Food-based Approaches for Ensuring Adequate Vitamin A Nutrition

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Carotenoids

- The colors of nature - leaves, fruits and vegetables.
- Over 600 have been identified in nature.
- About 50 give us vitamin A.
- The major carotenoids found in people: β-carotene, α-carotene, lycopene, β-cryptoxanthin and lutein.
Structures of provitamin A carotenoids

α-carotene

β-carotene

β-cryptoxanthin

retinol
Why study carotenoid bioavailability?

- Bioavailability of retinol is not an issue (80-95% absorbed).

- Foods high in provitamin A are not readily consumed by everybody, I.e. liver, eggs and fortified milk.

- There are more benefits to fruits and vegetables beside vitamin A.
A liver with too much vitamin A
LACK OF IMPROVEMENT IN VITAMIN A STATUS WITH INCREASED CONSUMPTION OF DARK-GREEN LEAFY VEGETABLES

de Pee S et al., Lancet 1995;346:75-81
Intestinal wall

Food $\beta$-C$_{total}$

$\beta$-C$_{freed}$

Bioaccessibility = $\beta$-C$_{freed}$ / $\beta$-C$_{total}$

Bioavailability = $\beta$-C$_{absorbed}$ / $\beta$-C$_{total}$

Bioconversion = Retinol / $\beta$-C$_{absorbed}$

Bioefficacy = Retinol / $\beta$-C$_{total}$

$\beta$-C$_{absorbed}$

Cleaved

Retinal

Reduced

Retinol

These studies measured BIOEFFICACY of foods.
Definitions

Bioaccessibility: how much carotenoid is released from the food matrix and available for absorption.
- 100% ~ all ingested β-carotene is released.

Bioavailability: fraction of ingested nutrient available for utilization or storage.
- 100% ~ all ingested β-carotene is absorbed.

Bioconversion: proportion of bioavailable carotene converted to retinol.
- 100% ~ all bioavailable β-carotene is converted.

Bioefficacy: efficiency ingested carotenoids are absorbed and converted to retinol.
- 100% ~ 1 μmol β-carotene = 2 μmol retinol.
The Question

Cacos = Animals = Humans?
The Answer!

Cacos ≠ Animals

Humans?
Animals
Anatomy of a gerbil study

- ~40 day old gerbils are placed on vitamin A depleted diets for a few weeks
- A small group of gerbils are killed at baseline
- Larger groups of gerbils are allocated to treatments, supplements, or control
- Period of time to build up and differentiate liver stores of vitamin A
Results: Conversion factors for carrots ranged from 9 to 11 µg β-carotene to 1 µg retinol for typical orange and 23 µg β-carotene to 1 µg retinol for biofortified carrots.
Bioefficacy of biofortified β-carotene maize on vitamin A status

- To determine the bioefficacy of maize with enhanced β-carotene
- To determine the effect of β-carotene and maize levels on vitamin A status
Results: Liver VA concentrations were higher ($P < 0.05$) in the VA group, lower in the control, but did not differ from the β-carotene group when compared with the high-β-carotene maize group. Bioconversion was ~3 μg β-carotene to 1 μg retinol.
**β-Carotene Assessment**

Liver βC is ~100% greater in maize treatment group.
Results: VA status with increasing β-carotene. Liver VA from orange maize was greater than yellow maize, regardless of percentage ($P < 0.05$).
Results:
Bioconversion for \( \beta \)-cryptoxanthin was 2.8 µg to 1 µg retinol and almost identical to \( \beta \)-carotene.
High β-cryptoxanthin maize

![Bar graph showing the comparison of β-cryptoxanthin levels in different conditions.](image)
You can get vitamin A from green leafy vegetables!
Conclusions:

- Provitamin A carotenoids from supplements and food provide adequate vitamin A to this animal model.

- Not all provitamin A carotenoids are equivalent, one conversion factor does not fit all foods but is related to vitamin A status.

- Biofortified maize and carrots not only maintained VA status in this model, but was as efficacious as \( \beta \)-carotene supplements.

- In populations consuming maize, using orange instead of white maize could dramatically impact vitamin A status.
Total liver reserves vs. conversions for foods analyzed to date

- Red carrots
- Maize
- Kale, spinach, and brussel sprouts

Conversion factor (μg/μg) vs. Total vitamin A liver reserves (μmol)

- Dark orange carrots
- Orange and purple carrots
- Kale, spinach, and brussels sprouts
- Red carrots and Cassava
- Maize

R = 0.88
Summaries of studies with vegetables in humans
Green and yellow vegetables can maintain body stores of vitamin A in Chinese children

Vitamin A assessment with 3 mg d₈ and d₄–retinol

The calculated equivalence was 26.7 μg β-carotene:1 μg retinol
Bioconversion of plant carotenoids to vitamin A varies inversely with vitamin A status (Filipino school children)

Vitamin A assessment with 5 mg d<sub>4</sub> and d<sub>8</sub>–retinol. Three-day sampling

Significant intervention effect – deworming?
Vitamin A status of Filipino schoolchildren at baseline and after 12 wk of feeding orange fruit and green vegetables (2258 µg RE/d) and 5.3 MJ/d from 33 g fat, 37 g protein, and 209 g carbohydrates

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>Post-intervention</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum (D:H)-retinol at 3 d after an oral dose of deuterated retinyl acetate</td>
<td>0.66 (0.12 - 2.90)</td>
<td>0.30 (0.09 - 0.58) *</td>
</tr>
<tr>
<td>Serum retinol (µmol/L)</td>
<td>0.68 (0.33 - 0.93)</td>
<td>1.06 (0.59 - 1.79) *</td>
</tr>
</tbody>
</table>
van Lieshout et al., JAFC, 2003

- Used fecal samples to estimate the bioavailability of β-carotene from pumpkin and spinach
- Pumpkin 1.8 times better than spinach
- Multiple doses of $^{13}\text{C}_{10}$-retinol and β-carotene; LCMS with positive ion atmospheric pressure chemical ionization (ACPI) detection
**Study Design**

1st isotope dose

-3 to 1 21 22

Plasma isotopic ratio, retinol carotenoids

control (corn oil)
sweet potatoes
Indian spinach
retinyl palmitate
β-carotene

2nd isotope dose

82 83 92 93 113

Day

- Basal, low vitamin A diet
- Supplementation period
Amount of vitamin A provided in supplements with noon and evening meals

<table>
<thead>
<tr>
<th>Treatment Group</th>
<th>Retinyl palmitate (µg)</th>
<th>β-carotene (mg)</th>
<th>βC:VA(^1) factor</th>
<th>µg RE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0</td>
<td>0</td>
<td>-</td>
<td>0</td>
</tr>
<tr>
<td>Swt Potato</td>
<td>0</td>
<td>2.25</td>
<td>6:1</td>
<td>375</td>
</tr>
<tr>
<td>Spinach</td>
<td>0</td>
<td>2.25</td>
<td>6:1</td>
<td>375</td>
</tr>
<tr>
<td>Vitamin A</td>
<td>687</td>
<td>0</td>
<td>-</td>
<td>375</td>
</tr>
<tr>
<td>β-carotene</td>
<td>0</td>
<td>2.25</td>
<td>6:1</td>
<td>375</td>
</tr>
</tbody>
</table>

\(^1\) β-carotene:retinol bioconversion factor
Estimated mean initial and final vitamin A pool size by treatment group

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Initial Pool Size (mmol)</th>
<th>Final Pool Size (mmol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>a</td>
<td>b</td>
</tr>
<tr>
<td>Sweet Potato</td>
<td>a,b</td>
<td>b</td>
</tr>
<tr>
<td>Indian Spinach</td>
<td>b</td>
<td>b</td>
</tr>
<tr>
<td>Vitamin A</td>
<td>b</td>
<td>b</td>
</tr>
<tr>
<td>β-carotene</td>
<td>b</td>
<td>b</td>
</tr>
</tbody>
</table>

Final means with different superscript letters are significantly different, \( p<0.034 \), analysis of covariance controlling for initial values.
Mean final vitamin A pool sizes were significantly higher in the spinach, vitamin A and β-carotene groups compared to the control group.

Vitamin A equivalency factors (β-carotene: vitamin A) were estimated as:

- ~13:1 for sweet potato
- ~10:1 for Indian spinach
- ~6:1 for β-carotene
Schoolchildren fed 4.2 mg provitamin A carotenoids for 9 wk (carrots, bok choy, squash and kangkong) with 2.4, 5 or 10 g fat/meal.

Low liver reserves (< 0.07 μmol/g) fell from 35% to 7% and the amount of fat did not influence the results.
Extrapolation from these studies

What does this mean for us?
Mean body weight

Women 2.4% BW

Estimated liver weight

Amount of Vitamin A in

Time

Loss

Expected improvement in liver reserves

Initial increase in liver reserves

Time

Estimated liver weight

Children 4% BW

Amount of Vitamin A in
Parameters for a reference male child using the 50\textsuperscript{th} percentile WHO weight-for-age and liver weight as 4\% of body weight and an adolescent girl using 2.4\% of body weight for liver weight for

<table>
<thead>
<tr>
<th>Age</th>
<th>Body weight</th>
<th>Liver weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>Boy</td>
<td>mo</td>
<td>kg</td>
</tr>
<tr>
<td>6</td>
<td>7.9</td>
<td>316</td>
</tr>
<tr>
<td>8</td>
<td>8.6</td>
<td>344</td>
</tr>
<tr>
<td>10</td>
<td>9.15</td>
<td>366</td>
</tr>
<tr>
<td>12</td>
<td>9.65</td>
<td>386</td>
</tr>
<tr>
<td>Boy</td>
<td>y</td>
<td>kg</td>
</tr>
<tr>
<td>2</td>
<td>12.15</td>
<td>486</td>
</tr>
<tr>
<td>3</td>
<td>14.35</td>
<td>574</td>
</tr>
<tr>
<td>4</td>
<td>16.35</td>
<td>654</td>
</tr>
<tr>
<td>Girl</td>
<td>y</td>
<td>kg</td>
</tr>
<tr>
<td>13</td>
<td>35</td>
<td>840</td>
</tr>
<tr>
<td>15</td>
<td>39</td>
<td>936</td>
</tr>
<tr>
<td>17</td>
<td>43</td>
<td>1032</td>
</tr>
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</table>
The amount of vitamin A estimated to be stored from provitamin A carotenoids in a vitamin A-depleted preschool boy or adolescent girl who had consumed biofortified maize for a day

<table>
<thead>
<tr>
<th>Age</th>
<th>Maize meal</th>
<th>Provitamin A (βCE)</th>
<th>Total βCE</th>
<th>Retained βCE</th>
<th>Conversion to VA</th>
<th>EAR</th>
<th>Extra VA stored</th>
</tr>
</thead>
<tbody>
<tr>
<td>Boy</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>100</td>
<td>10</td>
<td>1000</td>
<td>750</td>
<td>250</td>
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<td>40</td>
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<tr>
<td>2</td>
<td>150</td>
<td>10</td>
<td>1500</td>
<td>1125</td>
<td>375</td>
<td>210</td>
<td>165</td>
</tr>
<tr>
<td>3</td>
<td>200</td>
<td>10</td>
<td>2000</td>
<td>1500</td>
<td>500</td>
<td>275</td>
<td>225</td>
</tr>
<tr>
<td>Girl</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>200</td>
<td>10</td>
<td>2000</td>
<td>1500</td>
<td>500</td>
<td>420</td>
<td>80</td>
</tr>
<tr>
<td>15</td>
<td>200</td>
<td>10</td>
<td>2000</td>
<td>1500</td>
<td>500</td>
<td>485</td>
<td>15</td>
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<tr>
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<td>200</td>
<td>10</td>
<td>2000</td>
<td>1500</td>
<td>500</td>
<td>485</td>
<td>15</td>
</tr>
<tr>
<td>Girl</td>
<td></td>
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<td></td>
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<td></td>
</tr>
<tr>
<td>13</td>
<td>300</td>
<td>15</td>
<td>4500</td>
<td>3375</td>
<td>1125</td>
<td>420</td>
<td>700</td>
</tr>
<tr>
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<td>300</td>
<td>15</td>
<td>4500</td>
<td>3375</td>
<td>1125</td>
<td>485</td>
<td>640</td>
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<tr>
<td>17</td>
<td>300</td>
<td>15</td>
<td>4500</td>
<td>3375</td>
<td>1125</td>
<td>485</td>
<td>640</td>
</tr>
</tbody>
</table>
Infants eating sweet potato

Liver vitamin A accumulation (nmol/g) vs. Age of child (mo)

- Sweet potato: Linear increase with age
- Sachets or tablets: Linear decrease with age
Vitamin A status of Nicaraguan schoolchildren at baseline and 1 year after the start of the Nicaraguan national program of fortifying domestically-grown sugar with vitamin A.
A male child eating maize

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**Liver vitamin A accumulation**

<table>
<thead>
<tr>
<th>Age of child (y)</th>
<th>Liver vitamin A accumulation (µmol/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.5</td>
<td>0</td>
</tr>
<tr>
<td>1</td>
<td>0.1</td>
</tr>
<tr>
<td>1.5</td>
<td>0.2</td>
</tr>
<tr>
<td>2</td>
<td>0.3</td>
</tr>
<tr>
<td>2.5</td>
<td>0.4</td>
</tr>
<tr>
<td>3</td>
<td>0.5</td>
</tr>
<tr>
<td>3.5</td>
<td>0.6</td>
</tr>
<tr>
<td>4</td>
<td>0.7</td>
</tr>
<tr>
<td>4.5</td>
<td>0.8</td>
</tr>
</tbody>
</table>

- **Sugar**
- **Maize**
- **Supplements**

*Bioconversion slows*
Adolescent girl eating maize

Liver vitamin A accumulation (μmol/g)

Fortified sugar (high intake)
Biofortified maize (high intake)
Biofortified maize (low intake)
Fortified sugar (low intake)

Age of girl (y)
After two years of captivity!!!
Potential maize target level

Liver vitamin A accumulation (μmol/g) vs. Age of girl (y)

- 300 g maize
- 250 g maize
- 200 g maize
- Fortified sugar
Sherry’s top two factors for the general public

- The vegetable matrix is important. With processing and cooking, however, bioavailability can be improved.

- Absorption modifiers: one needs fat but not alcohol for improving bioavailability and overall bioefficacy.
Sherry’s top factor for the general public

- Vitamin A status of the host
How important are carotenoids and vitamin A?

- Autumn would be very boring in Wisconsin.
- We would not be able to see at night.
- We would not be able to fight off diseases.
- We would not be able to have children.
- We would cease to exist!
LET’S FEED PEOPLE: Jejunal Morphology

In the end will the broccoli win?

“If you really were my best friend, Howard, you’d eat my broccoli.”
YES!!! Eat those vegetables

“I’m probably going to test positive for broccoli.”
The bottom line...

All things in moderation except vegetables!

- Red Sweet Potato
- Cassava
- Orange Maize
- Carrots

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The Current VAAL Team

- Ashley Valentine
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- Ting Sun
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