Dietary restriction in humans

Dietary Restriction in Humans
George Sutphin
Kelly Gameski
Angela Poole
March 7, 2008
Dietary Restriction Extends Lifespan in a Variety of Organisms

(Yeast: (Jiang JC et al, FASEB, 2000))

(Worm: (Kaeberlein TL et al, Aging Cell, 2006))

And others: Rats, Spiders, Flies, etc…
Dietary Restriction Extends Lifespan in a Variety of Organisms

Yeast

WORM

MICE

Does it work in humans?
DR in humans is challenging...

- Humans as model organisms for aging:
  - Pros:
    - Findings in humans are always applicable to humans
    - Humans can tell you how they feel (easier to judge quality of life and other variables)
    - Wide genetic variation
  - Cons:
    - Humans live a long time
    - Hard to control environment and experimental inputs
      - Humans tend to have a mind of their own
      - You can’t (legally) keep them in cages
...but not impossible

• DR in non-human primates
• Natural DR (Okinawan Centaurians)
• Longevity studies not feasible → look at longevity markers with short term DR
  – Biological adaptation to DR
  – Risk factors for age-associated disease
  – Physiological variables known to deteriorate with age
• Human DR studies:
  – Biosphere 2
  – CALERIE
  – Calorie Restriction Society
Dietary Restriction in the Rhesus Monkey

- Average lifespan → 27 years in captivity
- DR improves numerous biomarkers of aging (insulin, metabolic rate, body temp, etc)
- Longevity data still 15 – 20 years out
DR in the Rhesus Monkey

CALORIE RESTRICTION DIET

Canto, 25
Although a senior citizen — the average rhesus monkey lifespan in captivity is 27 — Canto, above, is aging fairly well. Outwardly, he has a nice coat, elastic skin, a smooth gait, upright posture and an energetic demeanor. His bloodwork shows he is as healthy as he looks.

Human equivalent  Meals prepared by Mike Linksvayer, 36

NORMAL DIET

Owen, 26
He gets more food, but Owen, above, isn’t aging as well. His posture has been affected by arthritis. His skin is wrinkled and his hair is falling out. Owen is frail and moves slowly. His bloodwork shows unhealthy levels of glucose and triglycerides.

Diet of an average, active human male of 36

Breakfast  fermented soybeans and garlic
Lunch  tofu, konyakku and carrots
Dinner  vegan sausage, kale, tomato sauce and salad

Okinawan Centaurians

• Most natural caloric restriction goes along with malnutrition and disease (poverty)
• Exception: Older generation of Okinawans
  – Poverty reduced caloric intake
  – Public health measures provided nutrition and prevented disease
• Result:
  – More centaurians per capita in Okinawa than anywhere else in the world
  – However, oldest Okinawans are no older than oldest people elsewhere
DR in Biosphere 2

- 3 acre isolated artificial ecosystem
- 8 people enclosed for 2 years
- Not able to grow as much food as planned, lived on high-nutrient, low calorie foods (veggies, fruits, nuts, etc)
- All showed reduced BMI, blood pressure, blood glucose, insulin, cholesterol, T3, white blood cell count

→ Calorie restriction
CALERIE

• CALERIE = Comprehensive Assessment of Long-Term Effects of Reducing Calorie Intake

• Phase 1: Is DR feasible in humans?
  – 3 independent studies different institutions
  – 20-25% calorie restriction in overweight individuals
  – 6 – 12 months
  – Already complete

• Phase 2: Study effects of longer-term DR
  – 3 independent studies different institutions
  – 25% calorie restriction in normal to overweight individuals
  – 24 months
  – Scheduled to finish in 2011
Calorie Restriction Society

- People practicing self-imposed DR in the hope of extending lifespan and preventing disease
- Study looked at aging biomarkers:
  - 14 men, 4 women (most CRS members are men)
  - DR for 6 years
  - Control: 14 men, 4 women eating typical American diet
- Results:
  - Substantially lower blood pressure in DR group
  - BMI: 19.6 ± 1.9 kg/m² (DR), 25.9 ± 3.2 kg/m² (control)
  - Average body fat: 6.7% (DR), 22.4% (control)
  - DR group cholesterol all in lowest 10% of age group
  - DR group had improvements in all biomarkers studied
  - DR group all had low level chronic inflammation
Calorie Restriction: How-To Guide

• Reaching your target weight
  – 10-25% below “set point”
  – Plan to reduce weight over 1-2 years

• Changing your diet
  – Count calories, set a target and adjust to keep weight loss at 1-2% of total per month
  – Replace calorie-dense food with nutrient-rich, calorie sparse food first, then reduce quantity
  – Eat lots of vegetables and fruit
  – Add some lean protein (tuna, chicken) and healthy fats (nuts, avacado)

• Planning meals
  – Many books (“The Longevity Diet” is the most recent)
  – Nutrition software ensures proper nourishment
Calorie Restriction: How-To Guide

• Watch your biomarkers:
  – Tier 1: Daily/weekly at home measurements
    • Weight, Temperature, Resting Pulse
  – Tier 2: 6 – 12 month tests from doctor
    • Panels: Metabolic, Hepatic Function, Lipids, Blood, Blood Cell Count
  – Tier 3: Less frequent and more expensive
    • T3, insulin, rT3, cortisol, glycated hemoglobin, DHEA-S, etc…

• Know the risks:
  – Appearance Changes
  – Bone health
  – Cold sensitive
  – Age risks (don’t start before ~21)
  – Loss of “cushioning”
  – Pregnancy complications
  – Loss of stamina/strength
  – Decreased testosterone
  – Rapid weight loss
  – Slower wound healing
Overweight, non-obese, people who eat fewer calories lose more weight
One theory:

• If you lose weight, your resting energy expenditure goes down more than is expected simply as a function of body mass
• So-called “metabolic adaptation”
• Pros: calorie-restriction rhesus monkey study, quick weight loss by liquid diet in obese patients study
• Cons: much rodent literature
Study enrollment

Wait, I like food. Why did these people sign up for this?
Study design

• 4 groups:
  – **Baseline**: weight maintenance diet
  – **Calorie restriction**: 25% reduction in calories from weight maintenance diet
  – **Calorie restriction + exercise**: 12.5% reduction in calories from weight maintenance diet + exercise designed to burn 12.5% of calories from weight maintenance diet
  – **No soup for you**: 890 calories a day until 15% reduction in body weight, then weight maintenance diet
Study design cont’d

• The diet:
  – 2 weeks of baseline + 12 weeks:
    • 2 meals a day eaten at center
    • One sent home as takeout
  – Weeks 13-22
    • Meal self-selection based on calorie targets
  – Weeks 23-24
    • 2 meals at the center, one sent home as takeout

• What were they eating?
  – <30% fat
  – The very low calorie group: diet shakes + a “bolus” of fat for 8-11 weeks until reached the target weight
Study design cont’d

• **Groups:**
  – Weekly meetings
  – Regular follow-up to assess adherence

• **Exercise:**
  – 3x week supervised
  – 2x week unsupervised with HR monitor
Outcomes measured

• Weight loss
• Fasting glucose
• Fasting insulin
• Dehydroepiandrosterone sulfate
• Triiodothyronine
• Core body temp
• Absolute energy expenditure
• Protein carbonyls
Results: eat less, lose more weight
Results: proposed CR measures
Results: Core body temperature
24 hour energy expenditure
Results: Serum carbonyls & DNA damage
Authors’ conclusions:

- Reduction in fasting insulin level, reduction in core body temp, lower thyroid hormone levels (metabolic adaptation), and reduction in DNA damage strongly evidence benefits for CR
- The biomarkers (DHEAS, glucose) that were unchanged were simply not monitored long enough
- Cannot comment on oxidative stress
- Metabolic adaptation occurs
Calorie restriction increases muscle mitochondrial biogenesis in healthy humans

Anthony E Civitarese, Stacy Carling, Leonie K Heilbronn, Mathew H Hulver, Barbara Ukropcova, Walter A Deutsch, Steven R Smith, and Eric Ravussin, for the CALERIE Pennington Team
Hypothesis

Short-term caloric deficit will up-regulate the expression of genes involved in mitochondrial biogenesis in skeletal muscle resulting in increased mitochondrial content, improved whole body energy efficiency, and decreased DNA fragmentation in non-obese humans.

Similar Protocol To Previous Study

Control: 100% of energy requirements

Caloric Restriction (CR): 25% caloric restriction

Caloric Restriction with Exercise (CREX): 12.5% CR + 12.5% increased energy expenditure
### Metabolic Characteristics of Subjects Completing the Study 
\( n = 36 \)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control</th>
<th>Calorie Restriction</th>
<th>Calorie Restriction + Exercise</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex (M/F)</td>
<td>5/7</td>
<td>6/6</td>
<td>5/7</td>
</tr>
<tr>
<td>Age (y)</td>
<td>36.7 ± 2.1</td>
<td>38.9 ± 1.6</td>
<td>34.9 ± 1.6</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>82.3 ± 2.9</td>
<td>81.5 ± 3.0</td>
<td>81.1 ± 3.3</td>
</tr>
<tr>
<td>Body Fat (%)</td>
<td>32.3 ± 1.8</td>
<td>30.9 ± 2.1</td>
<td>31.0 ± 2.4</td>
</tr>
<tr>
<td>Fat Mass (kg)</td>
<td>25.5 ± 1.2</td>
<td>25.0 ± 1.7</td>
<td>24.9 ± 1.8</td>
</tr>
<tr>
<td>Fat-free mass (kg)</td>
<td>56.8 ± 3.1</td>
<td>56.5 ± 3.2</td>
<td>56.3 ± 3.5</td>
</tr>
<tr>
<td>24-h EE (kcal)</td>
<td>2,129 ± 97</td>
<td>2,092 ± 97</td>
<td>2,079 ± 102</td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td>89.6 ± 1.2</td>
<td>92.3 ± 2.0</td>
<td>89.3 ± 1.8</td>
</tr>
<tr>
<td>Insulin (uU/ml)</td>
<td>12.3 ± 0.9</td>
<td>12.6 ± 1.7</td>
<td>9.4 ± 1.5</td>
</tr>
<tr>
<td>Adiponectin (ug/ml)</td>
<td>3.0 ± 0.3</td>
<td>3.2 ± 0.3</td>
<td>3.2 ± 0.3</td>
</tr>
</tbody>
</table>

Baseline 6 months

*** Absolute 24-h Energy Expenditure was significantly reduced from baseline to month 6 in CR and CREX even after adjustment for metabolic body size changes in Fat Free Mass

*** Adiponectin tended to increase in both intervention groups
Mitochondrial Biogenesis Markers

- TFAM is a transcription factor that is a key activator of mitochondrial transcription
- PPARGC1A serves as a nutrient sensing system that increases mitochondrial biogenesis
- SIRT1 deacetylates and activates PPARGC1A
- eNOS, endothelial Nitric Oxide Synthase, regulates mitochondrial biogenesis in mouse skeletal muscle
Mitochondrial Biogenesis Markers

Skeletal muscle biopsies

Statistically significant increase in CR and CREX groups
⇒ Induction of mitochondrial biogenesis
Mitochondrial Enzyme Activity

- Oxidative enzyme activity was not increased in any of the groups.
- Several investigators have previously shown dissociation between mitochondrial mass and oxidative capacity after CR.

![Graph showing enzyme activity changes](image-url)
Same Pathway in Humans?

- **TFAM** is a transcription factor that is a key activator of mitochondrial transcription
- **PPARGC1A** serves as a nutrient sensing system that increases mitochondrial biogenesis
- **SIRT1** deacetylates and activates PPARGC1A
- **eNOS**, endothelial Nitric Oxide Synthase, regulates mitochondrial biogenesis in mouse skeletal muscle
NO and Mitochondrial Biogenesis

- Treat primary human muscle cells with NO donor (eNOS mimic) DETA-NO
- NO can induce mitochondrial biogenesis without increasing SIRT1 expression
- Adiponectin is an insulin-sensitizing adipokine with anti-diabetic and anti-inflammatory properties
- Previously showed adiponectin-AMPK interaction stimulates PPARGC1A protein expression and mitochondrial biogenesis

\[ g\text{AD}: \text{globular adiponectin} \]
\[ R1/R2: \text{adiponectin receptor} \]
Adiponectin signaling may be involved in regulation of SIRT1-PPARGC1A protein expression.

Some metabolic benefits from CR may result from improved secretion of adipokines such as adiponectin.
Summary

In overweight nonobese humans, short-term caloric restriction...

- Lowers whole-body energy expenditure
- Induces mitochondrial biogenesis of efficient mitochondria
- Decreases DNA damage
Increased Mitochondrial Content

⇒ Increase in mitochondrial mass
PPARGC1A vs SIRT1

Changes from baseline to 6-mo in PPARGC1A and SIRT1 were strongly correlated in all participants and in each group separately.