Functional Vitamin, Mineral and Antioxidant Assessment

J. F. Crawford, PhD.
SpectraCell Laboratories, Inc.
Houston, TX
Recent Publications

“Antioxidant supplementation increases morbidity and mortality.”

JAMA, 2007
“Micronutrient testing offers a unique, scientifically based evaluation of functional deficiencies that allows targeted treatment with nutritional supplements... improving patient compliance with tailored therapy and success in the treatment of a variety of diseases.”

Mark Houston, M.D.
Hypertension Institute
St Thomas Medical Center & Vanderbilt University
IMMUNOCOMPETENCE

CELL-MEDIATED IMMUNITY (Th1)
T-LYMPHOCYTES

HUMORAL IMMUNITY (Th2)
B-LYMPHOCYTES
T-LYMPHOCYTES

Th1 - (Cell Mediated Immunity)

- Attack Intracellular Pathogens
  - DTH skin responses to viral + bacteria Ag
  - CANCER cells

- Organ Specific Autoimmune Disease
  (Arthritis, MS, Type I Diabetes)
B-LYMPHOCYTES

Th2 (Humoral Immunity)

- Protection Against Extra Cellular Pathogens
- Antibody Production
- Allergy + Related IgE - based disease
  (Systemic Autoimmune Disease)
Improved Cellular Performance

Your cellular performance may also be improved after information obtained from micronutrient testing.
T-Lymphocytes are used for MNT
## Technology Summary

<table>
<thead>
<tr>
<th>Day 1</th>
<th>Isolation of Lymphocytes</th>
<th>Long term nutritional marker</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><strong>Incubation &amp; Growth in</strong></td>
<td><strong>Patented technology 15 yrs development at U.T. Austin</strong></td>
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<td><strong>defined culture media</strong></td>
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<tr>
<td>Days 2-3</td>
<td><strong>Mitogen stimulation &amp; growth</strong></td>
<td><strong>Vary components growth dependent on intracellular levels</strong></td>
</tr>
<tr>
<td>Day 4</td>
<td><strong>$^3$H Thymidine incorporation</strong></td>
<td><strong>200 growth measurements</strong></td>
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<tr>
<td>Day 5</td>
<td><strong>Growth response measurement</strong></td>
<td><strong>Deficiency, transport &amp; metabolic requirements</strong></td>
</tr>
</tbody>
</table>
Intracellular Micronutrient Testing

Nutrients Analyzed

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>Fructose utilization</th>
<th>Spectrox antioxidant function</th>
<th>Glutathione</th>
<th>Cysteine</th>
<th>Selenium</th>
<th>Vitamin C</th>
<th>Vitamin E</th>
<th>Vitamin D</th>
<th>Vitamin K</th>
<th>Carnitine</th>
<th>Copper</th>
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<tbody>
<tr>
<td>Thiamin (B1)</td>
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<td>Riboflavin (B2)</td>
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<td>Pantothenate (B5)</td>
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<td>Pyridoxine (B6)</td>
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<td>Cobalamins (B12)</td>
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<td>Insulin Sensitivity</td>
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</table>
Nutrient Deficiencies and Previous Supplementation

- **Multiple deficiencies with no previous supplementation**: 38%
- **Multiple deficiencies with previous supplementation**: 43%
- **Subjects showing no deficiency**: 19%
Intracellular Micronutrient Deficiencies

- B1
- B12
- B2
- CALCIUM
- FOLATE
- MAG
- GLUTATHIONE
- LIPOIC
- MAG
- SELENIUM
- D

Percentage Deficiencies:
- 0.0%
- 5.0%
- 10.0%
- 15.0%
- 20.0%
- 25.0%
- 30.0%
- 35.0%
# Factors Affecting Nutrient Status

<table>
<thead>
<tr>
<th>Category</th>
<th>Example Factors</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dietary Intake</td>
<td>Metabolism</td>
</tr>
<tr>
<td>Absorption</td>
<td>Excretion</td>
</tr>
<tr>
<td>Transport</td>
<td>Hormonal Status</td>
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<tr>
<td>Storage</td>
<td>Genetic Influences</td>
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<tr>
<td>Receptors</td>
<td>Disease</td>
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<td>Activation</td>
<td>Lifestyle Factors</td>
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<td>Inhibition</td>
<td>Pharmaceuticals</td>
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<td>Age</td>
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<td>Gender</td>
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<td>Socioeconomic</td>
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<td>Cultural/Ethnic</td>
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<td>Pregnancy</td>
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<td>Exercise</td>
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<td>Smoking</td>
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<td>Alcohol</td>
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</tbody>
</table>
“High-dose Vitamin Therapy Stimulates Variant Enzymes with Decreased Coenzyme Binding Affinity: Relevance to Genetic Diseases and Polymorphisms”

Bruce N. Ames, M.D., et al.,
The American Journal of Clinical Nutrition,
Vol 75, No 4, April 2002.
Medications & Nutrient Deficiency

**Anti-Depressants**
- Elavil, Tofranil, Sinequan, Aventyl
- B12, CoQ10

**Anti-Inflammatory**
- Aspirin, Advil, Motrin
- Vit C, Folate

- Prednisone, Cortisone
- D, Folic, Cal, Mag, Selenium, Zinc

**Statins**
- CoQ10, B12, D, E, Folic, A

**Hormone Replacement Therapy**
- Evista, Premarin, Estratab
- B2, B6, B12, C, Folate, Mag, Zinc
The graph illustrates the Proliferation Index over various years. It shows three categories: "Average," "< 25%," and "> 75%." The index decreases over time, indicating a decline in proliferation. The "Average" category starts at a higher index and continues to decline steadily. The "< 25%" category shows a slower decline compared to the "Average." The "> 75%" category displays the most significant decrease, starting from a lower index.
# Homocysteine & Vascular Disease

## Genetic and Dietary Determinants of Serum Homocysteine Concentrations

<table>
<thead>
<tr>
<th>Genetic</th>
<th>Nutritional *</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Cystathionine-beta-synthase deficiency</td>
<td>• Vitamin $B_6$</td>
</tr>
<tr>
<td>• Methionine synthase deficiency</td>
<td>• Vitamin $B_{12}$</td>
</tr>
<tr>
<td>• MTHFR deficiency</td>
<td>• Folate</td>
</tr>
<tr>
<td>• Defective absorption of $B_{12}$ or Folate</td>
<td></td>
</tr>
<tr>
<td>• Prevalence:</td>
<td></td>
</tr>
<tr>
<td>30% Female</td>
<td></td>
</tr>
<tr>
<td>25% Male</td>
<td></td>
</tr>
</tbody>
</table>

* 75% of cases of Hyperhomocysteinemia are nutritional in origin
Homocysteine & Vascular Disease

Effects of Treatment With N-Acetyl Cysteine (NAC)

O. Wiklund et al. / Atherosclerosis 119(1996)
Model of Inflammation and Inflammatory Disease

Classical Inflammatory Mediators

- Cellular attack (free $O_2$)
- Macrophage releases lymphokines producing IL-1
- T-lymphocytes produce IL-2
- IL-1 and IL-2 proliferation of T-lymphocytes produce interferon
- Cell-killing activity of T-cells and NKC enhanced and free radical production
The Magic Bullet
Major Cellular Antioxidants

- **Antioxidant Nutrients**
  - Vitamin C (Ascorbate)
  - Vitamin E (Tocopherols)
  - Selenium
  - Glutathione

- **Antioxidant Enzymes**
  - Superoxide Dismutase (Zn Cu, Mn)
  - Catalase (Fe)
  - Glutathione Peroxidase (Se)
Markers of Oxidative Stress

Thiobarbituric Acid Reactive Substances (Tbars)
  Lipid Peroxides
  Isoprostanes
  Guanosine Derivitives
  Selenium
  Protein Carbonyls
  Orac & Trap
  Lymphocyte Culture (Spectroxy)
Antioxidant Balance

Arachadonic acid peroxyl radical

12-HETE (alcohol)

12-HPETE (hydroperoxide endoperoxide)

tocopherol radical ox

tocopherol

Dehydroascorbate

Ascorbate

Glutathione (GSH)

Glutathione disulfide (G-S-S-G)

NADP

NADPH

Prostaglandins (E series) thromboxanes, leukotrienes
Spectrox - Total Antioxidant Function

Status

Result: 51.2 Percentile
Reference Range:
Desired > 65th percentile
Average 40th to 65th percentile
Deficient < 40th percentile

Desired          Average/ Deficient

65th
50th
40th

Desired Results
Average
Deficient

Total Antioxidant Function
Antioxidant Testing

100% Complete + Cells + Free Radicals = Spectrox 1

100% Complete + Cells + Selenium = Saturated Cells
Saturated Cells + Free Radicals = Spectrox 2

Measurement: % improvement in Spectrox 1
Figure 1

Pre-Treatment Spectrox Value Range:
1<25, 2= 25-40, 3=40-55, 4=55-70, 5>75
(p<0.05)
TSH

↓ Iodine ___

↓ Ferritin ___

↓ Vit D₃ ___

T₄ to T₃ decreased by rT₃, deficiencies of selenium, zinc, chromium, etc.

Zinc

T₄ to T₃ increased by abnormal cortisol, TPO antibodies, T₄ medication (Synthroid), estrogen

(Good) T₃

rT₃ (Bad) (Excess)
T₃ receptor density is influenced by cortisol, inadequate if too high or too low.
**Nutrition and Estrogen Metabolism**

- **Cholesterol**
- **Testosterone**
- **Estrogen**
- **3,4 Quinones**
- **GSH Mercapturates**

- 2-Methoxyestrogen  
  "Good" estrogen

- Insulin Mediated (Mg ++, Chromium, B-complex...)

- Oxidation
  Can be prevented with anti-oxidants

- Potential Carcinogens
  Selenium, Glutathione Mediated Conversion
Essential Hypertension

Diagnosis + treatment of intracellular nutrient deficiencies, oxidative stress, + insulin resistance will:

- Reduce Blood Pressure
- Improve Vascular Health
- Improve Endothelial Function

Mark Houston, MD, In Press, Therapeutic Advances in Cardiovascular Disease (2010)
### Percentage Deficiency in Hypertensive & Control Populations

<table>
<thead>
<tr>
<th></th>
<th>Hypertension</th>
<th>Control</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serine</td>
<td>10.9%</td>
<td>6.3%</td>
<td>0.0001</td>
</tr>
<tr>
<td>Insulin</td>
<td>18.9%</td>
<td>14.1%</td>
<td>0.0136</td>
</tr>
<tr>
<td>Calcium</td>
<td>21.6%</td>
<td>14.8%</td>
<td>0.0007</td>
</tr>
<tr>
<td>Vitamin D</td>
<td>32.2%</td>
<td>21.6%</td>
<td>0.0294</td>
</tr>
<tr>
<td>CoQ10</td>
<td>16.9%</td>
<td>9.1%</td>
<td>0.0294</td>
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</tbody>
</table>

Mark Houston, MD, In Press, Therapeutic Advances in Cardiovascular Disease (2010)
“Replacement of the micronutrient deficiencies, as well as high dose therapy of selected nutraceuticals in combination with optimal diet, exercise and weight management resulted in control of blood pressure to goal levels in 62% of the hypertensive population over a period of 6 months with complete tapering and discontinuation of anti-hypertensive drugs”.

Mark Houston, MD, In Press, Therapeutic Advances in Cardiovascular Disease (2010)
Coenzyme Q10 Deficiency in Patients on Statin Therapy

<table>
<thead>
<tr>
<th>Without Q10 Supplementation</th>
<th>Serum</th>
<th>4/15</th>
<th>26.6%</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>FIA</td>
<td>47/92</td>
<td>51.1%</td>
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</table>

<table>
<thead>
<tr>
<th>With Q10 Supplementation</th>
<th>Serum</th>
<th>2/15</th>
<th>13.3%</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>FIA</td>
<td>28/90</td>
<td>31.1%</td>
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</tbody>
</table>
## Disease Incidence Prevention by Serum 25(OH)D Level

| Serum 25(OH)D, ng/ml | 6  | 8  | 10 | 12 | 14 | 16 | 18 | 20 | 22 | 24 | 26 | 28 | 30 | 32 | 34 | 36 | 38 | 40 | 42 | 44 | 46 | 48 | 50 | 52 | 54 | 56 | 58 | 60 | 62 | 64 | 66 | 68 |
|---------------------|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| Studies of Individuals |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Cancers, all combined |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Breast Cancer | 30% | 12% | 8% | 2% |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Ovarian Cancer | 31% | 26% | 18% | 6% | 0% |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Colon Cancer | 12% | 23% | 33% | 33% | 66% | 66% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% |
| Non-Hodgkin's Lymphoma | 12% | 23% | 33% | 33% | 66% | 66% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% |
| Type 1 Diabetes | 12% | 23% | 33% | 33% | 66% | 66% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% |
| Fractures, all combined | 72% | 23% | 33% | 33% | 66% | 66% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% | 54% |
| Falls, women | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% | 30% |
| Multiple Sclerosis |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Heart Attack (Men) |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Natural Experiments |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Kidney Cancer |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Endometrial Cancer |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Rickets | 59% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% | 37% |

**Legend:**

All percentages refer to a common baseline of 25 ng/ml as shown on the chart.

% decreases reflect the disease prevention % at the beginning and ending of available data. Example: Breast cancer incidence is reduced by 30% when the serum level is 34 ng/ml vs the baseline of 25 ng/ml. There is an 83% reduction in incidence when the serum level is 50 ng/ml vs the baseline of 25 ng/ml.

The x's in the bars indicate 'reasonable extrapolations' from the data but are beyond existing data.

**References:**

- Rickets: Arnaud SB, Copyright GrassrootsHealth, 10/19/08 www.grassrootshealth.org
LABORATORY REPORT

Account Number: 123456
Dr. John Smith
123 Main St
Anystown, USA

Name: Jane Doe
Gender: Female
DOB: 11/13/1947
Accession Number: J71101
Requisition Number: 781864
Date of Collection: 05/19/2010
Date Received: 05/20/2010
Date Reported: 06/01/2010

Summary of Deficient Test Results

Testing determined the following functional deficiencies:

Vitamin D3 Calcium Zinc Glutathione
Spectroxe                

Borderline deficiencies include:

Vitamin B2 Panthothenic Acid

The Immune Response Index is an indication of the patient's T-lymphoproliferative response to mitogen stimulation relative to the response of a control population. A result greater than 65% indicates a healthy response, a measurement of cell-mediated immune function. A result between 40% and 64% indicates an average response that may improve with correction of the nutritional deficiencies determined by the micronutrient testing. Test results below 40% indicate a poor immune response and for better immune function requires improvement.

John F. Crawford, Ph.D.
Laboratory Director

All tests performed by SpectraCell Laboratories, Inc. * 10401 Town Park Drive Houston, TX 77072
Tel (713) 621-3101 * Toll-free (800)-227-LABS(5227) * Fax (713) 621-3234 * www.spectrancell.com
Immune Response Score (Th₁)
Borderline Deficiencies
Repletion Suggestions

1. Vitamin D3 (Cholecalciferol)  
   1000 IU b.i.d. (2000 IU daily) of Cholecalciferol  
   (Vitamin D3-1-alpha 25-dihydroxyvitamin D)

2. Calcium  
   500 mg b.i.d. (1000 mg daily) as citrate, malate, ascorbate or glycinate

3. Zinc  
   25 mg daily

4. Total Antioxidant Function  
   Based on Spectroxx and individual Antioxidant tests:
   * Glutathione Deficient: 600 mg t.i.d. (1800 mg daily) of N-Acetylcysteine (NAC) Take each dose with a meal
   * Cysteine: The daily dose of N-Acetylcysteine (NAC) listed for Glutathione is usually sufficient for Glutathione and/or Cysteine repletion.
   * Vitamin E: 200 IU daily of mixed tocopherols
   * Selenium: 50 mcg daily
   * Coenzyme Q10: 30 mg daily of CoQ10 Take each dose with a meal
   * Lipoic Acid: 50 mg daily
   * Vitamin C: 250 mg daily

Please note: Supplementation is usually required for four to six months to effect the repletion of a functional deficiency in lymphocytes

Suggestions for supplementation with specific micronutrients must be evaluated and approved by the attending physician. This decision should be based upon the clinical condition of the patient and the evaluation of the effects of supplementation on current treatment and medication of the patient.
The Modifiable Risk Factors for Optimal Aging

- Strengthen Immune Function
- Optimize Methylation Metabolism
- Limit Inflammatory Processes
- Improve Mitochondrial Function
- Reduce Chronic Stress
- Regulation of Glycemic / Insulin Function
THE BALANCE