial and ovarian cancers</td>
<td>Every year beginning at age 30–35 y (33); every 1–2 y starting at age 30–35 y (34); every 1–2 y starting between ages of 30 and 35 y (3)</td>
<td>Poor</td>
<td>Every year beginning at age 30–35 y. Role of serological markers for ovarian cancer screening is uncertain</td>
</tr>
<tr>
<td>Urinalysis with cytology</td>
<td>Every 1–2 y beginning at age 25–35 y (33) or beginning at age 50 y (34); every 1–2 y starting between ages of 30 and 35 y if urinary tract cancer runs in family (3)</td>
<td>Poor</td>
<td>Consider every 1–2 y beginning at age 40 y</td>
</tr>
<tr>
<td>Gastroduodenoscopy</td>
<td>“Could be offered periodically” (33); every 1–2 y starting at age 30–35 y if it occurs two or more times in the family (34); every 1–2 y starting between ages of 30 and 35 y if gastric cancer runs in family or in countries with high incidence of gastric cancer (3)</td>
<td>Poor</td>
<td>No evidence of increased risks except by analogy to other genes causing Lynch syndrome</td>
</tr>
<tr>
<td><strong>Surgical considerations</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Colorectal resection (segmental vs subtotal colectomy vs complete proctocolectomy)</td>
<td>For at-risk persons without colorectal cancer; generally not advised. Discuss as alternative, with preferences of well-informed patient actively elicited. For persons with a diagnosis of colorectal cancer or polyp</td>
<td>Poor</td>
<td>No change in recommendations</td>
</tr>
</tbody>
</table>

*Levels of certainty: High, Moderate, Poor.*
Human Populations (people and the tumors they develop)

Naturally existing mutations in tumors

Translation to patient care

in vitro and in vivo models
mouse cell line
Endometrial Cancer

- most common female genital tract malignancy, with 42,000 new diagnoses in 2010
- ~7,700 deaths

Abnormal proliferation of glands with loss of stroma
Proliferative

Estrogen driven proliferation of epithelium is regulated by growth factors produced by the stroma

Secretory

Progesterone driven process in which proliferation is terminated and both components differentiate
Cancers and Precancerous Tissues are Accessible and Available for Investigation

Estrogen

(Type I)

SH → CH → CAH → Endometrioid Ca

Histologically recognizable precancers

Normal epithelium

SH: simple hyperplasia
CH: complex hyperplasia
CAH: complex atypical hyperplasia
EIC: endometrial intraepithelial cancer
Molecular Genetic Alterations

<table>
<thead>
<tr>
<th>Oncogenes</th>
<th>DNA Repair</th>
<th>Suppressors</th>
<th>UEC</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>PTEN</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>MSI</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>KRAS2</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>TP53</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>FGFR2</td>
</tr>
</tbody>
</table>
Endometrial Tumorigenesis

- Normal epithelium

Estrogen

- PTEN
- MSI

SH → CH → CAH → Endometrioid Ca

FGFR2?

• loss of DNA mismatch repair (MSI) is strongly associated with oncogenic activation of FGFR2
## Endometrial Tumorigenesis

<table>
<thead>
<tr>
<th></th>
<th>Type I</th>
<th>Type II</th>
<th>p-value*</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PTEN</strong></td>
<td>62%</td>
<td>0</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>MSI</strong></td>
<td>28%</td>
<td>0</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>KRAS2</strong></td>
<td>26%</td>
<td>2%</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>TP53</strong></td>
<td>17%*</td>
<td>93%</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>FGFR2</strong></td>
<td>15%</td>
<td>2%</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>
FGFR2 Mutations in Endometrial Cancer

Frequent activating FGFR2 mutations in endometrial carcinomas parallel germline mutations associated with craniosynostosis and skeletal dysplasia syndromes

PM Pollock¹, MG Gartside¹, LC Dejeza², MA Powell³, MA Mallon⁴, Cancer Genome Project⁵, H Davies⁵, M Mohammadi⁶, PA Futreal⁵, MR Stratton⁵, JM Trent⁷ and PJ Goodfellow³⁴

- FGFR2 is a context-dependent oncogene (functions as a tumor suppressor in melanoma)
Somatic Mutations in Endometrial Cancer and Germline Mutations in Craniosynostosis Syndromes

• nature has already taught us a great deal about function of mutations
FGFR2 Signaling In the Endometrium

Estrogen driven proliferation of epithelium is regulated by growth factors produced by the stroma.

Progesterone driven differentiation of both stroma and epithelium.
FGFR2 Signaling In the Endometrium

Proliferative

Estrogen driven proliferation of epithelium is regulated by growth factors produced by the stroma

Secretory

Progesterone driven process in which proliferation is terminated and both components differentiate

- FGFs and FGFR important in this process
- Specificity of signaling is provided by tissue and cell-specific expression of receptors, ligands and heparan sulphate proteoglycans
IHC

- FGFR2 (IIIb isoform)
- FGF1

- cross talk between epithelial and mesenchymal components (paracrine signaling) through ERK pathway
Mutations in FGFR2 in 12-16% of endometrioid cancers.

Mutations in KRAS2 in ~15% of endometrial cancers. Mutations in KRAS2 are mutually exclusive with mutations in FGFR2.

Mutations in BRAF are extremely rare.

ERK is activated (phosphorylated) in >50% of endometrial cancers.

ERK targets - effectors
• most mutations activate the kinase domain: no ligand binding required
• S252W creates an autocrine loop in which FGFs produced by epithelium signal the same epithelial cells

S252W FGFR2 results in IIIc isoform ligand binding...making it the mesenchymal isoform
Inhibition of Activated Fibroblast Growth Factor Receptor 2 in Endometrial Cancer Cells Induces Cell Death Despite PTEN Abrogation

Sara A. Byron, Michael G. Gartside, Candice L. Wellens, Mary A. Mallon, Jack B. Keenan, Matthew A. Powell, Paul J. Goodfellow, and Pamela M. Pollock


Drug-sensitive FGFR2 mutations in endometrial carcinoma


*Department of Medical Oncology and †Center for Cancer Genome Discovery, Dana-Farber Cancer Institute, Boston, MA 02115; ‡Department of Medicine, Brigham and Women's Hospital and Harvard Medical School, Boston, MA 02115; §Department of Pathology, Harvard Medical School, Boston, MA 02115; ¶The Broad Institute of MIT and Harvard, Cambridge, MA 02142; †Department of Clinical Medicine and ‡The Gade Institute, Section for Pathology, University of Bergen, N-5020 Bergen, Norway; Departments of **Pathology and ††Obstetrics and Gynecology, Faukland University Hospital, N-5020 Bergen, Norway; and ‡‡Novartis Institutes for Biomedical Research, Cambridge, MA 02139

Communicated by R.L. Erikson, Harvard University, Cambridge, MA, April 9, 2008 (received for review December 29, 2007)

• oncogene addiction/dependence
Reversal of Phenotypes Associated with FGFR2 Mutation

RNA interference and inhibition of MEK-ERK signaling prevent abnormal skeletal phenotypes in a mouse model of craniosynostosis

Vivek Shukla¹,³, Xavier Coumoul¹–³, Rui-Hong Wang¹, Hyun-Seok Kim¹ & Chu-Xia Deng¹

• mouse model points to clear therapeutic potential for inhibiting FGFR2
Recurrent somatic mutation in endometrioid endometrial cancers identified by our group

Figure 4: ERK phosphorylation and gene expression of wild-type and R2-S252W mutant mice upon treatment with U0126. (a) ERK phosphorylation was reduced in wild-type and R2-S252W mutant mice after 18 h of treatment with U0126. (b-e) In response to the reduction of ERK phosphorylation, relative expression of Dusp6 (b), Spy1 (c), Spy2 (d) and Spy4 (f) were also downregulated, whereas there was no observed difference in the expression of Spy3 (e). The y-axis represents arbitrary units of relative expression based on real-time RT-PCR analysis. Each time point represents one mouse. All samples were from the thymi of P15 mice. At least three mice were analyzed for each genotype. Error bars refer to s.d.

• Shukla et al. 2007
We are on it!

Cancer drugs to treat birth defects

Andrew O M Wilkie

Identical mutations of the same genes can lead either to congenital malformations or to cancer, depending on their cellular and temporal context. The demonstration of activated RAS-ERK signaling in a mouse model of Apert syndrome suggests that drugs designed to inhibit this pathway in cancer may also delay the progression of several serious pediatric syndromes.

Until a few years ago, the title above might have seemed to belong only in the headlines.

Andrew O.M. Wilkie is at the Weatherall Institute of Molecular Medicine, University of Oxford, Oxford, UK. e-mail: awilkie@hammer.imm.ox.ac.uk

of the tabloid press. However, as the genes mutated in birth defects and cancer have been identified, and the details of how these mutations disturb the regulation of biochemical pathways have been explained, a remarkable convergence in their underlying cellular mechanisms has been uncovered. This is well
Human Populations
(people and the tumors they develop)

Cooperative Group recruitment of patients
with recurrent or persistent endometrial
carcinoma to a phase II trial

Translation to patient care
PROTOCOL GOG-0229I
A PHASE II EVALUATION OF BRIVANIB (BMS582664, IND#105029), AN ORAL, MULTITARGETED GROWTH FACTOR TYROSINE KINASE INHIBITOR IN THE TREATMENT OF RECURRENT OR PERSISTENT ENDOMETRIAL CARCINOMA (BMS Study CA182-029)

PROTOCOL GOG-0229H
A PHASE II EVALUATION OF AZD6244 (NSC#748727, IND #77782) IN THE TREATMENT OF RECURRENT OR PERSISTENT ENDOMETRIAL CARCINOMA
Genetics and Cancers

- 5% inherited mutations
- 15% familial clustering
- 80% sporadic acquired mutations
What Are We Missing?

Potential a great deal. Many cancers result from as yet unidentified large effect risk alleles

Why have we not found the missing big effect risk alleles?
- They are rare and we have not looked hard enough
- Wrong approaches

Biased assessment of genome
- Candidate genes
- Searches focused on exons and single base changes and/or small in/del alterations
- Sequence-centric searches

Looking in the wrong places
- ...phenotypes rule, but we do not know what the hallmarks are for these sorts of uncommon, high-penetrance risk alleles
- Similarly affected family members
- Early-onset of disease
- Synchronous and metachronous malignancies
Rare Large Effect Genetic Variants Explain Cancer Susceptibility
Investigation of The Cancer Genome

First observations that the material of inheritance was abnormal in cancer cells and consequent proposal that cancers are clones arising due to somatic changes

Identification of DNA as the material of inheritance

Description of the double helical structure of DNA

First recurrent chromosomal rearrangement in cancer

First somatic driver mutation and first cancer gene identified

Second-generation sequencing technologies

First sequence of all exons in a cancer

First complete cancer genome sequence

400 known cancer genes

Thousands of complete cancer genome sequences

Cancer genome sequences as a routine diagnostic?

Stratton et al., Science; 331:1553-8, 2010
Deep Sequencing Candidate Genes

Works when you know what you are looking for....

Detection of inherited mutations for breast and ovarian cancer using genomic capture and massively parallel sequencing

Tom Walsha, Ming K. Leea, Silvia Casadeia, Anne M. Thorntona, Sunday M. Straya, Christopher Penniib, Alex S. Norda, Jessica B. Mandella, Elizabeth M. Swisherb, and Mary-Claire Kinga,1

aDepartments of Medicine and Genome Sciences and bObstetrics and Gynecology, University of Washington, Seattle, WA 98195

www.pnas.org/cgi/doi/10.1073/pnas.1007983107
Exome Sequencing

Promising and low cost approach to discovery of susceptibility genes

“Genome-wide screening for mutations remains the most effective and unbiased way to discover genes involved in complex illnesses.”

“…although the power of sequencing is enormous, genetic heterogeneity remains a daunting challenge. With next-generation sequencing technology, the issue is not finding potentially deleterious mutations but rather determining which of many potential deleterious mutations in an individual play a role in disease”

McClellan and King, Cell;141:210-7, 2010
Start With What We Know

Cancer

Phenotypic extremes

constitutional genome
mutations that accumulate over a lifetime
environment
Identifying Critical Mutations

- Evolutionary conservation
- Predicted change in function
- Functional characterization
- Co-segregation in families
- Identification of additional alleles – sequence more?
Whole Genome Sequencing

Recent WGS from Link group, WUSM, identified a germline deletion in a known cancer gene in a patient with three cancers (Br, Ov and leukemia) who had tested negative for BRCA1 and BRCA1 mutations.
Whole Genome Sequencing....
Somatic Mutations
Acute myelogenous leukemia (AML) tumor and normal whole genome sequencing (focus on single base changes and small insertions/deletions)

10 mutations discovered, two of which were previously known

*Ley et al., Nature 2008*

A second AML case (AML2) sequenced at even higher coverage (23X)

Improvements in data analysis and bioinformatics made it possible to look more deeply at changes – and more insertions/deletions/breakpoints and large-scale rearrangements

12 mutations, three of which were seen when other cancers checked for these

- NRAS (G12D)
- NPMc (4 bp ins)
- IDH1 (R132C)

*Mardis et al. NEJM 2009*
Just Scratching The Surface

- A major data problem…a lot of sequences changes for a few specimens
- Crude phenotypes
- What is signal, what is noise….

paths, pools, functional studies…