Multiple-Frequency Bioelectrical Impedance Analysis (BIA)

- Fixed low and high frequency
  - 1 or 5 kHz to measure ECW
  - 50, 100, 200, or 500 kHz to measure TBW

\[ V = \rho L^2/R \]

Regression of \( Ht^2/R \) (or other) measured at low and high frequency against ECW and TBW measured by dilution methods
- Equations are population-specific

Ex: \( TBW \ (L) = m \ Ht^2/R_{200} + c \)
Ex: \( ECW \ (L) = m \ Ht^2/R_5 + c \)
EX: \( TBW - ECW = ICW \)

- Theoretically able to differentiate ECW vs. ICW, and to quantify BCM
Bioimpedance Spectroscopy (BIS)

- Range of frequencies (~5 - 1000 kHz)
- Biophysical modeling
  - Impedance data over the spectrum is fit to the Cole model through nonlinear least squares curve fitting
  - Cole model terms can then be:
    - Regressed vs. dilution volumes to derive equations (Cole)
      - Once cross-validated these equations can be used to estimate volumes
    - Applied to equations based on Hanai mixture theory (Cole/Hanai)
- Theoretically able to differentiate ECF vs. ICF, and to quantify body cell mass (BCM)
  - ICF ~ BCM
Bioimpedance spectroscopy is a unique bioimpedance approach that differs in underlying basis from the more readily recognized single-frequency bioelectrical impedance analysis in that it does not require the use of statistically derived, population-specific prediction equations. It has the potential advantage of not only measuring total body water, but also offering the unique capacity to differentiate between ECW and ICW and, thus, to provide an estimate of BCM.
Bioelectrical Impedance Analysis

• 1994 NIH Technology Assessment Conference
  • “BIA provides a reliable estimate of total body water under most conditions.”
  • “It can be a useful technique for body composition assessment in healthy individuals”
BIA: Advantages and Limitations

• Advantages
  – costs ($1,000-$15,000)
  – portable
  – non-invasive
  – fast

• Limitations
  – accuracy and precision
  – no better/worse than hydrodensitometry
BIA: Advantages and Limitations

- Best used for epidemiologic studies
- Choice of predictive equations important
- Influenced by fluid shifts
- Biggest drawback is the need for appropriately calibrated, cross-validated predictive equations (sme age, sex, ethnicity, and health)
BIA: Advantages and Limitations

• The accuracy has been questioned:
  – Skinfolds 2.4 % error
  – BIA 5% error
  – Visual 3.1% error

• Race cannot be entered into the machine

• Children distribute water differently than adults
Equations

• Each machine has its own equation (developed by the manufacturer and is proprietary)

• \( \% \text{fat} = 4.57 \div (1.1411 - ((BW \times \text{Resistance}) \div \text{Ht}^2)) - 4.142 \times 100 \)
  
  – \( \text{ht} = \) length of the conductor
BIA – Phase angle

The use of the phase angle has become more popular over the last few years because of its high association with clinical results, time of hospitalization and mortality in various diseases. Based on the principles of BIA, which mainly works by measuring body resistance and reactance in order to alternate an electric current, the storage of this current is thought to be able to create a change in phase which is considered to be the ratio between resistance and reactance and which is expressed geometrically as phase angle, being directly calculated as: arc tangent

$$\text{arc tangent} = \left( \frac{X_c}{R} \right) \times 180^\circ / \pi.$$
Graphic derivation diagram of the phase angle and its relation to resistance \( (R) \), reactance \( (Xc) \), impedance \( (Z) \) and frequency of the current applied.
Among all the direct measurements of BIA, the phase angle has proved to be a good predictor of prognosis and mortality regarding hemodialysis, cancer, human immunodeficiency syndrome (HIV), and liver and geriatric diseases. This measurement has attracted strong interest by being a noninvasive, objective and rapid (less than 2 minutes) tool for the determination of nutritional status and risk of patient morbidity, whereas other nutritional screening tools, although also noninvasive, require more time and / or are highly subjective.
Interpreting % Fat Values

• All methods of measuring % fat have a certain amount of inaccuracy! This inaccuracy is determined by the Standard Error of Estimate (SEE).

• The SEE tells you the amount of deviation from the true % fat you can expect from a particular method.
• There is a 67% probability that the true % fat is within + or - one SEE from the measured value.

• Example: Measure % fat = 20%; SEE = 3 % units of body fat
  There is a 67% probability that the true % fat is between + or - one SEE or 3 % units of fat or between 17 - 23 %.
• There is a 95% probability that the true % fat is within + or - two SEE from the measured value.
• Example: Measured %fat = 20%
SEE = 3 %
There is a 95% probability that the true %fat is within + or - two SEE or 6% of the measured value or 14 - 26 %.
SEE of Common Methods

- UWW – 1.5-2.5%
- Plethysmography – 2.2-3.7%
- Skinfolds – 3-4%
- Bioelectric Impedance
  - Whole Body – 3-4%
  - Segmental – 4-6%
Riassunto delle assunzioni fatte dai metodi bicompartimentali
Assumptions of Two-Component Models

• 1. The density of fat is 0.900 g/ml
• 2. The density of FFM is 1.100 g/ml
• 3. The densities of fat and FFM are the same for all individuals
Assumptions

• 4. The densities of the various components of FFM are constant within an individual.

• 5. The individual being measured differs from a reference body (73.8% water, 19.4% protein, 6.8% mineral) only in the amount of fat.
Dual-Energy X-ray Absorptiometry
Dual Energy X-Ray Absorptiometry (DXA)

- Uses x-rays to measure thickness, density and chemical composition of tissue.
DXA Technology

**X-ray Source**
(produces 2 photon energies with different attenuation profiles)

**Collimator**
(pinhole for pencil beam, slit for fan beam)

**Detector**
(detects 2 tissue types - bone and soft tissue)

**Patient**

Very low radiation to patient.
Very little scatter radiation to technologist

**Photons**
VARIAZIONE DEL COEFFICIENTE LINEARE DI ATTENUAZIONE PER L’OSSO, LA MASSA MAGRA E LA MASSA GRASSA
• Attenuation constant between individuals.

• Assesses total bone mineral content. Further, each extra-bone pixel contains information on % fat and % lean tissue. Area determinations that tell both the % fat and % lean mass in a single region of interest.

• Three component model
  – bone, fat, fat-free soft tissue mass
Dual-energy XA

Underlying principle: X-rays are attenuated by body tissue, each to a different degree depending on frequency (energy). DXA uses two beams at different energies. Ratio can accurately measure attenuation of each component.
DXA

• Two different energy level X-rays
• Lean, fat, and bone mass each reduce (attenuate) the X-ray signal in unique ways
• Computer analyzes scan point by point to determine body composition
• Method
  – 20-30/7-10/4-5 minutes
  – Applicable to young and old
TECNICA DUALE DI EMISSIONE RAGGI X

I due diversi livelli di energia per discriminare il tessuto molle da quello osseo, si possono ottenere con due tecniche differenti:

• Energia Pulsata

• Energia Filtrata
TECNICHE DI EMISSIONE RAGGI X

ENERGIA PULSATA

Il tubo radiologico (ad anodo fisso) di questa generazione di strumenti viene alimentato con due voltaggi differenti che si alternano per produrre i due livelli di alta e bassa energia. Tensione tipica: 100-140 kVp.

ENERGIA FILTRATA

Il tubo radiologico (ad anodo fisso o rotante) viene in questo caso alimentato con un solo voltaggio in cui si interpone sul fascio prodotto un filtro di terra rara per produrre i due livelli di diversa energia. Tensione tipica: 84 kVp.
TECNICHE DI EMISSIONE RAGGI X

ENERGIA PULSATA

ENERGIA FILTRATA
DXA

• Bone mineral density
  – Attenuation calibrated to phantoms of known calcium content
  – Precision approximately 1%

• Lean mass
  - Precision approximately 1-2%

- Fat mass
  - Precision approximately 2.5%
DXA

• Advantages

  – Scan the entire body (regions) for total % fat, % lean mass, and bone density

  – Relatively non-invasive, relatively inexpensive to manage, fast

  – Good for absolute measurement and following changes in an individual

  – Rapid

  – Minimal subject cooperation (just lay there)
Disadvantage

• Costly (when buying)
• Limited Access
COMPOSIZIONE CORPOREA

Valutazione della massa grassa, magra e ossea settoriale.
Campi di applicazione:
Medicina Sportiva, Dietologia, Riabilitazione Funzionale, Fisiatria.
DEXA BMD Caveats

- Assumed that there is no change in attenuation with thickness. This is not true over about 20 cm
- DEXA MEASURES PERCENT COMPOSITION NOT ABSOLUTE VALUES
- Despite labels, DXA does not measure true bone density. It is the attenuation in a particular surface, there is no depth to the measurement
Computed Tomography ("CT Scan" or "Cat Scan")

- The scanner device incorporates a moving table & a revolving X-ray tube
  - The table moves the patient back and forth through the revolving X-ray emissions
  - The X-ray emitter moves (revolves) in a 360° arc around the patient
- Instead of film, the CT scanner collects emitted X-rays via a collector
  - scintillator
- Collector transforms X-ray photons into a proportionally strong electric current
- The electric current is then converted into an image
  - Contrast dyes may be used for image enhancement
Normal CT scan (abdominal slice)
CT

- Area of interest outlined on the slice ($r = 0.94$ with planimetry on cadavers)
- Specific density for each tissue of interest derived from standards
- Allows true volume measurement and thus true density.
CT

• Excellent correlation between body fat mass and cross-sectional abdominal adipose tissue area
  – Men $r=0.92$
  – Women $r=0.97$

• Nine scans required

• Difficulty is radiation exposure and cost
Figure 8.2  Relation of total cross-sectional abdominal adipose tissue area measured by computed tomography to body fat mass assessed by hydrostatic weighing in a sample of 89 men.
Figure 8.3  Relation of total cross-sectional abdominal adipose tissue area measured by computed tomography to body fat mass assessed by hydrostatic weighing in a sample of 75 premenopausal women.
**Magnetic Resonance Imaging**

- **Nuclei paramagnetici sono presenti nel corpo** (1H, 13C, 23Na, 31P, 17O, 19F)
  - Il nucleo dell'idrogeno è prevalente perché presente nell'acqua e negli acidi grassi
- **Il soggetto è posto in un campo magnetico statico**
- **I nuclei magnetizzati (1H nuclei) nel soggetto si allineano a questo campo**
- **Un impulso di radiofrequenza (RF) crea un campo magnetico oscillante perpendicolare a quello statico**
- **I nuclei magnetizzati assorbono l'energia dell'impulso RF ed entrano in uno stato eccitato**
- **Quando la RF è spenta I nuclei eccitati ritornano allo stato basale ed emettono energia in forma di RF**
- **Differenti elementi assorbono ed emettono differenti quantità di energia RF (differenti risonanza)**
- **L'energia RF emessa è rilevata da un’antenna e trasformata in immagini**
### Magnetic Resonance Imaging
#### Tissues Composition & Signal Intensity

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Signal Intensity T1</th>
<th>Signal Intensity T2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fat</td>
<td>high (whitish)</td>
<td>intermediate</td>
</tr>
<tr>
<td>Muscle</td>
<td>intermediate (gray)</td>
<td>intermediate</td>
</tr>
<tr>
<td>Hyaline Cartilage</td>
<td>intermediate</td>
<td>intermediate - low (dull gray)</td>
</tr>
<tr>
<td>Ligaments &amp; Tendons</td>
<td>low (dark gray)</td>
<td>low</td>
</tr>
<tr>
<td>Cortical Bone</td>
<td>low</td>
<td>low</td>
</tr>
<tr>
<td>Granulation Tissue</td>
<td>intermediate</td>
<td>high</td>
</tr>
<tr>
<td>Fibrous Tissue</td>
<td>low</td>
<td>low</td>
</tr>
<tr>
<td>Hemorrhage / Edema</td>
<td>high - intermediate</td>
<td>high</td>
</tr>
<tr>
<td>Immature Scar</td>
<td>intermediate - low</td>
<td>low to high</td>
</tr>
<tr>
<td>Mature Scar</td>
<td>low</td>
<td>low</td>
</tr>
</tbody>
</table>
MRI

• Same principle as CT
• May need as few as 1 to 4 slices at the L4-L5 level (questionable benefit)
• Major drawback is cost
• Considered by some to be reference techniques for body composition
MRI

L4-L5 (#21)

Adipose Tissue Area (cm²)

Total

Visceral

0  4  8  12  16  20  24  28  32  36  40

0  100  200  300  400

[Graph showing distribution of adipose tissue area across different regions, with markers indicating total and visceral adipose tissue]
Near Infrared Red Spectroscopy (NIRS)

• Based on the premise that the degree of infrared light absorption is related to the composition of the substance through which light passes

• Fat and Fat-Free Mass absorb and reflect light differently
NIR

• Emit infrared light at wavelengths of 940-950 nm into a body part (ie., biceps) and measures the intensity of the re-emitted light

• More specific equations/machines are necessary
Advantages

• Non-invasive
• Safe
• Easy to administer
• Field technique
Disadvantages

• Cost? Is it worth it?
• Few Age/Gender Specific Equations
• Accurate?
  – Futrex 5000 3.1-4.2%
  – Futrex 5000A 6.3%
  – Futrex 1000 4.8-6.3%
  – Sum 3 2.4-3.6
Total Body Water

- Water in two compartments: body cells and extracellular fluids
- Dilution of known tracer – typically stable isotopes of water (tritium, deuterium, oxygen-18)
- Grams of tracer administered will be diluted and volume calculated
Total Body Water

Body water = \( w \times f \times \frac{C_{\text{dose}}}{C_{\text{body water}}} \)

\( W \) = moles of water in original sample
\( F \) = fractionation compared to water
\( C_{\text{dose}} \) = enrichment or concentration of dose
\( C_{\text{body water}} \) = enrichment or conc in body sample
Total Body Water

• Assumptions
  – Tracer only in body water – the labeled atoms can undergo exchange with other organics. 2-5% error for all isotopes
  – Equal distribution of tracer to all compartments – not usually a problem
  – Fast rate of equilibrium – IV takes 2-3 hours. Oral up to 6 or more
  – No tracer metabolism – constant excretion and dilution. Plateau method with several determinations after ingestion
Total Body Water

• Fat will not take up water
• Use a hydration constant to determine fat free mass
• Problems encountered: must assume the hydration of fat free mass equal in different individuals and disease states, measurement error, instrumentation required
Total Body Water – fat free mass

- Fat will not take up water
- Use a hydration constant (0.73) to determine fat free mass
- Problems encountered: must assume the hydration of fat free mass equal in different individuals and disease states, measurement error, instrumentation required
TOTAL BODY WATER

• Determined by introducing a marker fluid that moves freely in body water and is not metabolized. (isotope dilution)
• Deuterium Oxide, tritiated water
• % FAT PREDICTED FROM TOTAL BODY WATER
  – Assume 73.8% Water in Fat Free Mass
  – Even if no technical error in Body Water, there would still be S.E.E. = 3.6% Fat associated with biological variability
Whole-Body Counting

• Scintillation detectors were developed in the early 1950’s.
• They measure the body’s natural potassium as well as other radioactivity in the body.
Whole-Body Counting

• In 1958, Kulwich, Feinstein, and Anderson correlated natural potassium concentration with fat free mass.

• No longer in use
Whole-Body Counting

• There are an estimated 75 counters in the US.
• There are more than 180 whole-body counters worldwide.
• Two-thirds of these perform body potassium measurements in humans.
Whole-Body Counting

• Potassium is naturally distributed in three isotopic states.
• The isotope 40K is radioactive.
Whole-Body Counting

• Gamma rays from 40K are high-energy gammas, many of which exit the body and can be easily detected by external counting.

• The smaller the subject, the lower the 40K content and thus the weaker gamma signal.
Whole-Body Counting

- Factors such as age, fitness, or restricted mobility due to surgery or illness do not tend to affect the precision of total body potassium measurements.
- The 40K signal is natural and continuous, therefore the measurement can be interrupted as necessary, until counting is completed.
Whole-Body Counting

The three requirements for 40K whole body-counting include:

• Efficient gamma-ray detectors that can be placed close to the subject.

• Shielding for these detectors to reduce the natural background radiation levels
Whole-Body Counting

- Computer-based instruments that enable identification of the unique gamma rays.
Whole-Body Counting

Precision:

• For whole-body counters, precision is in the range of 2-5% for adults.

• In infants and very young children, precision is only 8-12% for 40 minute sample times.
Whole-Body Counting

• Total cost for an adult whole-body counter is $10,000-15,000.
• Cost of a special shielded room starts at $80,000.
• Start-up costs.
• Still in use for total body protein, body cell mass, skeletal mass
Neutron Activation Analysis

• IVNAA measures 11 elements from nuclear reactions.

• Protein, mineral, and fat can be estimated from these elements:
  – Carbon = Lipid
  – Nitrogen = Protein
  – Calcium = Bone
Neutron Activation Analysis

• IVNNA uses a whole-body counter.
  – It delivers a moderate beam of fast neutrons to the subject.
  – Atoms of target elements capture these neutrons.
  – This creates an unstable isotope.
Neutron Activation Analysis

• Unstable isotopes produce gamma rays when returning to a stable state.

• Gamma rays are measured:
  – energy level identifies the element
  – the activity indicates its abundance.
Prompt-Gamma Activation Analysis

• Isotope gets very excited with the added neurons.
  – Lasts only a fraction of a nanosecond before it returns to a stable state.
  – Measured simultaneously as the neutrons are exposed to the isotope.
Neutron Activation Analysis

Disadvantages:

• Radiation exposure.
• Must be performed by medical personnel
• Cost $30,000 - $300,000.
Figure 7

Neutron activation analysis determination of body composition.
Kehayias, Joseph; Valtuena, Silvia

Figure 7. C/O and C/H ratios for four chemical compartments. Carbon-to-hydrogen (C/H) and carbon-to-oxygen (C/O) ratios for four chemical compartments of the body. C/H is a measure of 'dryness', whereas C/O is an index of 'fatness'. Measurement errors are reduced when neutron activation is used for the simultaneous measurement of ratios of elements.
Equation 17

\[
\text{Skeletal muscle} = (0.188 \times \text{TBK}) + (0.00183 \times \text{TBN})
\]
Figure 8

Neutron activation analysis determination of body composition.
Kehayias, Joseph; Valtuena, Silvia


Figure 8. The elemental partition analysis (EPA) method applied to the assessment of skeletal muscle. Selection of element for skeletal muscle assessment by elemental partition analysis (EPA). Total body potassium (TBK) will require a large number of corrections for non-muscle potassium, whereas phosphorus (TBP) requires high precision measurements of TBP and bone.
Neutron activation analysis determination of body composition.
Kehayias, Joseph; Valtuena, Silvia

Figure 9. Irradiation set-up for muscle and protein assessment. Subject irradiation set-up for the fast neutron activation analysis of phosphorus and nitrogen. The height, h, of the stepping stool is adjusted so the head is shielded. A whole-body detector is used for subsequent gamma ray counting.
Metodi antropometrici associati alla composizione corporea
Height – Weight Tables

• Developed in 1940’s by INSURANCE companies.
• Based solely on mortality statistics.
  — Fatter people = increased risk of death
• Do not take into account body composition!!!
Body Mass Index

- BMI = Weight (kg) / Height (m) ^ 2
- Desirable
  - Men: 21.9 – 22.4
  - Women: 21.3 – 22.1
- Over weight: 25 - 30
- Obese: >30
Body Mass Index

• BMI’s above 27 associated with ↑ incidence of hypertension, diabetes, & CHD.

• Still used frequently by doctors and researchers.

• Does not take body composition into account either!
Body Mass Index

- Height: 5’10” = 1.77 m
- Weight: 221 lbs = 100.45 kg
- BMI = 32.09
- THIS GUY IS OBESE !!!!
Waist to Hip Ratio

• Indication of the pattern of body fat distribution

• Indicator of the health risks of obesity
  – excess trunk fat - increased risk of hypertension, type 2 diabetes, high cholesterol, CAD, premature death
Waist to Hip Ratio

• Risks increase with increasing ratios
  – very high risk >0.94 young men and 0.82 young women
  – very high risk >1.03 older (60-69 years) men and 0.90 for older women
### Ratings of % Fat (ages 20-29 yr)

<table>
<thead>
<tr>
<th>Rating</th>
<th>Men</th>
<th>Women</th>
</tr>
</thead>
<tbody>
<tr>
<td>Excellent</td>
<td>6-9</td>
<td>10-17</td>
</tr>
<tr>
<td>Good</td>
<td>10-14</td>
<td>17-21</td>
</tr>
<tr>
<td>Acceptable</td>
<td>15-19</td>
<td>21-25</td>
</tr>
<tr>
<td>Too Fat</td>
<td>20-22</td>
<td>27-32</td>
</tr>
<tr>
<td>Obese</td>
<td>&gt;22</td>
<td>&gt;32</td>
</tr>
</tbody>
</table>
Potential Uses on Methods

Found in Literature

• Bod Pod
  – FFM & FM
    • Adults
    • Infant model – tested

• DXA
  – BMC, BMD, FFM, and FM
  – Tissue Distribution

• CT and MRI
  – Site specific tissue analysis
Potential Uses on Methods
Found in Literature

• BIA
  – FFM & FM
    • Adult and Pediatric
  – Dialysis
  – Survival
    • Cancer, peritoneal dialysis, malnutrition, obesity
  – Congestive Heart failure
• MFBIA or BIS
  – FFM, FM, TBW, ECF
  – Pregnancy, HIV+ wasting
Which Method to Use?

• Depends on compartment of interest.
• Availability of techniques.
• Technical training of staff.
• Condition of patient.
• Location where assessment will be done:
  – Laboratory / clinic
  – Field / remote site