What is a biomarker and what can it be used for?
DSBS meeting February 21, 2011
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Outline
• Biomarker definition
• Classical examples of biomarkers
• Purposes of biomarkers (more examples)
• Clinical trial programs
• (Regulatory aspects)
• Gene expression biomarkers

Biomarker definition
• A characteristic that is objectively measured and evaluated as an indicator of healthy biological processes, pathological processes, or pharmacological responses to therapeutic intervention
• Note: “an indicator of” means “reflecting”
• “evaluated as” means “with the purpose of”, not “proven to be”

Properties of a biomarker?
• Constant (vs. Time-dependent)?:
  Yes (like genes), but constant biomarkers cannot be outcomes
• Binary? Yes, but outcome biomarkers are more useful if they are continuous, I think
• Multivariate? Yes (but often this is used to derive a univariate summary)

Examples that are not biomarkers
• Rating scales (MADRS)
• PRO (patient reported outcomes)
• Tolerability
• Pain
• Imaging interpretation (picture alone is a biomarker)

Surrogate endpoint definition
• A biomarker that is intended to substitute for a clinical endpoint. A surrogate endpoint is expected to predict clinical benefit (or harm or lack of benefit or harm) based on epidemiological, therapeutic, pathophysiological, or other scientific evidence
Weight

• Most classical example
• Doctors office: Light clothes, no shoes
• Home: very light clothes, morning
• Daily variation in the kg-range
• Does weight reflect health? Age-dependence; height adjustment; composition: fat versus muscles: apple-shape (abdominal fat)
• Does weight-change reflect health?
• Is weight-reducing treatments helpful? Water-reducing; metabolism-increasing

Related variables

• BMI (weight/height^2)
• LBM (lean body mass) formula
• LBM concept
• BSA (body surface area) formula
• Waist circumference
• Waist hip ratio

FDA (weight)

• Primary endpoint: %weight loss (mean difference to placebo stat. sign. and ≥5%) + response criteria
• Inclusion: BMI ≥ 30 or BMI ≥ 27 + comorbidity
• Reasonable sample size: 4500 for 1y
• DEXA scanning on some
• Safety important: Recently a drug was taken off the market due to increased cardiovascular risk

Temperature

• Circadian variation
• Core temperature versus surface temperature
• Measurements: rectal, mouth, ear
• Monthly variation (females)
• Fever (increased temperature) present with many infections

Blood pressure

• Systolic and diastolic
• Supine or sitting
• White coat hypertension
• High measurement error
• Office versus 24h measurements
• EMA guideline (2010): 3 baseline visits; systolic primary; diastolic secondary; sufficient number of patients for mortality (1y); no suspicion of adverse target organ effect

Cardiovascular markers

• Cholesterol (high-density versus low-density)
• Free fatty acids
• Triglycerides
Diabetes biomarker primer

- Blood glucose sensitive to meals.
  Strategies:
  - 1. Unrestricted (high upper normal limit)
  - 2. Fasting (overnight)
  - 3. Fasting supplemented with specific challenge (glucose tolerance test)
  - 4. HbA1c (reflecting average blood glucose over 6 weeks)

Diabetes biomarkers

<table>
<thead>
<tr>
<th>Biomarker</th>
<th>Type 1</th>
<th>Type 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urine glucose (UT)</td>
<td>↑</td>
<td>↓</td>
</tr>
<tr>
<td>Blood glucose (UT)</td>
<td>↑</td>
<td>↓</td>
</tr>
<tr>
<td>Hba1c (target)</td>
<td>↑</td>
<td>↓</td>
</tr>
<tr>
<td>Insulin (UT)</td>
<td>↓</td>
<td>↑</td>
</tr>
<tr>
<td>C-peptide</td>
<td>↓</td>
<td>↑</td>
</tr>
<tr>
<td>Antibodies (sel.)</td>
<td>↑</td>
<td>Normal</td>
</tr>
<tr>
<td>Insulin resistance</td>
<td>Normal</td>
<td>↑</td>
</tr>
<tr>
<td>Age</td>
<td>Flat incidence</td>
<td>Old</td>
</tr>
<tr>
<td>BMI</td>
<td>Normal</td>
<td>↑</td>
</tr>
<tr>
<td>pH/HCO3 (UT)</td>
<td>↓ (severe)</td>
<td>↑</td>
</tr>
</tbody>
</table>

Conclusion on classical biomarkers

- Many classical biomarkers are widely used and accepted, both to define diseases and for treatment decisions
- However, it is also well known that they are far from perfect

Types of biomarkers

- Diagnostic
- Early detection
- Monitoring
- Prognostic
- Predictive/Safety/dose

Examples-diagnostic

- Disease vs. healthy or disease vs. other diseases?
- Temperature
- Bacterial tests (streptococcal)
- Origin of cancer tissue

Examples – early detection

- Gene test for monogenic diseases (like cystic fibrosis)
- PSA (still controversial)
- Breast cancer screening
Examples – Monitoring

- HbA1c (diabetes)
- Immune markers (after transplantation)
- Temperature (during infection)
- Titre (after vaccination)
- Tumour size (cancer)
- Blood oxygen (heart and lung diseases)
- QTc (specific safety study)

Prognostic

- Mammaprint: 70 gene expression to split breast cancer patients into high vs. low risk of metastases

Predictive (which treatment; which dose)

- "Companion diagnostic", "personalized medicine": Measurable before/without treatment
- Body weight
- P450 enzymes (normal or poor metabolizers)
- Herceptin (Her2 positive breast cancer)

Biological understanding

- Useful
- But regulatory pathway does not require this
- Recommended to follow up on this in parallel to other activities

Practical issues

- Timing: A diagnostic needs fast turn-around time
- Other: Inconvenience to patient; risk to patient; circadian variation (incl. meals); sex, age, weight dependence; storage; stability; transportation; central lab.

Validation and qualification

- Validation:
  - The measurement is correct in the technical sense: Including sources of random error
- Qualification (clinical validation):
  - The measurement is related to the clinical state of the patient
Clinical trials to qualify a biomarker for monitoring

- Example: For HbA1c, DCCT (1993) was the turning point
- 1441 patients; stratified by retinopathy; followed for up to 9 years; price-tag 1 bill. $
- Intensified insulin treatment lead to both reduced HbA1c and reduced risk of complications (vs. conventional therapy)
- Relation between post-treatment HbA1c and complications in intensive group

Clinical trials to qualify a biomarker as diagnostic

- Trial in relevant population (meaning similar to the one where it will be applied)
- If multivariate diagnostic, formula derivation should be pre-specified
- Evaluate sensitivity/specificity compared to gold standard. Evaluate AUC for ROC curve

Evaluation as diagnostic (in targeted population!)

<table>
<thead>
<tr>
<th>Subject count</th>
<th>Diseased</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Biomarker+</td>
<td>A</td>
<td>B</td>
</tr>
<tr>
<td>Biomarker-</td>
<td>C</td>
<td>D</td>
</tr>
</tbody>
</table>

Diseased/healthy according to reference standard
Sensitivity: A/(A+C); Specificity: D/(B+D)
PPV: A/(A+B); NPV: D/(C+D)
Odds ratio: A*D / (B*C)
Agreement: (A+D)/(A+B+C+D) not relevant!

AUC - ROC

- Vary the threshold between Biomarker + and – subgroups
- Combined values of sensitivity and specificity gives ROC curve
- Evaluate performance by the AUC (more robust evaluation than selecting a single threshold)

Clinical trials to qualify a biomarker as predictive

- If multivariate biomarker, formula derivation should be pre-specified
- Positive subgroup effect checked according to standard practice (two adequate and well-controlled studies)
- Confirmed in later independent studies
- Negative subgroup effect checked, but potentially not as rigorous (interaction expected) depending on biological understanding

Potential trial designs I (FDA guideline)
Potential trial designs II (FDA guideline)

**Genes or proteins?**
- Genes (DNA): Constant, discrete, massively parallel
- Gene expressions (mRNA): time-varying, continuous, microarray (massively parallel), qPCR (parallel)
- Proteins: time-varying, continuous, difficult sample handling (freezing), separate measurements

- **Gene expression biomarkers**
  - Measure RNA for treatment choice
  - Multivariate so a selection step is necessary
  - Define subgroup to be treated:
  - Show effect in subgroup
  - Show/argue for no effect in complementary subgroup

- **Regulatory issues**
  - FDA: a biomarker is a device (handled by CDRH, center for devices and radiological health)
  - Derivation of a gene-based biomarker (selection based on many genes) require pre-specified gene selection method
  - Endpoint for diagnostic biomarkers is AUC-ROC
  - Only few have passed the regularity hurdle

- **Lundbeck biomarker examples**
  - Gene expressions in depression
  - Imaging (such as PET)
  - EEG
  - Various measurements known to reflect occupancy on specific receptors

- **Summary**
  - Classical well-known biomarkers are far from perfect
  - Many new biomarkers are suggested
  - Diagnostic; Prognostic; Predictive; Surrogate endpoints
  - Only few have passed the regularity hurdle
  - Biological understanding is key
  - Biomarkers are the future