I NETs: a che punto siamo?

I nuovi farmaci: associazione o superamento del trattamento con analoghi

Nicola Fazio
NET: possible targets for novel drugs

Angiogenesis
VEGF, EGF, IGF, PDGF, HGF, TGF-α
VEGFR, EGFR, IGFR, PDGFR
PI3K→Akt→mTOR
Ras→Raf→MAP-K
Proteasoma
Aurora kinase
HADC
# Rationale for Evaluating Novel Therapies in Clinical Trials in Patients with pancreatic NETs

From Capurso et al., Recent Patents on Endocrine, Metabolic & Immune Drug Discovery 2007

<table>
<thead>
<tr>
<th>Molecular Target</th>
<th>Expression in animal model</th>
<th>Expression in human tissue</th>
<th>Activation/mutations in human tissue</th>
<th>Efficacy of inhibitors in animal model</th>
<th>Efficacy of inhibitors in cell lines</th>
<th>Ongoing Clinical Trials</th>
</tr>
</thead>
<tbody>
<tr>
<td>EGFR</td>
<td>Not evaluated</td>
<td>+[42,43,47,48,52]</td>
<td>+ [48,52]</td>
<td>Not evaluated</td>
<td>+ [51]</td>
<td>-</td>
</tr>
<tr>
<td>e-Kit</td>
<td>Not evaluated</td>
<td>+[44,57]</td>
<td>Not evaluated</td>
<td>+ [17]</td>
<td>+ [60]</td>
<td>-</td>
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<tr>
<td>IGF-1</td>
<td>Not evaluated</td>
<td>+ [83,84,99]</td>
<td>Not evaluated</td>
<td>Not evaluated</td>
<td>+ [85]</td>
<td>-</td>
</tr>
<tr>
<td>mTOR</td>
<td>Not evaluated</td>
<td>+ [78]</td>
<td>+ [78]</td>
<td>Not evaluated</td>
<td>+ [73]</td>
<td>+</td>
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<tr>
<td>Agent</td>
<td>Study Design</td>
<td>Tumour type</td>
<td>Disease Stage</td>
<td>Histology</td>
<td>Behaviour before entry</td>
<td>TX duration (median)</td>
</tr>
<tr>
<td>------------------</td>
<td>-------------------------------</td>
<td>-------------</td>
<td>---------------</td>
<td>-----------</td>
<td>------------------------</td>
<td>---------------------</td>
</tr>
<tr>
<td>Bevacizumab</td>
<td>PII RT + OCT BVZ vs IFN</td>
<td>40 CT</td>
<td>Metastatic</td>
<td>NR</td>
<td>NR</td>
<td>18 weeks</td>
</tr>
<tr>
<td>Bevacizumab</td>
<td>PII + Temozolomide OCT allowed</td>
<td>18 PET 16 CT</td>
<td>Metastatic</td>
<td>27 WDEC 7 PDEC</td>
<td>NR</td>
<td>22 weeks</td>
</tr>
<tr>
<td>Sunitinib</td>
<td>PII OCT allowed</td>
<td>52 PET 41 CT</td>
<td>Advanced Unresectable</td>
<td>NR</td>
<td>NR</td>
<td>204 days (26-543)</td>
</tr>
<tr>
<td>Thalidomide</td>
<td>PII + Temozolomide OCT allowed</td>
<td>11 PET 15 CT, 3 PC</td>
<td>Metastatic</td>
<td>28 WDEC 1 PDEC</td>
<td>NR</td>
<td>7.3 months (1-23)</td>
</tr>
<tr>
<td>Endostatin</td>
<td>PII OCT allowed</td>
<td>20 PET 22 CT</td>
<td>Metastatic</td>
<td>38 WDEC 4 PDEC</td>
<td>PD 11/23 evaluated</td>
<td>6.4 months (10-45)</td>
</tr>
<tr>
<td>Gefitinib</td>
<td>PII OCT allowed</td>
<td>39 PET 57 CT</td>
<td>Metastatic</td>
<td>NR</td>
<td>PD</td>
<td>NR</td>
</tr>
<tr>
<td>Imatinib</td>
<td>PII OCT allowed</td>
<td>27 CT</td>
<td>Metastatic</td>
<td>27 G1/G2</td>
<td>PD 14/24 evaluated</td>
<td>NR</td>
</tr>
<tr>
<td>Everolimus + OCT</td>
<td>PII + OCT</td>
<td>13 PET 18 CT</td>
<td>Advanced</td>
<td>“low grade” PD in 21</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>Temsirolimus</td>
<td>PII Single agent</td>
<td>15 PET 21 CT</td>
<td>Metastatic</td>
<td>NR</td>
<td>PD</td>
<td>16 weeks (1-21)</td>
</tr>
</tbody>
</table>
Should we change the response evaluation criteria?

Sorafenib in HCC
“Tissue response” rather than tumor shrinkage

Pre-treatment
4 months later
Size and density at CT-scan vs SUV at PET-scan
Good correlation
Hypervascularized GEP-NET overexpress VEGF and VEGFR-1,2 in tumor cells and surrounding vasculature

Wiedenmann et al., Neuroendocrinology 2004

<table>
<thead>
<tr>
<th>Tissue Type</th>
<th>VEGF (%)</th>
<th>VEGFR (%)</th>
</tr>
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<tbody>
<tr>
<td>Carcinoids</td>
<td>100</td>
<td>70</td>
</tr>
<tr>
<td>Pancreatic</td>
<td>85</td>
<td>75</td>
</tr>
</tbody>
</table>

Kulke, ASCO 2006

High VEGF expression correlates with increased angiogenesis and decreased PFS in low-grade NET

Zhang et al., Cancer 2007
Targeting Vascular Endothelial Growth Factor in Advanced Carcinoid Tumor: A Random Assignment Phase II Study of Depot Octreotide With Bevacizumab and Pegylated Interferon Alfa-2b

Yao et al., J Clin Oncol 26:1316-1323. © 2008

Octreotide LAR not more than 30 mg q3w

BV 15 mg/Kg q3w

PEG-IFN 0.5 mcg/Kg/w
<table>
<thead>
<tr>
<th>Characteristic</th>
<th>No.</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Disease at entry</td>
<td></td>
<td></td>
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<tr>
<td>PD</td>
<td>23</td>
<td>52.3</td>
</tr>
<tr>
<td>SD</td>
<td>18</td>
<td>40.9</td>
</tr>
<tr>
<td>Unknown</td>
<td>3</td>
<td>6.8</td>
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<tr>
<td>Primary site</td>
<td></td>
<td></td>
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<tr>
<td>Foregut</td>
<td>6</td>
<td>13.6</td>
</tr>
<tr>
<td>Gastric</td>
<td>1</td>
<td>2.3</td>
</tr>
<tr>
<td>Lung</td>
<td>4</td>
<td>9.1</td>
</tr>
<tr>
<td>Thymus</td>
<td>1</td>
<td>2.3</td>
</tr>
<tr>
<td>Midgut</td>
<td>24</td>
<td>54.5</td>
</tr>
<tr>
<td>Ileum</td>
<td>11</td>
<td>25.0</td>
</tr>
<tr>
<td>Small intestine</td>
<td>12</td>
<td>27.3</td>
</tr>
<tr>
<td>Caecum</td>
<td>1</td>
<td>2.3</td>
</tr>
<tr>
<td>Hindgut</td>
<td>4</td>
<td>9.1</td>
</tr>
<tr>
<td>Rectum</td>
<td>4</td>
<td>9.1</td>
</tr>
<tr>
<td>Unknown</td>
<td>10</td>
<td>22.7</td>
</tr>
</tbody>
</table>
PFS at 18 w = 95% BV v 68% PEG-INF; p = 0.02
Overall PFS for 44 pts = 63 w (95%CI: 51-75)
PFS 66 w BV v 56 w PEG-IFN; p = 0.34

PFS shorter for pts with PD at study entry; p = 0.005

Yao et al., J Clin Oncol 26:1316-1323. © 2008
<table>
<thead>
<tr>
<th>Arm</th>
<th>pts</th>
<th>PR</th>
<th>SD</th>
<th>PD</th>
</tr>
</thead>
<tbody>
<tr>
<td>BV</td>
<td>22</td>
<td>18%</td>
<td>77%</td>
<td>5%</td>
</tr>
<tr>
<td>IFN</td>
<td>22</td>
<td>0</td>
<td>68%</td>
<td>27%</td>
</tr>
</tbody>
</table>

- 7 pts with PD during PEG-IFN with combination → PR 1
- SD 5
- 1 pt with PD during BV with combination → PD

Yao et al., J Clin Oncol 26:1316-1323. © 2008
Table 3. Selected Grade 3/4 Events During Stage I Monotherapy (first 18 weeks) by Treatment Arm According to Common Toxicity Criteria (version 2)

<table>
<thead>
<tr>
<th>Toxicity</th>
<th>Bevacizumab</th>
<th></th>
<th>PEG Interferon</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. of Patients</td>
<td>%</td>
<td>No. of Patients</td>
<td>%</td>
</tr>
<tr>
<td>Granulocytopenia</td>
<td>0</td>
<td>0</td>
<td>6</td>
<td>27</td>
</tr>
<tr>
<td>Hypertension</td>
<td>8</td>
<td>36</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Nausea</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>9</td>
</tr>
<tr>
<td>Fatigue</td>
<td>4</td>
<td>18</td>
<td>5</td>
<td>23</td>
</tr>
<tr>
<td>Vomiting</td>
<td>1</td>
<td>5</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>Myalgia</td>
<td>2</td>
<td>9</td>
<td>2</td>
<td>9</td>
</tr>
<tr>
<td>Headache</td>
<td>1</td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Anxiety</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>AlkPhos Inc</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>5</td>
</tr>
</tbody>
</table>

Abbreviation: PEG, pegylated.
Changes in BF and BV 48 hrs after Bevacizumab

Yao et al., J Clin Oncol 26:1316-1323. © 2008
Yao et al., J Clin Oncol 26:1316-1323. © 2008
Functional CT = 24 pts
Only 2 responders had fCT
lower day 2 BV = longer PFS (predictive?)

PEG-IFN → reduction bFGF, increase IL-18

Yao et al., J Clin Oncol 26:1316-1323. © 2008
SWOG phase III trial (study S0518)
OCT + IFNα vs OCT + BV

ongoing
<table>
<thead>
<tr>
<th>Study Description</th>
<th>Status/Details</th>
</tr>
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<tbody>
<tr>
<td>NCI phase I-II trial with FOLFOX plus BV in refractory carcinoids and pancreatic endocrine tumors</td>
<td>Ongoing (102 pts from Jun ‘05)</td>
</tr>
<tr>
<td>BV + RAD001 in Advanced Low or Intermediate Grade Neuroendocrine Carcinoma</td>
<td>Ongoing from Jan ‘08</td>
</tr>
<tr>
<td>XELBEVOCT: Xeloda 2000 mg/sm/day continuously + BV 5 mg/Kg/q2w + OCT LAR</td>
<td>Torino-Bologna-S.Giov Rot – 15 pts</td>
</tr>
<tr>
<td>Xeloda + BV + TMZ</td>
<td>Uppsala</td>
</tr>
<tr>
<td>XELOX + BV</td>
<td></td>
</tr>
<tr>
<td>Temozolomide + BV</td>
<td></td>
</tr>
<tr>
<td>Panzem + BV</td>
<td></td>
</tr>
</tbody>
</table>
Types of stimuli that modulate mTOR:
Growth Factors, hypoxia, amino acids, intracellular ATP concentrations
mTOR pathway is deregulated by mutations in cancer

- mTOR pathway
- mTOR inhibiting
- mTOR activating

**Growth signaling**
- EGF
- IGF
- VEGF

**Nutrients**
- Nutrients

**Cell Growth & Proliferation**
- Protein Synthesis
- Bioenergetics
- Angiogenesis

**Signaling Pathways**
- mTOR
- PTEN
- PI3K
- AKT
- Ras
- Abl
- ER
- NF1
- TSC1
- TSC2

**mTOR pathway**
- mTOR inhibiting
- mTOR activating
mTOR activation supports cancer cell growth

Nutrients

mTOR

S6K1

Protein Synthesis

4E-BP1

eIF-4E

Cyclin D1

HIF-1α

Glut1

LAT1

Cell growth

Angiogenesis

Nutrient uptake

Istituto Europeo di Oncologia
From Abraham, Expert Opin Ther Targets 2008
mTOR inhibitors

Easter island (Rapa Nui) soil

\[ \downarrow \]

Streptomyces hygroscopicus

\[ \downarrow \]

RAPAMYCIN (Sirolimus, Rapamune®)

\[ \downarrow \]

CCI-779 (Temsirolimus)

\[ \downarrow \]

AP23573

\[ \downarrow \]

RAD-001

(Everolimus, Certican®)
Phase II study of Temsirolimus in advanced neuroendocrine carcinoma

37 progressive pts

Efficacy

PR 5.6% (95% CI 0.6-18.7%)
TTP 6 months
1-y OS 71.5%

Toxicity all grades

Fatigue 78%
Hyperglycemia 69%
Rash/desquamation 64%

Duran et al., Br J Cancer 2006
Temsirsimimus effectively inhibited the phosphorylation of S6 ($P=0.02$).

Higher baseline levels of pmTOR ($P=0.01$) predicted for a better response.

Increases in pAKT ($P=0.041$) and decreases in pmTOR ($P=0.048$) after treatment were associated with an increased TTP.

Duran et al., Br J Cancer 2006
Expression and activation of mTOR in neuroendocrine tumors. Effects of mTOR inhibition by RAD001 upon growth, cell cycle regulation and signalling in neuroendocrine cell lines

D. Hörsch, et al. ASCO 2007

mTOR is expressed and activated in different NET and inhibition of mTOR by RAD001 induces growth inhibition, cell cycle arrest and decreased signaling by mTOR and ERK1/2 in neuroendocrine cell lines INS-1 and BON.
mTOR- inhibitor + IGFR-inhibitor

**IGF VEGF**
- **IGFR / VEGFR**
  - **PI3K**
  - **PKDI**
  - **PTEN**
- **AKT**
  - **TSC1**
  - **TSC2**
- **p53**

**sst-analog**
- **RAD001**
  - **mTOR**
  - **p70S6K**
  - **O’Reilly et al., Cancer Res 2006**
Octreotide and the mTOR Inhibitor RAD001 (Everolimus) Block Proliferation and Interact with the Akt-mTOR-p70S6K Pathway in a Neuro-Endocrine Tumour Cell Line

Simona Grozinsky-Glasberg

Octreotide and RAD001 share common endpoints
No additive effect when used together
Phase II study of RAD001 plus Octreotide LAR

Metastatic WD NET
Single-arm
Cohort 1 → 30 pts (RAD 5 mg/day)
Cohort 2 → 30 pts (RAD 10 mg/day)
Octreotide LAR 30 mg I.M. q 4 w

Yao et al., ASCO 2007
Phase II study of RAD001 plus Octreotide LAR

Baseline PD  65%
Baseline SD  27%
Most pre-treated
Pancreatic  29 (48%)
Midgut     16 (27%)
Liver mets  95%

Yao et al., ASCO 2007
# Phase II study of RAD001 plus Octreotide LAR

## G3-4 non-hematological toxicity

<table>
<thead>
<tr>
<th>Condition</th>
<th>5 mg</th>
<th>10 mg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mucositis</td>
<td>6%</td>
<td>9%</td>
</tr>
<tr>
<td>Fatigue</td>
<td>15%</td>
<td>6%</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>9%</td>
<td>13%</td>
</tr>
<tr>
<td>Skin rash</td>
<td>6%</td>
<td>3%</td>
</tr>
<tr>
<td>Pneumonitis</td>
<td>0</td>
<td>3%</td>
</tr>
</tbody>
</table>

Yao et al., ASCO 2007
Phase II study of RAD001 plus Octreotide LAR

G3-4 hematological toxicity

<table>
<thead>
<tr>
<th>Condition</th>
<th>5 mg</th>
<th>10 mg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thrombocytopenia</td>
<td>3%</td>
<td>6%</td>
</tr>
<tr>
<td>Anemia</td>
<td>3%</td>
<td>3%</td>
</tr>
<tr>
<td>Leukopenia</td>
<td>3%</td>
<td>6%</td>
</tr>
<tr>
<td><strong>Hyperglicemia</strong></td>
<td>3%</td>
<td>13%</td>
</tr>
<tr>
<td>Hypoglicemia</td>
<td>6%</td>
<td>0</td>
</tr>
<tr>
<td>Hypertrigliceridemia</td>
<td>0</td>
<td>6%</td>
</tr>
<tr>
<td>Hypophosphatemia</td>
<td>6%</td>
<td>16%</td>
</tr>
<tr>
<td>Hypokaliemia</td>
<td>3%</td>
<td>3%</td>
</tr>
<tr>
<td>Elevated AST</td>
<td>3%</td>
<td>3%</td>
</tr>
<tr>
<td>Elevated ALT</td>
<td>0</td>
<td>6%</td>
</tr>
<tr>
<td>Hyperbilirubinemia</td>
<td>0</td>
<td>3%</td>
</tr>
</tbody>
</table>

Yao et al., ASCO 2007
**Phase II study of RAD001 plus Octreotide LAR**

<table>
<thead>
<tr>
<th>Efficacy by dose level</th>
<th>Overall</th>
<th>5 mg</th>
<th>10 mg</th>
</tr>
</thead>
<tbody>
<tr>
<td>n=60</td>
<td>n=30</td>
<td>n=30</td>
<td></td>
</tr>
<tr>
<td>PR</td>
<td>12 (20%)</td>
<td>4 (13%)</td>
<td>8 (27%)</td>
</tr>
<tr>
<td>SD</td>
<td>43 (72%)</td>
<td>22 (73%)</td>
<td>21 (70%)</td>
</tr>
<tr>
<td>PD</td>
<td>5 (8%)</td>
<td>4 (13%)</td>
<td>1 (3%)</td>
</tr>
<tr>
<td>PFS</td>
<td>59 w</td>
<td>50 w</td>
<td>62 w</td>
</tr>
</tbody>
</table>

*Yao et al., ASCO 2007*
Phase II study of RAD001 plus Octreotide LAR

PFS in pts with PD at study entry = 50 w (95% CI 34-66)
PFS in pts with SD at study entry = 73 w (95% CI 65-80)
MOS not reached
2-year survival rate 78%

Yao et al., ASCO 2007
Phase II study of RAD001 plus Octreotide LAR

IHC on 25 samples
Pre- and on-treatment = 19 pts
PTEN expression, pAkt, pmTOR, p4EBP1, pS6

All tumors expressed pmTOR and almost all PTEN
Carcinoids more pAkt than pancreatic
Increase in pAkt not associated with PD

Meric-Bernstam et al., AACR 2008
RAD001 In Advanced Neuroendocrine Tumors
RADIANT

RADIANT-1: Phase 2 open label study of RAD001 in advanced pancreatic neuroendocrine tumors after failure of chemotherapy (2239) - CLOSED

RADIANT-2: Phase 3 double-blind placebo-controlled study of RAD001 in pts receiving Octreotide LAR for advanced carcinoid tumors (2325) - CLOSED

RADIANT-3: Phase 3 double-blind placebo-controlled study of RAD001 in advanced pancreatic tumors (2324) - ONGOING
Inhibition of mTOR pathway by everolimus cooperates with EGFR inhibitors in human tumours sensitive and resistant to anti-EGFR drugs.

Targeting mTOR pathway with everolimus overcomes resistance to EGFR inhibitors and produces a cooperative effect with EGFR inhibitors.
SUNITINIB MALATE (SU11248, SUTENT)

SUNITINIB

- VEGFR-1,2
- RET
- FLT-3
- c-Kit
- PDGFR-$\alpha,\beta$
Phase I study of Sunitinib (SU11248, Sutent) in advanced carcinomas

28 pts $\rightarrow$ 4 pts with NET (1 PR e 1 MR)

*Faivre et al., JCO 2006*
Phase II study of Sunitinib (SU11248, Sutent) in advanced carcinoid tumors

106 pretreated pts: pancreatic/carcinoid balanced

Sunitinib 50 mg/d po on schedule 4/2

15% PR and 75% SD in pancreatic
2% PR and 93% SD in carcinoids

Low rate of G3-4 toxicity: diarrhea, fatigue, hypertension, myelotox

*Kulke et al., ASCO 2005*

sVEGFR-3 as novel biomarker for the biological activity of sunitinib
IL-8 potential predictor of response

*Bello, ASCO 2006*
Sunitinib trials ongoing in NET

A phase III randomized, double-blind study of SUNITINIB (SU011248,SUTENT®) vs placebo in pts with progressive advanced WD pancreatic islet cell tumors

Sunitinib 37.5 mg daily starting dose, continuous daily regimen
SORAFENIB (BAY 43-9006)

- VEGFR-2,3
- Raf kinase
- FGFR-1
- FLT-3
- c-Kit
- PDGFR-β
MC044h, a phase II trial of sorafenib in patients (pts) with metastatic neuroendocrine tumors (NET): A Phase II Consortium (P2C) study

90 pts (50 intestinal, 43 pancreatic)

Sorafenib 400 mg po BID

10% PR in intestinal and 10% in islet cell
6m PFS in 8/20 intestinal and 14/23 islet cell

Grade 3-4 toxicity 43% of pts
- skin 20%
- GI 7%
- fatigue 9%

Hobday et al., ASCO 2007
Sorafenib trials ongoing in NET

An international multicenter phase II study of Sorafenib in pts with progressive metastatic neuroendocrine tumors (Mayo Clinic)

Ongoing: Jun ’05 – 90 pts
Oral sorafenib twice daily on days 1-28
Phase II trial of temozolomide plus thalidomide in NET

TMZ 150 mg/m2/day  po 1 week on 1 week off + THAL 200 mg/day po
29 pts  28 WD & 1 PD
13 pretreated with chemo
RR 25% (pancreas 45%, pheocrom 33%, carcinoids 7%)
SD 68%
Response duration 13 m (2-31 m) – PFS & OS not yet reached
2-y surv 60%

Tox: 60% G3-4 lymphopenia (3 cases of opportunistic infections)
55% pts withdrew due to toxicity within an average of 8 months

Kulke et al. JCO Jan 2006
36 pts with advanced NET (1 gastric, 7 thymic and 13 bronchial, 12 pancreatic, 1 paraganglioma, 1 foregut, and 1 cecal)

Temozolomide (200 mg/m^2) for 5 days every 4 weeks

PR 14%  SD 53%

Low MGMT = better response rate
The resistance to TMZ may be partially overcome by changing conventional to metronomic schedule.

Temozolomide 50 mg/m2/d continuously

Verhoeff et al., AACR 2008
Novel drugs in NET: open questions

- Homogeneous populations
- Status of disease at study entry
- RECIST
- PET with 5-HTTP or L-DOPA
- Surrogate biomarkers
- Combination of different biological agents
- SS analogs as control arm (?)
- Subgroup identification based on predictive factors
- TNM classification