Protein (and muscle)
Muscle Protein

Protein Synthesis Breakdown

Unloading-induced atrophy is a relatively uncomplicated form of muscle loss... most of the loss of muscle mass during disuse atrophy can be accounted for by a depression in the rate of protein synthesis.

whereas in disease states associated with inflammation (cancer cachexia, AIDS, burns, sepsis, and uremia), there is a procatabolic hormonal and cytokine environment...
It is imperative that these studies include examination of dynamic measures of muscle protein turnover and putative metabolic controllers... unless we have a clear idea of the basic responses to immobilization per se, the effects of such factors will not be easily teased out and therapeutic goals will remain largely unattainable.

Hypercatabolism

Hyper-catabolic conditions associated with upregulation of the ubiquitin-proteasome system:
- Cancer cachexia (Longo et al. 2001; Tisdale et al. 2009)
- Cachexia associated with heart failure (Filippatos et al. 2005, Freeman et al. 1998)
- Sepsis (Vivian et al. 1996; Tao et al. 1994)
- Starvation (Whitehouse 2001)
- Metabolic acidosis (Mitch et al. 1994)
- Stress-trauma associated with excess glucocorticoids (Wing et al. 1993; Bolli et al. 2000)
- Space flight (Hemph et al. 2001; Rikay et al. 1992)

Omega 3 (n3) Fatty Acids

- Eicosapentaenoic acid (EPA)
  - 20-C, omega-3 fatty acid
  - Dietary sources: fish oil, flaxseed, walnuts
  - Beneficial effects on cholesterol, lipid metabolism, and cardiovascular health
Omega-3 and Cancer

Vanamala et al., Carcinogenesis, 2008

Proportion of tumor bearing rats

Vitamin D

Sources
- UVB radiation
- Food: Seafood, mushrooms, egg yolk, Fortified foods

Nomenclature
- Vitamin D$_2$ (ergocalciferol)
- Vitamin D$_3$ (cholecalciferol)
- 25-OH vitamin D
- 1,25 (OH)$_2$ vitamin D

Vitamin D Intake Guidelines
- RDA (1997 IOM)
  - 19-50 y: 200 IU/d
  - 50-70 y: 400 IU/d
- The 2005 Dietary Guidelines for Americans recommendation advised older adults, people with dark skin, and people exposed to insufficient sunlight to consume 1000 IU/d.

Vitamin D: Review

Other metabolites:
- 24,25(OH)$_2$D$_3$
- 25,26(OH)$_2$D$_3$
- 35 others...

Vitamin D Status

Contributing Factors to Vitamin D Status
- Age
- Ethnicity
- Salt-sensitive hypertension
  - Increased protein excretion in salt-sensitive individuals and Dahl rats with salt loading
- Adiposity/obesity
Vitamin D is associated with:
- Calcium metabolism
- Fracture Risk/BMD

Smith et al., J Nutr, 2006
Smith et al., J Nutr, 2005

SN
Con
Control
Pre-flight
Post-flight

0
10
20
30
40
50
60
70
80
90
100

25 (OH) Vitamin D (nmol/L)

Vitamin D is associated with:
- Calcium metabolism
- Fracture Risk/BMD
- Muscle strength/function
- Cancer
- Cardiovascular health
- Immune function
- Diabetes
- Multiple Sclerosis
- Dementia
- Parkinson’s Disease
- Tuberculosis
- Incidence of C-section
- The common cold

Vitamin D status has been related to:
- Fractures, fracture risk, BMD
- Muscle strength/function, falls
- Cancer (prostate, breast, colon)
- Multiple sclerosis
- Blood pressure/heart disease
- Diabetes (type 1)

Bischoff-Ferrari, Am J Clin Nutr, 2006

Recommendations
Encourage adequate vitamin D:
- Intake
  - Fortified milk, orange juice
  - Fish (salmon, tilapia, tuna)
  - Few other sources...
- Sunlight
- Supplements

...the criterion for broad-based supplementation in the general population is not fulfilled, except for in high risk groups, such as the elderly...all other persons with negligible exposure to sunshine.

Space Food

<table>
<thead>
<tr>
<th>Food</th>
<th>Vitamin D (IU)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flight Requirement</td>
<td></td>
</tr>
<tr>
<td>per day</td>
<td>450 IU</td>
</tr>
<tr>
<td>Menu</td>
<td></td>
</tr>
<tr>
<td>Salmon</td>
<td>236</td>
</tr>
<tr>
<td>Tuna</td>
<td>152</td>
</tr>
<tr>
<td>Breakfast Drink</td>
<td>116</td>
</tr>
<tr>
<td>Tuna Noodle Casserole</td>
<td>68</td>
</tr>
<tr>
<td>Cornflakes</td>
<td>68</td>
</tr>
<tr>
<td>Tuna Salad Spread</td>
<td>64</td>
</tr>
<tr>
<td>Bran Chex</td>
<td>64</td>
</tr>
<tr>
<td>Scrambled Eggs</td>
<td>64</td>
</tr>
<tr>
<td>Bread Pudding</td>
<td>56</td>
</tr>
<tr>
<td>Granola w/Raisins</td>
<td>44</td>
</tr>
<tr>
<td>Tapioca Pudding</td>
<td>44</td>
</tr>
<tr>
<td>Teriyaki: Beef</td>
<td>36</td>
</tr>
<tr>
<td>Pork Chips</td>
<td>32</td>
</tr>
<tr>
<td>Vegetable Quiche</td>
<td>28</td>
</tr>
<tr>
<td>Polako Soup</td>
<td>23</td>
</tr>
</tbody>
</table>
Upper Limits

2000 IU/day is currently defined IOM no observed adverse events limit (NOAEL). Studies of higher levels have proven safe...

Sunlight does not result in toxicity
Watch multivitamins (vit A and other nutrients may be in excess)

Vitamin D Toxicity

Hypercalcemia, hypercalciuria, soft tissue calcification, kidney stones

Vitamin D

Vitamin D status goes down after long-duration spaceflight.

Questions:
Is the stability of vitamin D in the food system and supplement different during spaceflight?
Is the daily dose not high enough to maintain status?
Does vitamin D metabolism change during spaceflight?

Stability Study

Stability of vitamin D in food/supplement is not altered during spaceflight

Question:
Is the daily dose simply not high enough to maintain status in an environment with no sun exposure?
Polar Vitamin D

3 levels of vitamin D supplementation:
- 400 IU/d (n = 18)
- 1000 IU/d (n = 19)
- 2000 IU/d (n = 18)

3 blood collections and diet logs
- 25D, 1,25D, PTH, Ca, VDBP, NTX

Double blinded supplementation

Smith et al., 2009

Compliance
84% on average

Vitamin D status is related to body weight…
- what if we exclude subjects with BMI > 29 kg/m²?

1000 or 2000 IU/d was enough to reach 80 nmol/L and maintain vitamin D status

Residual Questions…

Could compliance be improved with a weekly dose instead of a daily dose?
Is vitamin D status related to observed changes in immune function during polar winters?
In addition to BMI, the efficacy of vitamin D supplementation is affected by baseline status.

Compliance:
- 2000 IU/d: 91%
- 10,000 IU/wk: 97%

Residual Questions...

Is a higher, less frequent dose as effective as a daily or weekly dose? Does a high dose result in a high serum concentration of 25-OH vitamin D (or metabolites) or alter serum or urine calcium?

Vitamin D Dosing Study

2,000 IU/d 10,000 IU/wk 50,000 IU weekly x4; then 1/mo
Vit D (and metabolites)
Ca, etc.
Diet, uv

Supervised administration of 50,000 IU vitamin D3 dose 24 h later:
- Fasted blood
- 24-h urine
1 subject in 2000 IU/d group had 2 values > 150 nmol/L.
2 subjects in 50000 IU group had 3-5 values > 150 nmol/L.

10000 IU/wk
50000 IU/mo
Placebo

Vitamin D Dosing Study
Baseline
60 Days

Nutrition SMO

Calcium

Human Body
Absorption
Secretion
Deposition
Resorption
Excretion
Feces

1 subject in 2000 IU/d group had 2 values > 150 nmol/L.
2 subjects in 50000 IU group had 3-5 values > 150 nmol/L.
Collagen Crosslinks

Bone Resorption

Space Flight:
- Urinary collagen crosslinks
- Urinary Ca
- Urinary OH-Proline
- Bone resorption is increased during flight

Bone Formation/Resorption

Calcium Isotopes

Higher δ44Ca = "heavier"
Lower δ44Ca = "lighter"
Current folate intakes do not maintain folate status
How much folate is in the food? If enough – then:
Is folate stable on orbit? If it is – then:
What is changing?

Vitamin B6
Vitamin B12
Folate
Biotin
Pantothenate
Vitamin A (β-carotene)
Vitamin D
Vitamin E
Vitamin K
Amino Acids
Fatty acids

Radiation

Vitamin E
Excess sodium intake (and related effects on acid/base physiology) is associated with a number of health issues:

- Bone loss
- Increased renal stone risk
- Impaired muscle performance/protein catabolism
- Altered glucose metabolism
- Altered vitamin D metabolism
- Hypertension

With the exception of hypertension, all of these other factors have been raised as concerns for space travelers.

**NOTE:** only a few JAXA food items are on the standard menu at this point (and no ESA or CSA). These are included in the bonus foods per crew request (along with other non-standard foods).

In 2005-2006, the average US intake of Na was estimated at 3,436 mg Na/d*.

In 1990-1999, the average US intake of Na was estimated at:
- 3,377 mg for 31-50 yo M**
- 3,539 mg for 31-50 yo F

The excess sodium is bound to glycosaminoglycans in skin, exchanging with a hydrogen ion.

**Mechanism**

Excess sodium intake leads to non-osmotic (i.e., non-fluid retaining) storage of sodium in skin, exchanging with a hydrogen ion.
Acidosis

Recap 3

- High acid load and the body is under threat.
- Natural defense mechanisms are activated to neutralize the acid load.
- Adenosine triphosphate (ATP) is produced to buffer the acid load.
- Increased respiratory rate (panting) and increased excretion of bicarbonate ions.

Pathophysiologic effects of acidosis

- Metabolites increase in the urine of individuals who are taking drugs for chronic kidney disease.
- Increased excretion of sodium and water.
- Increased risk of osteoporosis.
- Increased risk of bone fractures.

Iron

(RBCs, and oxidative damage)

Iron and Oxygen

Radiation/oxygen issues have implications for cataracts and other health issues.

Total Body Iron

- Males
- Females
Iron and Oxidative Damage

Bed Rest

NEEMO

Supplements

Outliers

EVA Pilot Study
Bone Formation

Nutrition and Bone

Dietary Protein

Animal vs. Vegetable

Nutrition and Bone

Vitamin K
**APro:K and Bone**

Controlled dietary intake

- High or Low APro:K
- Monitored dietary intake

Blood/Urine markers

**EVA Pilot Study**

**Pro K**

**Controlled dietary intake**

- High or Low APro:K
- Monitored dietary intake

Blood/Urine markers

**EXAMPLE Menu**

High APro/K Day 1 Example

- Oatmeal w/ Brown Sugar
- Seasoned Scrambled Eggs
- Grilled Pork Chop
- Smoked Turkey
- Bread Pudding

- Granola Bar
- Cheese
- Grits
- Pasta w/Pesto Sauce
- Butter Cookies

- Fruit Cocktail
- Green Beans & Mushrooms
- Broccoli au Gratin
- Almonds

- Apple Cider
- Cashews
- Peanuts
- Pineapple
- Drink

- Tea

Low APro/K Day 1 Example

- Oatmeal w/ Raisins & Spice
- Vegetarian Vegetable Soup
- Chicken Noodle Soup
- Waffles
- Grilled Cheese
- Chicken
- Peanut Butter

- Broccoli
- Carrot Sticks
- Creamed Spinach
- Macadamia Nuts

- Cocoa
- Carrot Coins
- Creamed Spinach

- Orange Juice
- Tofu w/ Hot Mustard Sauce

- Apples w/ Spice
- Water (250 mL)

**Nutrition and Bone**

**Design**

**Preflight**

- L-180
- L-45

**Inflight**

- FD15
- FD30
- FD60
- FD120
- FD180
- FD60

**Postflight**

- R+30
- R+180
- R+365

NOTE: the low ratio diet is NOT low protein, and NOT vegetarian.

NOTE: the pattern above (red or blue) is an example, your pattern may vary.

Blood/Urine (24-h F; 48-h G) will be collected at the end of each session, and on L-10 and R+6.
Acid/Base and Bone

High protein, low potassium diet

Acid Load >> Alkali Load

H⁺ >> Organic anions

Na⁺/H⁺ exchange in skin GAGs

Excess dietary sodium

CO₃²⁻ → Ca²⁺ → CO₃²⁻ → Ca²⁺

Na⁺

GAG⁻ ↔ GAG⁻ ↔ Na⁺

Arachidonic acid
Bed rest
Hindlimb unloading
Spaceflight
Ionizing radiation/UVC

NF-κB
κB Inhibitor

NF-κB
κB Inhibitor

NF-κB (active)

RANKL
LPS
TNF-α

Inflammation

Inflammatory Markers

Muscle proteolysis

Bone resorption

Inflammation/Bone

NF-κB
κB Inhibitor

NF-κB
κB Inhibitor

NF-κB (active)

Muscle proteolysis

Osteoclast differentiation

Inflammation

3-MH (µmol/g creat)