Biomarkers - Oncology and Inflammation

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Issues with drug discovery

According to the latest annual review of pharma R&D value by Deloitte and Thomson Reuters, Measuring the Return from Innovation, the average cost of bringing a new product successfully to market among the top 12 research-based pharmaceutical companies worldwide increased by 26.3% from US$830 million in 2010 to US$1,048 million in 2011.

Over the same period, the number of late-stage compounds in development dropped from 23 on average per company to 18 per company. Moreover, the average R&D Internal Rate of Return (IRR) among the companies analysed was down from 11.8% in 2010 to 8.4% this year.

The Critical Path Initiative

to ‘ensure that basic scientific discoveries translate more rapidly into new and better medical treatment by creating new tools to find answers of how the safety and efficacy of new medical products can be demonstrated in faster time frames, with more certainty, at lower cost and with better information’

[http://www.fda.gov].
CPI - biomarkers

Critical Path Initiative (FDA) / Innovative Medicine Initiative (EMEA)

- **Clinical endpoint**: how a patient feels, functions or survives
- **Biomarker**: objective measurement associated with pathological process / therapeutic intervention with prognostic quality
- **Surrogate**: validated biomarker that substitutes for a clinical endpoint

Biomarkers - imaging

- **Biomarkers** should be:
  - non-invasive
  - should allow for follow-up studies
  - focal
  - simple
  - low-cost

- **Non-invasive imaging** providing:
  - morphological
  - physiological
  - metabolic
  - cellular
  - molecular

- Readouts in tempo-spatially resolved manner

- Potential biomarkers for patient staging / stratification, proof of mechanism and therapeutic efficacy

Multimodal imaging inherently translational

Optimal method depends on question to be answered

- **Molecular constituents**: e.g. receptor expression
  - sensitivity (low concentrations)
  - specificity (minimal cross-contamination of signals)
  - temporal resolution

- **Physiology**: e.g. tumor perfusion
  - temporal resolution (analysis of bolus passage)

- **Morphology**: e.g. volume, shape, heterogeneity
  - spatial resolution

- **Optimal method** depends on the question to be answered

Tumor: structural imaging

Detection of space occupying lesions (native contrast or contrast enhancement)

Staging – texture / heterogeneity, infiltration of adjacent tissue, tumor volume

Therapy assessment – RECIST (response criteria of solid tumors)

Therapy effect on tumor volume

Effect of octreotide treatment on hormone dependent tumors expressing SST receptors

- Estradiol induced pituitary hyperplasia
- Duping R1227-H prostate tumor

Rudin et al. MM1 (1986)
information from imaging data

- drug biodistribution
- target expression
- receptor occupancy
- signaling pathways

hires structural imaging
- in vivo morphometry
- in vivo morphology
- disease phenotyping/dx

functional imaging
- physiological measurements
- functional MRI
- functional receptor imaging

cellular imaging
- cell migration & fate
- cell therapies

the hallmarks of cancer (1st generation)

Hanahan & Weinberg, Cell 144, 694 (2011)

intracellular signaling networks in cancer

Hanahan & Weinberg, Cell 144, 694 (2011)

the hallmarks of cancer (2nd generation)

Hanahan & Weinberg, Cell 144, 694 (2011)
Imaging cancer hallmarks

- Proliferation: FST-PET, choline
- Glucose metabolism: FDG-PET
- Apoptosis: P53, Cas 3
- Immune cells: TAMs
- ECM remodeling: proteases
- Permeability, TBV & vascular architecture

Assessment of tumor vascular architecture

In vivo tumor mouse

Excised tumor

collaboration with S. Lang, B. Müller (U. Basel)

How to characterize tumor vascular architecture?

- Normal tissue
  - Hierarchical architecture
  - Efficient perfusion
- Tumor
  - Chaotic
  - Insufficient perfusion

Vascular permeability and tumor blood volume maps in murine B16 melanoma model

- Primary tumors (ear)
- Anatomical
- Permeability [P*S]
  - Baseline
  - Post-treatment

Effect of VEGF-R inhibitor vatalanib on vascular permeability of cervical lymphnode metastases

Vascular permeability with DCE-MRI: translation

Enhancement of a liver metastasis at baseline and 30 hours after treatment with PTK 787

Baseline

After PTK treatment

Mean Ki (% Body) vs. Disease Status

- Progressive disease
- Stable disease

Vascular permeability biomarker - issues

- Approved CA (Gd-DTPA's) small hydrodynamic radius → highly leaky
- Reduced dynamic range
- Novel CAs: larger hydrodynamic radius

- Tumor heterogeneity not accounted for by conventional analysis
- Tissue classification (e.g. based on multiparametric MRI readouts)
- Pattern analysis techniques
**multiparametric analysis of vascular permeability with DCE-MRI**

- **Tissue classification**
- **Permeability Parameters**
  - ADC, T2, M0
- **Ktrans**

**Figure 4**

**multimodal imaging of angiogenesis: parameters/modeling**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Biological Information</th>
<th>Mod Process</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vascular permeability</td>
<td>Sites of angiogenesis</td>
<td>MRI</td>
</tr>
<tr>
<td>Tumor blood volume</td>
<td>Total vascularization</td>
<td>MRI</td>
</tr>
<tr>
<td>TBB and vessel size</td>
<td>Vascular architecture</td>
<td>MRI</td>
</tr>
<tr>
<td>Hypoxia</td>
<td>Tumor oxygenation</td>
<td>PET</td>
</tr>
<tr>
<td>HIF1a and HIF activity</td>
<td>Hypoxic signaling</td>
<td>OPT</td>
</tr>
<tr>
<td>VEGF</td>
<td>Proangiogenic signaling</td>
<td>OPT</td>
</tr>
<tr>
<td>Adhesion molecules</td>
<td>Activated endothelium</td>
<td>OPT</td>
</tr>
<tr>
<td>Proteases</td>
<td>ECM degradation</td>
<td>OPT</td>
</tr>
<tr>
<td>Inflammatory cells</td>
<td>Immune response / ECM</td>
<td>MRI</td>
</tr>
<tr>
<td></td>
<td>Degradation</td>
<td></td>
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</tbody>
</table>

**Imaging cancer hallmarks**

- **Proliferation**: FLT-PET, choline
- **Glucose metabolism**: FDG-PET
- **Apoptosis**: PdtSer, Cas-3
- **Permeability**: TBV & vascular architecture
- **ECM remodeling**: proteases
- **Immune cells**: TAMs

**therapy effects on tumor glucose metabolic activity**

- **18F-FDG PET** provides evidence of biological response to imatinib, within hours/days and earlier than classical CT measurements.
- Early post-treatment SUV values predict long-term response both in mouse model and human patient.

**therapy effects on FDG activity: translation to mouse**

murine tumors of FDC-P1 cell lines expressing c-KIT mutations that render the tumors either responsive (V560G) or resistant (D816V) to imatinib.
therapy effects on tumor glucose metabolic activity

clinical efficacy of PLX4032 (RG7204), a potent inhibitor of oncogenic B-RAF kinase activity, in patients with BRAF-mutant melanoma


comparison FDG- versus FLT-PET for tumor detection

lymphoma

Buck AK et al, Cancer Res. 2006;66:11055–11061

potential tumor therapy MRI biomarker

- DCE-MRI: alterations in vascular permeability → angiogenesis
- ADC mapping: changes in water diffusivity → cellularity / apoptosis
  - Evaluation of diffusion parameters as early biomarkers of disease progression in glioblastoma multiforme
- MRS: effects on tumor glucose or lipid metabolism
  - Gallagher JA et al. (2008) PNAS 105: 19801-4
  - Production of hyperpolarized [1,4-13C]glycerol from [1,4-13C]acetate is a marker of cell survival and treatment response in tumors

Gallagher FA et al. (2008) PNAS 105: 19801-4

deriving quantitative data from imaging data sets

how to translate imaging information into meaningful biological information?

→ morphometric analysis
→ densitometric analysis

1) translate intensity into concentration
   - PET: activity
   - MRI: relaxation change
   - FMT: intensity
   - amount of tracer in voxel: weighted sum of all compartments

2) correct for confounding contributions (scattering, absorption, transport/diffusion, chemical exchange/metabolism, bleaching,...)

3) dynamic modeling of concentrations in individual compartments

4) relate model parameters to biological process


Moffat BA et al. (2005) PNAS 102: 5524-9

Gallagher FA et al. (2008) PNAS 105: 19801-4

MRI Cologne, Germany

Glioblastoma multiformae

CE-MRI [18F] FDG [18F]-MET [18F]-FLT

MPI Cologne, Germany
Robustness / reproducibility of measurement

Estimates of various error sources

a) Experimental
   - Instrumental phantom studies
   - Biological: intraindividual, interindividual
   - Procedural: reproducibility of procedures automated procedures

b) Data Analysis
   - automated using validated procedures
   - semiautomated
   - operator-interactive

B16 melanomas: cervical lymphnode metastases

Robustness / reproducibility of measurement

Local activity proportional to local concentration of radioisotope

\[ Q(R) = \frac{dN(R)}{dt} = \lambda \cdot N(R) \]

Injected dose per gram of tissue: %ID/g

\[ \%ID/g = \frac{V \cdot \gamma}{D_m} \cdot 100\% \]

Standardized uptake value: SUV

\[ SUV = \frac{\%ID/g}{M} \cdot \frac{1}{\frac{V}{w}} \cdot M \]

\[ SUV^* = \frac{\%ID/g}{S} \cdot \frac{1}{\frac{V}{w}} \cdot S \]

Imaging yields concentration in a voxel

Voxel comprises multiple compartments, therefore \( c_i(t) \) corresponds to volume averaged concentration of CA across these compartments, i.e.

\[ c_i(t) = \frac{1}{V} \sum_{v} c_v(t) \]

Multicompartment models to deconvolve individual contributions

\[ \begin{align*}
 c_1(t) &\rightarrow c_2(t) \\
 c_3(t) &\rightarrow c_4(t) \\
 cb &\rightarrow c_5(t) \\
 cp &\rightarrow c_6(t)
\end{align*} \]

or the Scatchard equation:

\[ \frac{[R]}{[L]} + \frac{[L]}{[R]} = \frac{[L]}{[R]} \]

Receptor affinity from concentrations

Receptor Binding: \( L + R \leftrightarrow RL \)

Equilibrium constant:

\[ K_J = \frac{[L][R]}{[RL]} \]

(principle of microreversibility):

\[ K_{off} [RL] = K_{on} \cdot [R] \cdot [L] \]

Mass conservation for receptor:

\[ [R] = [R] + [L] \]

yields

\[ K_J = \frac{[L]}{[R]} \cdot \frac{[R]}{[L]} \]

or the Scatchard equation:

\[ \frac{[R]}{[L]} + \frac{[L]}{[R]} = \frac{[L]}{[R]} \]

Determination of \( c_i(t) \) for compartment of interest

Assumption: relaxivity depends linearly on amount of tracer in voxel

\[ R_i = R_0 + r \cdot c_{tr} \cdot V \]

Tissues:

- deviations from linearity
- mixed contrast e.g. \( R_1 \& R_2 \) (ambiguities)

deriving concentrations from signal intensities: PET

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**Phyiological Modeling**

Dynamic modeling of vessel formation

**Biomarkers: Facilitating Transition from Discovery to Clinics?**

Today there are only few examples of using imaging information to facilitate the translation from mouse to man

- Too early to tell
- Technical issues: tools used for mouse imaging not appropriate for clinical setting
  - Not approved
  - Not standardized (i.e. not suited for multi-center trials)
  - Not properly validated
  - Not quantitative
- Perceived issues: e.g. acceptance/expectations by drug developer
- A mouse is a model for man – not man!

**Translational Biomarkers – A Multimodality Approach**