Interpretation of Nutritional and Toxicological Diagnostics

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Background

- Highly specialized, complex diversity of requests
- Few laboratories available
- Expensive equipment  LC-MS
- Analytical chemistry + veterinary medicine
- False impressions in the media  CSI

Background

- WCVM – metals, mycotoxins, vitamins, few others
- Toxins/toxicants
  one perspective - toxicity only
- Nutrients
  two perspectives - deficiency or toxicity

Background

- Prairie Diagnostic Services (PDS)
  - Fee for service
  - 2800 cases per year
  - 50 000 analyses per year
  - Interpretation, consultation
  - Vitamins: HPLC
  - Minerals: ICP-MS
  - Mycotoxins: LC-MS
  - Other toxins: spectrophotometry, enzyme activity assays, etc.

Background

- My approach
  - Focus on the animal  Not possible in all situations
  - Chance of success ↑
  - Cost ↓

Interpretation of values

- Books  100s of pages
- Essential minerals and vitamins
  - Deficient, Marginal, Normal, High Normal, Toxic
- Toxic elements
  - Normal, High Normal, Toxic
- Diagnosis: analytical + clinical must be consistent
Interpretation of values

- Individual versus herd diagnosis
- Major variability
- Number of samples
- Tissue
- Representative population: age, sex, lactation

Sample submission

- Sometimes technically challenging
- Specific requirements (analysis dependent)
  - Tubes: plastic or glass
  - Blood: heparinized or whole
  - Tissue: where the agent concentrates
    - where the agent produces its effect
    - often toxin/nutrient specific
  - Fresh or frozen

Factors

1. **Season**
   - feed consumed (quality)
   - water quality
   - supplementation
   - e.g., Spring: low vitamin and mineral status

2. **Age**
   - fetus vs. calf vs. adult
   - 10-fold variation: vitamins, Se, Mn
   - sequestration: Se, Cu in fetus

3. **Species differences**
   - brain cholinesterase
   - Cu

4. **Breed differences**
   - Cu

Factors

5. **Disease state**

<table>
<thead>
<tr>
<th>Disease state</th>
<th>liver Zn, Fe</th>
<th>serum Zn, Fe</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre-existing infectious disease</td>
<td>↑ Zn, Cu</td>
<td>↑ Zn, Cu</td>
</tr>
<tr>
<td>Liver disease</td>
<td>abnormal mineral metabolism</td>
<td></td>
</tr>
<tr>
<td>Hemolysis</td>
<td>↑ serum Mg</td>
<td>↑ serum Fe</td>
</tr>
<tr>
<td>Starvation</td>
<td>↑ brain cholinesterase</td>
<td></td>
</tr>
<tr>
<td>Dehydration</td>
<td>↑ brain cholinesterase</td>
<td></td>
</tr>
</tbody>
</table>

Nutrient interactions

- Nutrient-nutrient: Cu, Mo, Cu, Zn, Vit E, Se
- Nutrient-toxic metal: Se, As, Fe, Pb
- Recommendation: multi-element analysis
- Implications: diagnosis, treatment, prevention
**Tissue submission**

- Varies with agent
- Liver more diagnostically appropriate
- Duration of exposure e.g., Pb blood vs. liver vs. bone
- Other considerations
  - Organ damage kidney, liver
  - Tissue binding As
  - Formalin-fixed tissues organics – not suitable
  - Autolysis metals – OK
  - Vitamins – lack stability
  - Enzymes – poor

**Vitamin D**

- Cholecalciferol (D3)
  - 25-hydroxy-D3 available (PDS)
  - 1,25-hydroxy-D3
  - 24,25-hydroxy-D3
- Analysis: serum, liver, milk

**Vitamin D - Factors**

- Species variation
- Age
- Stage of lactation
- Pregnancy
- Milk fever
- Plant sources
- Supplements
- Colostrum
- Suspected cases: monitor Ca status

**Vitamin E**

- Alpha-tocopherol
- Deficiency common in spring
- Poor feed stability
- PDS liver, plasma, serum

**Vitamin E - Factors**

- Marked age differences
- Colostrum major source
- Toxicity unlikely
- Consider in association with Se
- Many forms acetate, palmitate, dl mixtures (dl is active)
- Light sensitive
- Herd evaluations
- Marked seasonal variation

**VITAMINS**
Vitamin E – Normal values (Cattle)

<table>
<thead>
<tr>
<th>Age (days)</th>
<th>Plasma (mg/L)</th>
<th>Liver (mg/kg, wet weight)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fetus</td>
<td>0.2-0.4</td>
<td>1.2-2.8</td>
</tr>
<tr>
<td>Newborn</td>
<td>0.8-1.2</td>
<td>3.4-5.6</td>
</tr>
<tr>
<td>Infant</td>
<td>0.5-1.0</td>
<td>2.8-4.3</td>
</tr>
<tr>
<td>Juvenile</td>
<td>0.7-1.5</td>
<td>3.1-4.6</td>
</tr>
<tr>
<td>Yearling</td>
<td>1.2-2.2</td>
<td>3.4-5.1</td>
</tr>
<tr>
<td>Adult</td>
<td>3.0-10.0</td>
<td>4.0-8.6</td>
</tr>
</tbody>
</table>

Tissue concentrations exceeding upper limits reflect excessive supplementation, but unlikely toxicity.

Vitamin E – Deficiency (adult cattle)

<table>
<thead>
<tr>
<th>Status</th>
<th>Serum concentration (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Severe</td>
<td>0.1-0.15</td>
</tr>
<tr>
<td>Deficient</td>
<td>0.36-2.00</td>
</tr>
<tr>
<td>White muscle disease</td>
<td>0.60-1.60</td>
</tr>
<tr>
<td>Marginal</td>
<td>2.00-3.00</td>
</tr>
<tr>
<td>Adequate</td>
<td>3.00-22.0</td>
</tr>
</tbody>
</table>

Monitor status in association with Se.

Vitamin A

- Retinol
- Deficiency common in spring
- Poor stability, light sensitive
- PDS liver, plasma, serum

Vitamin A - Factors

- Marked age differences
- Colostrum major source
- Toxicity teratogenicity
- Many forms palmitate, acetate, propionate, carotenoids
- Herd evaluation essential
- Marked seasonal differences
- Interactions with nitrates

Vitamin A – Normal values

<table>
<thead>
<tr>
<th>Age (days)</th>
<th>Plasma (mg/L)</th>
<th>Liver (mg/kg wet weight)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fetus</td>
<td>--</td>
<td>0.17-0.22</td>
</tr>
<tr>
<td>Newborn</td>
<td>1-9</td>
<td>0.20-0.32</td>
</tr>
<tr>
<td>Infant</td>
<td>10-29</td>
<td>0.23-0.45</td>
</tr>
<tr>
<td>Juvenile</td>
<td>30-300</td>
<td>0.25-0.52</td>
</tr>
<tr>
<td>Yearling</td>
<td>301-700</td>
<td>0.30-0.45</td>
</tr>
<tr>
<td>Adult</td>
<td>&gt;700</td>
<td>0.30-0.70</td>
</tr>
<tr>
<td>Adult (def.)</td>
<td>&gt;0.05</td>
<td>&lt;8.5</td>
</tr>
<tr>
<td>Adult (marg.)</td>
<td>0.1-0.3</td>
<td>8.5-28.5</td>
</tr>
<tr>
<td>Adult (toxic)</td>
<td>&gt;2.0</td>
<td>?</td>
</tr>
</tbody>
</table>

MINERALS
Minerals

• Both deficiencies and toxicities are common
• Major regional differences
• Metal-metal interactions clinically relevant
• PDS: multi-element analysis
  4 panels
  24 metals
  Most tissues liver most diagnostically useful

Minerals

• Interpretation deficient marginal normal high normal toxic
• Factors age season disease interactions
• Concerns food safety, withdrawal times

Trace minerals in cattle (adult)

<table>
<thead>
<tr>
<th>Metal</th>
<th>Liver (mg/kg, wet weight)</th>
<th>Serum (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Deficient</td>
<td>Normal</td>
</tr>
<tr>
<td>As</td>
<td>&lt;0.4</td>
<td>7.0</td>
</tr>
<tr>
<td>Ca</td>
<td>&lt;40</td>
<td>40-200</td>
</tr>
<tr>
<td>Cu</td>
<td>&lt;10</td>
<td>25-100</td>
</tr>
<tr>
<td>Fe</td>
<td>&lt;26</td>
<td>45-300</td>
</tr>
<tr>
<td>Sr</td>
<td>&lt;0.5</td>
<td>&gt;5.0</td>
</tr>
<tr>
<td>Mg</td>
<td>&lt;100</td>
<td>100-250</td>
</tr>
<tr>
<td>Mn</td>
<td>&lt;1.0</td>
<td>2.5-10</td>
</tr>
<tr>
<td>Mo</td>
<td>&lt;0.2</td>
<td>0.15-1.5</td>
</tr>
<tr>
<td>Se</td>
<td>&lt;0.12</td>
<td>0.25-0.5</td>
</tr>
<tr>
<td>Zn</td>
<td>&lt;20</td>
<td>25-100</td>
</tr>
<tr>
<td>Co</td>
<td>&lt;0.05</td>
<td>0.02-0.085</td>
</tr>
</tbody>
</table>

Trace mineral interpretation comments

• All values should be used as a guideline
  Clinical disease is important
• Duration of exposure will influence interpretation
• Metal-metal interactions (Cu, Mo) are important
• Tissue concentrations often a poor indicator of status
  Mn, Co, Zn, Ca
• Infectious disease will alter Fe and Zn tissue values within hours
• Age alters Mn and Se status values

Ergot alkaloids

• Ergot moisture crop rotation
  no till farming ditch/weed control
• Invades all grass species
• PDS – 6 ergot alkaloid panel (feed)
  ergocornine ergometrine ergocristine ergosine ergocryptine ergotamine
Ergot

- Clinical manifestations
  - High dose: CNS excitation, gangrene
  - Low dose: agalactia (prolactin), poor performance, thermoregulation, abortion
- Major species differences
- Breed: beef vs. dairy

- Interpretation
  - Feed analysis: >200 ppb TMR potentially problematic
  - Tissue analysis: currently not available
- Should ergot-contaminated feed be consumed by lactating or pregnant cattle?
- Other concerns
  - Tissue residues: rapid elimination
  - Withdrawal times: short
  - Animal welfare: gangrene, limited recovery

Fusarium spp. mycotoxins

- Extensive crop contamination in Western Canada
- PDS mycotoxin panel (14 mycotoxins)
- Routine feed screening is commonplace
- Testing available on feed only

Fusarium mycotoxin guidance values for cattle feed

<table>
<thead>
<tr>
<th>Mycotoxin</th>
<th>Cattle guideline (ppm)</th>
<th>Relative potency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Deoxynivalenol</td>
<td>5.0</td>
<td>1</td>
</tr>
<tr>
<td>3-Ac-deoxynivalenol</td>
<td>5.0</td>
<td>1</td>
</tr>
<tr>
<td>15-Ac-deoxynivalenol</td>
<td>5.0</td>
<td>1</td>
</tr>
<tr>
<td>T-2 Toxin</td>
<td>1.0</td>
<td>5</td>
</tr>
<tr>
<td>HT-2 Toxin</td>
<td>0.1</td>
<td>50</td>
</tr>
<tr>
<td>Nivalenol</td>
<td>1.0</td>
<td>5</td>
</tr>
<tr>
<td>Diacetoxyscirpenol</td>
<td>1.0</td>
<td>5</td>
</tr>
<tr>
<td>Aflatoxin B1</td>
<td>0.02</td>
<td>—</td>
</tr>
<tr>
<td>Ochratoxin</td>
<td>1.0</td>
<td>—</td>
</tr>
<tr>
<td>Zearalenone</td>
<td>1.0</td>
<td>—</td>
</tr>
<tr>
<td>a-Zearalenol</td>
<td>1.0</td>
<td>—</td>
</tr>
<tr>
<td>b-Zearalenol</td>
<td>1.0</td>
<td>—</td>
</tr>
<tr>
<td>Fumonisin B1</td>
<td>30.0</td>
<td>—</td>
</tr>
<tr>
<td>Fumonisin B2</td>
<td>30.0</td>
<td>—</td>
</tr>
</tbody>
</table>

Fusarium mycotoxin comments

- The guidelines may vary considerably from region to region
- The effects of many mycotoxins are additive
  - Relative potencies must be considered with weighting factors
- The guidelines for dairy cattle are typically lower as compared to beef cattle

Mycotoxin calculator

- Saskatchewan Ministry of Agriculture
  - http://www.agriculture.gov.sk.ca/Mycotoxin-Calculator
  - Includes Western Canadian relevant information for several livestock species
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