Impact of biomarker in mCRC treatment with EGFR monoclonal antibody

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Illustration of “needs” in colorectal cancer

• Prognostic
  – Increased understanding of the natural history of individual cancers (stage II, III, IV)

• Predictive
  – Not all therapies work for everyone, efficacy only in subgroups (irinotecan, oxaliplatin, EGFR inhibitors, angiogenesis inhibitors)

• Search for Novel Therapeutic targets
  – Based on increased understanding of biology of disease
# New Agents Have Significantly Improved Treatment and Patient Outcomes

<table>
<thead>
<tr>
<th>Regimen</th>
<th>First Line</th>
<th>Second Line</th>
<th>FDA-Approval Year</th>
</tr>
</thead>
<tbody>
<tr>
<td>5-FU</td>
<td>√</td>
<td>√</td>
<td>1962</td>
</tr>
<tr>
<td>Irinotecan (monotherapy)</td>
<td></td>
<td>√</td>
<td>1996</td>
</tr>
<tr>
<td>IFL/irinotecan + infusional 5-FU/LV*</td>
<td>√</td>
<td></td>
<td>2000</td>
</tr>
<tr>
<td>Capecitabine (monotherapy)</td>
<td>√</td>
<td></td>
<td>2001</td>
</tr>
<tr>
<td>Oxaliplatin + infusional 5-FU/LV†</td>
<td>√</td>
<td>√</td>
<td>Second line, 2002 First line, 2004</td>
</tr>
<tr>
<td>Cetuximab (with or without irinotecan)</td>
<td>√</td>
<td>√</td>
<td>2004, 2008</td>
</tr>
<tr>
<td>Bevacizumab + IV 5-FU-based regimens‡</td>
<td>√</td>
<td>√</td>
<td>First line, 2004 Second line 2006</td>
</tr>
<tr>
<td>Panitumumab (single agent)</td>
<td></td>
<td></td>
<td>Salvage 2006</td>
</tr>
</tbody>
</table>

* FOLFIRI  
† FOLFOX  
‡ IFL, FOLFIRI, FOLFOX, and FU/LV  

More regimens provide more options for multiple lines of therapy to extend survival.  
New approaches to metastatic colon cancer

- Existence of clear subgroups

- Therapeutic « window of opportunity » to exploit

- Identify patients who may potentially be resected either before or after chemotherapy based on metastases that are limited in number, size, and sites of involvement.

- Maximize cure rates by optimal chemo choice and resection
  - « window of opportunity » to exploit
  - Chemo + biological based on pt selection

- 5-year and 10-year overall survival rates exceeding 40% and 25%, respectively.

The Oncologist 2007, Goldberg R et al
<table>
<thead>
<tr>
<th>Agent</th>
<th>Type of agent</th>
<th>Target</th>
<th>Proposed single-agent tumor markers</th>
<th>Proposed patient markers</th>
</tr>
</thead>
<tbody>
<tr>
<td>5-fluorouracil</td>
<td>Intravenous fluoropyrimidine</td>
<td>Thymidylate synthase (TS/TYMS)</td>
<td>Expression of TS, dihydropyrimidine dehydrogenase (DPD), thymidine phosphorylase (TP)</td>
<td>TS polymorphism, DPD activity and polymorphism, methylene tetrahydrofolate reductase polymorphism</td>
</tr>
<tr>
<td>Capecitabine</td>
<td>Oral fluoropyrimidine</td>
<td>TS Nucleotides in DNA for crosslinking</td>
<td>? same as 5-FU Expression of X-ray cross complementing factor 1 (XRCC1) and excision repair cross-complementation group 1 (ERCC1)</td>
<td>? same as 5-FU XRCC1 and ERCC1 polymorphism</td>
</tr>
<tr>
<td>Oxaliplatin</td>
<td>Oral fluoropyrimidine</td>
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<td>? same as 5-FU XRCC1 and ERCC1 polymorphism</td>
</tr>
<tr>
<td>Irinotecan</td>
<td>Camptothecin derivative</td>
<td>Topoisomerase I (Topo-1)</td>
<td>? expression of Topo-1, ? high levels of microsatellite instability (MSI-H)</td>
<td>Uridine diphosphate glucoronosyltransferase (UGT1A1) polymorphism</td>
</tr>
<tr>
<td>Bevacizumab</td>
<td>Humanized monoclonal antibody</td>
<td>Vascular endothelial growth factor (VEGF)</td>
<td>None identified</td>
<td>None identified</td>
</tr>
<tr>
<td>Cetuximab</td>
<td>Chimeric IgG1</td>
<td>Epidermal growth factor receptor (EGFR)</td>
<td>Expression of EGFR (FISH), KRAS mutation</td>
<td>Expression of EGFR (FISH), KRAS mutation</td>
</tr>
<tr>
<td>Panitumunab</td>
<td>Humanized IgG2</td>
<td>Epidermal growth factor receptor (EGFR)</td>
<td>Expression of EGFR (FISH), KRAS mutation</td>
<td>Expression of EGFR (FISH), KRAS mutation</td>
</tr>
</tbody>
</table>

Ref Hamilton S 2008
Monoclonal Ab against EGFR

Potential negative predictors

Potential positive predictors
• Evidence for oncogene dependency to EGFR in CRC

• First data on Erbitux is in advanced, chemorefractory disease
NCIC CO.17: randomized phase III trial

Failed or intolerant to all recommended therapies

Stratification:
- Center
- ECOG PS (0 or 1 vs 2)

ERBITUX + BSC

BSC alone

Disease progression
or
Unacceptable toxicity

NCIC CTG CO.17: Cetuximab vs BSC
Progression Free Survival

<table>
<thead>
<tr>
<th>Study arm</th>
<th>Med PFS (months)</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cetuximab + BSC</td>
<td>1.9</td>
<td>1.8 – 2.1</td>
</tr>
<tr>
<td>BSC alone</td>
<td>1.8</td>
<td>1.8 – 1.9</td>
</tr>
</tbody>
</table>

HR 0.68 (95% CI =0.57 – 0.80)

Stratified log rank p-value < 0.0001

NCIC CTG CO.17: Overall Survival

<table>
<thead>
<tr>
<th>Study arm</th>
<th>MS (months)</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cetuximab + BSC</td>
<td>6.1</td>
<td>5.4 – 6.7</td>
</tr>
<tr>
<td>BSC alone</td>
<td>4.6</td>
<td>4.2 – 4.9</td>
</tr>
</tbody>
</table>

HR 0.77 (95% CI = 0.64 – 0.92)

Stratified log rank p-value = 0.0046

Response Rate Disease Control

Percentage

<table>
<thead>
<tr>
<th></th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>cetuximab + irinotecan (n=218)</td>
<td>23%</td>
</tr>
<tr>
<td>cetuximab (n=111)</td>
<td>11%</td>
</tr>
</tbody>
</table>

* p=0.0074; ** p<0.001; [] = 95% CI

Figure 2. Time to Disease Progression in the Two Study Groups.
The hazard ratio for disease progression in the combination-therapy group as compared with the monotherapy group was 0.54 (95 percent confidence interval, 0.42 to 0.71) (P<0.001 by the log-rank test). The points on the curves represent the dates on which a patient’s data were censored.
Lessons learnt from early chemorefractory studies

• Paradigm shift

• Around 10% responses in monotherapy
  – EGFR dependency in subset of CRC

• Reversal of chemoresistance
  – All irinotecan refractory
<table>
<thead>
<tr>
<th>Clinical setting</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chemorefractory patients</td>
<td>Proven activity Cetuximab monotherapy (10%)</td>
</tr>
<tr>
<td></td>
<td>Cetuximab reverts chemoresistance in combi therapy (23%)</td>
</tr>
<tr>
<td>First-line treatment</td>
<td>Promising activity in randomized studies vs chemo alone (HR 0.8 to 0.85) Cetuximab</td>
</tr>
<tr>
<td>Crystal</td>
<td></td>
</tr>
<tr>
<td>Second line treatment</td>
<td>Promising activity in randomized studies vs chemo alone (HR 0.8 to 0.85) Cetuximab</td>
</tr>
<tr>
<td>Opus</td>
<td></td>
</tr>
<tr>
<td>Adjuvant treatment</td>
<td>Studies ongoing</td>
</tr>
</tbody>
</table>
Meanwhile: kras is a negative predictor for EGFR inhibition

- ........ trials were reanalyzed according to kras status.
  - Present efficacy data all patients versus kras wt
Monoclonal Ab against EGFR

Potential positive predictors

Potential negative predictors
<table>
<thead>
<tr>
<th>Cancer type</th>
<th>HRAS</th>
<th>KRAS</th>
<th>NRAS</th>
<th>BRAF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Biliary tract</td>
<td>0%</td>
<td>33%</td>
<td>1%</td>
<td>14%</td>
</tr>
<tr>
<td>Bladder</td>
<td>11%</td>
<td>4%</td>
<td>3%</td>
<td>0%</td>
</tr>
<tr>
<td>Breast</td>
<td>0%</td>
<td>4%</td>
<td>0%</td>
<td>2%</td>
</tr>
<tr>
<td>Cervix</td>
<td>9%</td>
<td>9%</td>
<td>1%</td>
<td>0%</td>
</tr>
<tr>
<td><strong>Colon</strong></td>
<td>0%</td>
<td>32%</td>
<td>3%</td>
<td>14%</td>
</tr>
<tr>
<td>Endometrial</td>
<td>1%</td>
<td>15%</td>
<td>0%</td>
<td>1%</td>
</tr>
<tr>
<td>Kidney</td>
<td>0%</td>
<td>1%</td>
<td>0%</td>
<td>0%</td>
</tr>
<tr>
<td>Liver</td>
<td>0%</td>
<td>8%</td>
<td>10%</td>
<td>3%</td>
</tr>
<tr>
<td>Lung</td>
<td>1%</td>
<td>19%</td>
<td>1%</td>
<td>2%</td>
</tr>
<tr>
<td>Melanoma</td>
<td>6%</td>
<td>2%</td>
<td>18%</td>
<td>43%</td>
</tr>
<tr>
<td>Myeloid leukaemia</td>
<td>0%</td>
<td>5%</td>
<td>14%</td>
<td>1%</td>
</tr>
<tr>
<td>Ovarian</td>
<td>0%</td>
<td>17%</td>
<td>4%</td>
<td>15%</td>
</tr>
<tr>
<td>Pancreas</td>
<td>0%</td>
<td>60%</td>
<td>2%</td>
<td>3%</td>
</tr>
<tr>
<td>Thyroid</td>
<td>5%</td>
<td>4%</td>
<td>7%</td>
<td>27%</td>
</tr>
</tbody>
</table>

The mutation data was obtained from the Sanger Institute Catalogue of Somatic Mutations in Cancer website.
### Evidence of correlation between KRAS wild-type and EGFR inhibitor efficacy in chemorefractory CRC: Response

<table>
<thead>
<tr>
<th>Reference</th>
<th>Treatment</th>
<th>No. of patients (wild-type: mutant)</th>
<th>Objective response, n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lièvre A, et al. (J Clin Oncol 2008)</td>
<td>ERBITUX ± CT</td>
<td>114 (78:36)</td>
<td>34 (44) 0 (0)</td>
</tr>
<tr>
<td>Benvenuti S, et al. (Cancer Res 2007)</td>
<td>Panitumumab or ERBITUX or ERBITUX + CT</td>
<td>48 (32:16)</td>
<td>10 (31) 1 (6)</td>
</tr>
<tr>
<td>DeRoock W, (Ann Onc 2008)</td>
<td>ERBITUX or ERBITUX + irinotecan</td>
<td>113 (67:46)</td>
<td>27 (41) 0 (0)</td>
</tr>
<tr>
<td>Finocchiaro G et al. (ASCO Proceedings 2007)</td>
<td>ERBITUX ± CT</td>
<td>81 (49:32)</td>
<td>13 (26) 2 (6)</td>
</tr>
<tr>
<td>Di Fiore F et al. (Br J Cancer 2007)</td>
<td>ERBITUX + CT</td>
<td>59 (43:16)</td>
<td>12 (28) 0 (0)</td>
</tr>
<tr>
<td>Khambata-Ford S et al. (J Clin Oncol 2007)</td>
<td>ERBITUX</td>
<td>80 (50:30)</td>
<td>5 (10) 0 (0)</td>
</tr>
<tr>
<td>NCIC.CO17 study, NEJM 2008</td>
<td>ERBITUX + BSC or BSC</td>
<td>188 (111:77)</td>
<td>8 (14.8) 1 (5)</td>
</tr>
<tr>
<td>Amado R, (J Clin Oncol 2008)</td>
<td>Panitumumab</td>
<td>208 (124:84)</td>
<td>21 (17) 0 (0)</td>
</tr>
</tbody>
</table>
NCIC CTG CO.17 K-Ras Analysis

- Genomic DNA extracted from FFPET slides or sections
- Assessed by bidirectional sequencing for codon 12/13 mutations
- No difference between K-ras mutated and WT patients re: demographics, previous treatment or other variables

N=572 randomized: ITT subset

N=394: K-ras assessed subset (69%)

N=164 (42%) mutant
N=230 (58%) wild-type

Karapetis C et al, NEJM 2008
NCIC CTG C0.17: PFS in the *K-ras* Wild-Type Patients

### Study arm | Med PFS (months) | 95% CI |
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Cetuximab + BSC</td>
<td>3.8</td>
<td>3.1 - 5.1</td>
</tr>
<tr>
<td>BSC alone</td>
<td>1.9</td>
<td>1.8 - 2.0</td>
</tr>
</tbody>
</table>

HR 0.40, 95% CI (0.30,0.54)

Log rank p-value: <0.0001

Karakpetis C et al, NEJM 2008
NCIC CTG C0.17: PFS in the Mutant K-ras Subgroup

<table>
<thead>
<tr>
<th>Study arm</th>
<th>Med PFS (months)</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cetuximab + BSC</td>
<td>1.8</td>
<td>1.7 - 1.8</td>
</tr>
<tr>
<td>BSC alone</td>
<td>1.8</td>
<td>1.7 - 1.8</td>
</tr>
</tbody>
</table>

HR 0.99 95% CI (0.73,1.35)

Log rank p-value: 0.96

Karapetis C et al, NEJM 2008
ERBITUX every second week regimen

First-line therapy with cetuximab followed by cetuximab plus FOLFIRI in patients with metastatic colorectal cancer: KRAS mutation status correlates with clinical outcome

Updated information presented at WCGIC 2008
ERBITUX every second week regimen
Study design

Group A (control arm)
Cohort of 10 patients
ERBITUX 400 mg/m² initial dose then 250 mg/m² weekly

Group B (experimental arm)
Cohorts of 10 patients
ERBITUX at escalating doses for successive cohorts; 400, 500, 600, 700 mg/m² every 2 weeks

PART I
6 weeks' treatment complete PK profile obtained during this period
1° endpoint DLT assessment

PART II
FOLFIRI added to patients’ current ERBITUX regimen
Evaluate best overall response
Progression-free survival
2° endpoints

Updated information presented at WCGIC 2008
ERBITUX every second week regimen

Correlation of KRAS status with response

**Response rate (%)**

- **KRAS wild-type (n=29)**
  - Monotherapy phase: 28 (%), p=0.015
  - Combination therapy phase: 55 (%), p=0.144

- **KRAS mutant (n=19)**
  - Monotherapy phase: 0 (%)
  - Combination therapy phase: 32 (%)


Updated information presented at WCGIC 2008
Study design Crystal (first line)

Stratification by:
- Region
- ECOG PS

Primary endpoint
- Progression-free survival time
  (as assessed by blinded independent review)

Secondary endpoints
- ORR (independent review)
- OS
- Quality of life (EORTC QLQ-C30)
- Safety

ERBITUX + FOLFIRI
- ERBITUX (IV 400 mg/m² on day 1,
  then 250 mg/m² weekly)
  + irinotecan (180 mg/m²)
  + 5-FU (400 mg/m² bolus + 2400 mg/m²
    as 46-h continuous infusion)
  + LV (every 2 weeks)

FOLFIRI
- irinotecan (180 mg/m²)
  + 5-FU (400 mg/m² bolus + 2400 mg/m²
    as 46-h continuous infusion)
  + LV (every 2 weeks)
CRYSTAL trial: Primary endpoint PFS

Progression-free survival time (months)

1-year PFS rate
23% vs 34%

HR = 0.851
P = 0.0479

RR
FOLFIRI + Cetuximab 46.9%
FOLFIRI 38.7%
P = 0.0038

Subjects at risk
FOLFIRI alone 599
Cetuximab + FOLFIRI 599
FOLFIRI 499
8.9 mo
8.0 mo
Van Cutsem et al. ASCO 2007
Application of $K$-RAS hypothesis on
The phase III CRYSTAL trial
Relating KRAS status to efficacy

- Efficacy analysis repeated to evaluate the influence of KRAS status in study population
- Genomic DNA isolated from archived tumor material
- KRAS mutation status of codons 12/13 determined using quantitative PCR-based assay
  - 45% of study population with evaluable samples (540/1198)*
  - 64.4% of evaluable population showed the KRAS wild type (348/540)

*The population available for KRAS analysis was representative of the overall ITT population
PFS: Wild-type KRAS

- Median PFS cetuximab + FOLFIRI: 9.9 months
- Median PFS FOLFIRI alone: 8.7 months

**Progression-free survival**

- 1-year PFS rate: 43% vs 25%

**HR=0.68; p=0.017**

(n=348)

Median PFS cetuximab + FOLFIRI: 9.9 months
Median PFS FOLFIRI alone: 8.7 months
Relating KRAS status to efficacy: PFS – KRAS mutant

**KRAS mt HR=1.07**

- mPFS Erbitux+Folfiri: 7.6 mo
- mPFS Folfiri: 8.1 mo

**Subjects at Risk**

<table>
<thead>
<tr>
<th></th>
<th>Progressed</th>
<th>Censored</th>
<th>Median PFS</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>A: ERBITUX + FOLFIRI</td>
<td>105</td>
<td>43</td>
<td>7.6 Mon</td>
<td>[6.7, 9.4]</td>
</tr>
<tr>
<td>B: FOLFIRI</td>
<td>87</td>
<td>44</td>
<td>8.1 Mon</td>
<td>[7.5, 9.4]</td>
</tr>
</tbody>
</table>

**Hazard Ratio 1.07 95% CI [0.710, 1.610]**
Response rate

Wild-type KRAS
- Cetuximab + FOLFIRI: 59%
- FOLFIRI alone: 43%

Mutant KRAS
- Cetuximab + FOLFIRI: 40%
- FOLFIRI alone: 36%

Odds ratio\(^a\)=1.91; p=0.003
Odds ratio\(^a\)=0.80; p=0.46

\(^a\)Cochran–Mantel–Haenszel test
Median OS cetuximab + FOLFIRI: 24.9 months
Median OS FOLFIRI alone: 21.0 months

OS: Wild-type KRAS

HR=0.84; p=0.22 (n=348)

2-year OS rate: 51% vs 44%

Median OS cetuximab + FOLFIRI: 24.9 months
Median OS FOLFIRI alone: 21.0 months
Conclusions (1)

• Adding cetuximab to FOLFIRI results in a **significant prolongation of PFS** and increased ORR for pts with KRAS wild type tumors

• Increase in OS was suggested in KRAS wild type, but study was not powered to demonstrate a significant difference
KRAS and ERBITUX + oxaliplatin-based regimen in first-line treatment of mCRC: The phase II OPUS trial
## Summary of Efficacy Data

<table>
<thead>
<tr>
<th></th>
<th>ITT</th>
<th>KRAS wt</th>
<th>KRAS mt</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ERBITUX + FOLFOX</td>
<td>ERBITUX + FOLFOX</td>
<td>FOLFOX</td>
</tr>
<tr>
<td>N</td>
<td>168</td>
<td>169</td>
<td>61</td>
</tr>
<tr>
<td>ORR (%)</td>
<td>46</td>
<td>36</td>
<td>61</td>
</tr>
<tr>
<td>p-value</td>
<td>0.064</td>
<td>0.011</td>
<td>0.110</td>
</tr>
<tr>
<td>PFS (months)</td>
<td>7.2</td>
<td>7.2</td>
<td>7.7</td>
</tr>
<tr>
<td>HR</td>
<td>0.931</td>
<td></td>
<td>0.57</td>
</tr>
<tr>
<td>p-value</td>
<td>0.62</td>
<td></td>
<td>0.016</td>
</tr>
</tbody>
</table>
Primary Endpoint:
Overall response rate

- KRAS wt:
  - 61 (n=52)
  - Overall response rate 65% (odds ratio: 2.54; p=0.011)

- KRAS mt:
  - 49
  - Overall response rate 33% (odds ratio: 0.51; p=0.106)

- No benefit for Erbitux (p = 0.106)
Relating KRAS status to efficacy:
PFS – KRAS wild-type

- ERBITUX + FOLFOX: 7.7 months (mPFS)
- FOLFOX: 7.2 months (mPFS)

HR 0.57, 95% CI [0.351, 0.894]

43% risk reduction for progression
Relating KRAS status to efficacy:
PFS – KRAS mutant

KRAS mt HR=1.83
mPFS ERBITUX+FOLFOX: 5.5 months
mPFS FOLFOX: 8.6 months
KRAS and prediction of chemotherapy efficacy

**FOLFIRI HR=0.97 (p=0.87)**
Median PFS: Wild-type (n=176) 8.7 months
vs mutant (n=87) 8.1 months

Response Rates

KRAS wild-type Liver Limited
p=0.025*

KRAS wild-type
p=0.0025*

43
59
77

FOLFIRI
ERBITUX + FOLFIRI
ERBITUX + FOLFIRI

Response rate (%)

Response Rates


*Cochran–Mantel–Haenszel (CMH) test
Patients with non-resectable CRC liver metastases
(technically non-resectable / ≥ 5 liver metastases)
without extrahepatic metastases

Biopsy:
EGFR screening

Randomization

FOLFOX6 + ERBITUX
FOLFIRI + ERBITUX

Therapy: 8 cycles (~ 4 months)

Evaluation of resectability

Technically (non-)resectable

4 additional CTX cycles

Technically resectable

Resection

Therapy continuation for 6 cycles (~ 3 months)

Primary endpoint: Response

From: Folprecht et al. ESMO 2008, ASCO-GI 2009
## Patient characteristics

<table>
<thead>
<tr>
<th></th>
<th>FOLFOX6 + ERBITUX</th>
<th>FOLFIRI + ERBITUX</th>
<th>All patients</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n=56</td>
<td>n=55</td>
<td>n=111</td>
</tr>
<tr>
<td>Median age, years</td>
<td>65.1</td>
<td>62.0</td>
<td>63.3</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>64%</td>
<td>64%</td>
<td>64%</td>
</tr>
<tr>
<td>Stratified characteristics</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>≥5 metastases</td>
<td>45%</td>
<td>46%</td>
<td>45%</td>
</tr>
<tr>
<td>Technically non-resectable</td>
<td>55%</td>
<td>55%</td>
<td>55%</td>
</tr>
<tr>
<td>Staging using PET *</td>
<td>16%</td>
<td>20%</td>
<td>18%</td>
</tr>
<tr>
<td>EGFR-positive (IHC)</td>
<td>71%</td>
<td>75%</td>
<td>73%</td>
</tr>
</tbody>
</table>

*PET is not generally reimbursed in Germany*

From: Folprecht et al. ESMO 2008, ASCO-GI 2009
## Response rates

<table>
<thead>
<tr>
<th></th>
<th>FOLFOX6 + ERBITUX</th>
<th>FOLFIRI + ERBITUX</th>
<th>KRAS wild-type</th>
<th>All patients</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>n</strong></td>
<td>52</td>
<td>53</td>
<td>67</td>
<td>105*</td>
</tr>
<tr>
<td><strong>CR/PR</strong></td>
<td>85% (44 pts)</td>
<td>66% (35 pts)</td>
<td>79% (53 pts)</td>
<td>75% (79 pts)</td>
</tr>
<tr>
<td><strong>95% CI</strong></td>
<td>71.9-93.1%</td>
<td>51.7-78.5%</td>
<td>67.4-88.1%</td>
<td>65.9-83.1%</td>
</tr>
<tr>
<td><strong>SD</strong></td>
<td>11% (6 pts)</td>
<td>23% (12 pts)</td>
<td>13% (9 pts)</td>
<td>17% (18 pts)</td>
</tr>
<tr>
<td><strong>PD</strong></td>
<td>4% (2 pts)</td>
<td>11% (6 pts)</td>
<td>8% (5 pts)</td>
<td>8% (8 pts)</td>
</tr>
</tbody>
</table>

* 105 pts evaluable for efficacy
Responses are not yet confirmed
Data regarding response confirmation are still pending for 14 patients

From: Folprecht et al. ESMO 2008, ASCO-GI 2009
Resections by patient subgroup

<table>
<thead>
<tr>
<th></th>
<th>Technically non-resectable</th>
<th>≥ 5 liver metastases</th>
<th>KRAS wild-type</th>
</tr>
</thead>
<tbody>
<tr>
<td>n=57</td>
<td>40%</td>
<td>44%</td>
<td>43%</td>
</tr>
<tr>
<td>(23 pts)</td>
<td>(21 pts)</td>
<td>(29 pts)</td>
<td></td>
</tr>
<tr>
<td>All resections</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>R0 resections</td>
<td>32%</td>
<td>40%</td>
<td>34%</td>
</tr>
<tr>
<td>(18 pts)</td>
<td>(19 pts)</td>
<td>(23 pts)</td>
<td></td>
</tr>
</tbody>
</table>

Comparison of R0 resections between strata technically non-resectable and ≥ 5 liver mets: p=0.4

From: Folprecht et al. ESMO 2008, ASCO-GI 2009
### Resections according to tumor response

<table>
<thead>
<tr>
<th>Best responses</th>
<th>PR/CR</th>
<th>SD</th>
<th>PD</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>n=79</td>
<td>n=18</td>
<td>n=8</td>
</tr>
<tr>
<td>R0 resections</td>
<td>35 pts</td>
<td>2 pts</td>
<td>0</td>
</tr>
<tr>
<td>Without R0 resection</td>
<td>44 pts</td>
<td>16 pts</td>
<td>8 pts</td>
</tr>
</tbody>
</table>

35/37 of resected patients (95%) had a tumor response. The 2 remaining patients had minor responses.

From: Folprecht et al. ESMO 2008, ASCO-GI 2009
CELIM randomised phase II study: Response and Resection rates

FOLFOX + ERBITUX

or

FOLFIRI + ERBITUX

as Neoadjuvant treatment of non-resectable colorectal liver metastases

(CELIM: Folprecht et al. ESMO 2008)

“Being able to achieve complete resection rates as high as 34% in these patients is remarkable and offers them a real chance of cure”

Dr. Gunnar Folprecht

Next EORTC Trial?

From: Folprecht et al. ESMO 2008, ASCO-GI 2009
KRAS: An important biomarker for mCRC

- KRAS wild-type status is predictive of response to ERBITUX
  - Associated with survival benefit
  - Associated with response

- Early screening for KRAS mutation is mandatory for all mCRC patients
In July 2008, the EMEA granted approval of ERBITUX for use in all lines of mCRC in patients with KRAS wild-type tumors

- FDA expected to follow
- New standard of care
- Setting standards
Wanting to do individualized therapy, but how?
KRAS test methodology

• What is needed: sample
  – Archival paraffin-embedded tumor specimen
  Alternatives:
  – Fresh frozen tumor specimen
• Laboratory: pathology and molecular diagnostics
Tissue:

• Primary Tumor
  – Around 95% concordance between primary sample and metastasis reported (Artale et al, JCO 2008)

• Metastasis

• Endoscopic biopsy
  – For unresected patients
  – If verified contains carcinoma
What do you test for? (specificity)

- Wild type versus mutant

<table>
<thead>
<tr>
<th>Mutation</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>GGT -&gt; GTT</td>
<td>G12V</td>
</tr>
<tr>
<td>GGT -&gt; GAT</td>
<td>G12D</td>
</tr>
<tr>
<td>GGC -&gt; GAC</td>
<td>G13D</td>
</tr>
<tr>
<td>GGT -&gt; TGT</td>
<td>G12C</td>
</tr>
<tr>
<td>GGT -&gt; GCT</td>
<td>G12A</td>
</tr>
<tr>
<td>GGT -&gt; AGT</td>
<td>G12S</td>
</tr>
<tr>
<td>GGT -&gt; CGT</td>
<td>G12R</td>
</tr>
</tbody>
</table>

Other codon 12 and 13 mutation

- Codon 61, 146, 154

7 most frequent variants: >98%

Rare variants: <2%
60% kras wt

40% of kras wt respond

40% kras mut

Bond like setting
Monoclonal Ab against EGFR

Potential positive predictors

Potential negative predictors
60% kras wt

40% of kras wt respond

40% kras mut

12% braf, nras, kras

Bond like setting
<table>
<thead>
<tr>
<th>Gene</th>
<th>AA Mut</th>
<th>Nucl Mut</th>
<th>Mutations distribution</th>
</tr>
</thead>
<tbody>
<tr>
<td>p.G12S</td>
<td>c.34G&gt;A</td>
<td>7.3%</td>
<td></td>
</tr>
<tr>
<td>p.G12R</td>
<td>c.34G&gt;C</td>
<td>1.2%</td>
<td></td>
</tr>
<tr>
<td>p.G12C</td>
<td>c.34G&gt;T</td>
<td>8.7%</td>
<td></td>
</tr>
<tr>
<td>p.G12D</td>
<td>c.35G&gt;A</td>
<td>36.0%</td>
<td></td>
</tr>
<tr>
<td>p.G12A</td>
<td>c.35G&gt;C</td>
<td>6.1%</td>
<td></td>
</tr>
<tr>
<td>p.G12V</td>
<td>c.35G&gt;T</td>
<td>22.3%</td>
<td></td>
</tr>
<tr>
<td>p.G13S</td>
<td>c.37G&gt;A</td>
<td>0.3%</td>
<td></td>
</tr>
<tr>
<td>p.G13R</td>
<td>c.37G&gt;C</td>
<td>0.3%</td>
<td></td>
</tr>
<tr>
<td>p.G13C</td>
<td>c.37G&gt;T</td>
<td>0.6%</td>
<td></td>
</tr>
<tr>
<td>p.G13D</td>
<td>c.38G&gt;A</td>
<td>15.2%</td>
<td></td>
</tr>
<tr>
<td>p.G13A</td>
<td>c.38G&gt;C</td>
<td>0.2%</td>
<td></td>
</tr>
<tr>
<td>p.G13V</td>
<td>c.38G&gt;T</td>
<td>0.10%</td>
<td></td>
</tr>
<tr>
<td>p.G13G</td>
<td>c.39C&gt;A</td>
<td>0.02%</td>
<td></td>
</tr>
<tr>
<td>p.G13G</td>
<td>c.39C&gt;G</td>
<td>0.02%</td>
<td></td>
</tr>
<tr>
<td>p.G13G</td>
<td>c.39C&gt;T</td>
<td>0.00%</td>
<td></td>
</tr>
<tr>
<td>p.A146T</td>
<td>c.436G&gt;A</td>
<td>0.12%</td>
<td></td>
</tr>
<tr>
<td>p.A146P</td>
<td>c.436G&gt;C</td>
<td>0.02%</td>
<td></td>
</tr>
<tr>
<td>p.A146V</td>
<td>c.437C&gt;T</td>
<td>0.04%</td>
<td></td>
</tr>
<tr>
<td>p.A59T</td>
<td>c.175G&gt;A</td>
<td>0.04%</td>
<td></td>
</tr>
<tr>
<td>p.Q61K</td>
<td>c.181C&gt;A</td>
<td>0.02%</td>
<td></td>
</tr>
<tr>
<td>p.Q61E</td>
<td>c.181C&gt;G</td>
<td>0.02%</td>
<td></td>
</tr>
<tr>
<td>p.Q61P</td>
<td>c.182A&gt;C</td>
<td>0.00%</td>
<td></td>
</tr>
<tr>
<td>p.Q61R</td>
<td>c.182A&gt;G</td>
<td>0.10%</td>
<td></td>
</tr>
<tr>
<td>p.Q61L</td>
<td>c.182A&gt;T</td>
<td>0.18%</td>
<td></td>
</tr>
<tr>
<td>p.Q61H</td>
<td>c.183A&gt;C</td>
<td>0.25%</td>
<td></td>
</tr>
<tr>
<td>p.Q61H</td>
<td>c.183A&gt;T</td>
<td>0.08%</td>
<td></td>
</tr>
</tbody>
</table>

Total: 99.21%
Pi 3kinase-PTEN-AKT axis

- 15% pi3K mutations in colorectal cancer
- PTEN loss of function in ...%
- « proof of concept in breast cancer »
60% kras wt

40% of kras wt respond

PI3K/PTEN/AKT?

12% braf, nras, kras

40% kras mut

Cancer Research and Clin Cancer Res 2009
Pi3kinase: not as central as KRAS for EGFR/HER1?
• Positive predictors
  – Ihc: not correlated (technique, patterns)
  – Gene copy number Fish: rare amplifications, polysomy heterogenous in the tumor
  – Mutations: very rare activating mutations in colorectal cancer
  – Skin toxicity
  – Ligand expression
Epiregulin and amphiregulin expression predict outcome
High Epiregulin and Amphiregulin is seen mostly in KRAS wt pts.

- KRAS wild type (WT) status and high ligand expression were significantly correlated ($\chi^2; p=0.03$ for AR and $p=0.02$ for ER).

- High ligand expression is found in half of kras wt tumors
60% kras wt

40% of kras wt respond

High ligand expression

PI3K/PTEN/AKT?

12% braf, nras, kras
Future question

- Downstream activation of the pathway
  - Negative predictor

- Upstream activation
  - Positive predictor

- Acquired resistance
Future

• Precise molecular prediction
• Improve the therapeutic index of EGFR inhibitors

• Combining with inhibitors of molecules involved in acquired resistance
  – Mek, pan-Her, IGF1R, NFKB, ........
  – Data driven
Conclusions

- Biomarkers will lead to tailored therapy

- What do we want to achieve?
  - Resectability and cure (response rates)
  - Disease control (progression-free survival)
Options 1st-line systemic treatment of CRC in patients with wild-type KRAS

- **Chemotherapy**
  - Fluoropyrimidine
  - Oxaliplatin + FU
  - Irinotecan + FU
  - Triple combination

- **Biologic agents**
  - ERBITUX
  - Bevacizumab
Response rate with standard chemotherapy (FOLFOX/FOLFIRI) according to pivotal trials for the European label

- **FOLFOX + ERBITUX KRAS wt³**
  - Response rate: 61%

- **FOLFIRI + ERBITUX KRAS wt⁴**
  - Response rate: 59%

- **FOLFIRI + ERBITUX ITT⁴**
  - Response rate: 47%

- **FOLFOX + ERBITUX ITT³**
  - Response rate: 46%

- **FOLFIRI⁴**
  - Response rate: 39%

- **FOLFOX³**
  - Response rate: 36%

- **FOLFOX/XELOX + bevacizumab⁵**
  - Response rate: 38%

- **FOLFOX/XELOX⁵**
  - Response rate: 38%

**Tailored therapy – new era in mCRC**

ITT, intent-to-treat population; wt, wild-type; LLD, liver-limited disease

References:
High response rates correlate with high resection rates in mCRC

Curative liver resections

ERBITUX + FOLFIRI

Significant

+118%

R0 resection rate (%)

CRYSTAL

ERBITUX + CT resection rate = 43% and R0 rate = 34%

Improving overall survival in mCRC: impact of tailored therapy

1. Van Cutsem et al. ESMO 2008

ITT, intent-to-treat population; wt, wild-type

Tailored therapy – new era in mCRC

1. Van Cutsem et al. ESMO 2008
Conclusions

KRAS wild type is strongly predictive of responsiveness to ERBITUX

- In up to 65% of patients with mCRC tumor cells carry the wild type KRAS gene
- Every patient should be tested at the time of diagnosis for KRAS mutations
- In patients with mCRC with KRAS wt ERBITUX offers outstanding efficacy, irrespective of chemotherapy partner (FOLFIRI, FOLFOX)