Construction of a Family of Tissue Simulating Phantoms for Glucose Determination Using Diffuse Reflectance Near-IR Spectroscopy

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What is a Tissue Phantom?

- **A tissue phantom models one or more properties of an *in-vivo* sample**
  - Allow studies with an in-*vitro* sample to represent an *in-vivo* sample
  - Analogous to computer modeling
- **Useful for:**
  - Time
  - Expense
  - Convenience
  - Understanding (quantification)

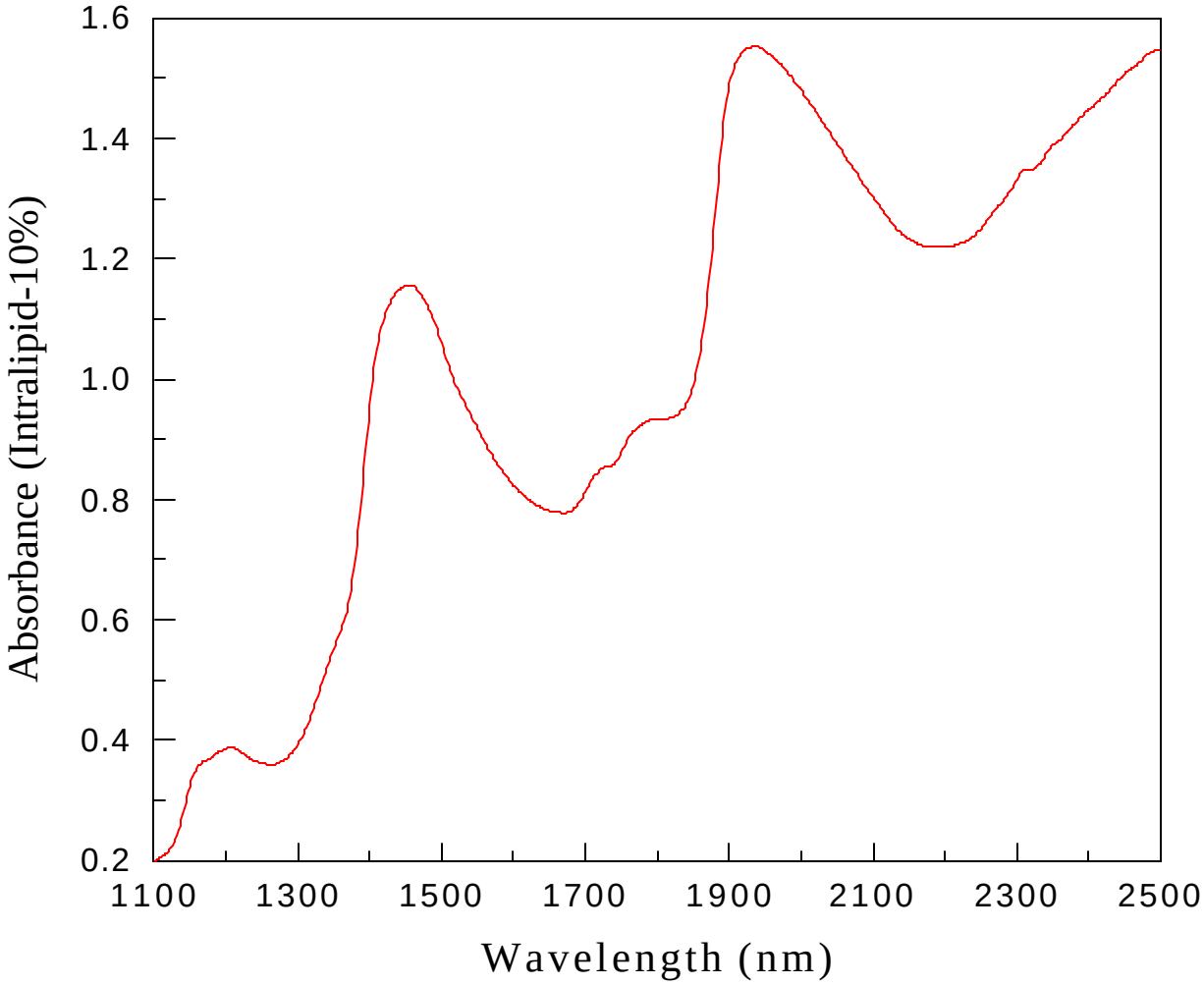
Need for a Tissue Phantom

- **Noninvasive Glucose Studies**
  - **Glucose determination in DR**
    - Combination, 1st Overtone, 2nd Overtone
  - **Performance specification/ testing**
    - Signal to Noise
    - Resolution
    - Wavelength repeatability
    - MCR
    - Clinical costs
  - **Photon Distribution**
    - Total optical pathlength
    - Depth
    - Radial diffusion
  - **Non-homogenous (Layering)**
  - **Utilize as standard (PDS)**

Tissue Phantom Requirements

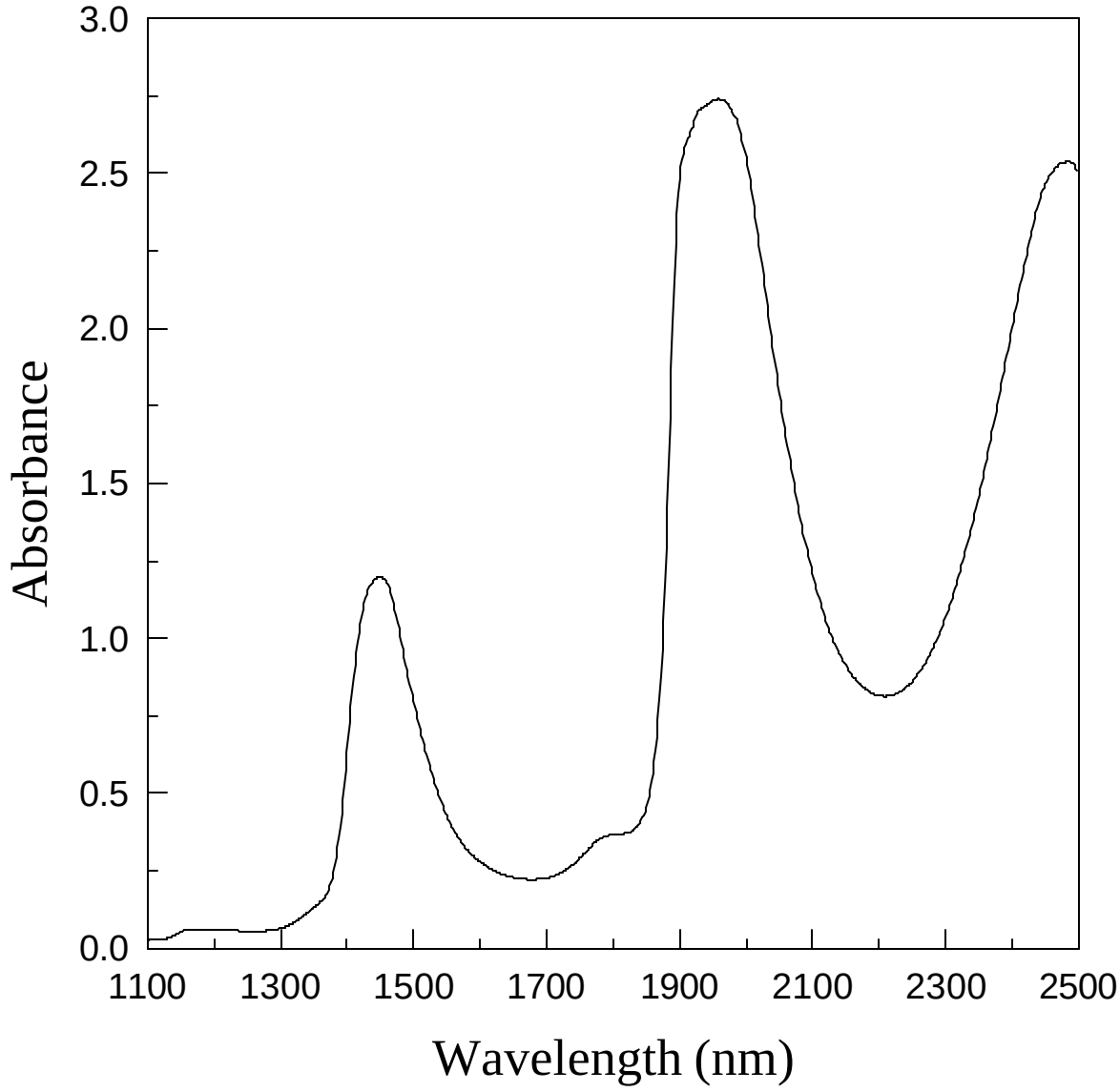
- **Sample constituents**
  - **Presence of major near-IR skin absorbers**
    - Physiological concentration
  - **Absence of extraneous absorbers**
- **Freedom of experimental design**
  - **Quantitative**
  - **Flexibility sample constituent**
    - Presence
    - Concentrations
- **Diffuse reflectance sampling**
  - **No specular reflectance**
  - **μ_a, μ'_s approximating skin**
- **Usable**
  - **Ease of preparation**
  - **Stability**
  - **Cost effective**

Intra-lipid: Commercial



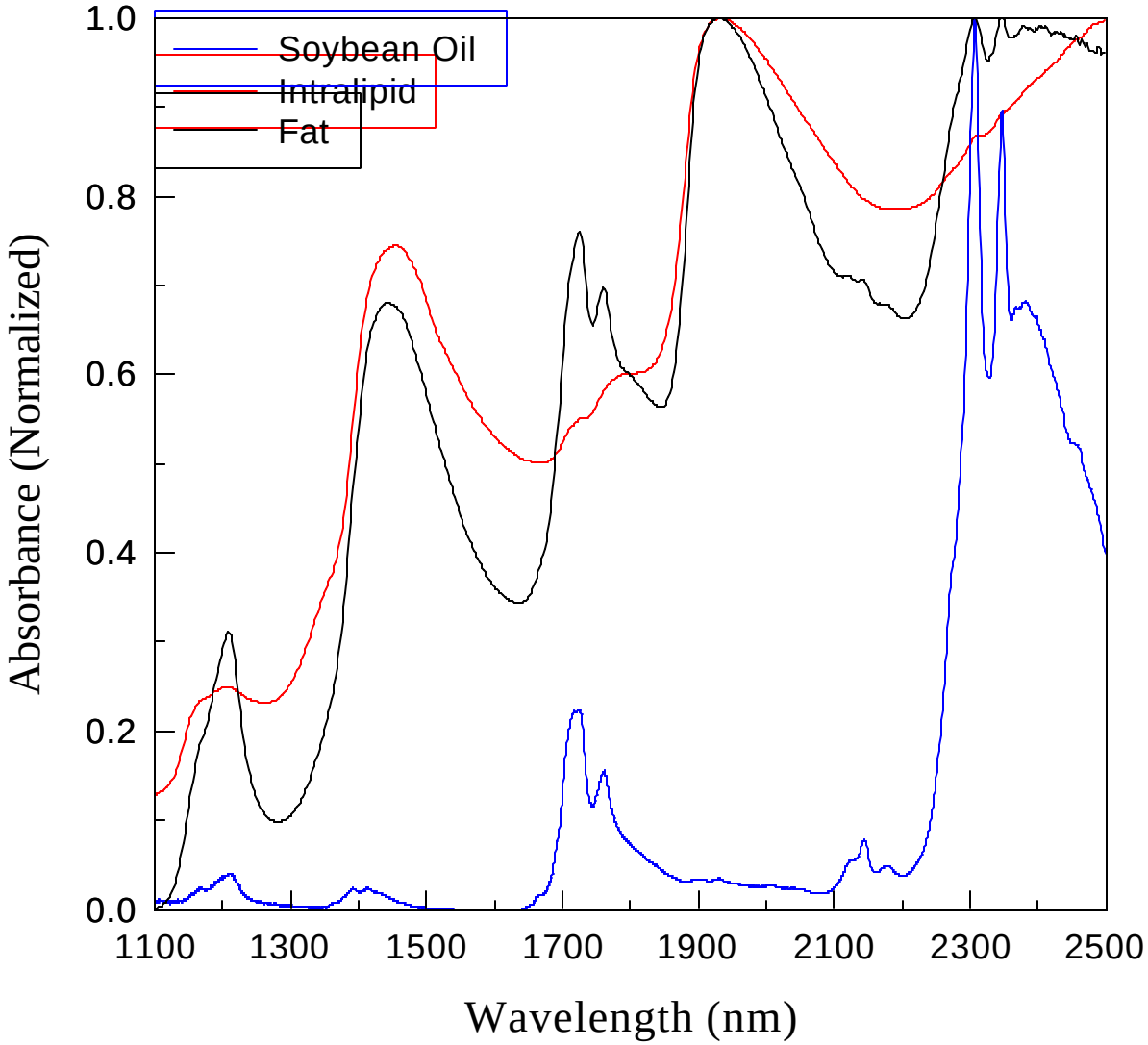
- **Intra-lipid**
  - Fat emulsion used clinically as intravenous nutrient
  - Composed of water, soybean oil, lecithin and glycerol
- **Starting place for tissue phantom**
  - Utilized in visible and short wavelength near-IR

Intra-lipid Components: Water



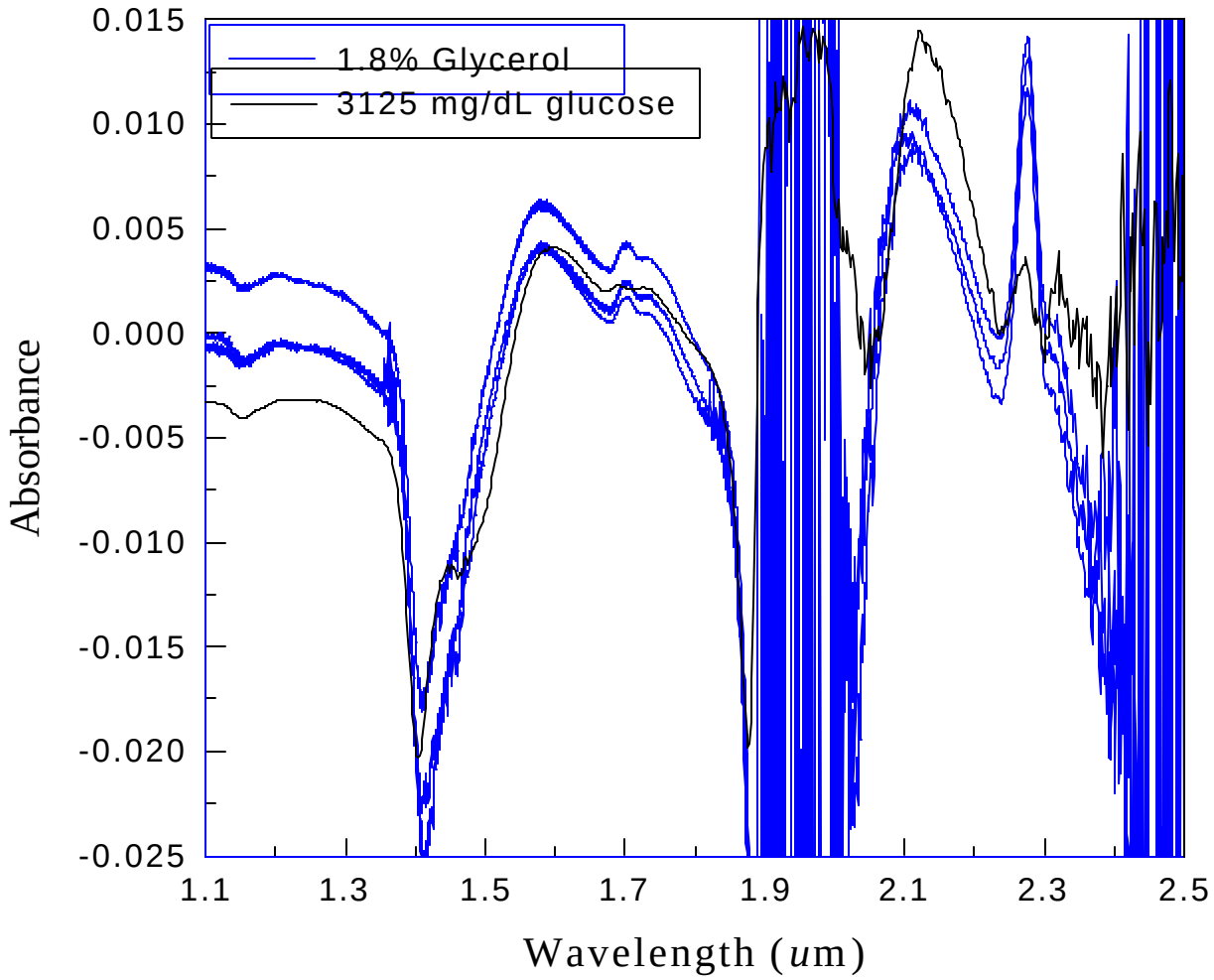
- **Major component of skin**
- **Excellent base absorber for phantom**

Intra-lipid Components: Soybean Oil



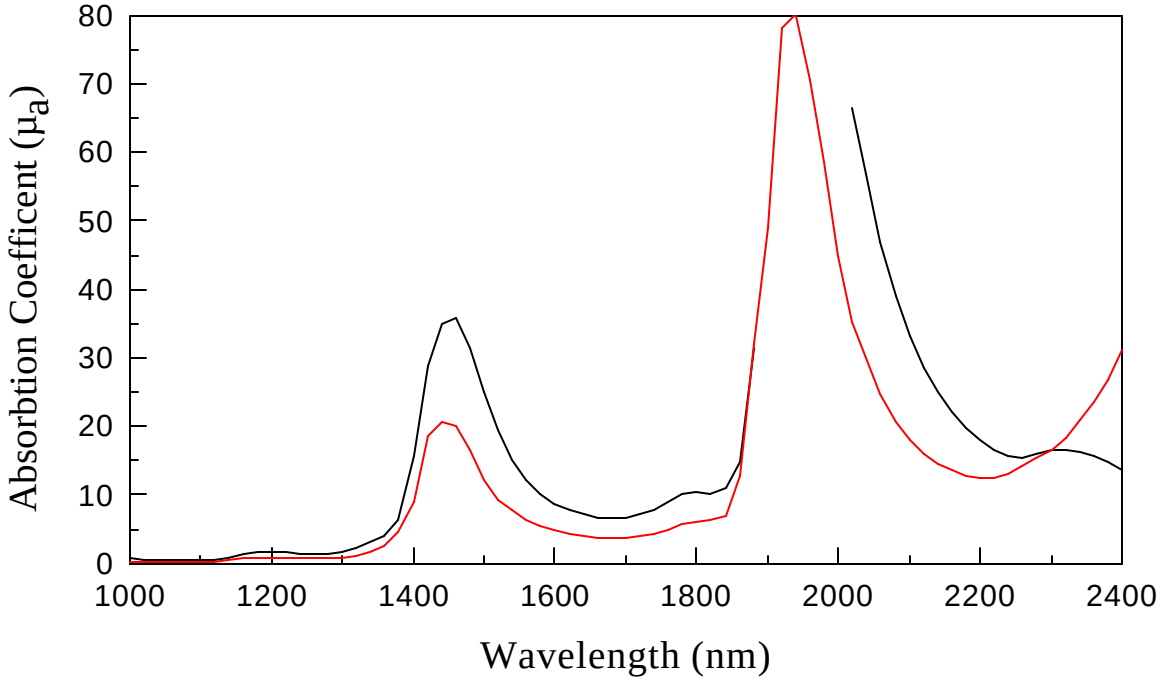
- **Large component of skin**
  - Absorbance of fat and soybean are comparable
  - Lecithin: phosphatidyl choline
    - Same as long chain oil
- **Excellent base scatterer for phantom**

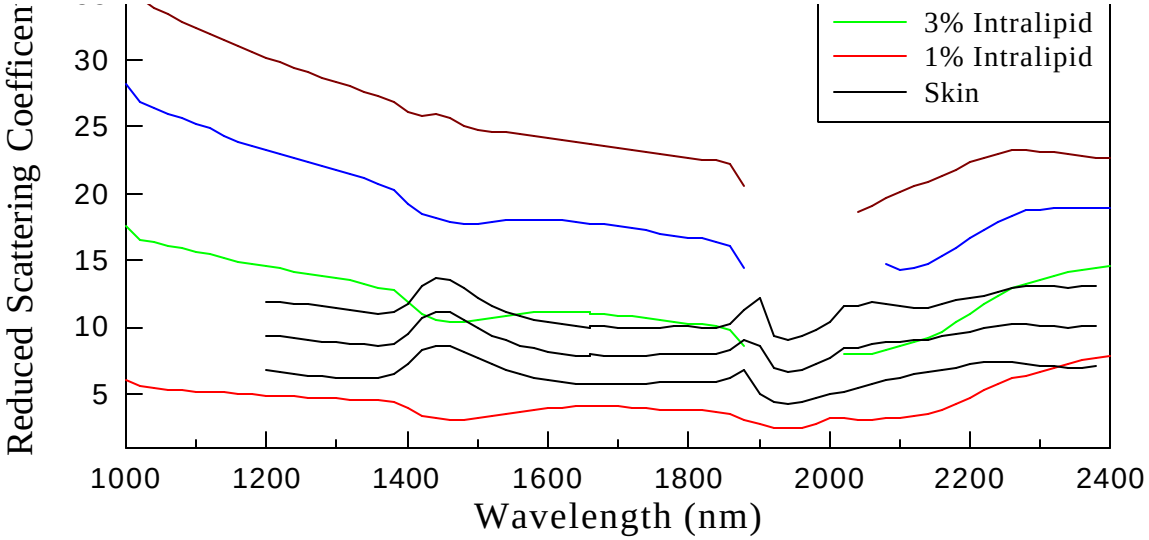
Intra-lipid Components: Glycerol



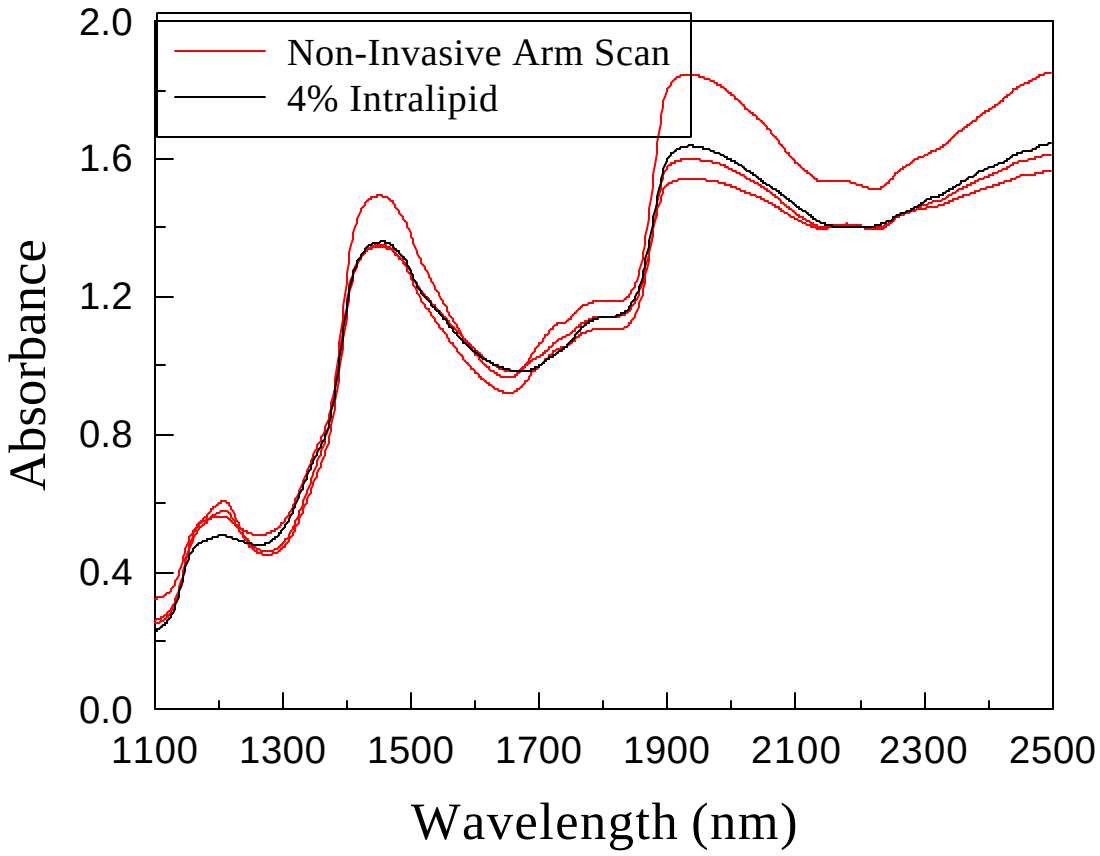
- **Minor component of skin**
  - Interferes with glucose
  - Present in Intra-lipid as nutrient
  - Present in body at low concentration
- **Not significant source of scattering**

Absorbance and Scattering Coefficients





Intra-Serum vs. Arm

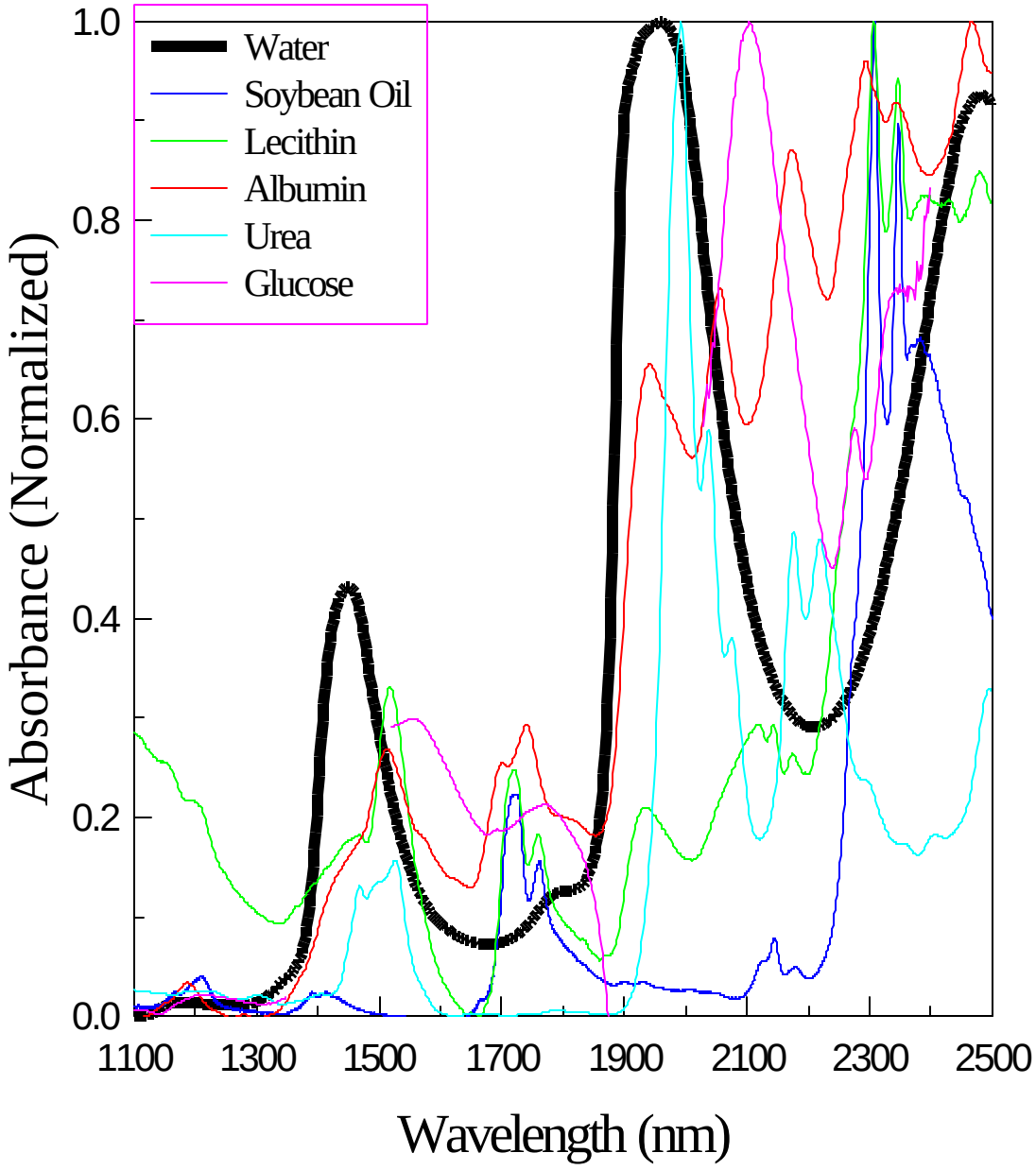


- **Major absorbance bands present**
- **Scattering and total returned light approximates arm**

Creation of Intra-Serum

- **Utilize modified Intra-lipid base**
  - Water
  - Soybean oil (castor oil)
  - No glycerol
- **Serum components**
  - Albumin
  - Urea
  - Glucose
  - Readily expandable
    - Globulin
    - Salts
      - Na^+, K^+, Cl^-, …
- **Intra-gel**
  - Crosslink with collagen
- **Gravimetric preparation**

Intra-Serum Component Spectra



Experimental Design

Objective: DR Glucose in Near-IR all 3 regions

- **Time correlation**
- **Sample constituent correlation** (Requires multiple constituents)
  - Water
  - Soybean Oil
  - Lecithin
  - Albumin
  - Urea
  - Glucose
- **128 sub-cubes of 4-D space**
- **Physiological concentrations**

Constituent	Low concentration	High concentration
Intralipid	3 %	8 %
Albumin	500 mg/dL	2000 mg/dL
Urea	5 mg/dL	70 mg/dL
Glucose	40 mg/dL	600 mg/dL

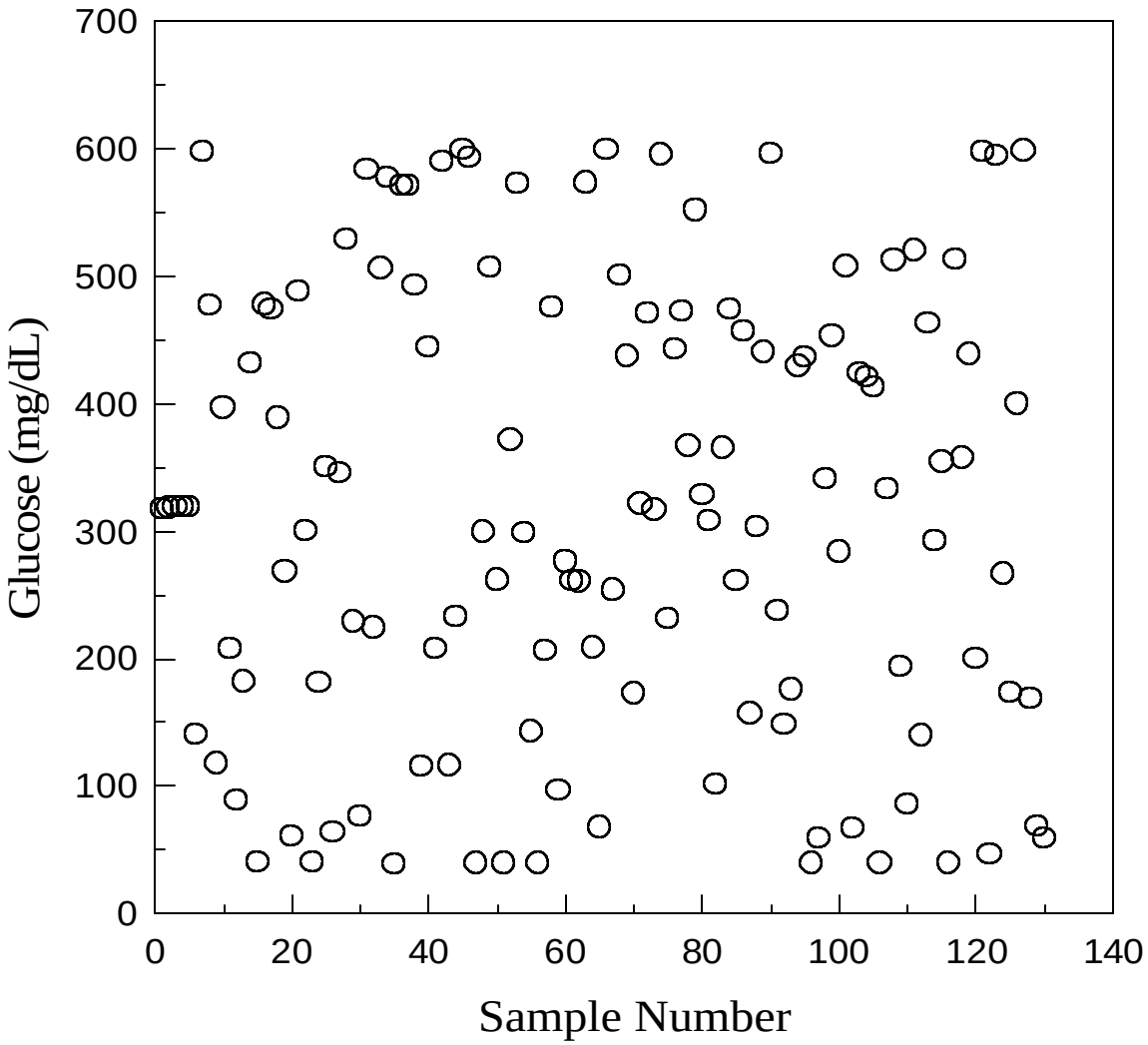
Instrumentation

- **Modified Nicolet 860**
  - **High resolution**
  - **S/N**
    - Optical pathlength approximating skin
    - µA noise
- **Auxiliary external source**
  - **ARA Engineering & Oriel**
- **Fiber-optic interface**
  - **Fiber stationary: two configurations**
  - **Bundle 261 Excitation and 72 collection in 57x9 array**
- **Sample holder**
  - **Stirred**
  - **Depth controlled**
  - **no specular reflectance**
- **Components**
  - **1.1 and 1.45 µm LP filter**
    - Study #1: 2nd overtone
    - Study #2: 1st overtone and combination band
  - **2.6 µm InGaAs**

Correlation & Multivariate Analysis

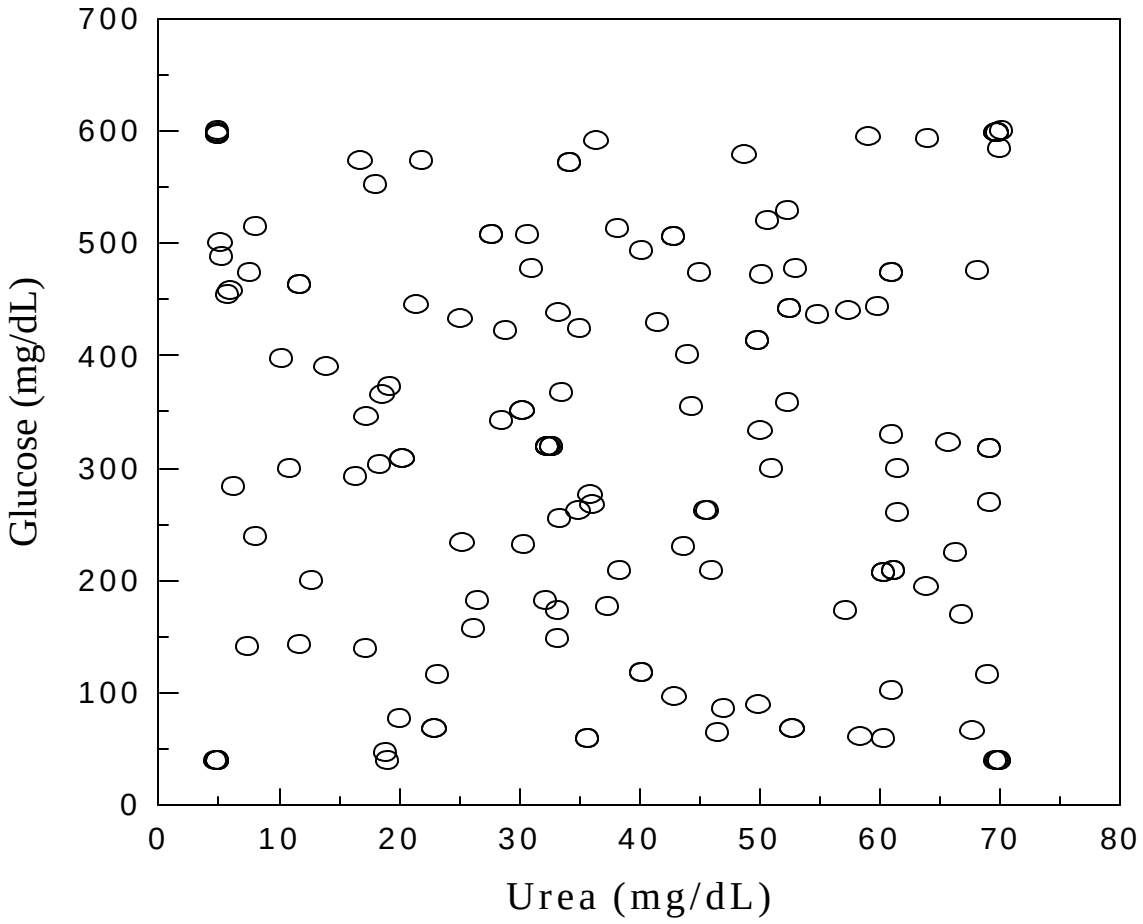
- **Instrument correlation**
  - **Address with experimental design**
    - Time
  - **Address with reference**
    - Temperature
    - Instrument drift
- **Sample constituent correlation**
  - **Address with experimental design**
  - **Address with analysis**

Correlation: Time



- **Removal of all time correlation**
- **Collection and prediction on references also demonstrates no instrument correlation**

Correlation: Analyte/Analyte



- Removal of all analyte/analyte correlation
- Urea/glucose concentration correlation
  - Concentration Correlation = -0.07

Data Preprocessing

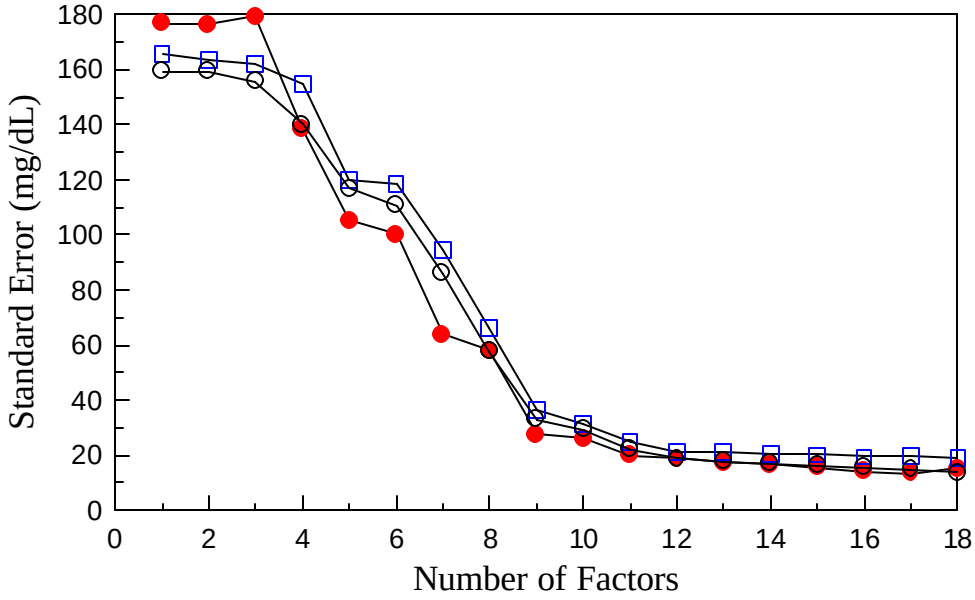
- **Goal:** Demonstration of glucose determination in DR mode
  - Therefore, simple preprocessing
- **Preprocessing**
  - Absorbance
  - Multiplicative Scatter Correction
  - Savitsky-Golay smoothing
  - Range selection
    - 2nd Overtone (1025 to 1400 nm)
    - 1st Overtone (1500 to 1840 nm)
    - Combination Band (2025 to 2375 nm)

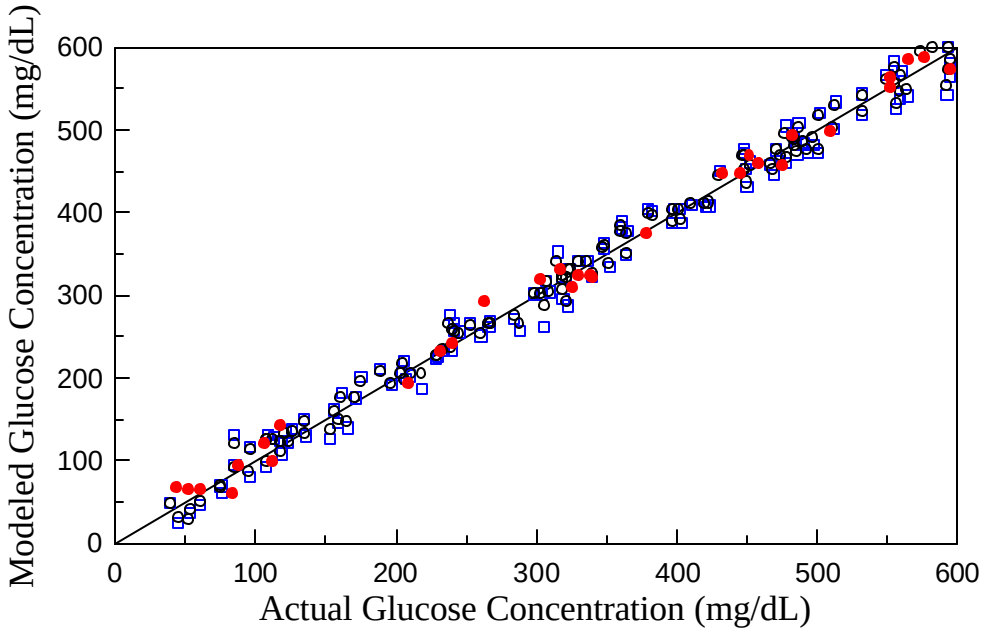
Data Analysis

- Data Sets
  - Calibration / Monitoring
    - First 75% of samples
  - Prediction Set:
    - Forward looking 25% of samples
- PLS Analysis
- Reference Analysis
  - Identical processing on reference spectra
  - SEP equal to 1 SD of glucose concentrations
- Results

Spectral Range	SEC	SEP	# of PLS Factors
2nd Overtone	20.8	40.0	22
1st Overtone	14.9	13.5	17
Combination	29.1	29.6	13
All 3 Regions	8.83	11.7	22

Results: 1st Overtone Region





Phantom Limitations

- **Water concentration**
  - Intra-gel
- **Layering** (non-homogeneous)
  - Intra-gel slabs
- **Scattering coefficient**
  - Castor oil
- **Stability**
- **Sample preparation**
  - Time
  - Cost

Conclusions

- **Developed Tissue Phantom**
  - Presence of near-IR skin absorbers
    - Absence of extraneous absorbers
  - Optically similar to skin
  - Freedom in experimental design
    - Quantitative
    - Flexible components
    - Flexible concentration
  - Usable
    - Cost
    - Preparation time
- **Demonstration of glucose determination in diffuse reflectance mode in near-IR region**
  - Physiological concentrations
  - No correlation with time
  - No correlation between components

Future Directions

- **Tighter match to skin**
  - **Castor oil:**
  - **Intra-gel:** crosslink with collagen
    - Decrease water concentration
  - **Layers**
    - Stratum corneum
    - Epidermis
    - Dermis
- Optical Properties
  - Total optical pathlength
  - Depth of penetration
  - Radial diffusion
- **Instrument Specification**
  - S/N
  - Resolution
  - Wavelength axis

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