Risk Assessment of Metals in Food Utilizing Mode of Action Analysis

Samuel M. Cohen, MD, PhD
Department of Pathology and Microbiology
University of Nebraska Medical Center
Omaha, NE 68198-3135

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## Exogenous Metals

<table>
<thead>
<tr>
<th>Essential</th>
<th>Non-essential</th>
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</thead>
<tbody>
<tr>
<td>Iron</td>
<td>Arsenic</td>
</tr>
<tr>
<td>Manganese</td>
<td>Chromium</td>
</tr>
<tr>
<td>Copper</td>
<td>Lead</td>
</tr>
<tr>
<td>Selenium</td>
<td>Mercury</td>
</tr>
<tr>
<td>Cobalt</td>
<td>Nickel</td>
</tr>
<tr>
<td>Molybdenum</td>
<td>Aluminum</td>
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</tbody>
</table>
Essential and Non-Essential Metals

• Oxidation state influences biology
  • Change in oxidation state may be chemical or enzymatic
• Elemental form not biologically active
• Active cellular transport
• Trace amounts present in most foods
  • Usually at non-detectable levels using routine analytical methods
• Usually present in protein-bound form once in body
Essential Metals

- Minimum daily requirement
  - Deficiencies produce adverse effects
- Excessive amounts are toxic
- Blood and tissue levels tightly regulated to avoid deficiency or excess
- Extensive redundancy in transport mechanisms
Non-essential Metals

- No requirement
- Transporters vary
- Binding to protein varies
- Absorption, blood levels, excretion are not regulated
Iron

• Requirements vary by age and gender
  • Required for cellular metabolism and aerobic respiration
  • Free iron or as heme
  • Recycled in body

• Deficiency – Anemia

• Excess – Hemosiderosis, hemochromatosis
Iron


http://www.slideshare.net/hira_rahman/iron-11905611
Manganese

• Deficiency
  • Dermatitis, abnormalities of serum calcium, phosphorus, and alkaline phosphatase, bone abnormalities

• Toxicity
  • CNS abnormalities
    • Acute: psychiatric symptoms, hallucinations
    • Chronic: Parkinson-like syndrome
    • Less common: effects on other organs

• Exposure
  • Dietary, inhalation, Intravenous

• Numerous transporters of Mn$^{+2}$ into and out of cells
  • Not specific for Mn$^{+2}$, but for divalent metals
  • To maintain specific intracellular levels
Manganese

Manganese Toxicity

• Absorption from GI tract, by inhalation, or IV (total parenteral nutrition)

• Accumulation in basal ganglia, substantia nigra, putamen and related parts of the brain

• Effects on dopamine, GABA and glutamate neurotransmitter levels and function, possibly interfering with cellular efflux

• Neuronal toxicity

• Parkinsonism
Chromium Toxicity

• Cr\textsuperscript{VI} toxic, not Cr\textsuperscript{III}
• Induces duodenal tumors in mice but not rats (NTP)
• Some evidence of genotoxicity \textit{in vitro}, negative \textit{in vivo} if orally administered.
Chromium-induced Duodenal Tumors in Mice

- Absorption of Cr$^{VI}$ from GI tract into villous cells
  - Do not enter crypt cells (intestinal stem cells)
- Toxicity to intestinal villi
  - Macrophages in lamina propria
  - Blunting of villi
- Crypt regeneration
  - Elongation of crypt compartment
  - Increased number of crypt cells, increased number of stem cell DNA replications
Cullen JM et al, Toxicol Pathol, November 4, 2015, epub ahead of print
Inorganic Arsenic Toxicity

- Cancer
  - Bladder
  - Lung
  - Skin

- Non-Cancer
  - Skin arseniasis
  - Ischemic heart disease
  - Diabetes mellitus
  - Others
Classic Pathway for Arsenic Metabolism

Putative Pathway for Arsenic Metabolism

\[ \text{As}^V \rightarrow \text{As}^{III} \rightarrow \text{As(V)} \rightarrow \text{As}^{III} \rightarrow \text{As}^V \]

\[ \text{OH} \rightarrow \text{As}^{V} \rightarrow \text{CH}_3^+ \rightarrow \text{MMA}^{V} \rightarrow \text{DMA}^{V} \rightarrow \text{TMAO}^{V} \]

\[ \text{OH} \rightarrow \text{As}^{III} \rightarrow \text{CH}_3^+ \rightarrow \text{MMA}^{III} \rightarrow \text{DMA}^{III} \rightarrow \text{TMA}^{III} \]

Critical (Reactive) Forms of Arsenic

<table>
<thead>
<tr>
<th>Arsenite</th>
<th>iAs$^{\text{III}}$</th>
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</thead>
<tbody>
<tr>
<td>Monomethylarsonous Acid</td>
<td>MMA$^{\text{III}}$</td>
</tr>
<tr>
<td>Dimethylarsinous Acid</td>
<td>DMA$^{\text{III}}$</td>
</tr>
</tbody>
</table>
Inorganic Arsenic and Cancer

• Epithelial: urinary bladder, skin, lung

• Possible Modes of Action:
  • DNA Reactive: direct binding to DNA (does not occur, Nesnow et al., 2002)
  • Indirect Genotoxicity: micronuclei, DNA repair, oxidative damage, mitotic spindle (tubulin) (genotoxic effects in vitro, only at high concentrations not attainable in vivo)
  • Non-genotoxic: cytotoxicity and regeneration vs. direct mitogenicity
Mode of Action of Inorganic Arsenic Intracytoplasmic Inclusions of Urothelium

These inclusions were mistaken for micronuclei:
Mode of Action for Inorganic Arsenic Carcinogenesis

• Key Events
  • Ingestion of significant amounts of arsenic
  • Generation of trivalent forms ($\text{iAs}^{\text{III}}$, $\text{MMA}^{\text{III}}$, $\text{DMA}^{\text{III}}$)
  • Reaction with critical cellular thiols (glutathione, proteins)
  • Cytotoxicity and cell death
  • Regenerative proliferation
  • Tumors
Urothelial Cytotoxicity and Proliferation Induced By Inorganic Arsenic
Biological Effects of Arsenic

Pentavalent arsenicals → Thioarsenicals

Trivalent arsenicals → Thioarsenicals

Biodonal effects

Trivalent arsenicals + Protein-SH
Implications for Risk Assessment

Non-cancer Biological Effects

- Skin arseniasis
- Bronchial toxicity
- Urothelial toxicity
- Cancer

Other non-cancer toxicities

Trivalent arsenicals

Threshold

Sulfhydryl groups
Conclusions

• Metals are ubiquitous in the environment
• Toxicity of metals (essential and non-essential) involves complex metabolism and transport systems
• Toxicity usually specific for oxidation state
• Toxicity dose response is non-linear and has threshold
• Knowledge of mode of action critical to rational risk assessment
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