Session 2: Biomarkers of epigenetic changes and their applicability to genetic toxicology

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Workshop: “Genetic Toxicology: Opportunities to Integrate New Approaches”
WHY A SESSION ON EPIGENETICS

• **Mutations**: heritable changes to the sequence of bases of the genome

• **Epimutations**: heritable non-sequence changes to the cellular genome which affect the capacity of its constituent genes to be expressed.

• Genetic toxicology tests are routinely used to predict mutagenic and carcinogenic potential of chemicals
  – Predictive performance good but not great!

• Should we be paying more attention of *epimutagenesis* in safety assessment studies?

EPIGENETICS

Heritable modifications superimposed on DNA base sequence that regulate gene expression

DNA Methylation

Gene Expression

Histone Modification

Non-Coding RNAs
DNA Methylation

• Normal and essential biological process
  • X-chromosome inactivation, development, cellular differentiation, chromatin structure, imprinting, genome stability
• Imprinting maternal vs. paternal expression
• Demethylation in post-fertilization zygote
• Remethylation during development

Hypermethylation = less transcription
Hypomethylation = more transcription
Histone Modifications

- Core histones H2, H2B, H3, H4 can be modified (e.g., methylated, acetylated, phosphorylated, sumolated, ubiquitinated)
- Some examples…

<table>
<thead>
<tr>
<th>Acetylation</th>
<th>H3K4</th>
<th>H3K9</th>
<th>H3K27</th>
<th>H2BK5</th>
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<tbody>
<tr>
<td>Methylation</td>
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Non-coding RNA

Particular interest in microRNA

• Influence gene expression transcriptionally, post-transcriptionally (mRNA degradation and translational inhibition), and by chromatin conformation

• Increase genomic integrity by silencing transposons and stabilizing centromeres
Session 2: Biomarkers of epigenetic changes and their applicability to genetic toxicology

1:45PM   A new paradigm for epigenetic control of cell phenotype: Dynamic reprogramming of tRNA modifications and ribosomes controls selective translation of stress response proteins
   *Dr. Peter Dedon (Massachusetts Institute of Technology)*

2:15PM   Epigenomics and impact for drug safety sciences
   *Dr. Jennifer Marlowe (Novartis)*

2:45PM   Epigenetic traits as biomarkers of carcinogenesis
   *Dr. Igor Pogribny (U.S. Food and Drug Administration, NCTR)*

3:15PM   MIR-34 prevents in vivo lung tumor initiation and progression in the therapeutically resistant *KRAS;TRP53* mouse model
   *Dr. Andrea Kasinski (Yale University)*
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• Questions for the discussion
  – Can biomarkers of epigenetic changes improve/complement the safety assessment process?
  – Is there a need to design screening systems to identify “epimutations” with potential trans-generational inheritance?
  – Can new, less invasive trans-species biomarkers of exposure, susceptibility and effect be identified from accumulated knowledge on epigenetics phenomena?
  – What do we know about the dose-response relationship for epigenetic effects?
Can biomarkers of epigenetic changes improve/complement the safety assessment process?

- Current biomarkers are based on limited # of chemicals
  - Daunting to define baseline
- Are they sensitive enough to detect changes at low doses?
Is there a need to design screening systems to identify “epimutations” with potential trans-generational inheritance?
Can new, less invasive trans-species biomarkers of exposure, susceptibility and effect be identified from accumulated knowledge on epigenetics phenomena?
What do we know about the dose-response relationship for epigenetic effects?
Discussion

• How to differentiate between biomarkers of epigenetic changes – adverse/adaptive
  – Not a lot of mechanistic meaning yet in our understanding
  – Still need to mine current biomarkers data for meaningful correlations – need prospective human study
• Epigenetic markers useful for safety eval prospectively?
  – miRNAs? (precedence in cancer research)
  – Do we know enough to use any right now?
    • Use epigenetic tests as screening (?) tests for large numbers of chemicals (e.g. follow-up positives with 2-year bioassay)
    • Biomarkers for mechanistic insight vs. predictive risk (as long as measurement correlates well with outcome)
• How predictive are in vitro cell lines for in vivo situation?
• miRNAs downstream from p53 as biomarkers
• Human biomarkers preferably assessed from blood
  – Need better understanding of how biomarkers get in urine/blood
• Regulatory eval – based on dose; need better dose-response evaluation with biomarkers, histopath, other markers (AST), etc.
• Epigenetic changes in germ cells (transgenerational effects)
Recent Safety Assessment Reviews


Priestley et al. (2012): Epigenetics – relevance to drug safety science, Toxicology Research (In Press; DOI: 10.1039/c2tx00003b)