Clinical Colorectal Cancer Genetics for the General Surgeon

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No financial disclosures

Cancer is a Genetic Disease

- Cancer is in essence a genetic disease
- Cancer is not an event, it is a multistep process
- Genetic mutations contribute to, rather than cause cancer
- Alterations in cancer cell DNA
  - ↑ cell proliferation
  - ↓ cell death (apoptosis)
  - Local invasiveness
  - Metastatic spread
- Most cancers result from mutations in somatic cells
  - Sporadic colorectal cancer
- Some cancers result from mutations in germline cells
  - Inherited colorectal cancer (Lynch, FAP, MAP, JPS, PJS)

Colorectal Cancer Genetics

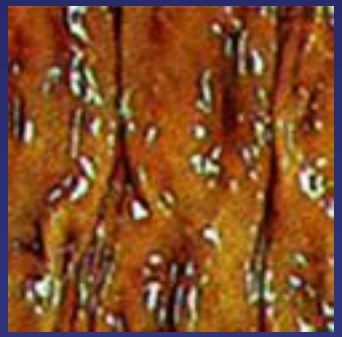
- Pubmed: colorectal AND cancer AND genetics = >10,000 articles in the past 5 years

Enormous topic

Today's talk:

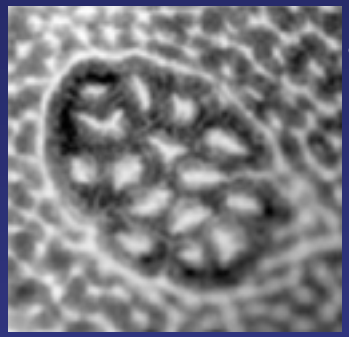
- Selected, clinically relevant aspects of colorectal cancer molecular genetics:
  - Cancer treatment – medical & surgical issues
  - Cancer prognosis
  - Response to therapy
- Genetic emphasis:
  - Microsatellite instability & DNA mismatch repair
  - EGFR & VEGF signaling pathways

The Adenoma to Carcinoma Sequence: Multiple Genetic Alterations



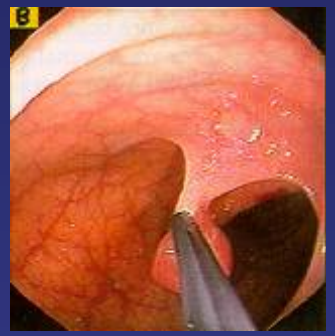
Normal Mucosa

- APC mutation & 5q loss
- MMR-deficiency
- K-Ras or BRAF mutation



Aberrant Crypt Focus

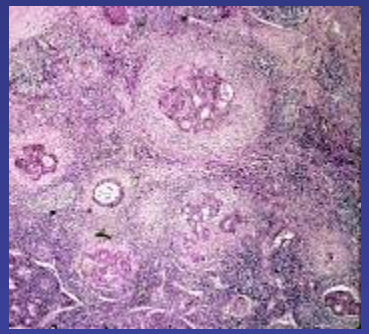
- Global Hypomethylation
- COX-2 overexpression



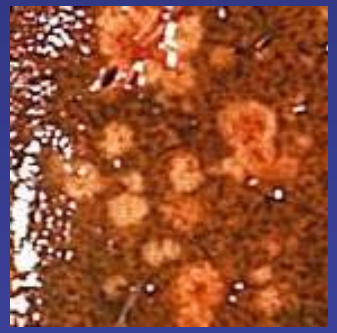
Adenomatous Polyp

- Loss of 18q SMAD2,4,DCC?
- p53 mutation & 17p loss

Invasive Colorectal Cancer



Lymph Node Metastasis



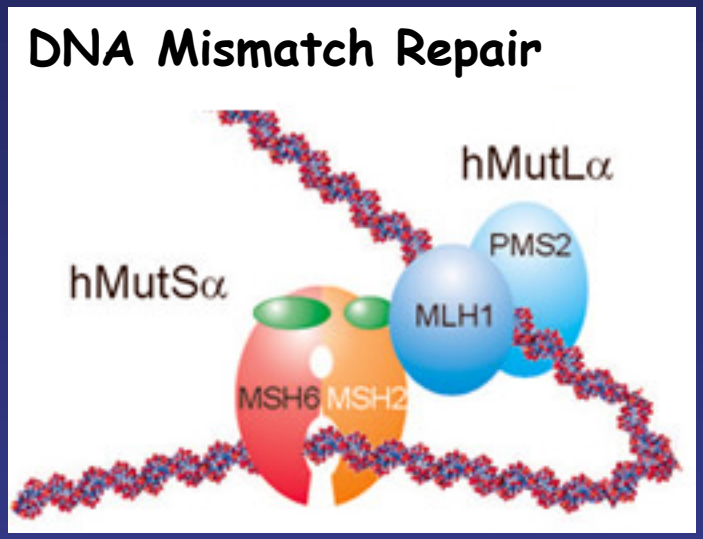
Distant (Liver) Metastasis

Mutator predisposition pathways → multiple genetic alterations

Microsatellite Instability (MSI)
DNA Mismatch Repair (MMR)

Lynch Syndrome &
Sporadic Colorectal Cancer

The MSI Mutator Pathway



Microsatellite DNA

CA

---CACACA CACA---
---GTGTGT GTGT---

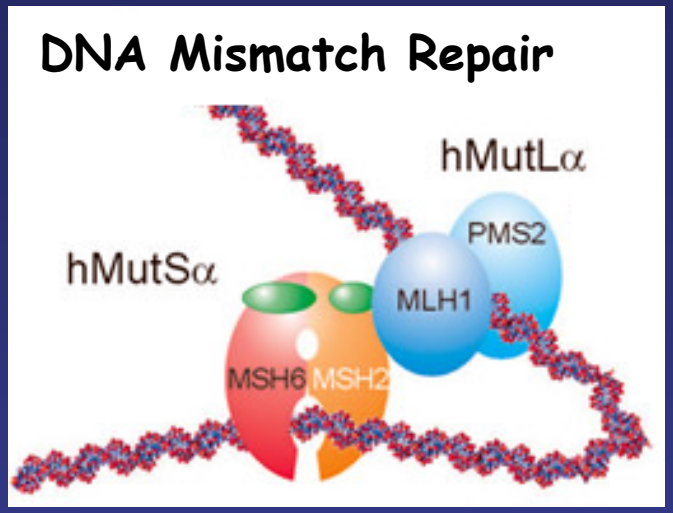
Mismatch repair deficiency

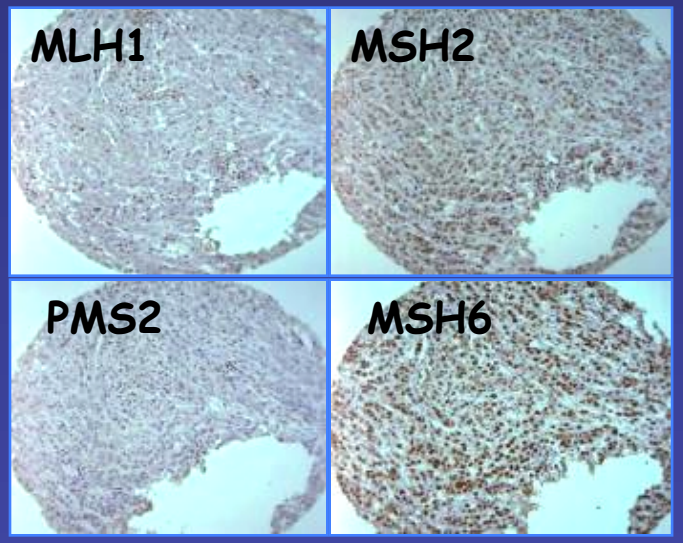
- Loss of function of one MMR gene:
  MLH1, MSH2, MSH6, PMS2
- → MSI
- 15% of sporadic colorectal cancer
- Lynch syndrome (2-4%)
- ↑ ↑ specific mutations
  (i.e. BRAF, TGFBRII, CTNNB1)

	MSI	MSS
Proximal to SF	80%	42%
AJCC I/II	77%	52%
Poor grade	32%	6%
Mucinous	30%	10%
Signet ring	26%	8%

all p<0.0001
Yamuachi Gut 2012
1,443 colorectal cancers

MMR Immunohistochemistry

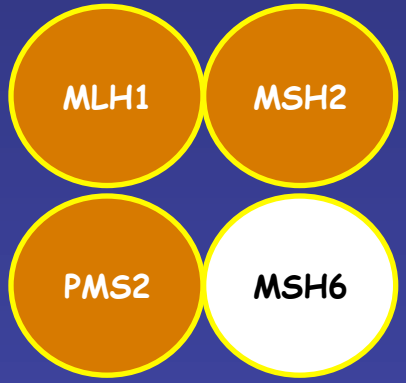




MLH1-deficient
15% of sporadic CRC
45% of Lynch syndrome

MSH2-deficient
45% of Lynch syndrome

PMS2-deficient
<5% of Lynch syndrome

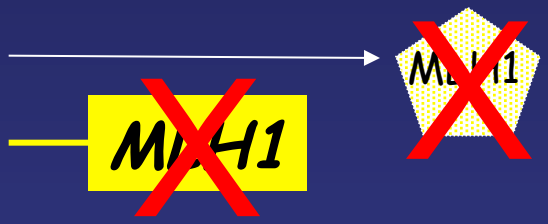


MSH6-deficient
<10% of Lynch syndrome

MMR IHC can help guide genetic testing & clinical management

What is Sporadic MSI Colorectal Cancer?

Normal
- promoter unmethylated
- gene transcribed & translated

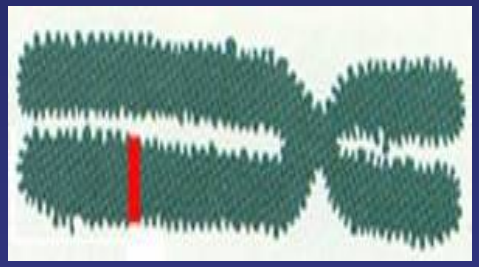


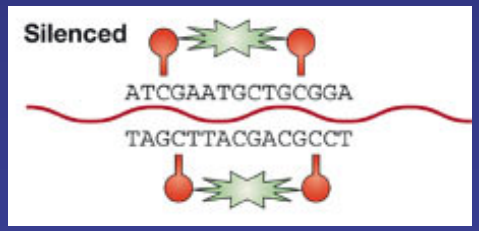
Lynch MSI CRC
- MLH1 mutated
- ~45% of Lynch (MSH2, MSH6, PMS2)

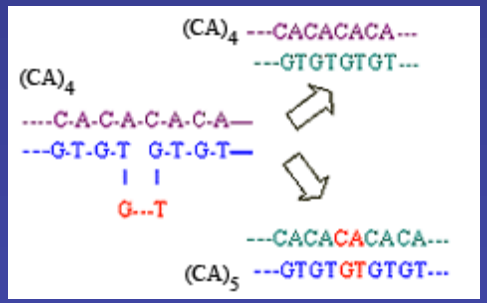
Sporadic MSI CRC
- promoter hypermethylated
- transcription blocked
- ~15% of sporadic CRC

CpG Island Methylator Phenotype
- Epigenetic CIMP pathway
- 20-30% of colorectal cancers
- often older, female, right-sided
- often BRAF mutations

Colorectal Cancer Mutator Pathways







Chromosomal Instability Pathway (CIN)

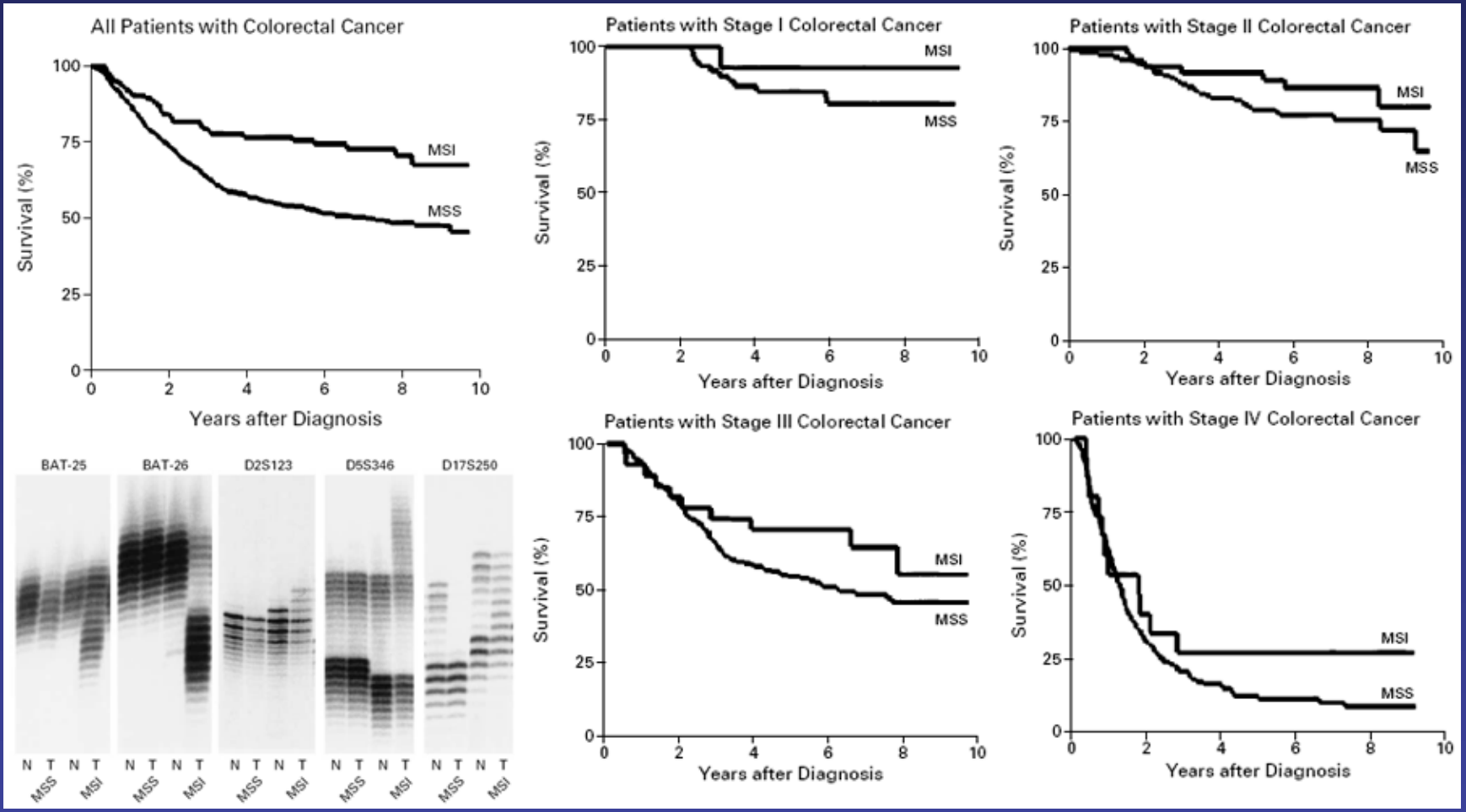
CpG Island Methylator Pathway (CIMP)

Microsatellite Instability Pathway (MSI)

	Sporadic	Inherited
CIN/MSS	80%	<1% FAP
CIN/CIMP	5%	not reported
MSI/CIMP	15%	rare?
MSI	rare	2-4% Lynch

Clinical Implications of MSI Beyond Lynch Syndrome

MSI & Colorectal Cancer Survival

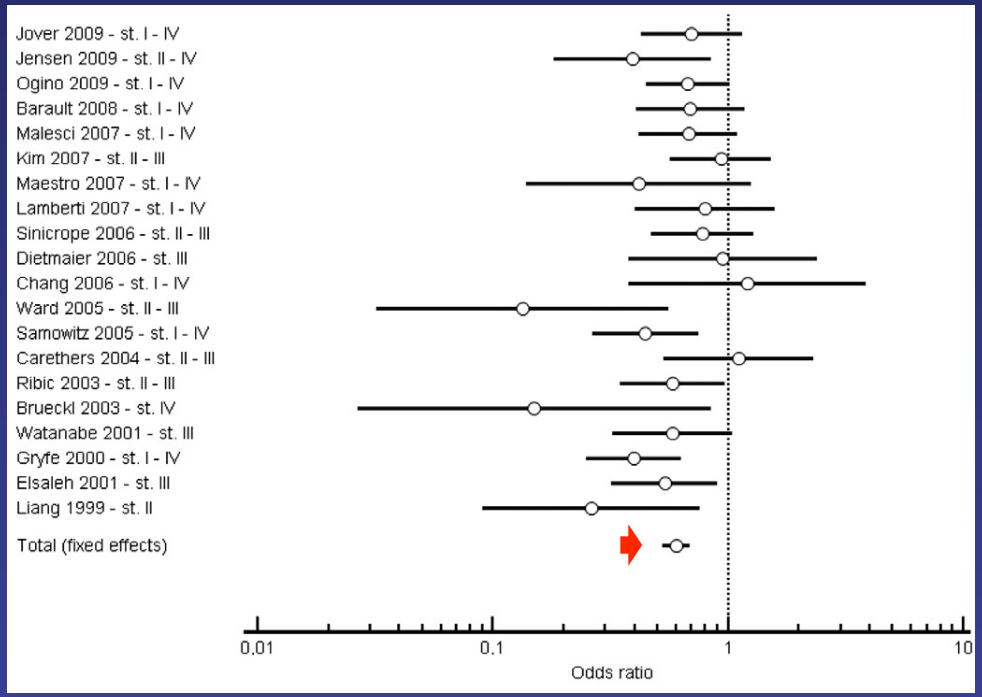


Cox Proportional Hazards MSI vs MSS = 0.45 (0.30-0.68), p<0.001

MSI is prognostic of independent, multivariate improved survival

Gryfe NEJM 2000

MSI: Colorectal Cancer Prognosis



Guastadisegni EJC 2010

- meta-analysis
- 20 studies
- 9,243 patients

Hazard Ratio

MSI-H vs MSS = 0.60 (0.53-0.69)

- association maintained across cancer stages
- no evidence of:
  - publication bias
  - study heterogeneity

MSI is associated with an improved prognosis in colorectal cancer

Colorectal Cancer Molecular Genetics and Therapy

Why Do We Need Predictive Biomarkers?

Stage II/III	DFS	OS	
Surgery alone	55%	64%	
• 5yr benefit FULV	+12%	+ 7%	Gill JCO 2004
• additional 5yr benefit FOLFOX	+ 6%	+ 0-5%	Andre JCO 2009
	+18%	+7-17%	

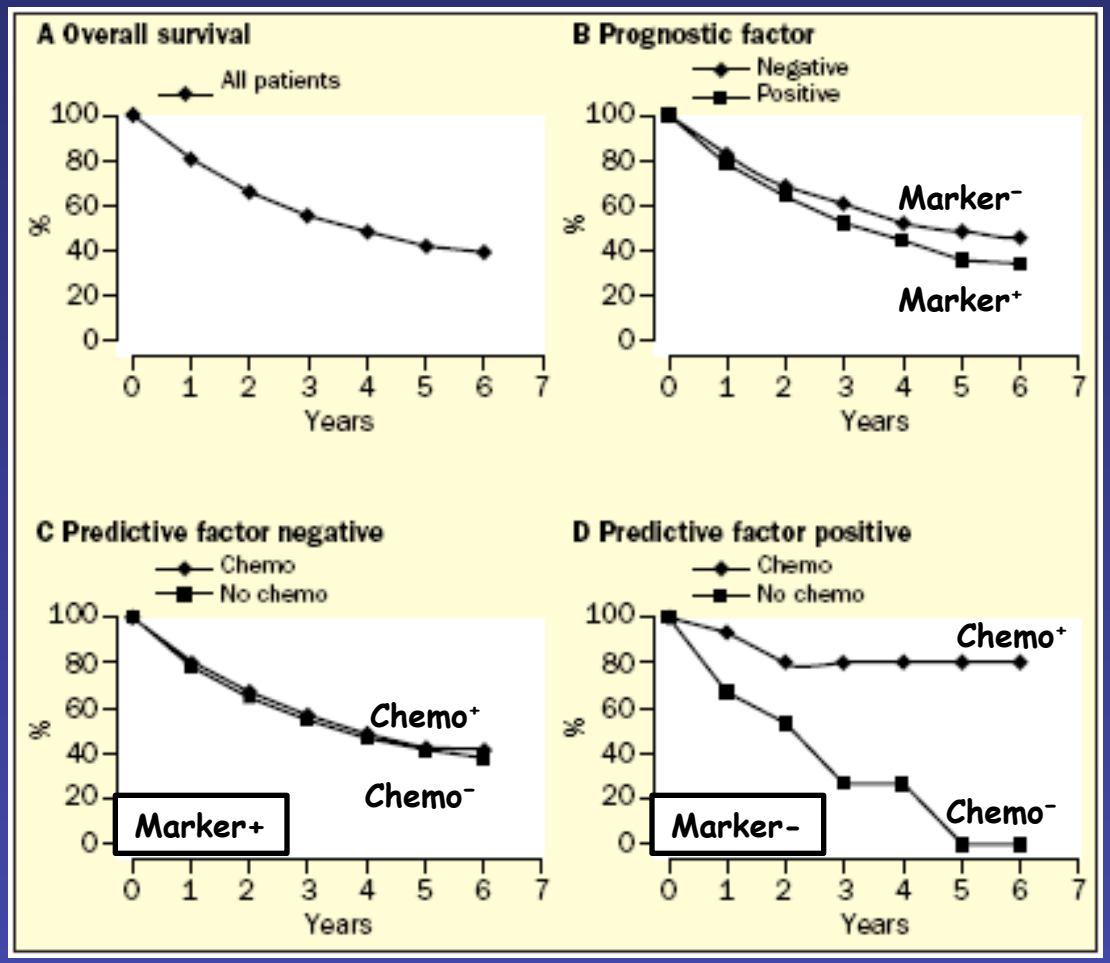
82-93% do not benefit from adjuvant chemotherapy

- 55-64% cured by surgery alone & will never benefit
- Toxicity (grade III/IV): FULV >20%, FOLFOX >40%
- ↑ complications & cost with FOLFOX

Prognostic & Predictive Biomarkers

Prognostic – marker status is associated with a difference in clinical outcome
- cancer characteristic

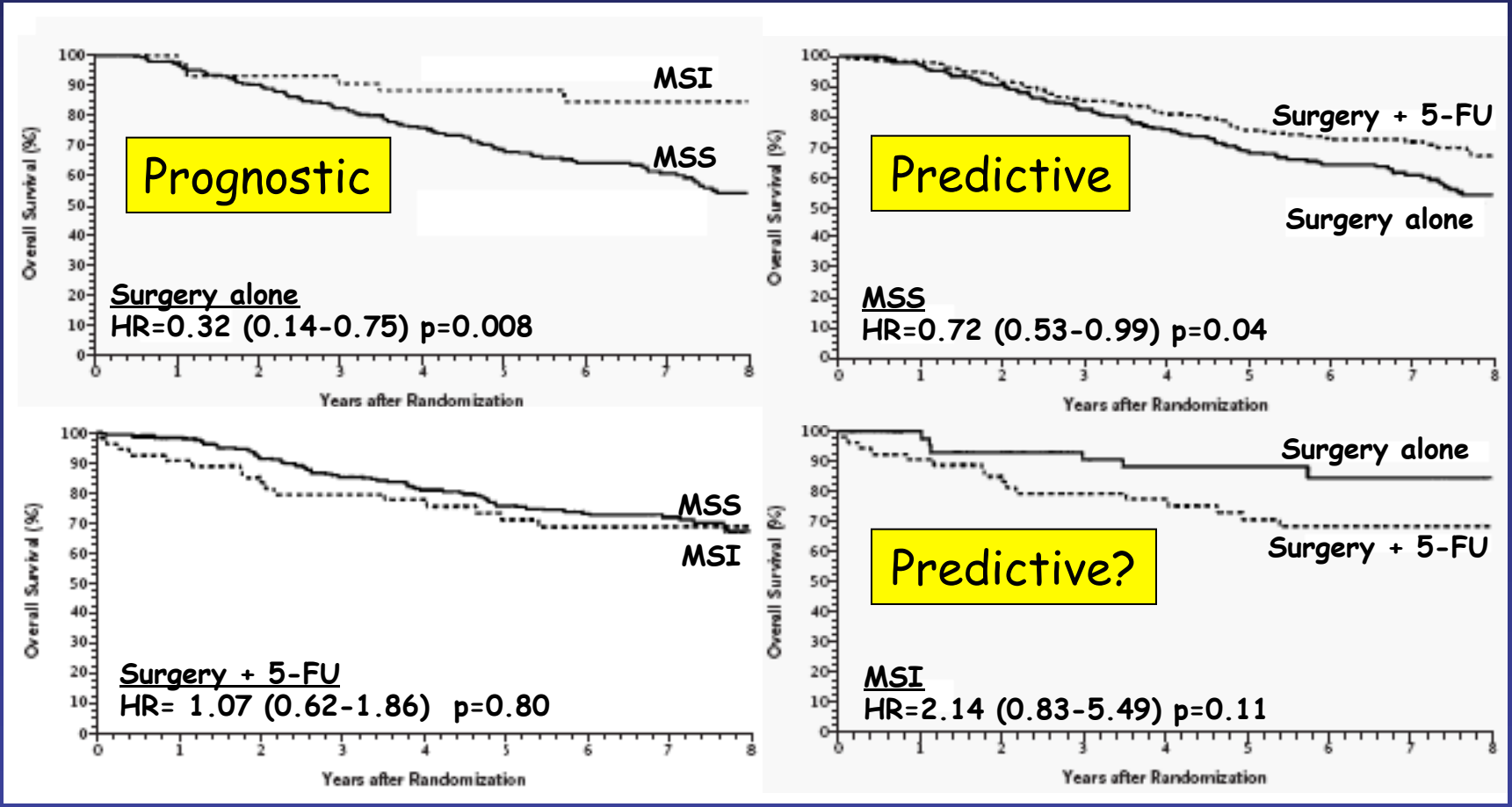
Predictive – marker status is associated with a difference in response to treatment
- more complex cancer-treatment characteristic



Predictive Studies:

- Both treated & untreated patients are necessary
- Surgery only arm required to determine which patients benefit from FULV (7-12%)
- Ethical dilemma in an era where FOLFOX is the standard of care (but benefits 7-18% of patients)

MSI & Predicting 5-FU Response



MSS, but not MSI, is predictive of improved survival with adjuvant 5-FU compared to surgery alone

Ribic NEJM 2004

MSI: Predicting 5-FU Response

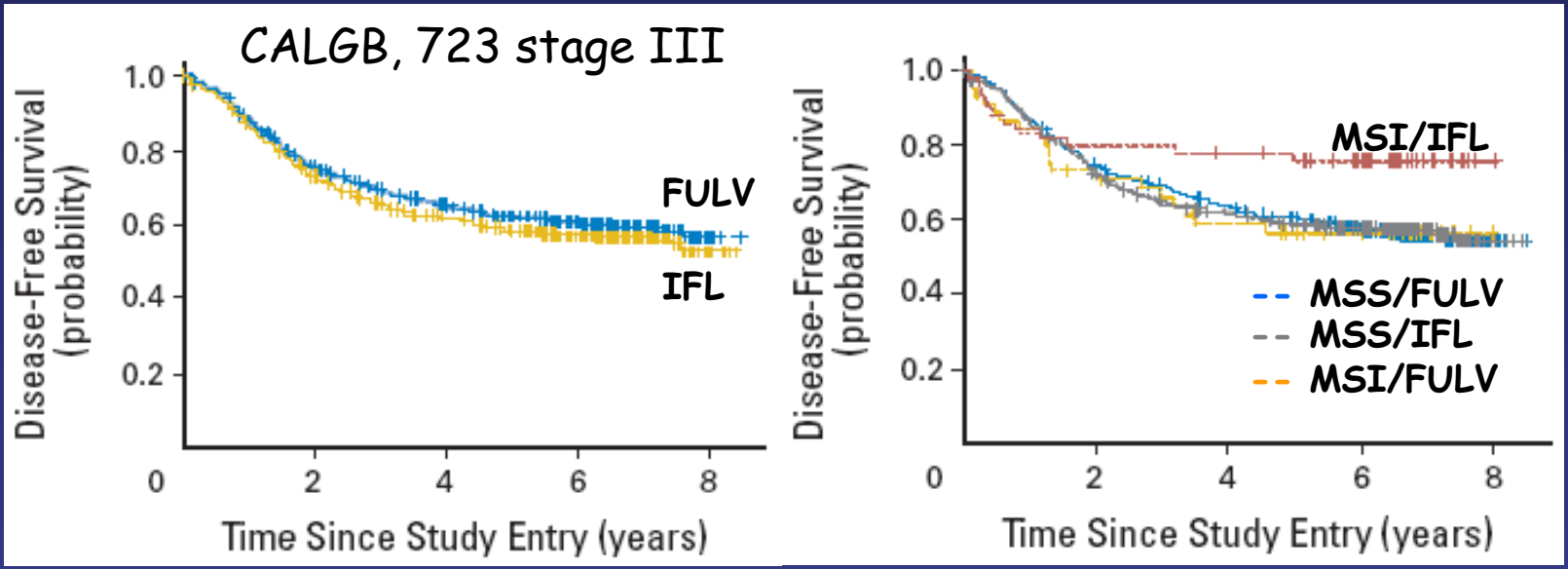
Study	Journal	Patients	MSI (%)	Good Prognosis	Predicts 5-FU Benefit
Sinicrope, 2011	JNCI	2,141	16	MSI	MSS/LS MSI**
Hutchins, 2011	JCO	1,913	11	MSI	No
Ohrling, 2010	Acta Oncol	1,006	16	No	No**
Ribic, 2004	NEJM	570	17	MSI	MSS
Kim, 2007	JCO	542	18	No	No
Halling, 1999	JNCI	508	15	MSI	No
Sargent, 2010	JCO	457	15	MSI	MSS
Barratt, 2002	Lancet	368	24	No	MSS***
Storojeva, 2005	Onc Rep	160	NA	No	No

* MSS & LS MSI benefit, not sporadic MSI
** Negative 5-FU RCT
*** trend

MSS predicts 5-FU benefit: 2-4 of 9 RCTs
MSI benefit from 5-FU: 0 of 9 RCTs

Adjuvant FOLFOX not recommended for MSI AJCC II

MSI & Adjuvant Irinotecan?



Bertagnolli, *JCO* 2009

MSI-Status	Rx	5-yr DFS	HR (95% CI)
MSI	FULV	57%	
	IFL	76%	0.52 (0.25-1.07), p=0.07
MSS	FULV	61%	
	IFL	59%	1.01 (0.79-1.29)

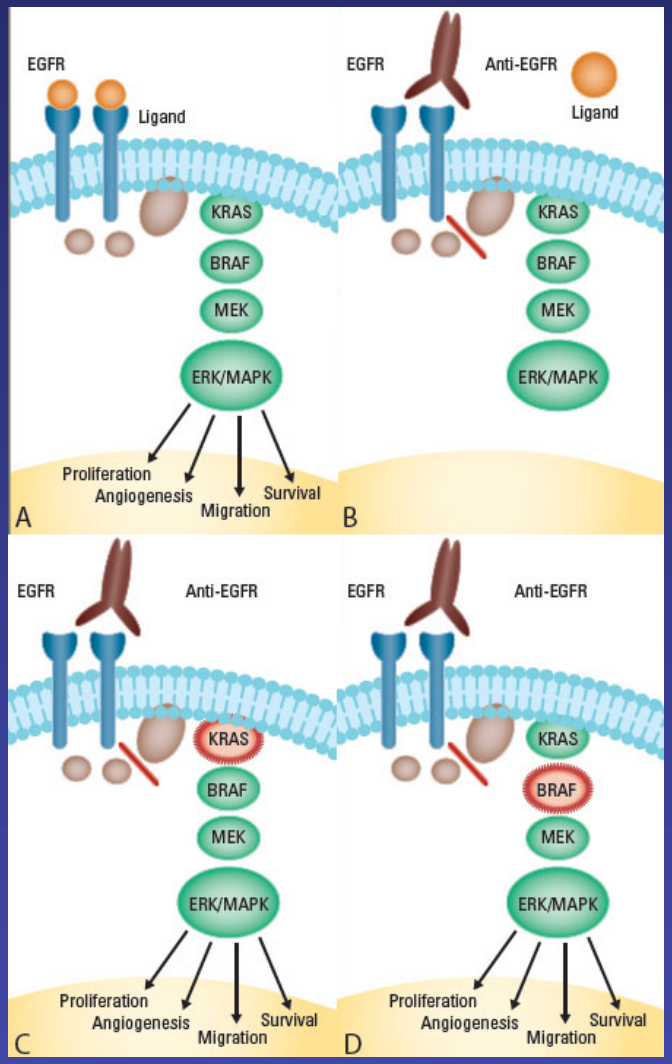
- Suggests that MSI is predictive of improved survival with irinotecan
- Not validated by PETACC-3 RCT of FU vs IFL in 1,254 stage II/III

Tejpar, *JCO* 2009

Molecular Genetics-based Therapeutics

anti-EGFR therapy
anti-VEGF therapy

EGFR Targeted Colorectal Cancer Therapy: Cetuximab (Erbitux) & Panitumumab (Vectibix)



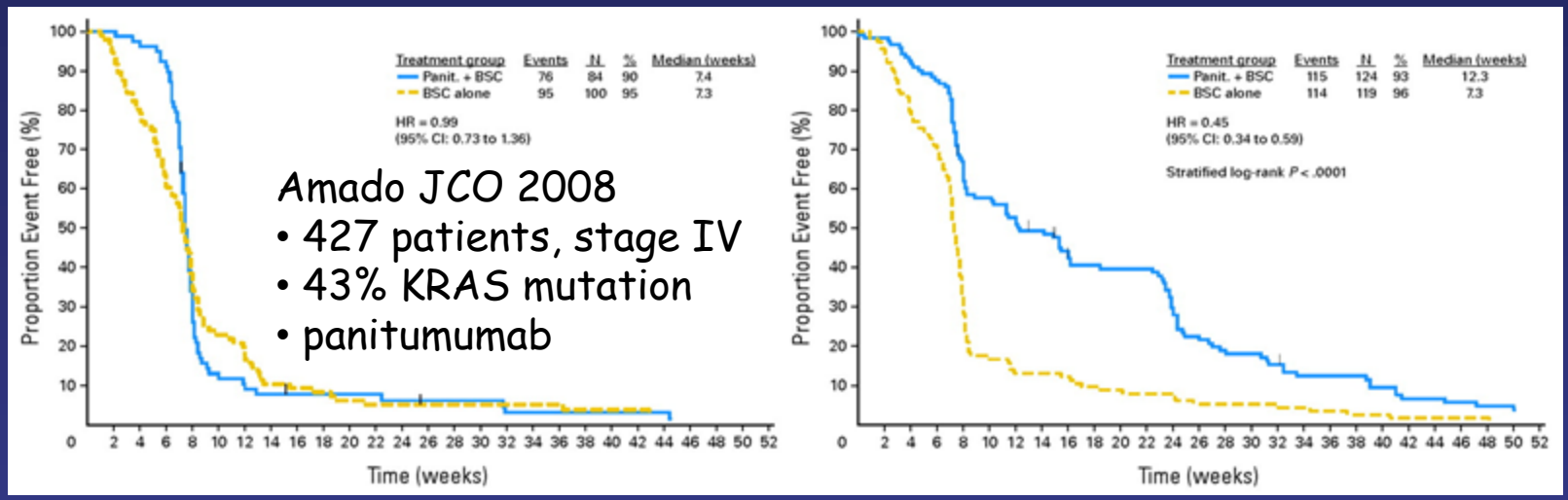
Cetuximab & Panitumumab:

- Anti-EGFR monoclonal antibodies

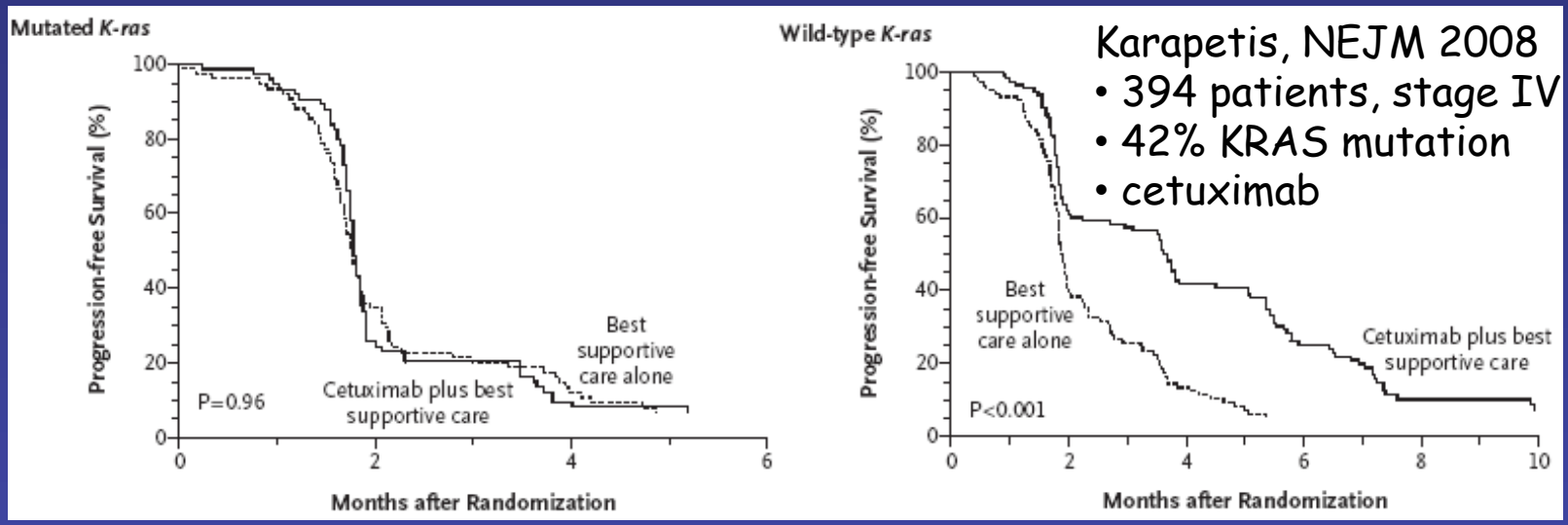
K-Ras & BRAF:

- Oncogenes
- Downstream of EGFR
- Circumvent anti-EGFR therapy
- Activating mutations:
  KRAS 40% of colorectal cancers
  BRAF 15% of colorectal cancers

$KRAS^{WT}$: Predicting Anti-EGFR Response



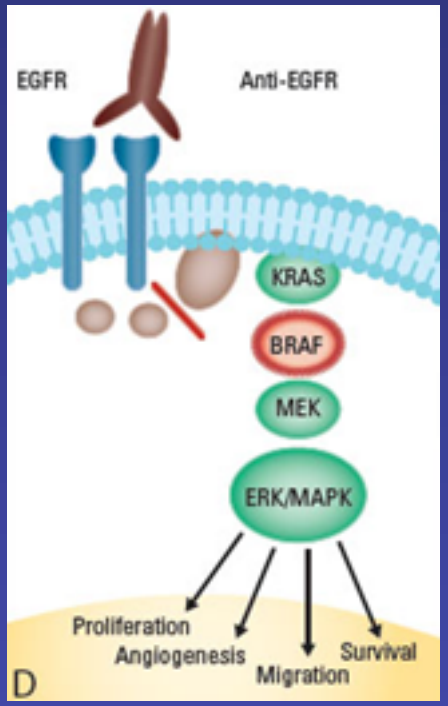
Amado JCO 2008
- 427 patients, stage IV
- 43% KRAS mutation
- panitumumab



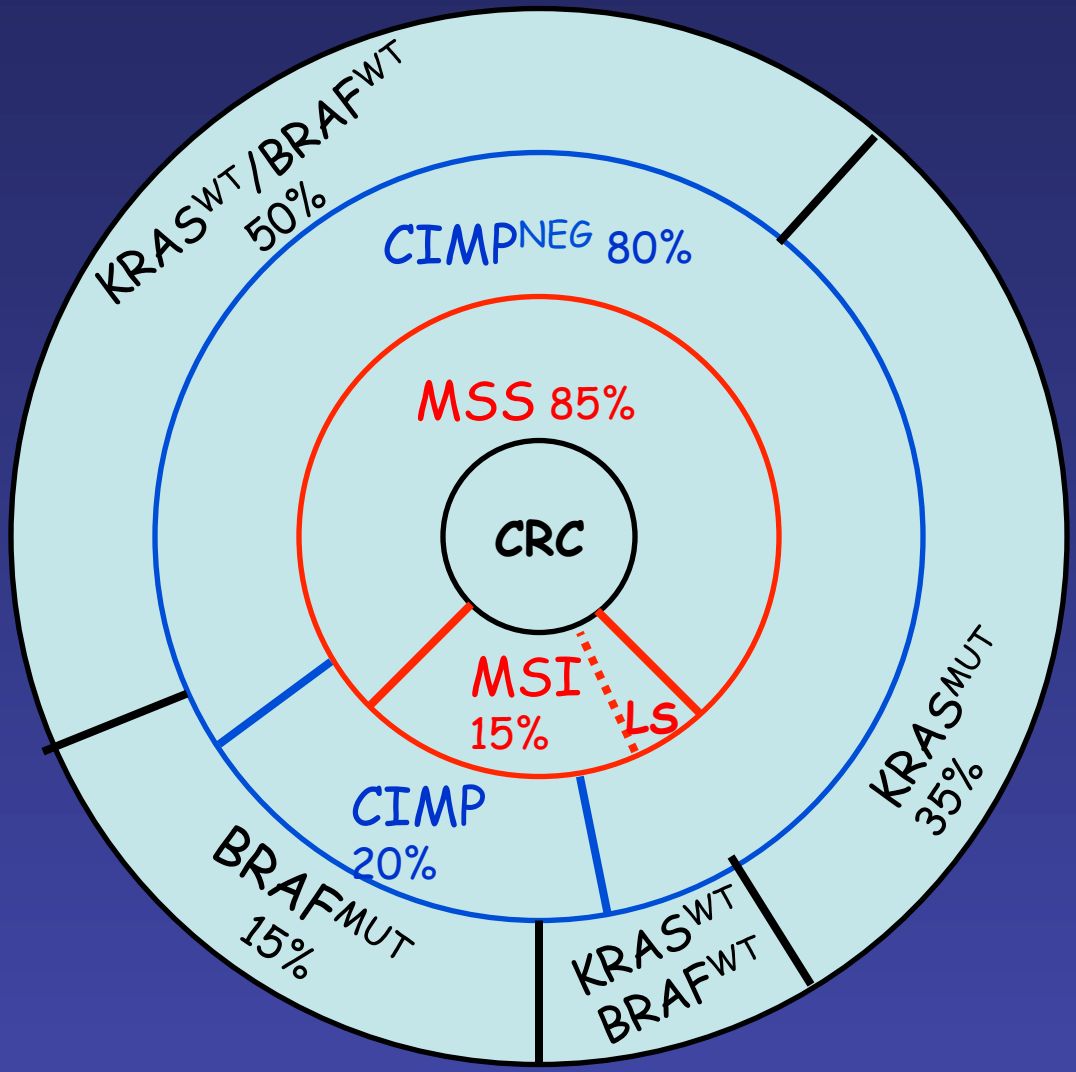
Karapetis, NEJM 2008
- 394 patients, stage IV
- 42% KRAS mutation
- cetuximab

$KRAS^{WT}$ is predictive of improved survival with anti-EGFR palliative therapy

What about BRAF mutation & anti-EGFR therapy?



Complicated interelationships: MSI, CIMP & EGFR



CIMP (20%):
- 67% MSI (sporadic)
- 33% MSS

BRAF (15%):
- 70% CIMP
- 55% MSI (70% of sporadic)

KRAS (35%):
- 90% CIMP-negative
- 95% MSS
- 5% MSI (35% of LS)
- BRAF & KRAS mutually exclusive

Based on Yamauchi Gut 2012
1,443 colorectal cancers

Significant associations of mutator pathways & somatic mutations

BRAF Mutation and Prognosis

Study	Journal	RCT	Patients	BRAF (%)	Poor Prognosis
Hutchins, 2011	JCO	Adjuvant	1,584	8	MSS/BRAF
Roth, 2009	JCO	Adjuvant	1,307	8	MSS/BRAF
Ogino, 2011	Clin Cancer Res	Adjuvant	506	15	MSS/BRAF
Maughan, 2011	Lancet	Palliative*	1,269	8	BRAF
Van Cutsem 2011	JCO	Palliative*	999	6	BRAF
Richman 2009	JCO	Palliative	711	8	BRAF
Tol, 2010	EJC	Palliative*	559	9	BRAF
Tveit, 2012	JCO	Palliative*	498	12	BRAF
Price, 2011	JCO	Palliative	315	11	BRAF

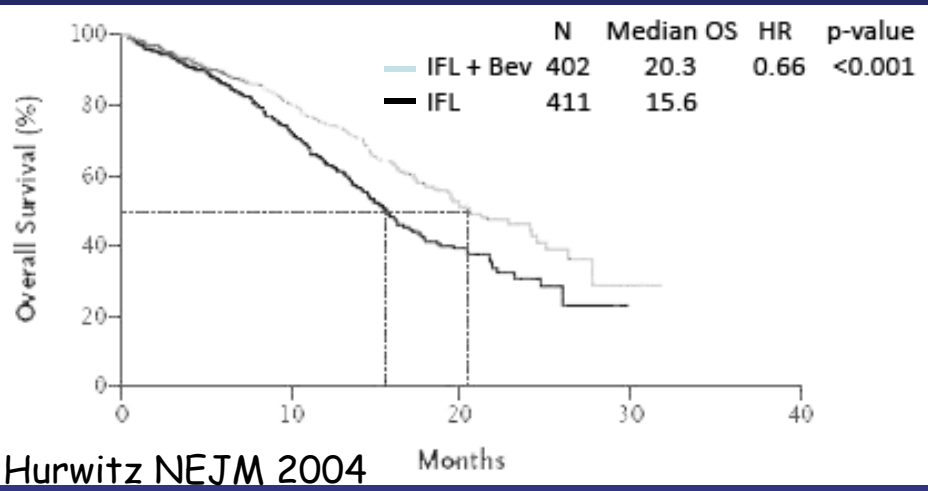
* anti-EGFR RCT

Oncogenic BRAF mutation is associated with poor prognosis

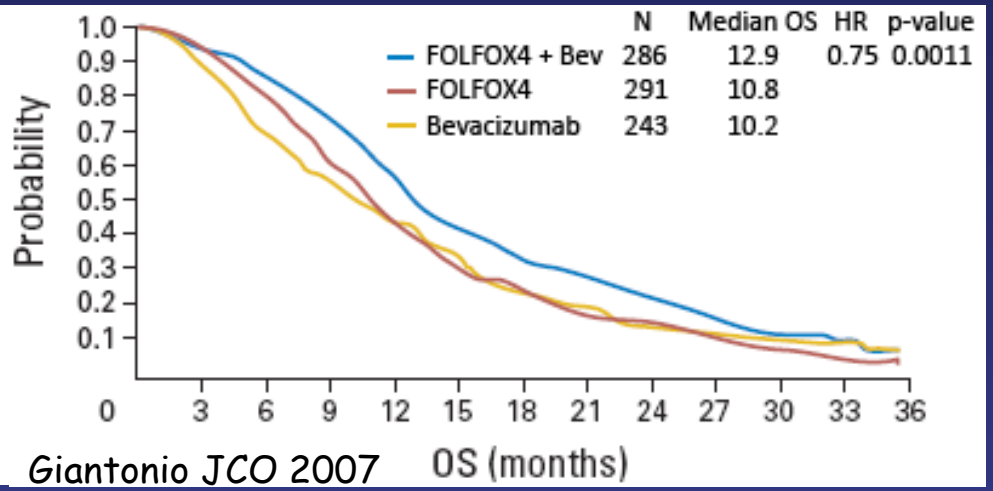
- Poor survival prognosis MSS/BRAF specific
- MSI/BRAF (sporadic MSI) not associated with poor prognosis
- BRAF not predictive of response to therapy, including anti-EGFR

Anti-VEGF, Angiogenesis-inhibition therapy

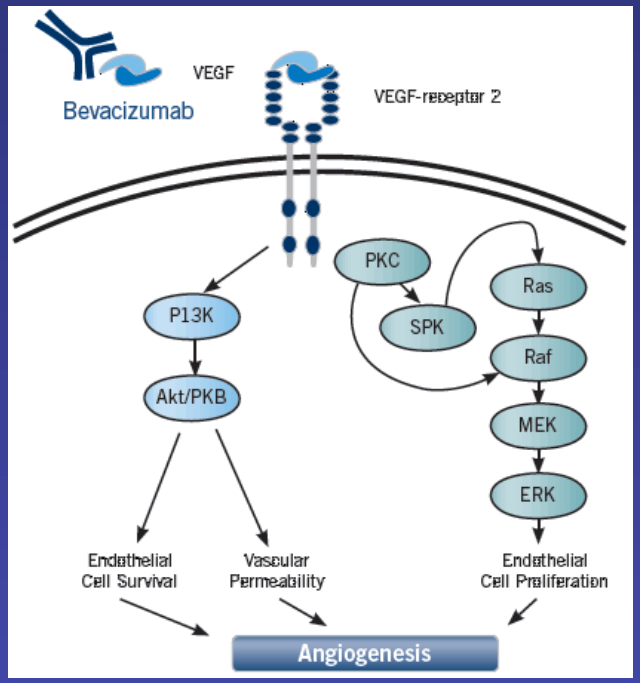
Bevacizumab in Stage IV CRC



Hurwitz NEJM 2004



Giantonio JCO 2007



Bevacizumab (Avastin)

- Monoclonal antibody VEGF inhibitor
- Inhibits angiogenesis
- Potentially complicates wound healing

Galfrascoli Dig Liver Dis 2011

- systemic review 6 RCTs, 3,385 stage IV CRC pt's
- OS = 0.80 (0.71-0.91)
- PFS = 0.62 (0.52-0.74)

Bevacizumab and Surgery

Galfrascoli Dig Liver Dis 2011

- HTN (Gr III/IV) = 2.98 (2.32-3.84)
- Bleeding (Gr III/IV) = 2.07 (1.19-3.62)
- GI perforation = 5.04 (1.72-14.79)
- GI perforation = 1-4% in CRC
  = 3-11% in ovary
  = 1% in others

Manufacturer Warning:

- Half life = 11-50 days
- Do not give Avastin within 28d of surgery
- Hold Avastin at least 28d for elective surgery
- Discontinue Avastin in patient with wound dehiscence or wound healing complications

Grade III / IV wound healing or bleeding complications within 60d postop

- 528/1,132 in phase II/III had surgery

Time of Surgery	Chemo	Chemo + Avastin
Before study		
Surgery	194	230
Complications	(1) 0.5%	(3) 1.3%
During study		
Surgery	29	75
Complications	(1) 3.4%	(10) 13.3%

Scappaticci J Surg Onc 2005

Anti-VEGF molecular therapy ↑ cost & associated with ↑ toxicity & ↑ surgical complication rates

Summary

MMR-deficiency → MSI

- Lynch syndrome & 15% sporadic colorectal cancer
- ↑ prognosis
- MSS, not MSI likely predictive of ↑ 5-FU response
- Sporadic MSI associated with CIMP & BRAF mutation

EGFR signaling

- KRAS mutation in 40% of colorectal cancer circumvents cetuximab / panitumumab anti-EGFR therapy
- MSS/BRAF mutation associated with ↓ prognosis (not MSI/BRAF)

Anti-VEGF angiogenesis inhibitors

- Bevacizumab ↑ palliative prognosis, but associated with spontaneous GI perforations, hemorrhage & ↑ surgical complications