### Poster Discussion:

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Discussant: Matt Seymour, University of Leeds & NCRN, UK
we may have a **prognostic marker** – correlates with underlying cancer biology

we may have a **predictive marker** – correlates with response to drug X
survival

randomised trial

marker status (a)  
marker status (b)

prognostic factor

predictive factor

survival

100%
- **pharmacology/pathways:** identify candidate molecules
- **‘-omic’ studies:** identify candidate molecules
- **initial lab or clinical observation:** which markers correlate with biology?
- **screen multiple candidates in RCTs:** vs **incremental outcome** (drug effect)
- **validation of screen-positive markers:** was the initial observation false-positive?
- **prospective testing of strategy:** does the biomarker improve patient care?
- **if no RCT available:** marker vs **unequivocal drug outcome**
- **ORR**
- **specific tox**
- **PFS**
- **survival**
hsa-mir31-3p Expression as a Predictor of Anti-EGFR response in WT KRAS Patients with mCRC

microRNA

• ~22-base RNAs
• bind to and regulate mRNA
• more stable than mRNA
• profiling arrays (e.g. Illumina);
• specific assays (e.g. qPCR - Taqman)
hsa-mir31-3p Expression as a Predictor of Anti-EGFR response in WT KRAS Patients with mCRC


Global miRNA expression profiling of 43 WT KRAS CR tumor frozen tissue samples using the Illumina Human microRNA Expression Profiling Assay v2®: 1145 miRNAs

Survival information is associated with miRNA hsa-mir-31-3p expression.
Predictive ability of hsa-miR31-3p

• Training group: retrospective series of 33 patients treated with cetuximab and irinotecan:

  hsa-miR31-3p exhibits significant different expression levels* between tumor samples from patients with bad or good PFS (25 weeks PFS)

• Validation groups*: 2 prospective series of 19 patients treated with cetuximab and of 19 patients treated with panitumumab

  Predictive ability of hsa-miR31-3p expression is validated on the 2 series

* Taqman technology
Nomogram

- Construction of a nomogram for PFS to predict the likelihood of progression

Multivariate Cox proportional hazards models with BRAF mutational status and has-miR31-3p expression as covariates.

Nomogram for PFS constructed based on BRAF mutational status and log. miR expression (miR). For each patient, points are allocated to each of the variable by selecting the corresponding points from the points scale.
• What have we learned?
  – miRNAs are promising new markers
  – miR31-3p is prognostic for PFS in KRAS-wt patients receiving chemo+EGFRmAb

• What next?
  – use RCT material to see if miR31-3p predicts which KRAS-wt patient benefit or do not benefit from the addition of EGFR-mAb
Development of a predictive score using amphiregulin (AREG), epiregulin (EREG) and EGFR-FISH expression levels to determine treatment efficacy in mCRC patients receiving cetuximab-based therapy. - Analysis of the German AIO CRC-0104 trial -

S. Stintzing1, R.P. Laubender2, D.P. Modest1, C. Kapaun1, A. Jung3, L. Fischer von Weikersthal4, H.Hass5, U. Vehling-Kaiser6, C.Giessen1, T. Kirchner3, U. Mansmann2, V. Heinemann1

1Department of Medical Oncology and Comprehensive Cancer Center, Hospital Grosshadern, University of Munich; 2Institute of Medical Informatics, Biometry and Epidemiology, University of Munich; 3 Institute of Pathology, University of Munich; 4 Gesundheitszentrum St. Marien, Amberg; 5 Marienhospital Stuttgart; 6 Onkologische Praxis, Landshut
**KRAS** mutation

**NRAS** mutation

EGFr

- ras
- raf
- MEK
- ERK
- PI3K
- AKT
- PTEN
- Jun

Myc

p53

SMAD

ligands

therapeutic mAb
**BRAF** mutation

- **EGFr**
- **ras**
- **raf**
- **MEK**
- **ERK**
- **PI3K**
- **AKT**
- **PTEN**
- **SMAD**
- **Myc**
- **p53**
- **Jun**

**Ligands**
- **therapeutic mAb**
PI3K mutation
PTEN loss

- raf
- MEK
- ERK
- PI3K
- AKT
- PTEN
- ligands
- therapeutic mAb

- ras
- Myc
- SMAD
- p53
- Jun
PI3K
AKT
PTEN
p53
Jun
EGFr
therapeutic mAb
ligands
ras
MEK
ERK
raf
PI3K
PTEN
SMAD
Myc
p53
Jun
Myc
Receptor increase

therapeutic mAb

ligands

EGFr

ras

raf

MEK

PI3K

AKT

PTEN

SMAD

Jun

Myc

p53
Ligand upregulation

EGFr

Ligands: epieregulin, amphiregulin

Therapeutic mAb

Signaling pathway:
- EGFr
- ras
- raf
- MEK
- ERK
- PI3K
- AKT
- SMAD
- p53
- Jun
- Myc
- PTEN
PFS (A) and OS (B) according to EREG expression (blue, red) in KRAS-wt patients receiving cetuximab (n=112), and compared with KRAS-mut patients (yellow)

Amphiregulin

Epiregulin

Overall Survival according to AREG and EREG expression in KRAS-wt patients receiving cetuximab (n=75)

CiOx Study Design

Metastatic colorectal cancer
61 pts (of 185 in trial)

R

CAPIRI + Cetuximab
CAPOX + Cetuximab

Endpoints: ORR, PFS, OS

Multivariate analysis for association with ORR

AREG & EREG
EGFR FISH + age sex BRAF-mut
Single Markers

EGFR-FISH: AUC = 0.651

AREG: AUC = 0.661

EREG: AUC = 0.615

Combined Model (AREG+EGFR)

Model correctly identifies patients with better outcomes in this test set (as expected)

- ORR
- PFS
- OS
Conclusions

- In this retrospective and exploratory analysis it was possible to predict ORR probability with the help of molecular markers.

- There is an acceptable discriminatory performance as measured by the AUC for predicting ORR by AREG and EGFR-FISH.

- Both molecular markers might be promising candidates for predicting ORR under cetuximab-based treatment regimens.

- Prospective evaluation of these markers is needed for replicating and validating our findings.
What have we learned?
- the combined use of AREG and EGFR FISH may be useful

What next?
- validate the predictive model in independent dataset(s)
- establish assays for FFPE material
- consider alternative ways of combining data – e.g. [AREG↑ or EREG↑ or EGFR↑] –vs– not
- if still interesting, explore in RCT material
Pharmacogenetic predictors of severe chronic peripheral neuropathy in stage II-III colon cancer patients treated with oxaliplatin-based adjuvant chemotherapy: A GEMCAD study

Toxicity predisposition

• Potential uses:
  – drug avoidance
  – dose reduction [and escalation]
  – targeted preventive measures

• Challenges:
  – pharmacological pathway led –vs– genomic approach to marker identification
  – wide and reliable risk discrimination needed to alter clinical management
The oxaliplatin problem

- High risk in relation to gain for many patients
- Extent of the problem under-estimated initially

original article

Capecitabine combined with oxaliplatin (CapOx) in clinical practice: how significant is peripheral neuropathy?

D. J. Storey¹,²*, M. Sakala², C. M. McLean², H. A. Phillips², L. K. Dawson², L. R. Wall², M. T. Fallon¹ & S. Clive²

“...Concerning, while the majority (94%) experienced acute PN (43% with functional impairment), at least 11% (possibly up to 30%) of adjuvant patients still had unresolved chronic PN which impaired function 12 months after CapOx completion.”
## CLINICAL AND PATHOLOGICAL CHARACTERISTICS

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>All patients (n=379)</th>
<th>Training cohort (n=202)</th>
<th>Validation cohort (n=177)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Median age (range)</strong></td>
<td>61.94 (23-85)</td>
<td>63.82 (23-85)</td>
<td>59.8 (23-76)</td>
</tr>
<tr>
<td><strong>Grade</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Grade 1</td>
<td>60 (15.8%)</td>
<td>19 (9.4%)</td>
<td>41 (23.2%)</td>
</tr>
<tr>
<td>Grade 2</td>
<td>274 (72.3%)</td>
<td>160 (79.2%)</td>
<td>114 (64.4%)</td>
</tr>
<tr>
<td>Grade 3</td>
<td>45 (11.9%)</td>
<td>23 (11.4%)</td>
<td>22 (12.4%)</td>
</tr>
<tr>
<td><strong>Stage</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>II</td>
<td>105 (27.7%)</td>
<td>60 (29.7%)</td>
<td>45 (25.4%)</td>
</tr>
<tr>
<td>III</td>
<td>274 (72.3%)</td>
<td>142 (70.3%)</td>
<td>132 (74.6%)</td>
</tr>
<tr>
<td><strong>Sex</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Men</td>
<td>194 (51.2%)</td>
<td>115 (56.9%)</td>
<td>79 (44.6%)</td>
</tr>
<tr>
<td>Women</td>
<td>185 (48.8%)</td>
<td>87 (43.1%)</td>
<td>98 (55.4%)</td>
</tr>
<tr>
<td><strong>Tumour site</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cecum-right colon</td>
<td>115 (30.34%)</td>
<td>60 (29.7%)</td>
<td>55 (33.1%)</td>
</tr>
<tr>
<td>Transverse colon</td>
<td>23 (6.1%)</td>
<td>15 (7.4%)</td>
<td>8 (4.5%)</td>
</tr>
<tr>
<td>Left colon-sigma</td>
<td>52 (13.7%)</td>
<td>28 (13.9%)</td>
<td>24 (13.6%)</td>
</tr>
<tr>
<td><strong>Adjuvant CT</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FOLFOX</td>
<td>173 (45.6%)</td>
<td>51 (25.24%)</td>
<td>77 (43.5%)</td>
</tr>
<tr>
<td>CAPOX</td>
<td>206 (54.4%)</td>
<td>151 (74.75%)</td>
<td>100 (56.5%)</td>
</tr>
</tbody>
</table>

### PERIPHERAL NEUROPATHY

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<th>Validation cohort (n=177)</th>
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<tr>
<td>No neuropathy</td>
<td>59 (29.2%)</td>
<td>23 (13%)</td>
<td></td>
</tr>
<tr>
<td>Grade 1</td>
<td>95 (47.02%)</td>
<td>82 (46.32%)</td>
<td></td>
</tr>
<tr>
<td>Grade ≥2</td>
<td>48 (23.76%)</td>
<td>72 (40.68%)</td>
<td></td>
</tr>
</tbody>
</table>
# SELECTED GENES AND SNPs

<table>
<thead>
<tr>
<th>Category</th>
<th>Gen</th>
<th>SNP</th>
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<th>Gen</th>
<th>SNP</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Oxaliplatin sensibility/ resistance</strong></td>
<td>ERCC1</td>
<td>rs11615, rs3212964</td>
<td><strong>Drug transport</strong></td>
<td>MDR1/ABC B1</td>
<td>rs1045642</td>
</tr>
<tr>
<td></td>
<td>ERCC2/XPD</td>
<td>rs13181, rs1799793, rs238404</td>
<td></td>
<td>ABCG2</td>
<td>rs2231142</td>
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<tr>
<td></td>
<td>ERCC5/XPG</td>
<td>rs4150279, rs4150360</td>
<td></td>
<td>SULT1A1</td>
<td>rs1968752</td>
</tr>
<tr>
<td></td>
<td>ERCC6</td>
<td>rs2228527, rs7907557</td>
<td></td>
<td>XPA</td>
<td>rs3176639, rs3176751</td>
</tr>
<tr>
<td></td>
<td>XRCC1</td>
<td>rs25489, rs12611088, rs3213255</td>
<td></td>
<td>XPC</td>
<td>rs2607739, rs2733534</td>
</tr>
<tr>
<td></td>
<td>XRCC2</td>
<td>rs3218536, rs3218408, rs3111471</td>
<td></td>
<td>E-selectine</td>
<td>rs3917412, rs3917436</td>
</tr>
<tr>
<td></td>
<td>RAD23B</td>
<td>rs10759225, rs2147072</td>
<td></td>
<td>CCNH</td>
<td>rs2230641, rs3093816</td>
</tr>
<tr>
<td></td>
<td>GSTP1</td>
<td>rs749174</td>
<td></td>
<td>MMP 1</td>
<td>rs498186</td>
</tr>
<tr>
<td></td>
<td>MGMT</td>
<td>rs1803965, rs656639</td>
<td></td>
<td>mTOR</td>
<td>rs2295080, rs357278, rs6895953</td>
</tr>
</tbody>
</table>
CCNH rs2230641+ ABCG2 rs3114018 SNPs and NEUROPATHY (training cohort)

- **CCNH C/C and/or ABCG2 A/A**
  - Neuropathy grade <2: n=33 (63.5%)
  - Neuropathy grade ≥2: n=111 (80.4%)
- **CCNH any T and ABCG2 any C**
  - Neuropathy grade <2: n=19 (36.5%)
  - Neuropathy grade ≥2: n=27 (19.6%)

RR=2.36; p=0.022

CCNH rs2230641+ ABCG2 rs3114018 SNPs and NEUROPATHY (validation cohort)

- **CCNH C/C and/or ABCG2 A/A**
  - Neuropathy grade <2: n=20 (40.8%)
  - Neuropathy grade ≥2: n=74 (59.2%)
- **CCNH any T and ABCG2 any C**
  - Neuropathy grade <2: n=29 (59.2%)
  - Neuropathy grade ≥2: n=38 (33.9%)

RR=2.82; p=0.003
Genes containing or adjacent to SNPs correlating with severe neuropathy in discovery and validation sets (n=96 discovery; 247 validation; 657K SNP profile)

- TAC1
- FOXC1, GMDS
- ITGA1, PELO
- ACYP2, TSPYL6
- BTG4, POU2AF1
- CAMK2N1
- FARS2, LYRM4
- DLEU7
527PD

• What have we learned?
  – convincing data: effect in validation cohort very similar to training set.
  – however CAUTION: hypothesis led SNPs but post-hoc combination of data from the best performing SNPs.

• What next?
  – risk discrimination not wide enough to be clinically applicable ‘as is’ (34% vs 60% grade ≥2)
  – combine with other risk factors (other SNPs, comorbidities)
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