Energetics: Scientific Foundations of Obesity and Other Health Aspects

Energetics at the Molecular Level

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Overview of Metabolism

Metabolism Summary

Proteins
amino acids

Carbohydrates
glucose, fructose, galactose

Fats and Lipids
fatty acid, glycerol

Organic Compounds:

Hydrogen: H
Oxygen: O
Carbon: C
Nitrogen: N
Phosphorous: P
Sulfur: S

Water
Vitamins
Minerals

Nitrogen Pool
tissue protein

Glycogen
glycogenogenesis

glycogenolysis

gluconeogenesis

Glucose-6-Phosphate

Pyruvic Acid
Lactic Acid

Fatty Acid Spiral

Lipogenesis

Urea Cycle
urea

CO₂

CO₂

Citric Acid Cycle

2H⁺

2e⁻

ADP

ADP

ADP

ATP

O₂

H₂O

Electron Transport Chain
Energy Metabolism Overview

(a) Pyruvate produced in glycolysis is oxidized in (b) the tricarboxylic acid (TCA) cycle. (c) Electrons liberated in this oxidation flow through the electron-transport chain and drive the synthesis of ATP in oxidative phosphorylation. In eukaryotic cells, this overall process occurs in mitochondria.

‘1937’  
Krebs cycle =  
TCA cycle =  
Citric acid cycle
Complete oxidation of glucose:

\[ \text{C}_6\text{H}_12\text{O}_6 + 6\text{O}_2 \rightarrow 6\text{CO}_2 + 6\text{H}_2\text{O} \quad \Delta G^o' = -2823 \text{ kJ mol}^{-1} \]

Electron transfer:

1. **Oxidation of glucose carbon atoms**
   \[ \text{C}_6\text{H}_12\text{O}_6 + 6\text{H}_2\text{O} \rightarrow 6\text{CO}_2 + 24\text{H}^+ + 24\text{e}^- \]

2. **Reduction of molecular oxygen**
   \[ 6\text{O}_2 + 24\text{H}^+ + 24\text{e}^- \rightarrow 12\text{H}_2\text{O} \]

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**Figure 22-1** The sites of electron transfer that form NADH and FADH\textsubscript{2} in glycolysis and the citric acid cycle. Page 798

Voet *Biochemistry* 3e. © 2004 John Wiley & Sons, Inc.; Ch 22, pgs 797-840
Where does Metabolism Start?
We Burn Food

\[
\text{ADP} + P_i \rightarrow \text{CO}_2 \quad \text{H}_2\text{O} \quad \text{ATP}
\]
**Cellular Metabolism**

- **Cellular metabolism** refers to all of the chemical processes that occur inside living cells.
Energy can exist in two states:

- **Kinetic energy** – energy of motion.
- **Potential energy** – stored energy.
  - Chemical energy – potential energy stored in bonds, released when bonds are broken.

Energy can be transformed from one state to another.
Energy

- The ultimate source of energy for most living things is the sun.
Laws of Thermodynamics

- **First law of thermodynamics** – energy cannot be created or destroyed – only transformed.

- **Second law of thermodynamics** – a closed system moves toward entropy, increasing disorder.
  - Living systems are open systems that maintain organization and increase it during development.
Metabolism involves:

- **Catabolic reactions** that break down large, complex molecules to provide energy and smaller molecules.
- **Anabolic reactions** that use ATP energy to build larger molecules.
Importance of ATP

- **Endergonic** reactions require energy to proceed.
- Coupling an energy-requiring reaction with an energy-yielding reaction can drive endergonic reactions.
- **ATP** is the most common intermediate in coupled reactions.
Importance of ATP

- ATP consists of adenosine (adenine + ribose) and a triphosphate group.
  - The bonds between the phosphate groups are high energy bonds.
  - A-P~P~P
Importance of ATP

- Phosphates have negative charges.
  - Takes lots of energy to hold 3 in a row!
  - Ready to spring apart.
- So, ATP is very reactive.
Cellular Respiration - 3 Stages

- Food is digested to break it into smaller pieces – no energy production here.

- **Glycolysis** – coupled reactions used to make ATP.
  - Occurs in cytoplasm
  - Doesn’t require O$_2$

- **Oxidation** – harvests electrons and uses their energy to power ATP production.
  - Only in mitochondria
  - More powerful
Catabolic reactions:

**Stage 1: Digestion and hydrolysis**
break down large molecules to smaller ones that enter the bloodstream.

**Stage 2: Degradation**
Further breaking and some oxidation of molecules to 2 & 3-carbon compounds.

**Stage 3: Oxidation**
of small molecules to CO₂ & H₂O in the citric acid cycle and electron transport provides energy for ATP synthesis.
Glycolysis

- Glycolysis – the first stage in cellular respiration.
  - A series of enzyme catalyzed reactions.
  - Glucose converted to pyruvic acid.
  - Small number of ATPs made (2 per glucose molecule), but it is possible in the absence of oxygen.
  - All living organisms use glycolysis.
Glycolysis: General Functions

- Provide ATP energy
- Generate intermediates for other pathways
  - Hexose monophosphate pathway
  - Glycogen synthesis
  - Pyruvate dehydrogenase
    - Fatty acid synthesis
    - Krebs’ Cycle
  - Glycerol-phosphate (TG synthesis)
Glycolysis

- **Uphill portion primes the fuel with phosphates.**
  - *Uses 2 ATPs*
- **Fuel is cleaved into 3-C sugars which undergo oxidation.**
  - **NAD**⁺ accepts e⁻s & 1 H⁺ to produce NADH
  - NADH serves as a carrier to move high energy e⁻s to the final electron transport chain.
- **Downhill portion produces 2 ATPs per 3-C sugar (4 total).**
  - Net production of 2 ATPs per glucose molecule.
Glycolysis

Summary of the enzymatically catalyzed reactions in glycolysis:

$$\text{Glucose} + 2\text{ADP} + 2\text{P}_i + 2\text{NAD}^+ \rightarrow 2\text{Pyruvic acid} + 2\text{NADH} + 2\text{ATP}$$

http://www.youtube.com/watch?v=3GTjQTqUuOw&list=FL9N_Px072WuVorSwDfqf-9w&index=4&feature=plpp
Figure 13.11 Overview of the regulation of glycolysis

From Mathews and van Holde: Biochemistry 2/e. © The Benjamin/Cummings Publishing Co., Inc.
Regulation of Glycolysis

Regulatory mechanisms controlling glycolysis include allosteric and covalent modification mechanisms.

Glycolysis is regulated reciprocally from gluconeogenesis. Molecules, such as F2,6BP, that turn on glycolysis, turn off gluconeogenesis. Conversely, acetyl-CoA turns on gluconeogenesis, but turns off glycolysis. See Figure 16.6

The principle enzymes of glycolysis involved in regulation are hexokinase (reaction 1), phosphofructokinase (reaction 3), and pyruvate kinase (reaction 10):

1. **Hexokinase** is allosterically inhibited by glucose-6-phosphate (G6P). That is, the enzyme for the first reaction of glycolysis is inhibited by the product of the first reaction. As a result, glucose and ATP (in reactions 1 and 3) are not committed to glycolysis unless necessary.

2. **Phosphofructokinase** (PFK) is a major control point for glycolysis. PFK is allosterically inhibited by ATP and citrate, allosterically activated by AMP, ADP, and F2,6BP. Thus, carbon movement through glycolysis is inhibited at PFK when the cell contains ample stores of ATP and oxidizable substrates. Additionally, PFK is activated by AMP and ADP because they indicate low levels of ATP in the cell. F2,6BP is the major activator, though, because it reciprocally inhibits fructose 1,6 bisphosphatase, which is the gluconeogenic enzyme that catalyzes the reversal of this step.

3. **Pyruvate kinase** is allosterically inhibited by acetyl-CoA, ATP, and Alanine; allosterically activated by F1,6BP, and inhibited by cAMP-dependent phosphorylation. Note that several of the allosteric regulators are products of other metabolic pathways or are made in other metabolic pathways. These include acetyl-CoA, AMP, F2,6BP, and G1P, (readily converted into G6P). By having regulation dependent on other pathways, glycolysis is coordinately controlled with these pathways as well.

http://www.pearsonhighered.com/mathews/ch13/c13rog.htm
Figure 16.6: Major control mechanisms affecting glycolysis and gluconeogenesis

Major Points of Hormone Regulation of Carbohydrate Metabolism

Glucagon Activation

Fructose bisphosphate phosphatase
Fructose-1,6-bisphosphate
Phosphoenolpyruvate
Pyruvate Carboxylase
Oxaloacetate
Pyruvate
Pyruvate Dehydrogenase
Acetyl-CoA

G-6-P
PK-1, PK-2
Glycogen
Insulin Activation

P_i
Where is the bulk of this energy (ATP) produced?

Mitochondria
Harvesting Electrons form Chemical Bonds

- When oxygen is available, a second oxidative stage of cellular respiration takes place.
  - First step – oxidize the 3-carbon pyruvate in the mitochondria forming Acetyl-CoA.
  - Next, Acetyl-CoA is oxidized in the Krebs cycle.
Multi-enzyme complexes:

• Groups of noncovalently associated enzymes that catalyze 2 or more sequential steps in a metabolic pathway

• Advantages:
  – Enhanced reaction rates
  – Reduction of side reactions
  – Reactions can be coordinately controlled
The PDH complex contains multiple copies of 3 enzymes

**Eukaryotic PDH:**

- ~10,000 kDa Dodecahedron (12 pentagonal faces)
- **E₁** Pyruvate Dehydrogenase (30 E₁ \(\alpha_2\beta_2\) heterotetramers)
- **E₂** Dihydrolipoyl transacetylase (20 E₂ trimers - core)
- **E₃** Dihydrolipoyl dehydrogenase (12 E₃ dimers)

**Mammals also have:**

- E₃ binding protein (a catalytically inactive E₂-like protein that may help bind E₃ to the complex)
- 1-3 copies each of PDH kinase and PDH phosphatase
PDH Multienzyme Complex

Catalyzes 5 sequential reactions – 5 coenzymes required

Pyruvate + CoA + NAD$^+$ → acetyl CoA + CO$_2$ + NADH
**PDH Co-Factors**

TPP: Thiamine PyroPhosphate \((E_1)\) – (Vitamin B1 Thiamin)

Lipoamide: Lipoic Acid linked to \(\varepsilon\)-amino group of Lysine \((E_2)\) – (\(\alpha\)-lipoic acid)

FAD: Flavin Adenine Dinucleotide \((E_3)\) – (Vitamin B2 Riboflavin)

NAD: Nicotinamide Adenine Dinucleotide \((E_3)\) – (Vitamin B3 Niacin)

\(E_3\) Thiols: Protein bound thiols oxidized to form a disulfide.

---

CoA-S-H

Ac

CoA-S-Ac

Coenzyme A
Producing Acetyl-CoA

- The 3-carbon pyruvate loses a carbon producing an acetyl group.
- Electrons are transferred to NAD$^+$ forming NADH.
- The acetyl group combines with CoA forming Acetyl-CoA.
- Ready for use in Krebs cycle.
The Krebs Cycle

- The **Krebs cycle** is the next stage in oxidative respiration and takes place in the **mitochondria**.
  - Acetyl-CoA joins cycle, binding to a 4-carbon molecule to form a 6-carbon molecule.
  - 2 carbons removed as CO₂, their electrons donated to NAD⁺, 4-carbon molecules left.
    - 2 NADH produced.
  - More electrons are extracted and the original 4-carbon material is regenerated.
    - 1 ATP, 1 NADH, and 1 FADH₂ produced.
1. The cycle begins with 2C acetyl-CoA condensing with 4C oxaloacetic acid to form 6C citric acid. CoA is released to react again with pyruvic acid.

2. An isomer of citric acid is oxidized by NAD⁺, yielding 5C α-ketoglutaric acid, NADH, and a molecule of CO₂.

3. Oxidation by NAD⁺ occurs again, yielding 4C succinic acid, NADH, and CO₂.

4. One molecule of ATP is formed directly with each cycle.

5. Another oxidation by FAD yields FADH₂.

6. A final oxidation by NAD⁺ yields NADH and returns the cycle to its start point.
The Krebs Cycle

- Each glucose provides 2 pyruvates, therefore 2 turns of the Krebs cycle.
- Glucose is completely consumed during cellular respiration.
The Krebs Cycle

Acetyl unit $+ 3 \text{NAD}^+ + \text{FAD} + \text{ADP} + P_i \rightarrow 2 \text{CO}_2 + 3 \text{NADH} + \text{FADH}_2 + \text{ATP}$

http://www.youtube.com/watch?v=-cDFYXc9Wko
Current Dietary Reference Intake (DRI) composed of:

Setting DRIs

**EAR:**
- 50% risk of inadequacy

**EAR** (estimated Average Requirements)

**RDA:**
- 2-3% risk of inadequacy

**UL:**
- Upper Limit with no risk of inadequacy or adverse effects

Adequate Intake (AI):
- no RDA established,
- the amount established is somewhat less firmly believed to be adequate for everyone in the demographic group.

Between RDA and UL:
- Risk of inadequacy and of excess are both close to 0

The RDA is used to determine the Recommended Daily Value (RDV) which is printed on food labels in the U.S. and Canada.
**Vitamin B complex:**

* 8 water-soluble vitamins:

- Vitamin B1 (thiamine)
- Vitamin B2 (riboflavin)
- Vitamin B3 (niacin or niacinamide)
- Vitamin B5 (pantothenic acid)
- Vitamin B6 (pyridoxine, pyridoxal, or pyridoxamine, or pyridoxine hydrochloride)
- Vitamin B7 (biotin)
- Vitamin B9 (folic acid)
- Vitamin B12 (various cobalamins; commonly cyanocobalamin in vitamin supplements)

- each B vitamin has distinguishing character and chemical composition.

- They work in a group as well as individually to help and regulate numerous body functions including metabolizing glucose to release energy.
B-complex vitamins are absolutely essential to your body’s metabolism.

• Without even just one of them, don’t expect the food you’re eating to go down the drain anytime soon.

• Vitamin B1 / Thiamine – for proper metabolism of starch and sugars. Found in eggs, whole grain flour, potatoes, oranges and asparagus.

• Vitamin B2 / Riboflavin – for proper metabolism of fats, proteins and carbohydrates. Found in milk, cheese, leafy green vegetables and almonds.

• Vitamin B5 / Pantothenic Acid – for the generation of energy to be used in the body. Found in green vegetables, whole grain flour, chicken, eggs, beans and green vegetables.

• Vitamin B6 / Pyridoxine – for the metabolism of unsaturated fatty acids. Found in eggs, beef, chicken, bananas and avocados.

• Vitamin B12 / Cyanocobalamin – for the whole gamut of metabolism: digestion, protein synthesis, food absorption and general metabolism. Found in meat, fish and eggs.

While the B-complex family is the star of the “vitamins for weight loss,” there are some other vitamins that can help you reach your goal of losing weight in the long run. Vit C, Vit E, Choline
Vitamins: Cooking, Storage, other considerations

• Vitamins A, D, E and K, riboflavin and beta carotene are destroyed when exposed to light.

• Vitamins C, A, B12, folic acid and thiamin are destroyed by heat.

• Vitamins C, A, D, E, K, B12 and folic acid are destroyed by exposure to air.

• Vitamins C, B6, thiamin, riboflavin, niacin, selenium, potassium and magnesium leach into cooking water.

• Vitamins C, B12, folic acid, thiamin and riboflavin are destroyed when combined with acid or alkaline substances.
### Recommended dietary allowances and suggested optimal intakes

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<th>Nutrient</th>
<th>Men</th>
<th>Women</th>
<th>Suggested intake</th>
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<td>Vitamin A</td>
<td>1000 mcg RE</td>
<td>800 mcg RE</td>
<td>1500 mcg RE</td>
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<td>Niacin</td>
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<td>Vitamin B6 (under 50)</td>
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<td>Vitamin B6 (over 50)</td>
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<tr>
<td>Vitamin B12</td>
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<td>Pantothenic acid</td>
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## Dietary Reference Intakes (DRIs): Recommended Dietary Allowances and Adequate Intakes, Vitamins

### Food and Nutrition Board, Institute of Medicine, National Academies

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<tr>
<th>Life Stage Group</th>
<th>Vitamin A (μg retinol eq)</th>
<th>Vitamin C (mg)</th>
<th>Vitamin D (μg)</th>
<th>Vitamin E (μg)</th>
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<th>Folate (μg)</th>
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<td>&gt; 70 yrs</td>
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</tr>
<tr>
<td>9-13 yrs</td>
<td>600*</td>
<td>45</td>
<td>15</td>
<td>11</td>
<td>60*</td>
<td>0.9</td>
<td>0.9</td>
<td>12</td>
<td>1.0*</td>
<td>300</td>
<td>1.8*</td>
<td>4*</td>
<td>20*</td>
</tr>
<tr>
<td>14-18 yrs</td>
<td>700*</td>
<td>65</td>
<td>15</td>
<td>12</td>
<td>75*</td>
<td>1.0</td>
<td>1.0</td>
<td>14</td>
<td>1.2*</td>
<td>400</td>
<td>2.4*</td>
<td>5*</td>
<td>25*</td>
</tr>
<tr>
<td>19-50 yrs</td>
<td>700*</td>
<td>75</td>
<td>15</td>
<td>12</td>
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<td>5*</td>
<td>30*</td>
</tr>
<tr>
<td>51-70 yrs</td>
<td>700*</td>
<td>75</td>
<td>15</td>
<td>12</td>
<td>75*</td>
<td>1.1</td>
<td>1.1</td>
<td>14</td>
<td>1.5*</td>
<td>400</td>
<td>2.4*</td>
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<tr>
<td>&gt; 70 yrs</td>
<td>700*</td>
<td>75</td>
<td>15</td>
<td>12</td>
<td>75*</td>
<td>1.1</td>
<td>1.1</td>
<td>14</td>
<td>1.5*</td>
<td>400</td>
<td>2.4*</td>
<td>5*</td>
<td>30*</td>
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<tr>
<td><strong>Pregnancy</strong></td>
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<tr>
<td>14-18 yrs</td>
<td>750*</td>
<td>80</td>
<td>15</td>
<td>15</td>
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</tr>
<tr>
<td>19-50 yrs</td>
<td>750*</td>
<td>85</td>
<td>15</td>
<td>15</td>
<td>75*</td>
<td>1.4</td>
<td>1.4</td>
<td>18</td>
<td>1.9*</td>
<td>600</td>
<td>2.6*</td>
<td>6*</td>
<td>30*</td>
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<tr>
<td>51-70 yrs</td>
<td>750*</td>
<td>85</td>
<td>15</td>
<td>15</td>
<td>75*</td>
<td>1.4</td>
<td>1.4</td>
<td>18</td>
<td>1.9*</td>
<td>600</td>
<td>2.6*</td>
<td>6*</td>
<td>30*</td>
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<tr>
<td><strong>Lactation</strong></td>
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</tr>
<tr>
<td>14-18 yrs</td>
<td>1,100</td>
<td>115</td>
<td>15</td>
<td>19</td>
<td>75*</td>
<td>1.4</td>
<td>1.5</td>
<td>17</td>
<td>2.0*</td>
<td>500</td>
<td>2.3*</td>
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<tr>
<td>19-50 yrs</td>
<td>1,100</td>
<td>115</td>
<td>15</td>
<td>19</td>
<td>75*</td>
<td>1.4</td>
<td>1.5</td>
<td>17</td>
<td>2.0*</td>
<td>500</td>
<td>2.3*</td>
<td>7*</td>
<td>35*</td>
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<tr>
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<td>1,100</td>
<td>115</td>
<td>15</td>
<td>19</td>
<td>75*</td>
<td>1.4</td>
<td>1.5</td>
<td>17</td>
<td>2.0*</td>
<td>500</td>
<td>2.3*</td>
<td>7*</td>
<td>35*</td>
</tr>
</tbody>
</table>

**Note:** This table (taken from the DRI reports, see [www.nap.edu](http://www.nap.edu)) presents Recommended Dietary Allowances (RDAs) in bold type and Adequate Intakes (AIs) in ordinary type followed by an asterisk (*). An RDA is the average daily dietary intake level sufficient to meet the nutrient requirements of nearly all (97-98 percent) healthy individuals in a group. It is calculated from an Estimated Average Requirement (EAR). If sufficient scientific evidence is not available to establish an EAR, and thus calculate an RDA, an AI is usually developed. For healthy breastfed infants, an AI is the mean intake. The AI for other life stages and gender groups is believed to cover the needs of all healthy individuals in the group, but lack of data or uncertainty in the data prevent being able to specify with confidence the percentage of individuals covered by this intake.

---

1. As retinol activity equivalents (RAE). 1 RAE = 1 μg retinol, 12 μg β-carotene, 24 μg α-carotene, or 24 μg δ-cryptoxanthin. The RAE for dietary provitamin A carotenoids is two-fold greater than retinol equivalents (RE), whereas the RAE for preformed vitamin A is the same as RE.
2. As cholecalciferol. 1 μg cholecalciferol = 40 IU vitamin D.
3. Under the assumption of maximal sunlight.
4. As α-tocopherol. α-Tocopherol includes RRR-α-tocopherol, the only form of α-tocopherol that occurs naturally in foods, and the 2R, stereoisomeric forms of α-tocopherol (RRR, RSR, RSS, and RRS-α-tocopherol) that occur in fortified foods and supplements. It does not include the 2S-stereoisomeric forms of α-tocopherol (SSR, SSR, SRS, and SSS-α-tocopherol), also found in fortified foods and supplements.
5. As niacin equivalents (NE). 1 mg of niacin = 60 μg of erythronol, 64 μg of converted to niacin equivalents (NE).
6. As dietary folate equivalents (DFE). 1 DFE = 1 μg food folate + 0.6 μg folic acid from fortified food or as a supplement consumed with food = 0.5 μg of a supplement taken on an empty stomach.
7. Although AIs have been set for choline, there are few data to assess whether a dietary supply of choline is needed at all stages of the life cycle, and it may be that the choline requirement can be met by endogenous synthesis at some of these stages.
8. Because 10 to 30 percent of older people may malabsorb food-bound B12, it is advisable for those older than 50 years to meet their RDA mainly by consuming foods fortified with B12 or a supplement containing B12.
9. In view of evidence linking folate intake with neural tube defects in the fetus, it is recommended that all women capable of becoming pregnant consume 400 μg from supplements or fortified foods in addition to intake of folate from dietary sources.

### Dietary Reference Intakes (DRIs): Recommended Dietary Allowances and Adequate Intakes, Total Water and Macronutrients

<table>
<thead>
<tr>
<th>Life Stage Group</th>
<th>Total Water* (L/d)</th>
<th>Carbohydrate (g/d)</th>
<th>Total Fiber (g/d)</th>
<th>Fat (g/d)</th>
<th>Linoleic Acid (g/d)</th>
<th>α-Linolenic Acid (g/d)</th>
<th>Protein* (g/d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infants</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0 to 6 mo</td>
<td>0.7*</td>
<td>60*</td>
<td>ND</td>
<td>31*</td>
<td>4.4*</td>
<td>0.5*</td>
<td>9.1*</td>
</tr>
<tr>
<td>6 to 12 mo</td>
<td>0.8*</td>
<td>95*</td>
<td>ND</td>
<td>30*</td>
<td>4.6*</td>
<td>0.5*</td>
<td>11.0</td>
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<tr>
<td>Children</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1–3 y</td>
<td>1.5*</td>
<td>130</td>
<td>19*</td>
<td>ND*</td>
<td>7*</td>
<td>0.7*</td>
<td>13</td>
</tr>
<tr>
<td>4–8 y</td>
<td>1.7*</td>
<td>130</td>
<td>25*</td>
<td>ND*</td>
<td>10*</td>
<td>0.9*</td>
<td>19</td>
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<tr>
<td>Males</td>
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<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>9–13 y</td>
<td>2.4*</td>
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<td>ND*</td>
<td>12*</td>
<td>1.2*</td>
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<tr>
<td>14–18 y</td>
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<td>130</td>
<td>38*</td>
<td>ND*</td>
<td>16*</td>
<td>1.6*</td>
<td>52</td>
</tr>
<tr>
<td>19–30 y</td>
<td>3.7*</td>
<td>130</td>
<td>38*</td>
<td>ND*</td>
<td>17*</td>
<td>1.6*</td>
<td>56</td>
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<tr>
<td>31–50 y</td>
<td>3.7*</td>
<td>130</td>
<td>38*</td>
<td>ND*</td>
<td>17*</td>
<td>1.6*</td>
<td>56</td>
</tr>
<tr>
<td>51–70 y</td>
<td>3.7*</td>
<td>130</td>
<td>30*</td>
<td>ND*</td>
<td>14*</td>
<td>1.6*</td>
<td>56</td>
</tr>
<tr>
<td>&gt; 70 y</td>
<td>3.7*</td>
<td>130</td>
<td>30*</td>
<td>ND*</td>
<td>14*</td>
<td>1.6*</td>
<td>56</td>
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<tr>
<td>Females</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>9–13 y</td>
<td>2.1*</td>
<td>130</td>
<td>26*</td>
<td>ND*</td>
<td>10*</td>
<td>1.0*</td>
<td>34</td>
</tr>
<tr>
<td>14–18 y</td>
<td>2.5*</td>
<td>130</td>
<td>26*</td>
<td>ND*</td>
<td>11*</td>
<td>1.1*</td>
<td>46</td>
</tr>
<tr>
<td>19–30 y</td>
<td>2.7*</td>
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<td>25*</td>
<td>ND*</td>
<td>12*</td>
<td>1.1*</td>
<td>46</td>
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<tr>
<td>31–50 y</td>
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<td>25*</td>
<td>ND*</td>
<td>12*</td>
<td>1.1*</td>
<td>46</td>
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<td>51–70 y</td>
<td>2.7*</td>
<td>130</td>
<td>21*</td>
<td>ND*</td>
<td>11*</td>
<td>1.1*</td>
<td>46</td>
</tr>
<tr>
<td>&gt; 70 y</td>
<td>2.7*</td>
<td>130</td>
<td>21*</td>
<td>ND*</td>
<td>11*</td>
<td>1.1*</td>
<td>46</td>
</tr>
<tr>
<td>Pregnancy</td>
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<td></td>
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<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>14–18 y</td>
<td>3.0*</td>
<td>175</td>
<td>28*</td>
<td>ND*</td>
<td>13*</td>
<td>1.4*</td>
<td>71</td>
</tr>
<tr>
<td>19–30 y</td>
<td>3.0*</td>
<td>175</td>
<td>28*</td>
<td>ND*</td>
<td>13*</td>
<td>1.4*</td>
<td>71</td>
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<tr>
<td>31–50 y</td>
<td>3.0*</td>
<td>175</td>
<td>28*</td>
<td>ND*</td>
<td>13*</td>
<td>1.4*</td>
<td>71</td>
</tr>
<tr>
<td>Lactation</td>
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<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>14–18</td>
<td>3.8*</td>
<td>210</td>
<td>29*</td>
<td>ND*</td>
<td>13*</td>
<td>1.3*</td>
<td>71</td>
</tr>
<tr>
<td>19–30 y</td>
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</tr>
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<td>29*</td>
<td>ND*</td>
<td>13*</td>
<td>1.3*</td>
<td>71</td>
</tr>
</tbody>
</table>

**NOTE:** This table (taken from the DRI reports, see www.nap.edu) presents Recommended Dietary Allowances (RDA) in bold type and Adequate Intakes (AI) in ordinary type followed by an asterisk (*). An RDA is the average daily dietary intake level sufficient to meet the nutrient requirements of nearly all (97%–98 percent) healthy individuals in a group. It is calculated from an Estimated Average Requirement (EAR). If sufficient scientific evidence is not available to establish an EAR, and thus calculate an RDA, an AI is usually developed. For healthy breastfed infants, an AI is the mean intake. The AI for other life stage and gender groups is believed to cover the needs of all healthy individuals in the group, but lack of data or uncertainty in the data prevent being able to specify with confidence the percentage of individuals covered by this intake.

* Total water includes all water contained in food, beverages, and drinking water.

* Based on g protein per kg of body weight for the reference body weight, e.g., for adults 0.8 g/kg body weight for the reference body weight.

* Not determined.

## Dietary Reference Intakes: Electrolytes and Water

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>Function</th>
<th>Life Stage Group</th>
<th>AI (L/d)</th>
<th>UL*</th>
<th>Selected Food Sources</th>
<th>Adverse Effects of Excessive Consumption</th>
<th>Special Considerations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water</td>
<td>Maintains homeostasis in the body and allows for transport of nutrients to cells and removal and excretion of waste products of metabolism.</td>
<td>Infants 0–6 mo</td>
<td>0.7</td>
<td>No UL.</td>
<td>All beverages, including water, as well as moisture in foods (high moisture foods include watermelon, meats, soups, etc.).</td>
<td>No UL because normally functioning kidneys can handle more than 0.7 L (24 oz) of fluid per hour; symptoms of water intoxication include hypokalemia which can result in heart failure and rhabdomyolysis (skeletal muscle tissue injury) which can lead to kidney failure.</td>
<td>Recommended intakes for water are based on median intakes of generally healthy individuals who are adequately hydrated; individuals can be adequately hydrated at levels below as well as above the AIs provided. The AIs provided are for total water in temperate climates. All sources can contribute to total water needs: beverages (including tea, coffee, juices, sodas, and drinking water) and moisture found in foods. Moisture in food accounts for about 20% of total water intake. Thirst and consumption of beverages at meals are adequate to maintain hydration.</td>
</tr>
</tbody>
</table>

|         |          | 7–12 mo         | 0.8      |     |                       |                                          |                        |
|         |          | Children 1–3 y   | 1.3      |     |                       |                                          |                        |
|         |          | 4–8 y           | 1.7      |     |                       |                                          |                        |
|         |          | Males 9–13 y    | 2.4      |     |                       |                                          |                        |
|         |          | 14–18 y         | 3.3      |     |                       |                                          |                        |
|         |          | 19–30 y         | 3.7      |     |                       |                                          |                        |
|         |          | 31–50 y         | 3.7      |     |                       |                                          |                        |
|         |          | 50–70 y         | 3.7      |     |                       |                                          |                        |
|         |          | > 70 y          | 3.7      |     |                       |                                          |                        |
|         |          | Females 9–13 y  | 2.1      |     |                       |                                          |                        |
|         |          | 14–18 y         | 2.3      |     |                       |                                          |                        |
|         |          | 19–30 y         | 2.7      |     |                       |                                          |                        |
|         |          | 31–50 y         | 2.7      |     |                       |                                          |                        |
|         |          | 50–70 y         | 2.7      |     |                       |                                          |                        |
|         |          | > 70 y          | 2.7      |     |                       |                                          |                        |
|         |          | Pregnancy       |          |     |                       |                                          |                        |
|         |          | 14–18 y         | 3.0      |     |                       |                                          |                        |
|         |          | 19–50 y         | 3.0      |     |                       |                                          |                        |
|         |          | Lactation       |          |     |                       |                                          |                        |
|         |          | 14–18 y         | 3.8      |     |                       |                                          |                        |
|         |          | 19–50 y         | 3.8      |     |                       |                                          |                        |

**NOTE:** The table is adapted from the DRI reports. See [www.nap.edu](http://www.nap.edu). Adequate Intakes (AIs) may be used as a goal for individual intake. For healthy breastfed infants, the AI is the mean intake. The AI for other life stage and gender groups is believed to cover the needs of all individuals in the group, but lack of data prevent being able to specify with confidence the percentage of individuals covered by this intake; therefore, no Recommended Dietary Allowance (RDA) was set.

*UL = The maximum level of daily nutrient intake that is likely to pose no risk of adverse effects. Unless otherwise specified, the UL represents total intake from food, water, and supplements. Due to lack of suitable data, ULs could not be established for potassium, water, and inorganic sulfate. In the absence of ULs, extra caution may be warranted in consuming levels above recommended intakes.

*ND = Not determinable due to lack of data of adverse effects in this age group and concern with regard to lack of ability to handle excess amounts. Source of intake should be from food only to prevent high levels of intake.

**SOURCE:** Dietary Reference Intakes for Water, Potassium, Sodium, Chloride, and Sulfate. This report may be accessed via www.nap.edu.
Coenzyme NAD$^+$

NAD$^+$ (nicotinamide adenine dinucleotide)

- Participates in reactions that produce a carbon-oxygen double bond (C=O).
- Is reduced when an oxidation provides 2H$^+$ and 2e$^-$.  

\[
\text{Oxidation} \quad \begin{array}{c}
\text{CH}_3\text{CH}_2\text{OH} \quad \rightarrow \quad \text{CH}_3\text{C} \parallel \text{H} + 2\text{H}^+ + 2\text{e}^-
\end{array}
\]

\[
\text{Reduction} \quad \begin{array}{c}
\text{NAD}^+ + 2\text{H}^+ + 2\text{e}^- \quad \rightarrow \quad \text{NADH} + \text{H}^+
\end{array}
\]
Structure of Coenzyme NAD$^+$

NAD$^+$

- Is nicotinamide adenine dinucleotide.
- Contains ADP, ribose, and nicotinamide.
- Reduces to NADH when the nicotinamide group accepts H$^+$ and 2e$^-$. 

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Niacin (B3). Biochemistry:

Nicotinamide Coenzymes:
- Nicotinamide
- Adenine
- Dinucleotide (Phosphate)

NAD and NADP roles: **Redox Reactions**
1. Hydrogen acceptors in most dehydrogenations
2. Hydrogen donor in Electron Transport Chain
3. NADPH is a reducing agent in biosynthesis (ie. Fatty acids, cholesterol) & involved in protection against toxicity of ROS (ie. GSH from GSSG reduction by glutathione reductase)
Coenzyme FAD

FAD (flavin adenine dinucleotide)
- Participates in reactions that produce a carbon-carbon double bond (C=C).
- Is reduced to FADH$_2$.

**Oxidation**

\[ \text{---CH}_2\text{---CH}_2\text{---} \rightarrow \text{---CH}=\text{CH}--- + 2\text{H}^+ + 2\text{e}^- \]

**Reduction**

\[ \text{FAD} + 2\text{H}^+ + 2\text{e}^- \rightarrow \text{FADH}_2 \]
Structure of Coenzyme FAD

FAD

- Is flavin adenine dinucleotide.
- Contains ADP and riboflavin (vitamin B₂).
Riboflavin (B2). Chemistry & Biochemistry:

- Isoalloxazine (tricyclic ring) derivative:
  
  **7,8-di-methyl-10-(1’-D-ribityl)isoalloxazine**

- “Ribo” refers to the ribityl side chain and “flavin” is now synonymous with any substituted isoalloxazine.
  - bright yellow, fluorescent (UV), slightly water soluble
  - decomposed by light but is heat stable

Converted to 2 Co-enzymes: **FMN** & **FAD**

FMN: Flavin Mononucleotide

FAD: Flavin adenine dinucleotide
Riboflavin. Biochemistry: **Oxidation/Reduction Rxs**

**FMN / FAD**

**FMNH2 / FADH2**

**FMN (riboflavin-5’-phosphate):**
- produced from riboflavin by riboflavin kinase functions as prosthetic group of various oxidoreductases including NADH dehydrogenase.
- It is the principal form in which riboflavin is found in cells and tissues. It requires more energy to produce, but is more soluble than riboflavin.

**“Energy carriers”**

**FAD (flavin adenine dinucleotide):**
- derived from riboflavin bound to phosphate of ADP
- redox cofactor involved in metabolism.
- two different redox states that provide its function.
- FAD can be reduced to the FADH2, whereby it accepts two hydrogen atoms:
  - Many oxidoreductases, called flavoenzymes or flavoproteins, require FAD as a prosthetic group which functions in electron transfers.
  - reduced coenzyme FADH2 (energy-carrying), is a substrate for OxPhos in the mitochondria. FADH2 is reoxidized to FAD, generating proton gradient across the inner mitochondrial membrane for ATP synthase to produce 2.0 equivalents ATP.
- primary sources of reduced FAD in eukaryotic metabolism are the TCA (citric acid cycle). FAD is a prosthetic group in the enzyme succinate dehydrogenase → succinate to fumarate; whereas in beta oxidation - coenzyme in the reaction of acyl CoA dehydrogenase.
Coenzyme A

Coenzyme A.

- Consists of vitamins B₃, pantothenic acid, and ADP.
- Activates acyl groups such as the two-carbon acetyl group for transfer.

\[
\text{O} \\
\text{CH}_3\text{C} \quad + \quad \text{HS-CoA} \quad \rightarrow \quad \text{CH}_3\text{C-S-CoA}
\]

acetyl group \hspace{1cm} \text{acetyl CoA}
Structure of Coenzyme A

Aminoethanethiol  Pantothenic acid  Phosphorylated ADP

Coenzyme A
Fuel molecule entry points in Oxidative Metabolism

Using Electrons to Make ATP

- NADH & FADH₂ contain energized electrons.
- NADH molecules carry their electrons to the inner mitochondrial membrane where they transfer electrons to a series of membrane bound proteins – the electron transport chain.
Building an Electrochemical Gradient

- In eukaryotes, aerobic metabolism takes place in the mitochondria in virtually all cells.
- The Krebs cycle occurs in the **matrix**, or internal compartment of the mitochondrion.
- Protons (H\(^+\)) are pumped out of the matrix into the **intermembrane space**.
Electron Transport Review

http://www.youtube.com/watch?v=kN5MtqAB_Yc&list=FL9N_Px072WuVorSwDfqf-9w&index=2&feature=plpp
Factors Governing Electron Flow Through the Transport Chain

Membrane potential ($\Delta \Psi$)

Inner membrane

Matrix

NADH → NAD$^+$

$\frac{1}{2}O_2 + 2H^+ \rightarrow H_2O$

Slide provided by Darrell Neufer, PhD; East Carolina University
Factors Governing Electron Flow Through the Transport Chain

State IV

Membrane potential ($\Delta \Psi$)

Point 1: The rate of respiration (electron flow) is determined by the rate at which protons enter back into the matrix.

Slide provided by Darrell Neufer, PhD; East Carolina University
Factors Governing Electron Flow Through the Transport Chain

State IV

Membrane potential ($\Delta \Psi$)

Inner membrane

Matrix

NADH $\rightarrow$ NAD$^+$

$H^+$

$e^-$

$O_2$

$O_2^-$

$\frac{1}{2}O_2 + 2H^+ \rightarrow H_2O$

Point 2: Electron leak to oxygen is favored when membrane potential is high (i.e., state 4 conditions).

Point 2: Electron leak to oxygen is favored when membrane potential is high (i.e., state 4 conditions).

Membrane potential ($\Delta \Psi$)
**Metabolism of Lipids**

- Triglycerides are broken down into glycerol and 3 fatty acid chains.
- Glycerol enters glycolysis.
- Fatty acids are oxidized and 2-C molecules break off as acetyl-CoA.
  - Oxidation of one 18-C stearic acid will net 146 ATP.
  - Oxidation of three glucose (18 Cs) nets 108 ATP.
  - Glycerol nets 22 ATP, so 1 triglyceride nets 462 ATP.
Metabolism of Proteins

- Proteins digested in the gut into amino acids which are then absorbed into blood and extracellular fluid.
- Excess proteins can serve as fuel like carbohydrates and fats.
  - Nitrogen is removed producing carbon skeletons and ammonia.
    - Carbon skeletons oxidized.
Metabolism of Proteins

- Ammonia is highly toxic, but soluble.
  - Can be excreted by aquatic organisms as ammonia.
- Terrestrial organisms must detoxify it first.
Rate of cellular respiration slows down when your cells have enough ATP.

Enzymes that are important early in the process have an allosteric (regulating) site that will bind to ATP.

When lots of ATP is present, it will bind to this site, changing the shape of the enzyme, halting cellular respiration.
Regulating Cellular Respiration

- Enzyme activity is controlled by presence or absence of metabolites that cause conformational changes in enzymes.
  - Improves or decreases effectiveness as catalyst.
Control of ATP production

**Coordinated control of ATP production**
- Glycolysis, the citric acid cycle and the oxidative phosphorylation
  Control of \([\text{NADH}] / [\text{NAD}^+]\)
- Aerobic vs. anaerobic metabolism
  Aerobic ATP production is far more efficient than anaerobic

- Disease conditions:
  - Cancer: Coordinated controls broken down and increased ATP utilization
  - Cardiovascular disease: oxygen deprivation, decreased \(O_2\) supply to cells → reduced ATP synthesis, increased ROS production → cell damage.

**Figure 22-48** Diagram depicting the coordinated control of glycolysis and the citric acid cycle by ATP, ADP, AMP, \(P_i\), \(Ca^{2+}\), and \([\text{NADH}] / [\text{NAD}^+]\).
Regulation of blood glucose levels

Glucagon

- Catabolic, in response to hypoglycemia
- Liver
  - Activates glycogen degradation, gluconeogenesis
- Adipose
  - Stimulates lipolysis and release of fatty acids
Regulation of blood glucose levels

Insulin

Anabolic in response to hyperglycemia

- Liver
  - Stimulates glycogen synthesis, glycolysis, and fatty acid synthesis
- Muscle
  - Stimulates glycogen synthesis
- Adipose
  - Stimulates lipoprotein lipase resulting in uptake of fatty acids from chylomicrons and VLDL
  - Stimulates glycolysis for glycerol phosphate synthesis (precursor to triglycerides)
Diagram of the action of insulin on liver, adipocyte and muscle cells
Metabolic changes occurring in the fasted-to-fed cycle

Review: When Food Meets Man: the Contribution of Epigenetics to Health
by Emma De Fabiani, Nico Mitro, Federica Gilardi, Andrea Galmozzi, Donatella Caruso and Maurizio Crestani.
Nutrients 2010, 2(5), 551-571; doi:10.3390/nu2050551
Fasted State

- **Glycogenolysis**
  - Glucose (simple sugar)
  - Glycogen
  - Glucose

- **Gluconeogenesis**
  - Proteins, fats
  - Glucose
**Insulin in Body and Brain**

**INSULIN’S ROLE IN BODY AND BRAIN**
Insulin, long recognized as a primary regulator of blood glucose, is now also understood to play key roles in neuroplasticity, neuromodulation, and neurotrophism, the process of neuronal growth, stimulated by neuronal differentiation and survival.

**METABOLIC INFLUENCE**
Insulin is one of the primary hormones involved in blood glucose regulation. Its dysregulation is associated with obesity and diabetes.

**NEUROLOGIC INFLUENCE**
Insulin activates insulin receptors and downstream signaling molecules in the brain and spinal cord, as well as insulin-sensitive glucose transporters in the peripheral insulin-sensitive tissues (liver, muscle, fat). Through these mechanisms, insulin participates in feeding behavior, reward pathways, whole body metabolism, and normal emotional and cognitive brain functions. The dysregulation of insulin-mediated signaling pathways in the brain is implicated in neurodegenerative diseases such as Alzheimer’s and psychiatric disorders such as schizophrenia.
Different Cell Types Require Different Fuel Molecules

Glycemic Index or Glycemic Load, are they the same?

**Glycemic Index:** David Jenkins, Thomas Wolever and colleagues 1981, University of Toronto – wanted a way to classify foods for diabetics,
- uses a scale from 0-100
- GI of 100 given to area under 2-hour blood glucose curve in response to oral intake of 50g of glucose.

How quickly carbohydrates enter the blood stream after intake *(proteins and fats are not on the glycemic index). Carbohydrates are chosen 1st for breakdown and metabolism over proteins and fats.

Original posited that 1) Starches produce a similar after meal blood glucose responses – Low AND ALL Sugars produce similar blood glucose responses – High – all based on chemical structures.

Low - 0-55; Moderate - 56-69; High - 70-100

Found: white bread (starch) = GI value of 69 = High
    ice cream (w sugar) = GI value of 36 = Low

Use of GI: UK, Europe, Australia, USA, Canada, WHO, – all for Diabetes

www.EatGoodCarbs.com ; www.GlycemicIndex.com
**Glycemic Index or Glycemic Load, are they the same?**

High GI carbs – **Gushers** – quickly digested, rapid rise in blood glucose, GI = 70+

**PROMOTE FAT STORAGE**

Low GI carbs – **Tricklers** – slowly digested, slow/gradual rise in blood glucose, GI = 0-55

**DISCOURAGES FAT STORAGE**

QUALITY of carbohydrate = GI

**Glycemic Load:** BOTH the quality and quantity of the carbohydrate amount in 1 number

\[
GL = \frac{\text{carbohydrate (grams/serving) x GI}}{100}
\]

Eg: GL of small versus large apple, since GI is the same

- Apple Small \[(13 \text{ grams x 38}) / 100 = 5 \text{ grams}\]
- Apple Large \[(26 \text{ grams x 38}) / 100 = 10 \text{ grams}\]

GL: how much insulin would need to be released by the pancreas into the blood stream to allow tissues to absorb that specific amount of that specific carbohydrate and reduce blood sugar levels effectively

www.EatGoodCarbs.com ; www.GlycemicIndex.com

Walter Willett, Professor at Harvard School of Public Health
High GI vs Low GI Foods

Blood Glucose Levels

Time/Hours

Graph adapted from: www.gisymbol.com (University of Sydney). Images from Microsoft Clipart.
Tissue-tissue cross-talk in glucose and lipid homeostasis
Diabetes Mellitus - Insulin Insufficiency

Characterized by: - high blood-glucose level
- Glucose overproduced by liver
- glucose underutilized by other organs
- shift in fuel usage from carbohydrates to fats → keton bodies (shortage of oxaloacetate)
- high level of keton bodies → kidney cannot balance pH any more → lowered pH in blood and dehydration → coma

Type I diabetes: insulin-dependent diabetes (requires insulin to live)
- caused by autoimmune destruction of β-cells
- begins before age 20
- insulin absent → glycagon present
- person in biochemical starvation mode + high blood-glucose level
- entry of glucose into cells is blocked
- glucose excreted into urine → also water excreted → feel hungry + thirsty

Type II diabetes: insulin-independent diabetes
- have a normal-high level of insulin in blood → unresponsive to hormone
- develops in middle-aged, obese people
Development of Systemic Insulin Resistance

Modified from: http://michaelscally.blogspot.com/2013/02/obesity-induced-insulin-resistance.html
Energy Intake

- Taken up 83%
- Passed 17%

A significant proportion of standard metabolic rate is devoted to driving mitochondrial proton leak which is a futile cycle.

SMR 36%
- External Work 8%
- Growth & Reproduction 15%
- Thermogenesis 5%
- Digestion 10%
- Muscle use 26%
"Metabolism of fuel generates a stoichiometric amount of NADH and FADH₂. Oxidation of NADH and FADH₂ results in 10 and 6 protons, respectively, being pumped out of the mitochondrial matrix. Three protons enter via ATP synthase in order to synthesize one molecule of ATP from ADP and Pi. One additional proton enters the matrix as it is co-transported with Pi via the phosphate carrier. ATP is then utilized to perform a fixed amount of work. The major consumers of ATP are shown above. Muscle relaxation, ion leaks, protein degradation and dephosphorylation create the possibility for "futile cycles". See Rolfe and Brown (Rolfe, D. F. & Brown, G. C. Cellular energy utilization and molecular origin of standard metabolic rate in mammals. *Physiol. Rev.* 77, 731–758 (1997)) for a complete analysis of the concept of coupling with respect to reactions in energy metabolism.

Towards a molecular understanding of adaptive thermogenesis
Bradford B. Lowell and Bruce M. Spiegelman
doi:10.1038/35007527
Thermodynamic perspective of energy expenditure

Resting Metabolic Rate and Energy Balance

Total energy expenditure = Heat produced + work on environment (when organism is at rest, all energy expenditure is equal to heat produced, that is, thermogenesis)

Adaptive thermogenesis
- variable, regulated by the brain
- responds to temperature and diet
- occurs in brown adipocyte mitochondria, skeletal muscle and other sites

Physical activity
- variable

Obligatory energy expenditure
- required for performance of cellular and organ functions

Towards a molecular understanding of adaptive thermogenesis
Bradford B. Lowell and Bruce M. Spiegelman
Nature 404, 652-660(6 April 2000)
doi:10.1038/35007527
Identify molecular and functional changes in skeletal muscle and mitochondria which contribute to the development of obesity, insulin resistance, and diabetes.
Human Oxidative Metabolism, IR and T2DM

Altered glycolytic and oxidative capacities of skeletal muscle contribute to insulin resistance in NIDDM.

Simoneau JA, Kelley DE.

Published online: 15 June 2003; doi:10.1038/ng1188

PGC-1α-responsive genes involved in oxidative phosphorylation are coordinately downregulated in human diabetes

Vamsi K Moorthy1, 2, 3, 10, Cecilia M Lindgren1, 4, 10, Karl-Fredrik Eriksson1, Aravind Subramanian1, Smita Sihag1, Joseph Lehar1, Pere Puigserver1, Emma Carlsson1, Martin Ridderstråle1, Esa Laurila3, Nicholas Houstis1, Mark J Daly1, Nick Patterson1, Jill P Mesirov1, Todd R Golub1, 5, Pablo Tamayo1, Bruce Spiegelman1, Eric S Lander1, 6, Joel N Hirschhorn1, 7, 8, David Altshuler1, 2, 7, 9, 11
& Leif C Groop1, 4, 11


Coordinated reduction of genes of oxidative metabolism in humans with insulin resistance and diabetes: Potential role of PGC1 and NRF1.


Family history of diabetes links impaired substrate switching and reduced mitochondrial content in skeletal muscle.

Ukropcova B, Sereda O, de Jonge L, Bogacka I, Nguyen T, Xie H, Bray GA, Smith SR.


Mitochondrial dysfunction and type 2 diabetes.

Lowell BB, Shulman GI.


Skeletal muscle "mitochondrial deficiency" does not mediate insulin resistance.

Holloszy JO.
Factors Governing Electron Flow Through the Transport Chain

Membrane potential ($\Delta \Psi$)

Inner membrane

Matrix

Factors Governing Electron Flow Through the Transport Chain

Membrane potential ($\Delta \Psi$)

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Membrane potential ($\Delta \Psi$)

Inner membrane

Matrix
Factors Governing Electron Flow Through the Transport Chain

**PUSH – State 4 - LEAK**

Membrane potential ($\Delta\Psi$)

- Inner membrane
- Matrix
- NADH $\rightarrow$ NAD$^+$
- $\frac{1}{2}O_2 + 2H^+ \rightarrow H_2O$
- ADP

**PULL - State III - Activity**

The rate of respiration (electron flow) is determined by the rate at which protons enter back into the matrix

DRM Modified slide: original by Darrell Neufer, PhD; East Carolina Diabetes and Obesity Institute; ECU
Factors Governing Electron Flow Through the Transport Chain

**State IV - LEAK**

Membrane potential ($\Delta \Psi$)

Electron leak to oxygen is favored when membrane potential is high (i.e., state 4 conditions).

Slide provided by Darrell Neufer, PhD; East Carolina University
Mitochondria make “ROS”

ROS = Reactive oxygen species (a.k.a. free radicals)…
Complex I → ROS from the FMN site, facing in.
Complex III → ROS from semiquinone, on both sides of the membrane.
Some debate as to whether $\text{O}_2^\cdot-$ or $\text{HO}_2^\cdot-$ is formed.
$\text{O}_2^\cdot-$ cannot leave mitochondrial matrix. $\text{H}_2\text{O}_2$ can freely diffuse out.
Role in aging process (Harman theory) vs. role in cell signaling?
mtDNA has no histones → greater mutation frequency than nDNA.
Mitochondrial $\text{H}_2\text{O}_2$ is Higher in Obese and HF-fed Lean Males

\[ \text{mH}_2\text{O}_2 \text{ emission (pmol/min/mg dry wt)} \]

Succinate (mM)

\[ \text{H}_2\text{O}_2 \text{ emitted/O}_2 \text{ consumed} \]

Succinate  PCM

\[ \text{Pre-HF meal} \]
\[ 4 \text{ h post-HF meal} \]
\[ 5 \text{ d HF diet/12 h fasted} \]

Lean Males

\[ J \text{ Clin Invest.} 2009; 119(3):573–581 \text{ doi:10.1172/JCI37048} \]
Mitochondrial superoxide regulates insulin sensitivity in vivo.

(A and B) Mice fed a standard lowfat (LFD, A) or high fat diet (HFD, B) ± 30 or 50 mg/kg MnTBAP 6h before i.p. injection of 1.5 g glucose/kg body weight. n≥8 mice per group.

(C and D) Glucose disposal into muscle and gonadal adipose tissue was measured by GTT with 3H-2DOG tracer. Mice were fed LFD or 2 weeks HFD ± 50 mg/kg MnTBAP (HFD+MnT) 6 h before glucose tolerance testing. n = 5 mice per group.
Mitochondrial superoxide regulates insulin sensitivity in vivo.

(E and F) GTTs (1.5 g glucose/kg body weight) were performed on MnSOD transgenic (MnSOD-TG) and age matched control (WT) mice fed a LFD then switched to HFD for 1 week. The same mice were used in both tests, n=7–8 mice.

(G) For the experiment in E-F above, insulin levels were measured after 6 h fasting and 15 min after glucose injection. N=7.

(H) GTT of MnSOD-TG and age matched WT mice fed a LFD or HFD for 12 weeks. n = 3–4 for LFD and 5–6 for HFD.

(I) Insulin tolerance test (ITT) of MnSOD-TG and age matched WT mice fed a LFD or HFD for 24 weeks. n = 7–8 in each group.
Insulin resistance is a cellular antioxidant defense mechanism.

**Metabolic Syndrome**

**WHO**
- High Insulin
  - Fasting/postpr
  +
  - (any 2 below)
- Wst/Hip > 0.9
- BMI ≥ 30kg/m²
- Waist > 37in
- TG ≥ 150mg/dL
- HDL < 40mg/dL
- BP – Trtment Htn or ≥ 140/90

**2001 ATP III**
(any 3 below)

**Men:**
- Waist ≥ 40in
- TG ≥ 150mg/dL
- HDL < 40mg/dL
- BP ≥ 130/85 mm Hg
- FBG ≥ 100mg/dL

**Women:**
- Waist > 35in
- TG ≥ 150mg/dL
- HDL < 50mg/dL
- BP > 130/85 mm Hg
- FBG > 100 mg/dL

Source of Oxidative Stress [RO(N)s] include fat overloaded adipose cells in viscera (VAT), liver and muscle: ‘metabolically triggered inflammation - meta-inflammation’.
Proposed Regulation of Superoxide Production in Skeletal Muscle

The greater the rate of entry of e⁻ from ETF, the greater the competition for oxidized Q → increased rate of superoxide production at complex I

Slide provided by Darrell Neufer; Trends in Endo. & Metab. 23(3), 2012, 142–153 & Int. J. Obesity Suppl., 2,S31-S36, 2012
Factors Governing Electron Flow Through the Transport Chain

Membrane potential ($\Delta\Psi$)

Inner membrane

Matrix

NADH $\rightarrow$ NAD$^+$

Expend energy

Slide courtesy of Darrell Neufer modified DRM
Metabolic integration by PPARs.
Now onto:
Mitochondria in Sickness and in Health

The End
and
Thank you for your attention.