Turbid tissue optics III: Instrumentation and measurements

Andrew Berger

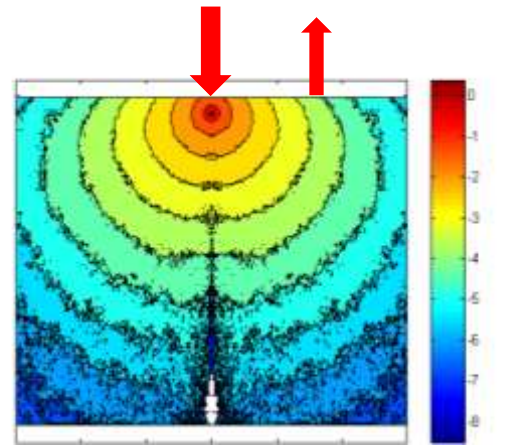
Abbe lecture #4

21.01.2014

Roadmap from last time

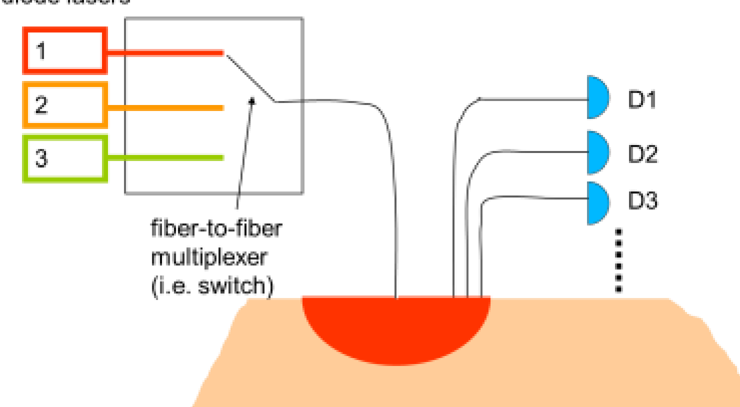
review of basic concepts from last time

the Virtual Tissue Simulator



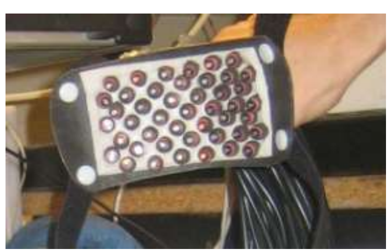
reflectance measurements: three types

- steady-state
- pulsed
- sinusoidally-modulated *("frequency domain")*



instrument design considerations

various applications



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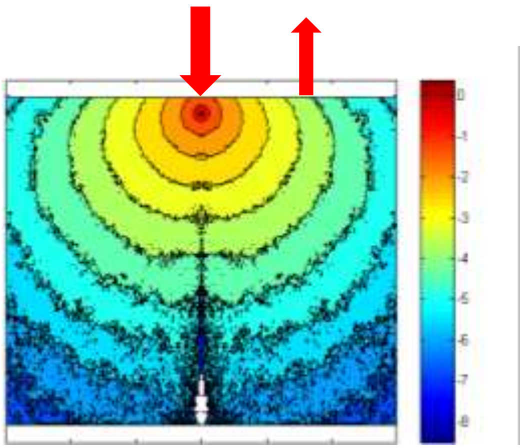

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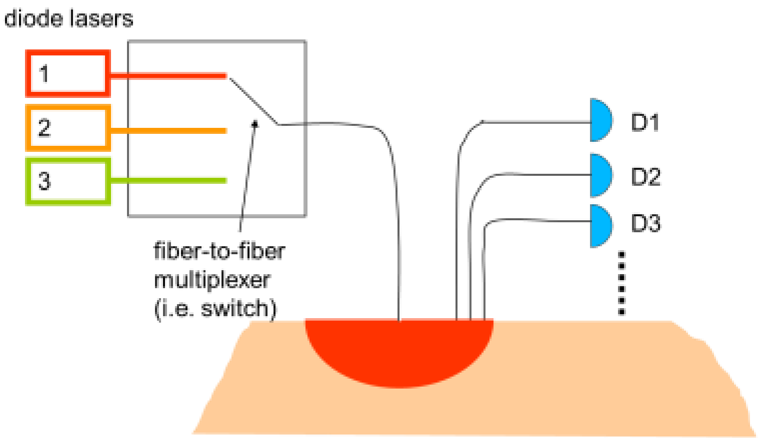

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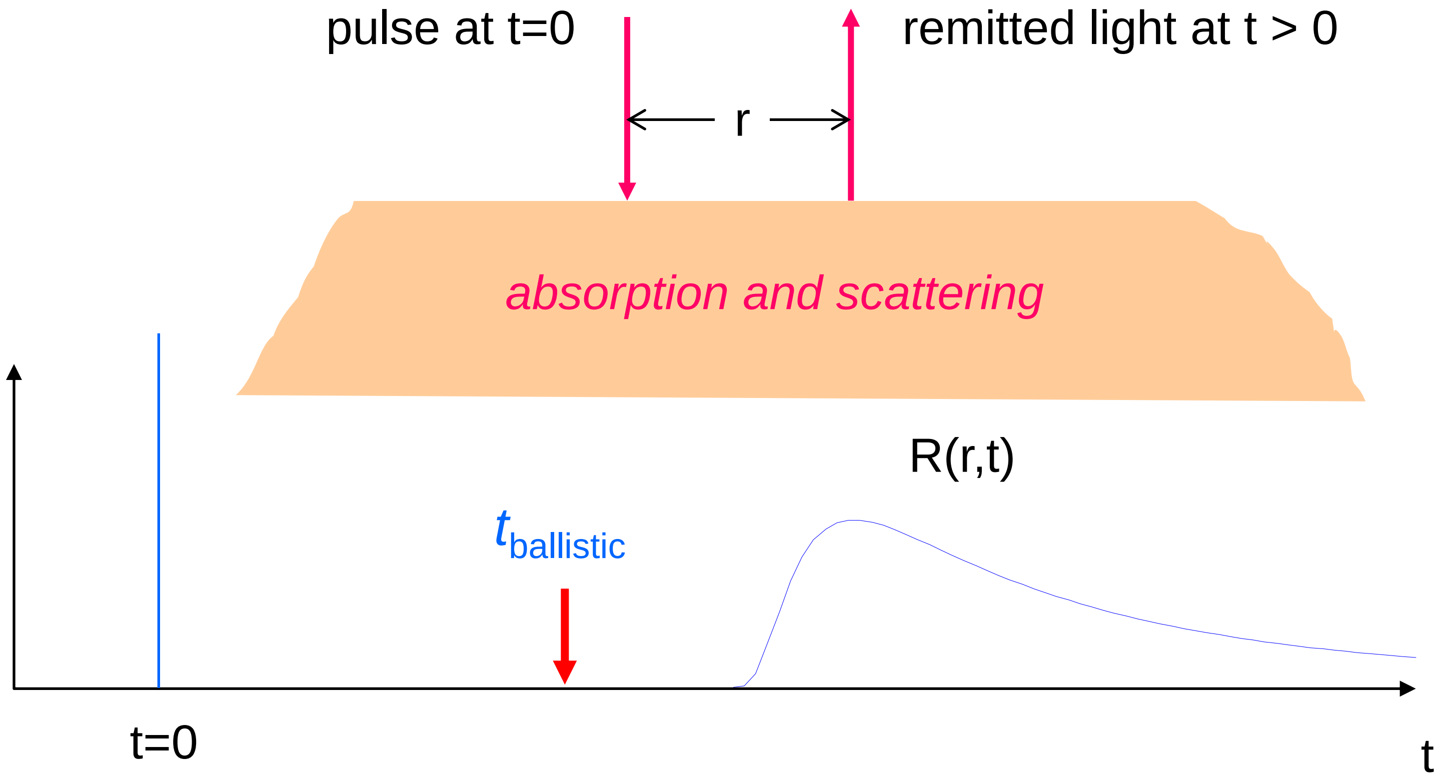
Time-resolved measurements



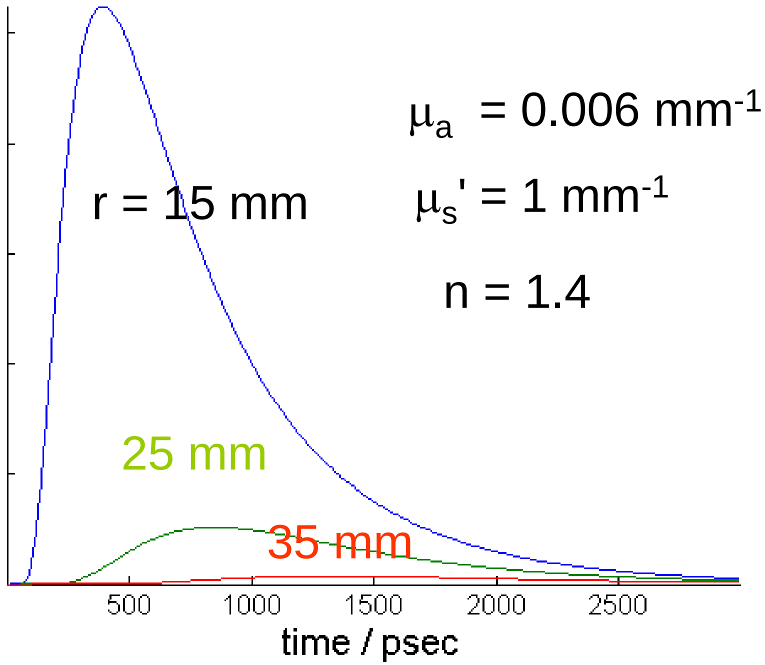
Infinite geometry: $$\Phi(\mathbf{r}, t) = c(4\pi Dct)^{-3/2} \exp\left(-\frac{r^2}{4Dct} - \mu_a ct\right)$$

Time-resolved measurements

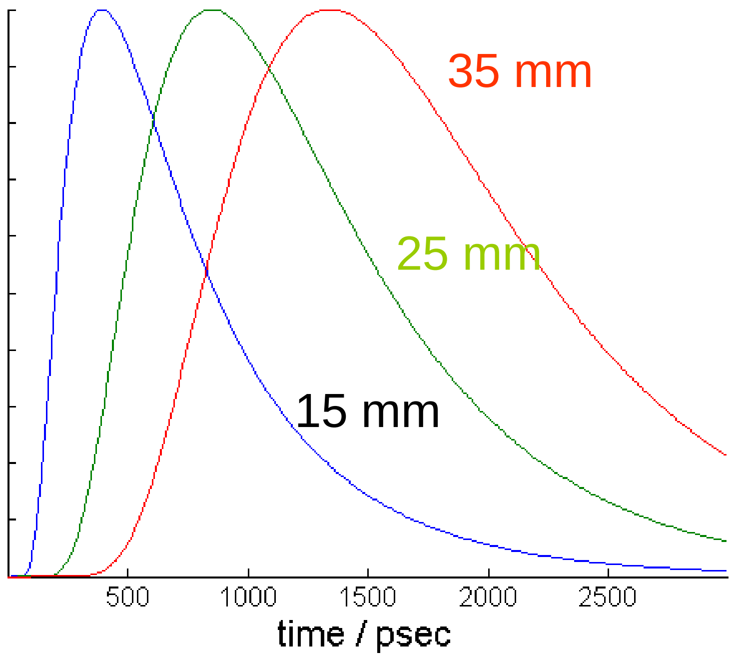
Infinite geometry: $$\Phi(\mathbf{r},t) = c(4\pi Dct)^{-3/2} \exp\left(-\frac{r^2}{4Dct} - \mu_a ct\right)$$

(similar curves for semi-infinite reflectance)

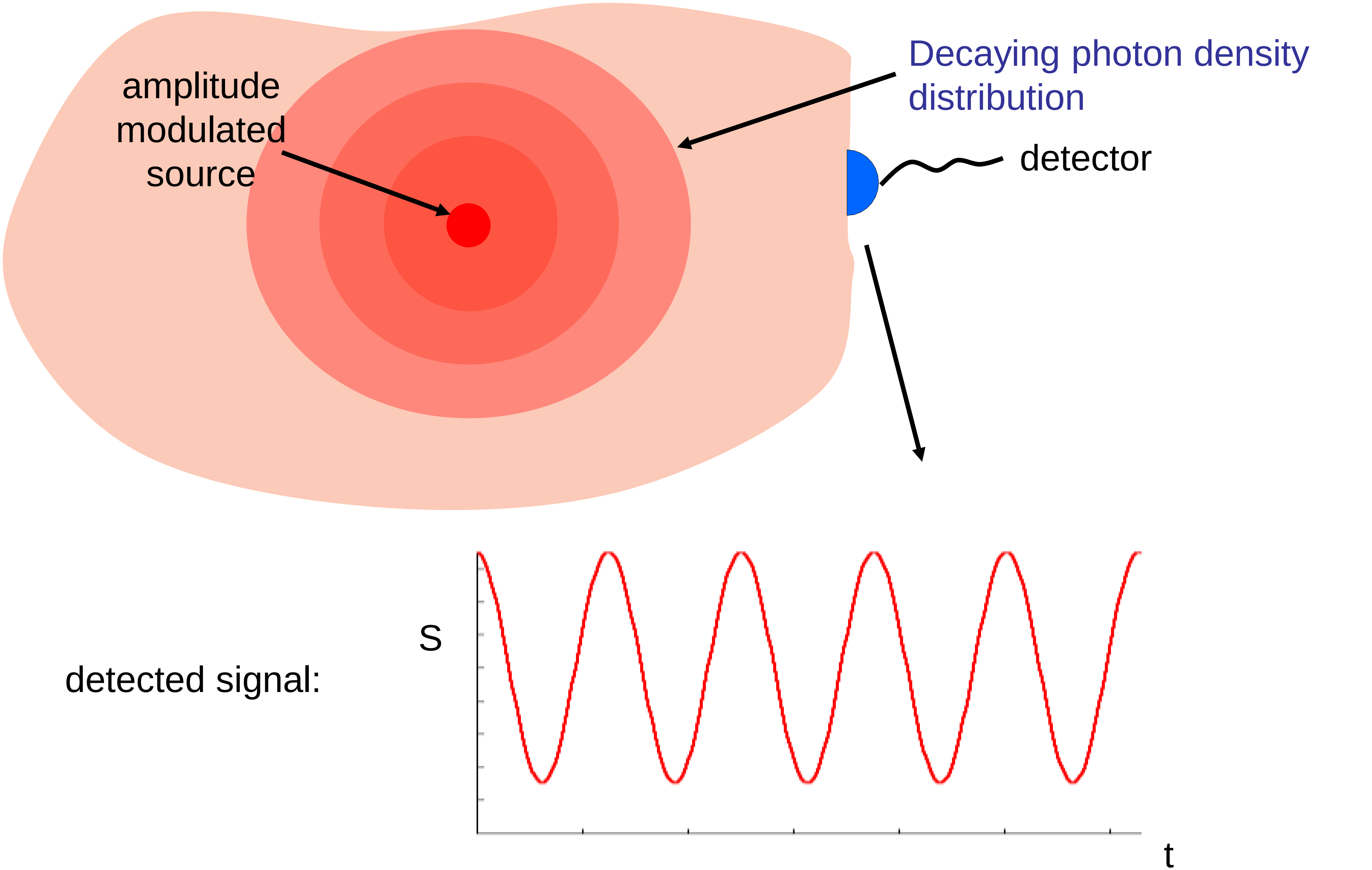
different source-detector separations



normalized



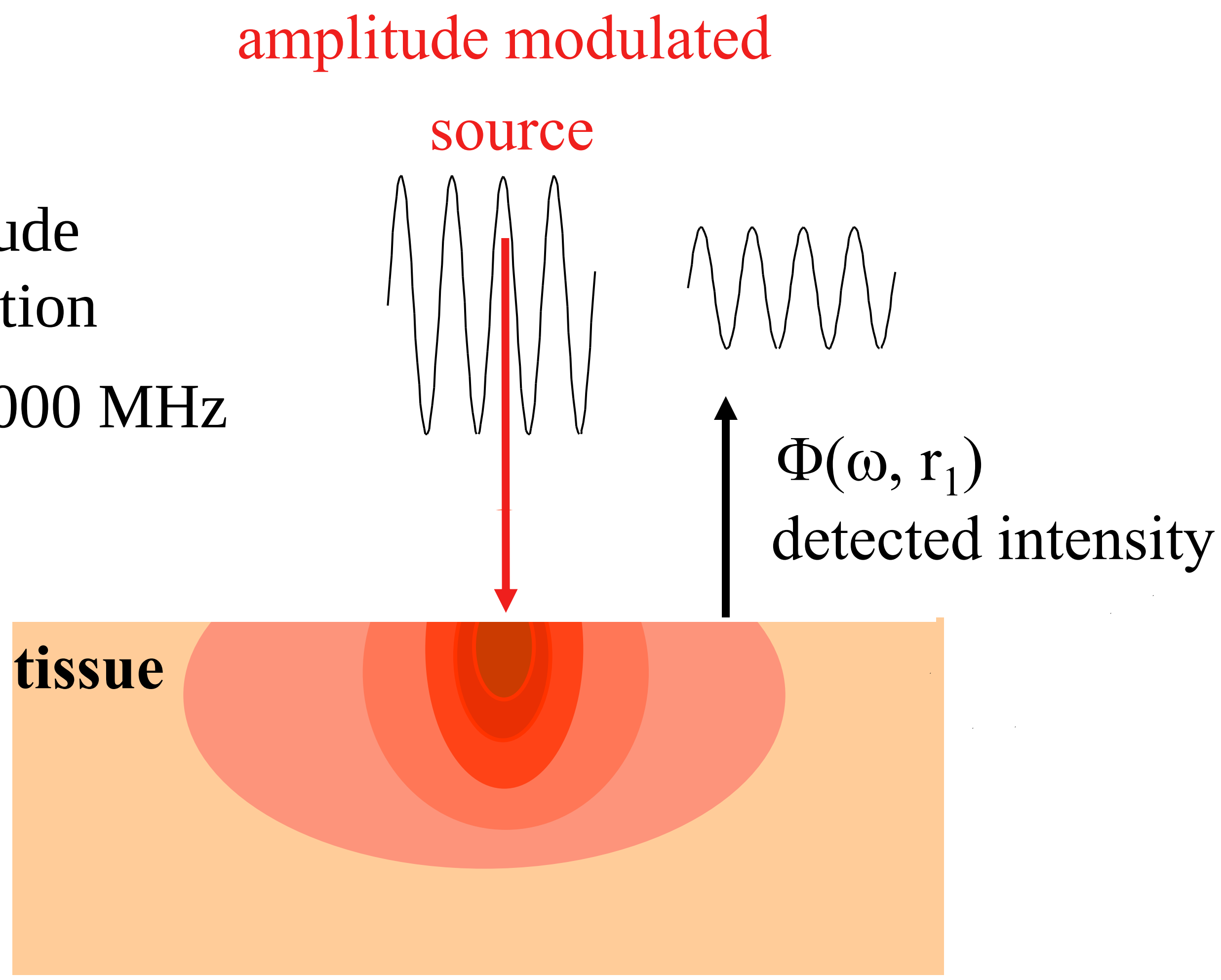
Frequency domain diffusion



Frequency domain diffusion

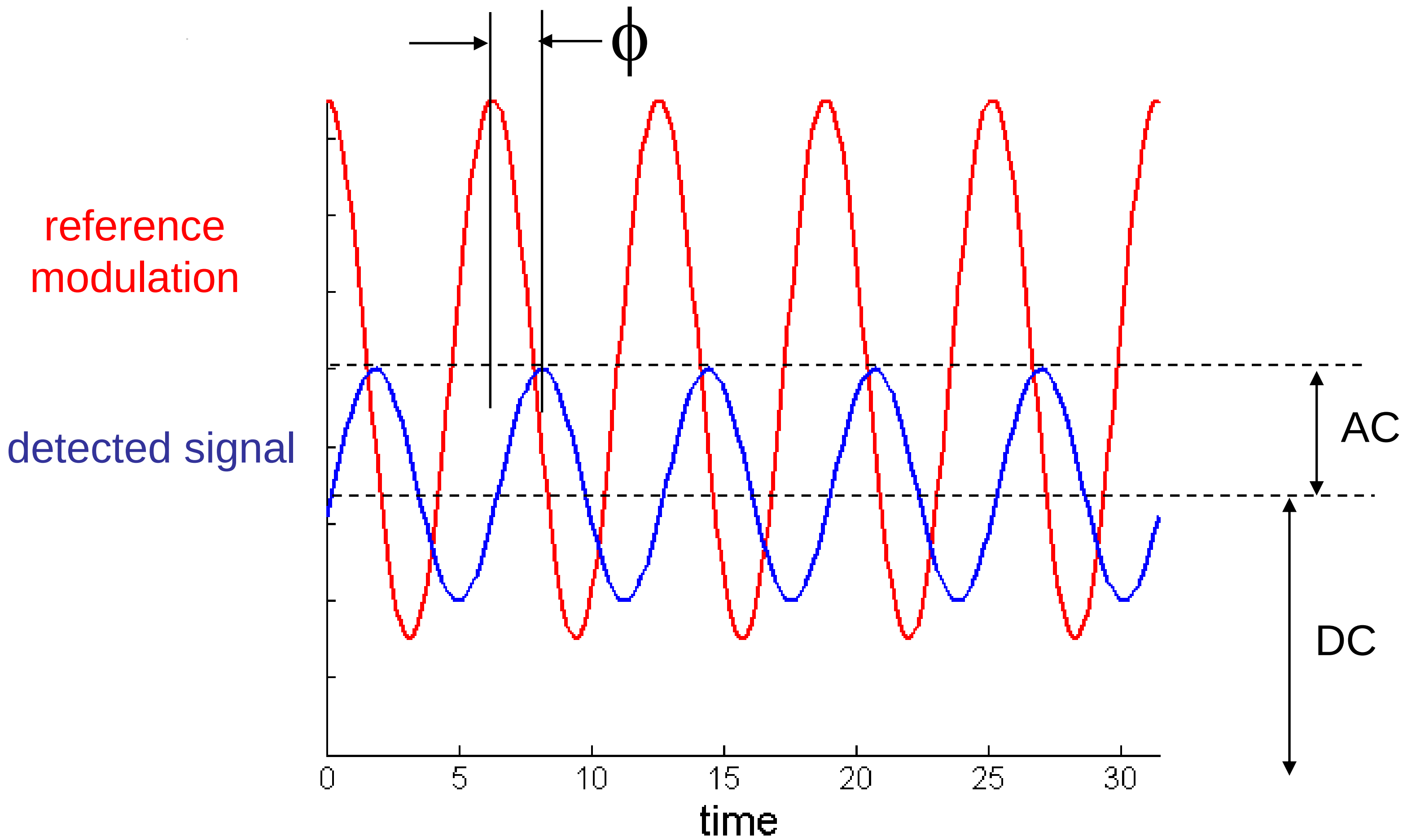
Amplitude modulation

50 MHz - 1000 MHz

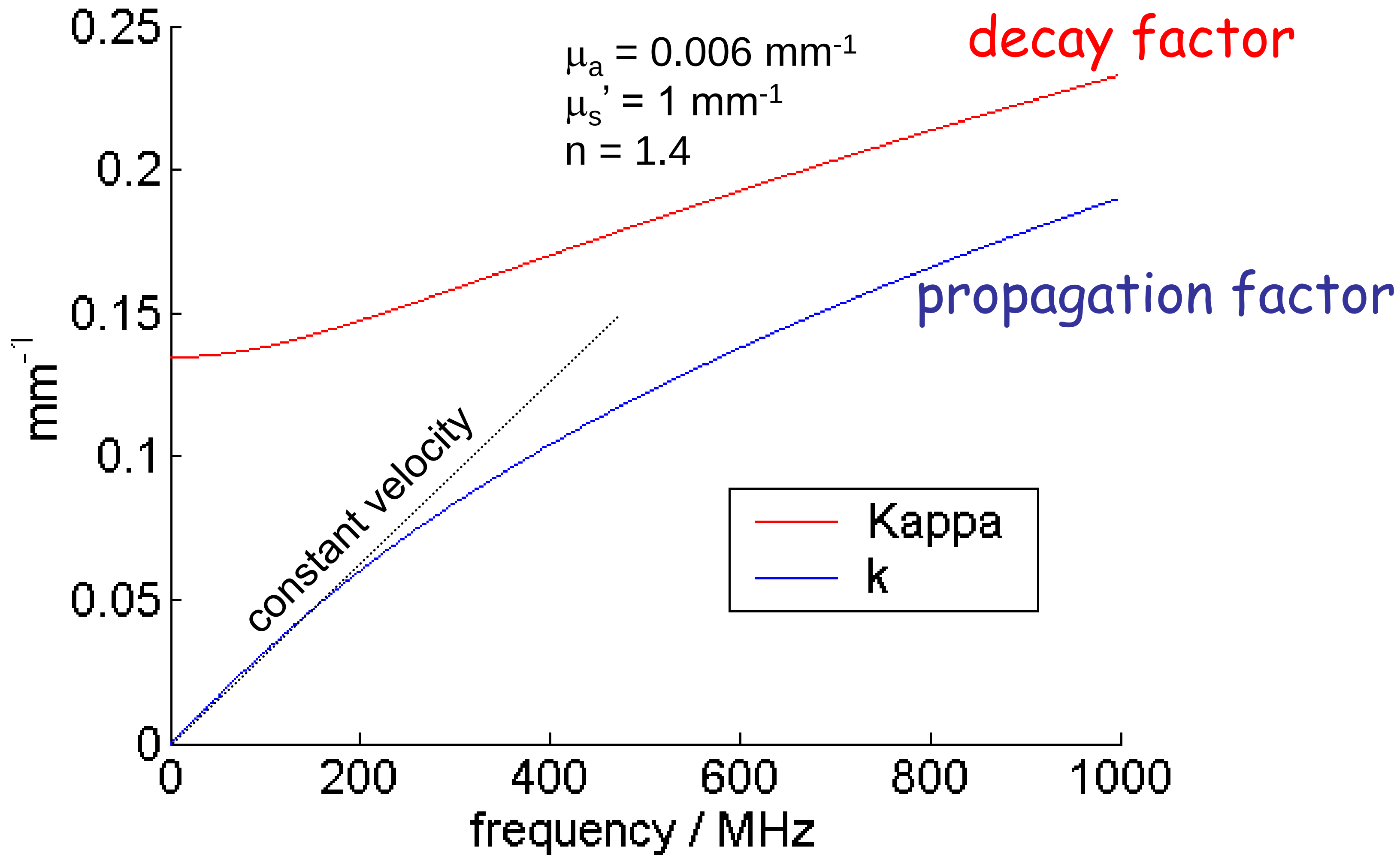


Frequency-resolved

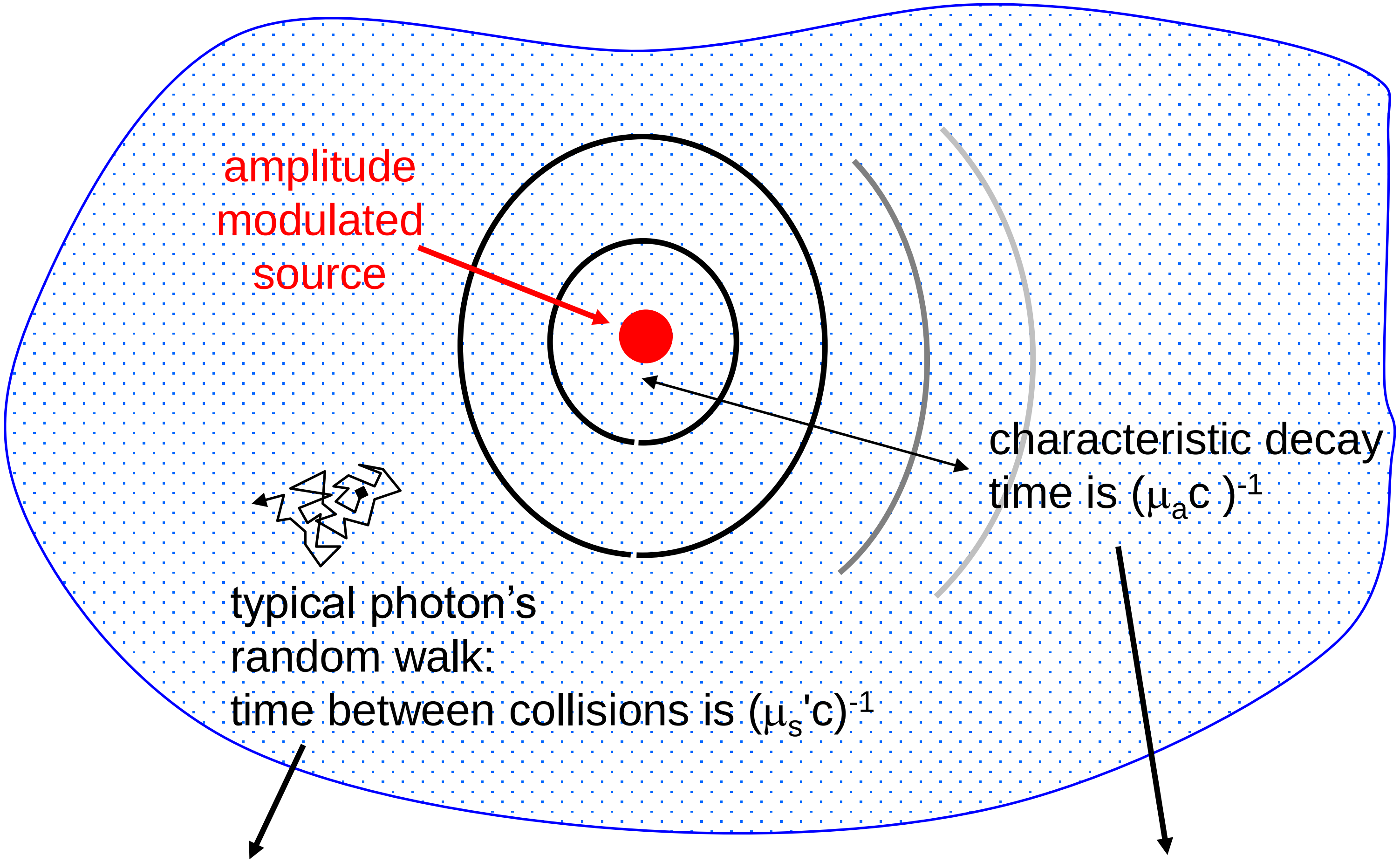
The observables:



Frequency-dependent wave properties: i.e, dispersion!



Validity of photon density wave picture



if photon scatters only once during a source modulation cycle, diffusion model does not approximate reality (>10 GHz)

if launched photons "survive" for less than an oscillation cycle, AC effects are no different from DC effects (<10 MHz)

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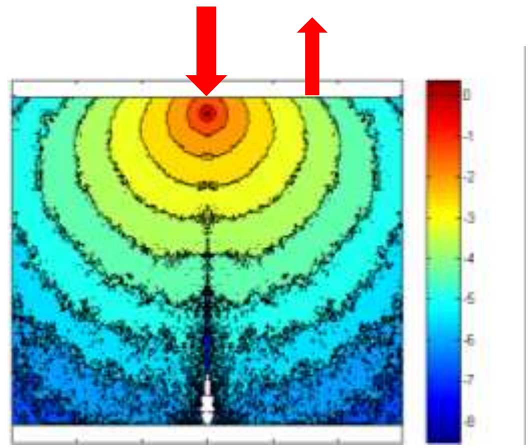

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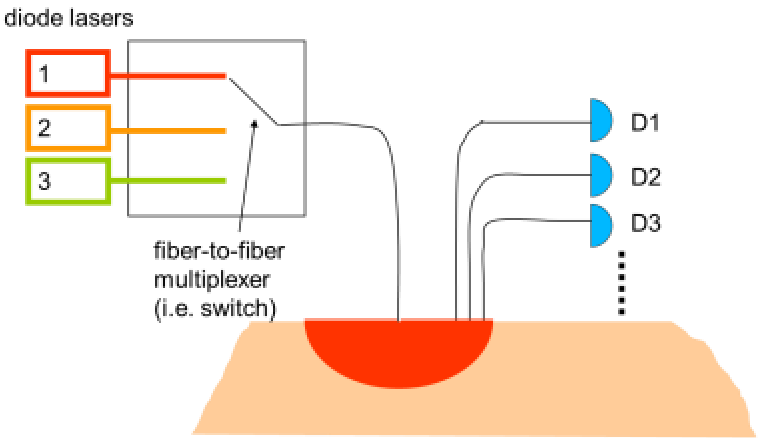
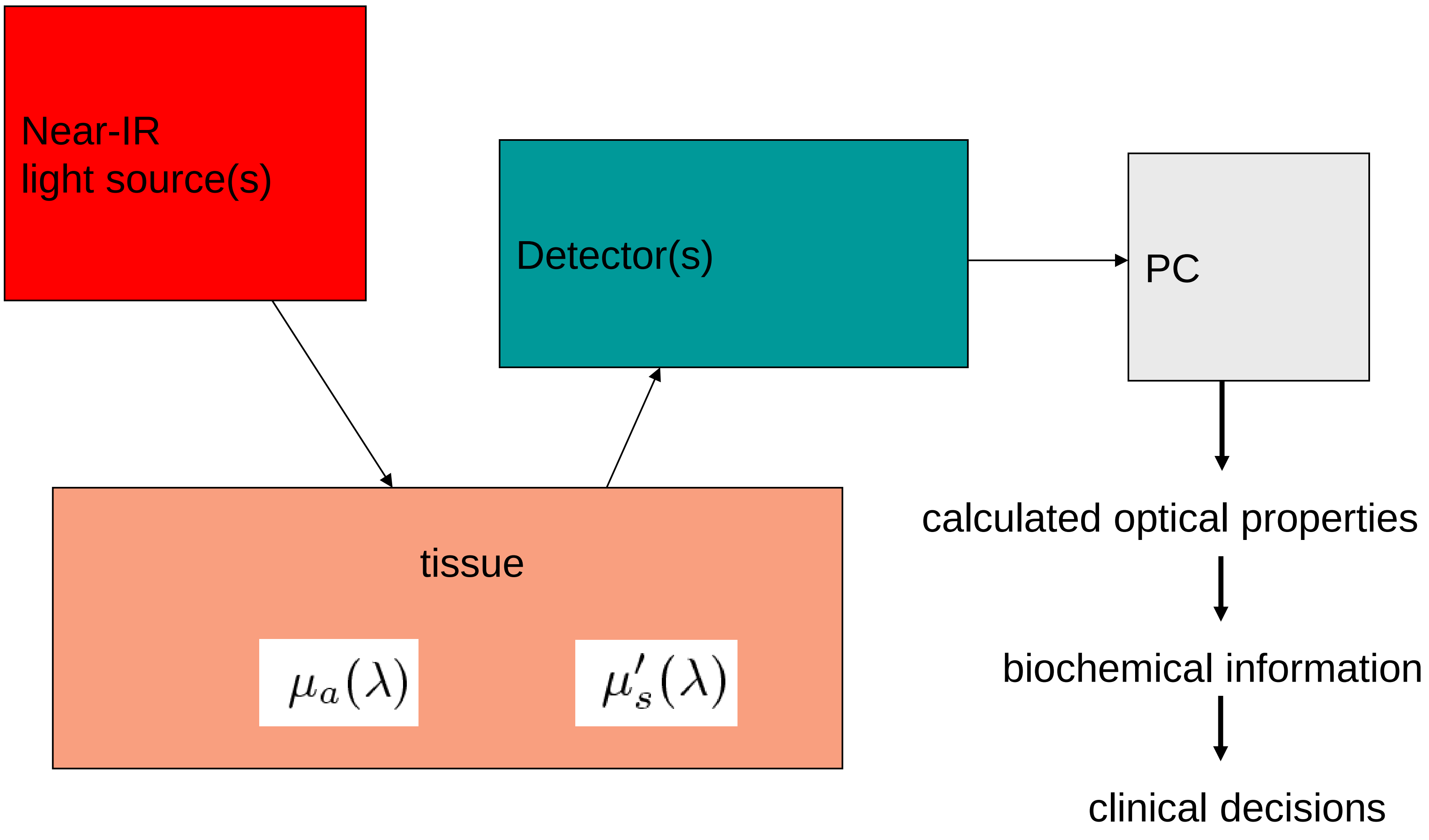


various applications

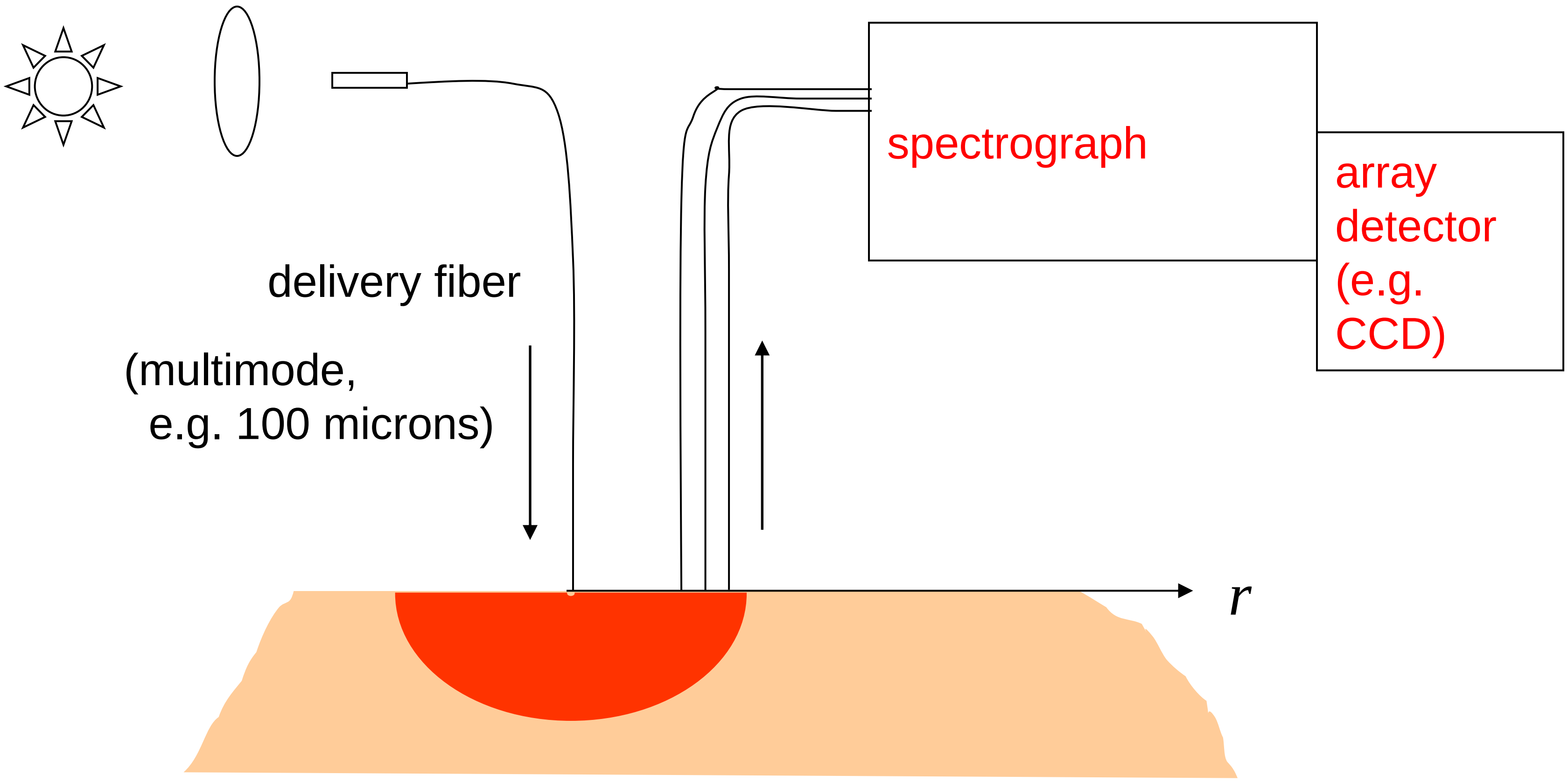
Basic instrumentation for all cases



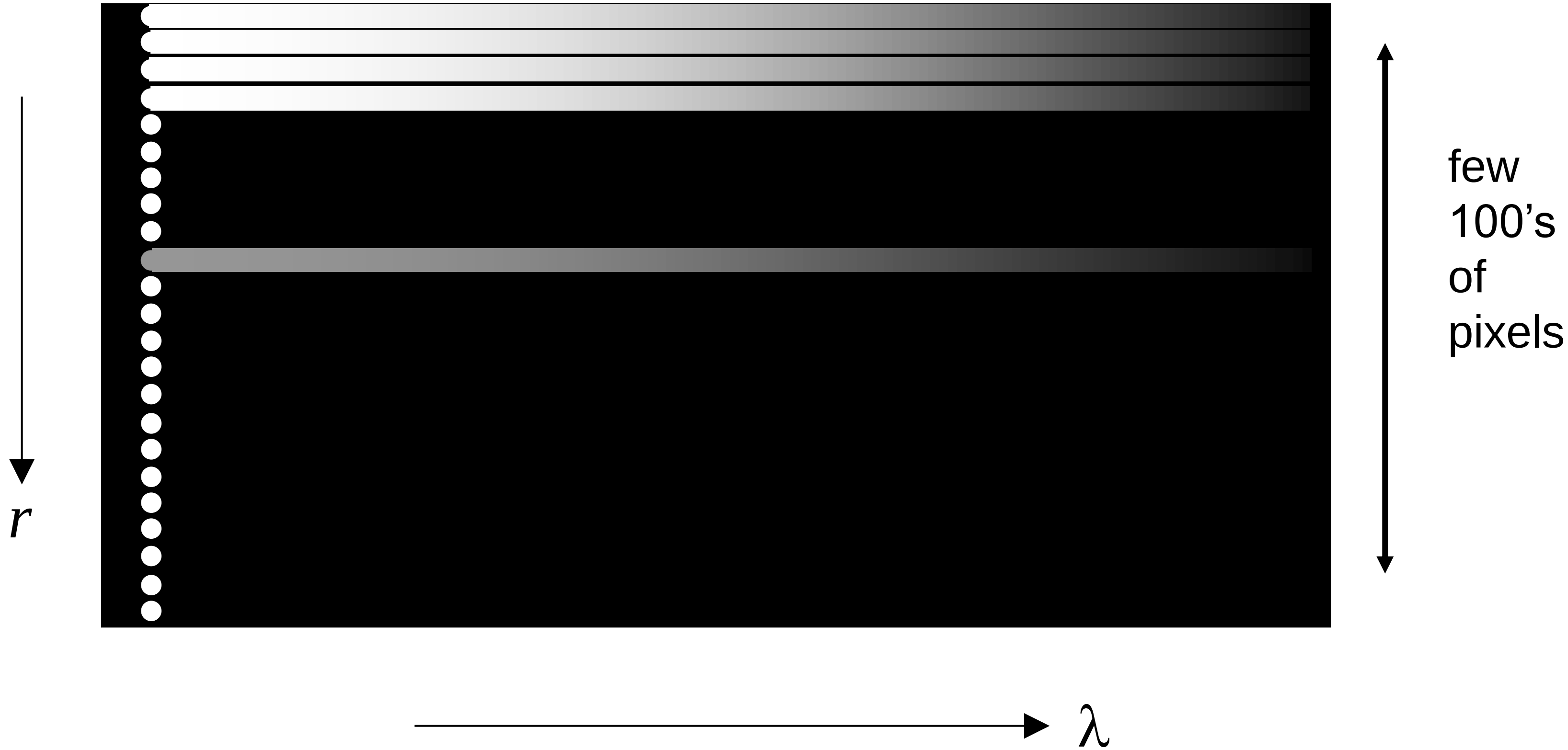
Steady state (“CW”) + spectrometer

broadband source (lamp)

~250 μW/10nm



Spectrographic CCD display

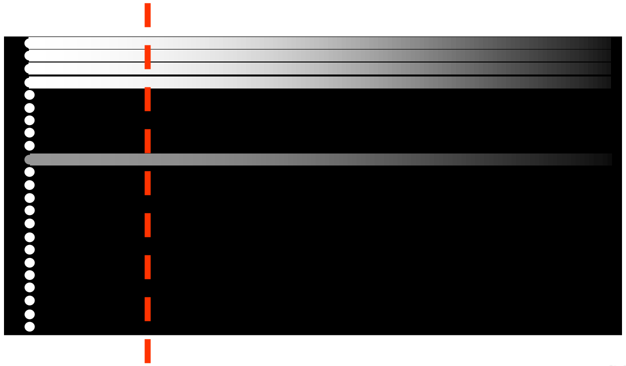


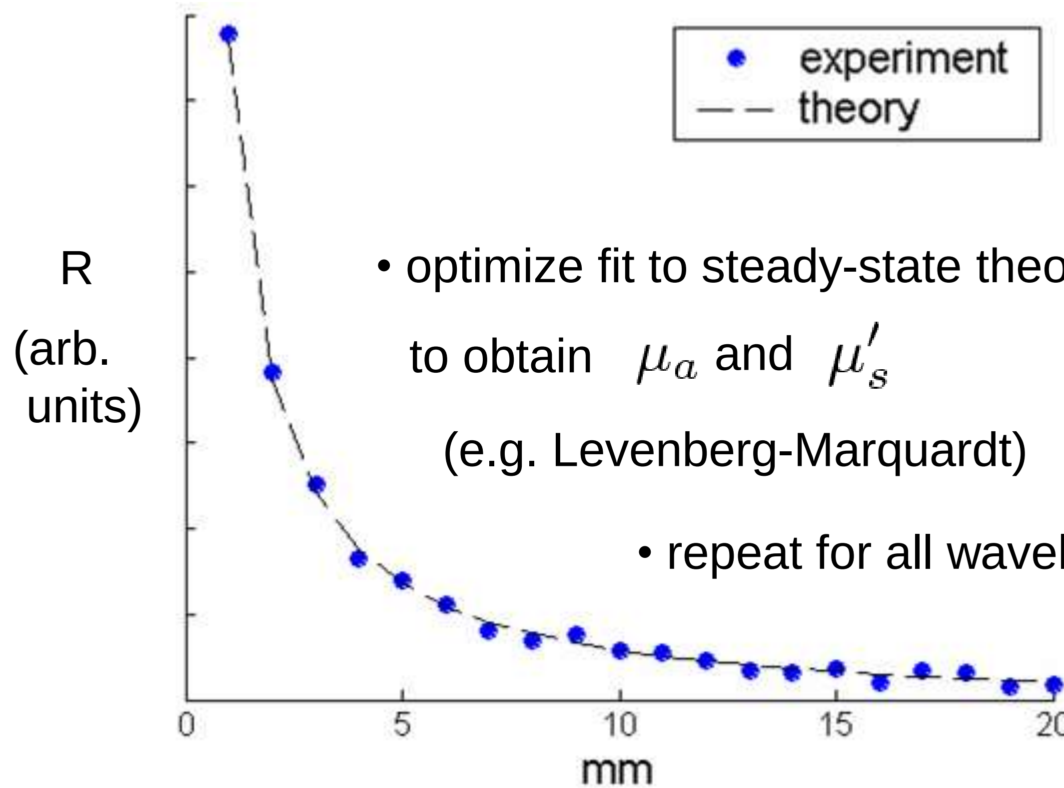
$$\text{fiber image:} \quad \frac{150\,\mu\text{m}}{\text{fiber}} \cdot \frac{1\,\text{pixel}}{25\,\mu\text{m}} = 6\,\text{pixels}/\text{fiber}$$

integration time: 10's of seconds

calibration: shine equal light into all channels

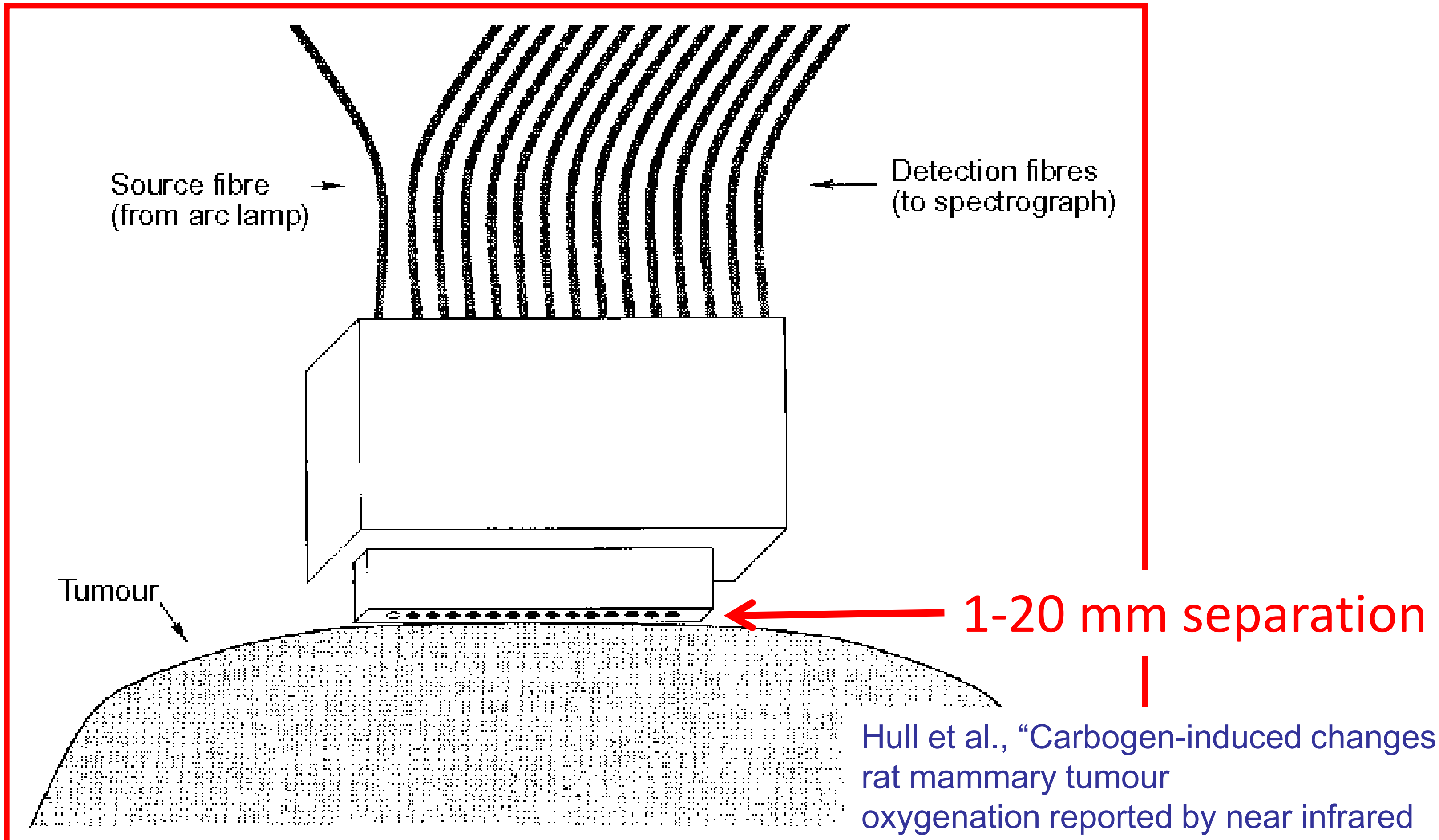
Steady state reflectance data





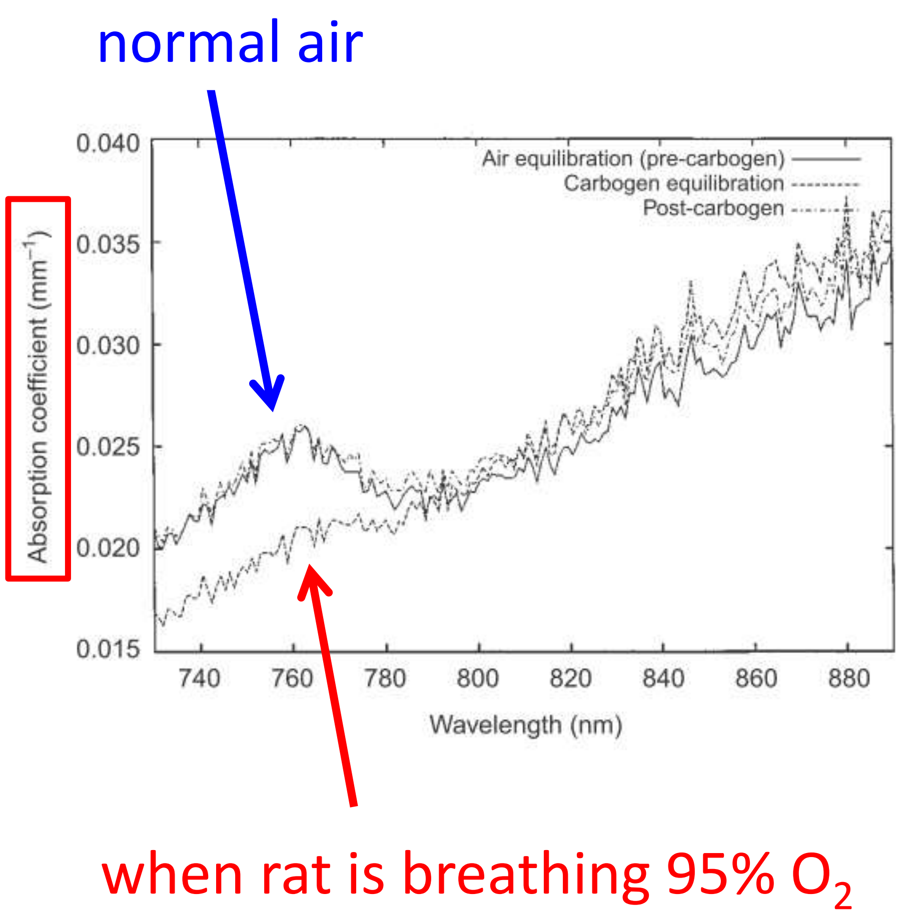
- optimize fit to steady-state theory to obtain μ_a and μ_s' (e.g. Levenberg-Marquardt)
- repeat for all wavelengths

Linear probe for *in vivo* diffuse reflectance spectroscopy



Hull et al., "Carbogen-induced changes in rat mammary tumour oxygenation reported by near infrared spectroscopy," *Br. J. Cancer*, **79**(11/12), 1709-1716 (1999).

Seeing blood oxygenation change



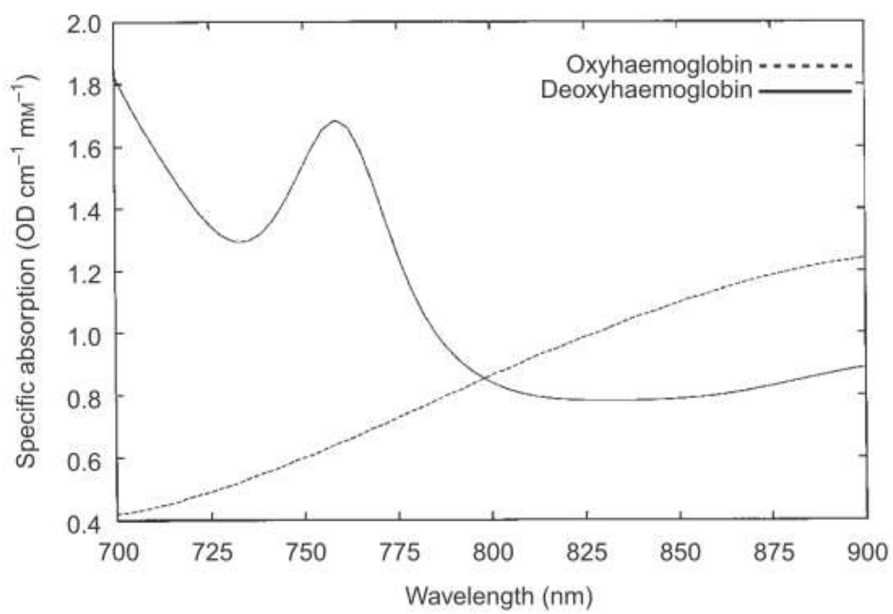
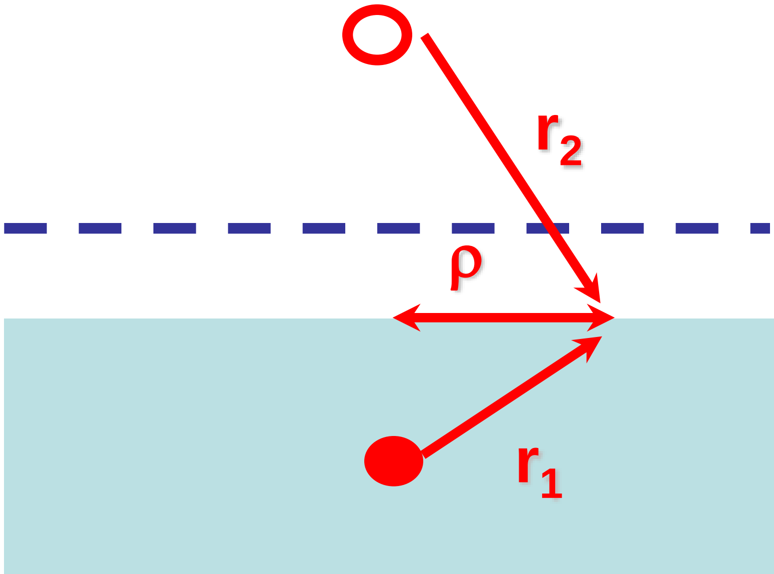
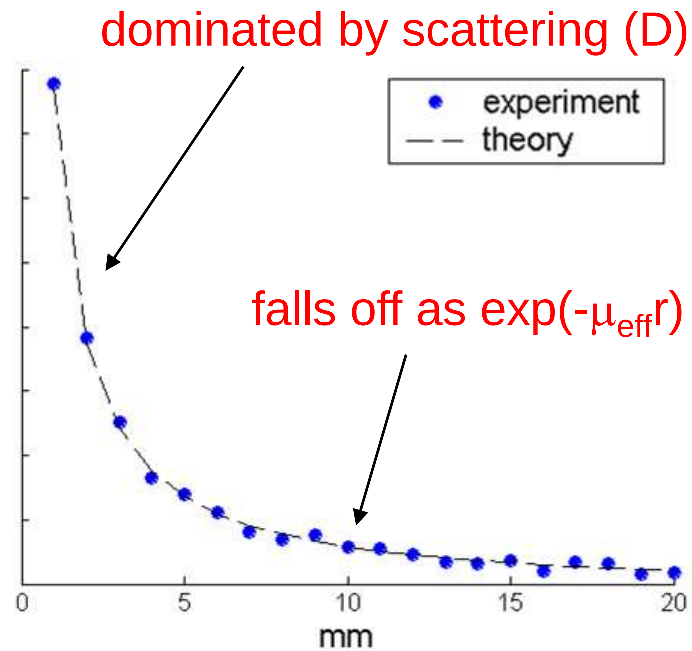


Figure 2 Near infrared absorption spectra of deoxy- (—) and oxyhaemoglobin (- - -). The oxyhaemoglobin spectrum is from Wray et al (1988); the deoxyhaemoglobin spectrum is from Matcher et al (1995)

Hull et al., *Br. J. Cancer* , **79**(11/12), 1709-1716 (1999).

Need to measure both “near” and “far”

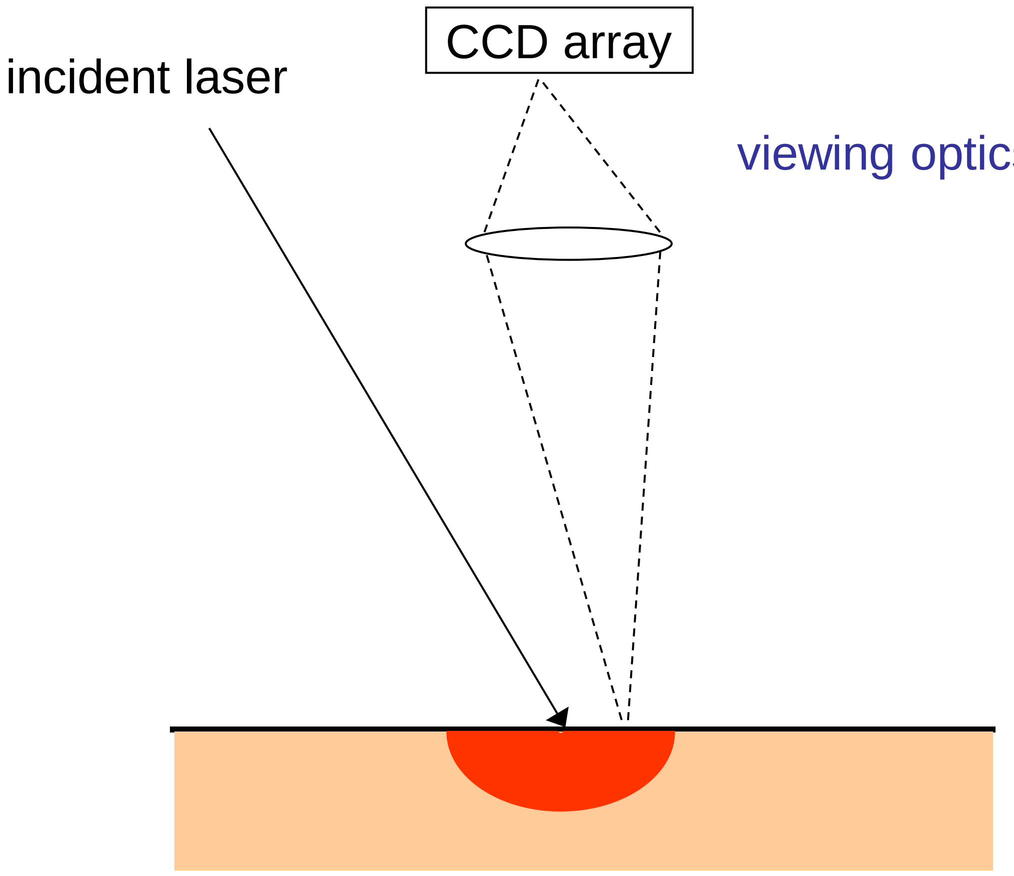




$$\Phi(\rho) = \frac{1}{4\pi D}\left(\frac{\exp(-\mu_{eff}\, r_1(\rho))}{r_1(\rho)} - \frac{\exp(-\mu_{eff}\, r_2(\rho))}{r_2(\rho)}\right)$$

Dynamic range requirements: VTS calculations

Non-contact alternative

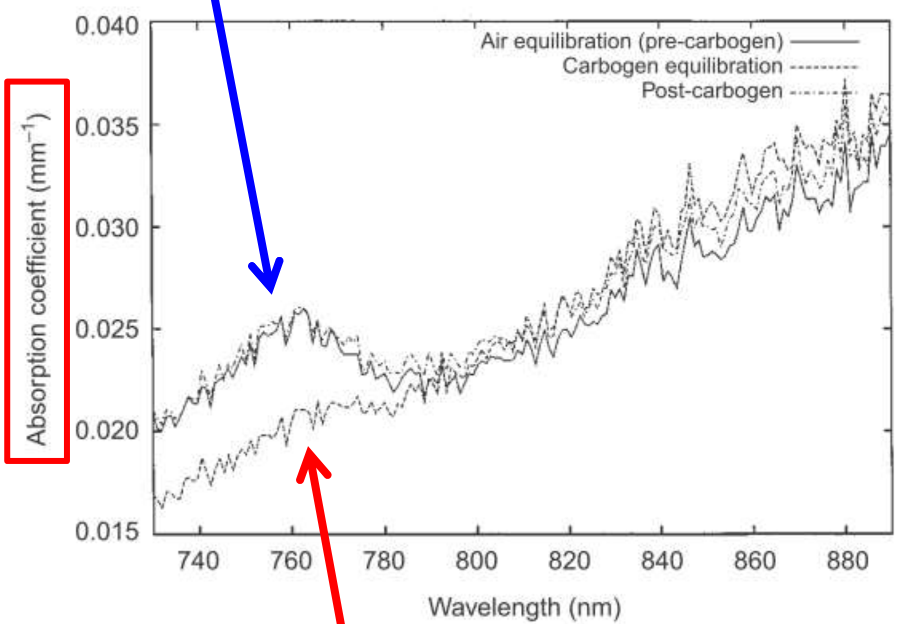


- absolute reflectance measurable
- fit shape AND height of reflectance curve

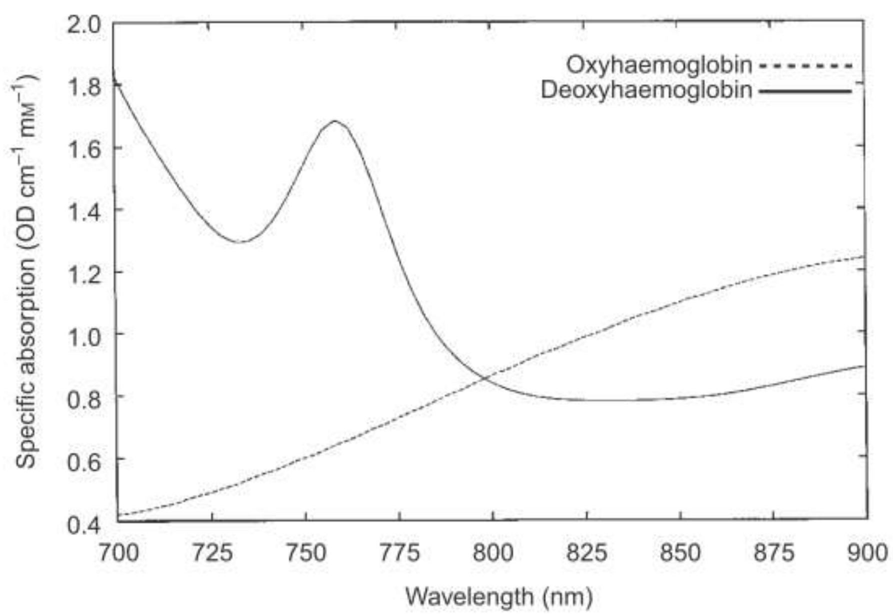
Typical power levels: VTS/Matlab calculation

Not so many wavelengths are needed!

normal air



when rat is breathing 95% O_2



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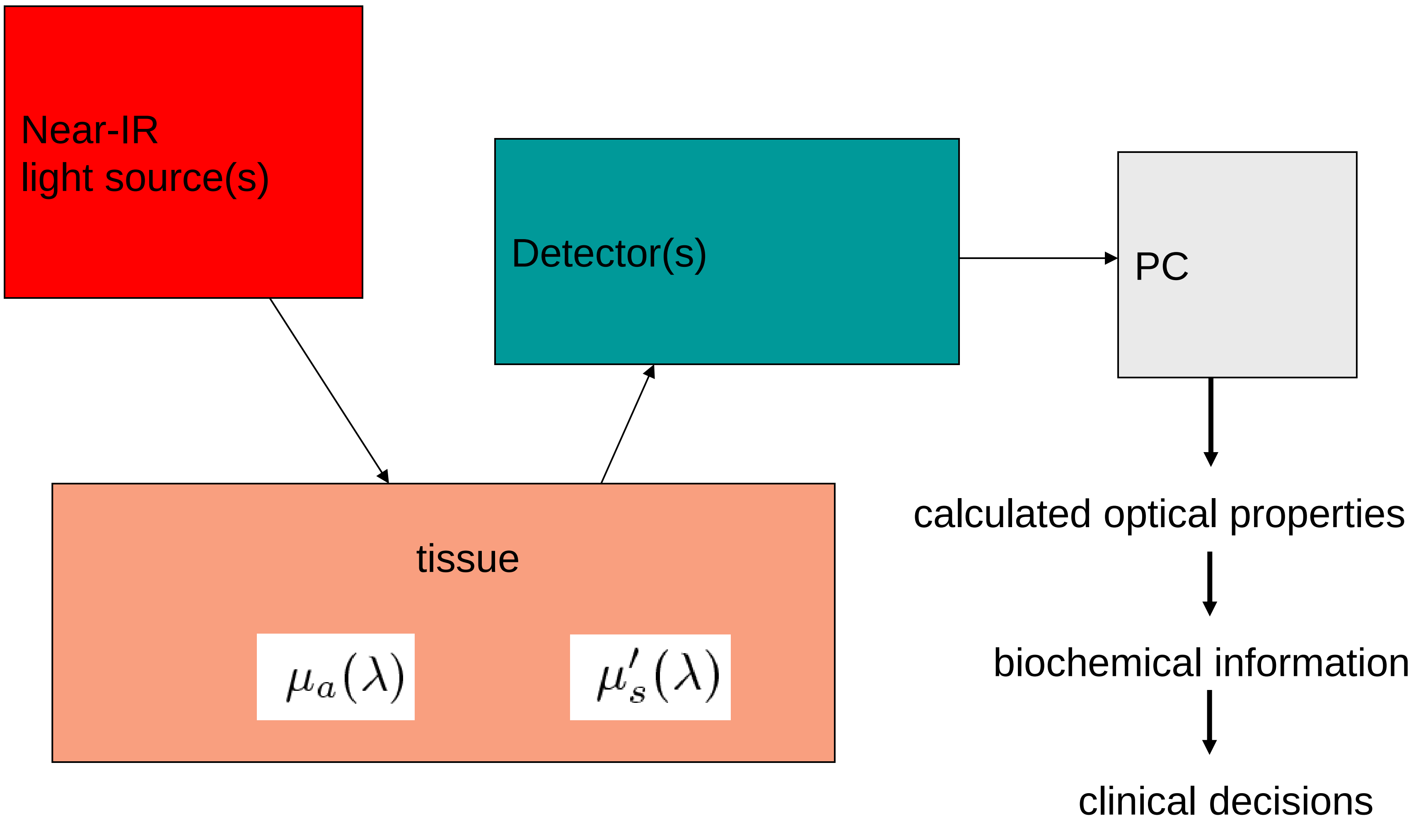
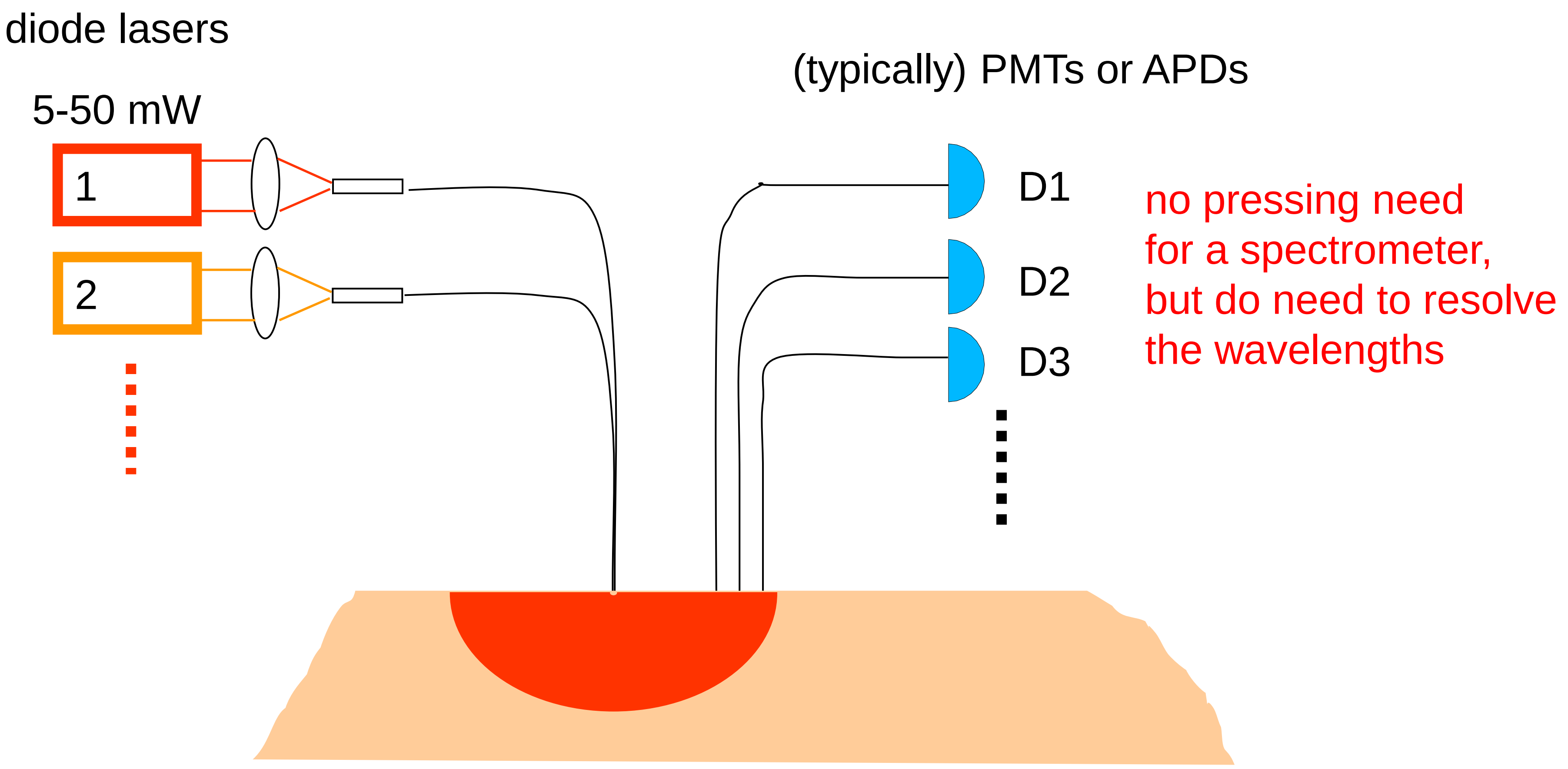
Basic instrumentation for all cases

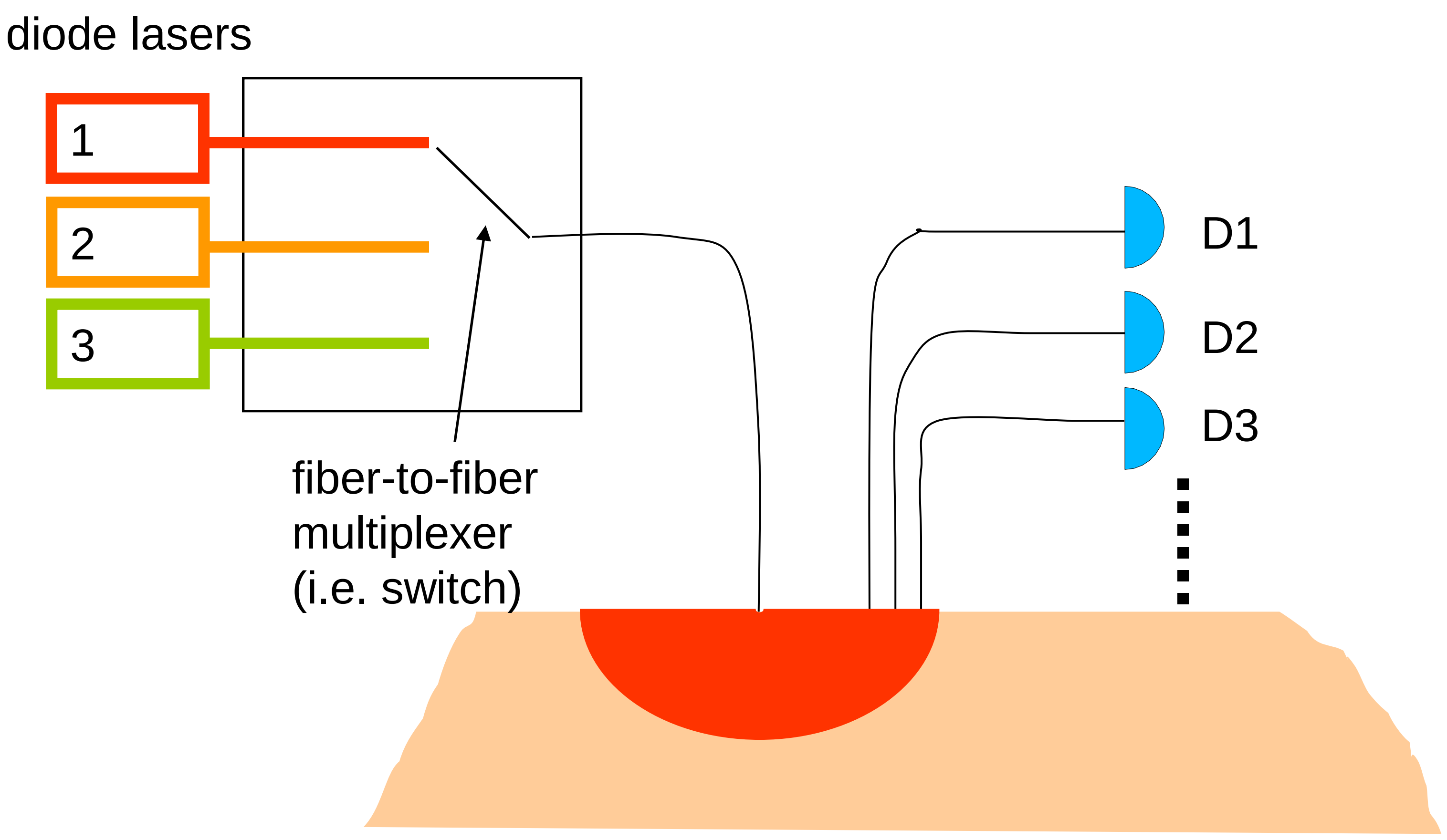


Steady state measurements: no spectrometer



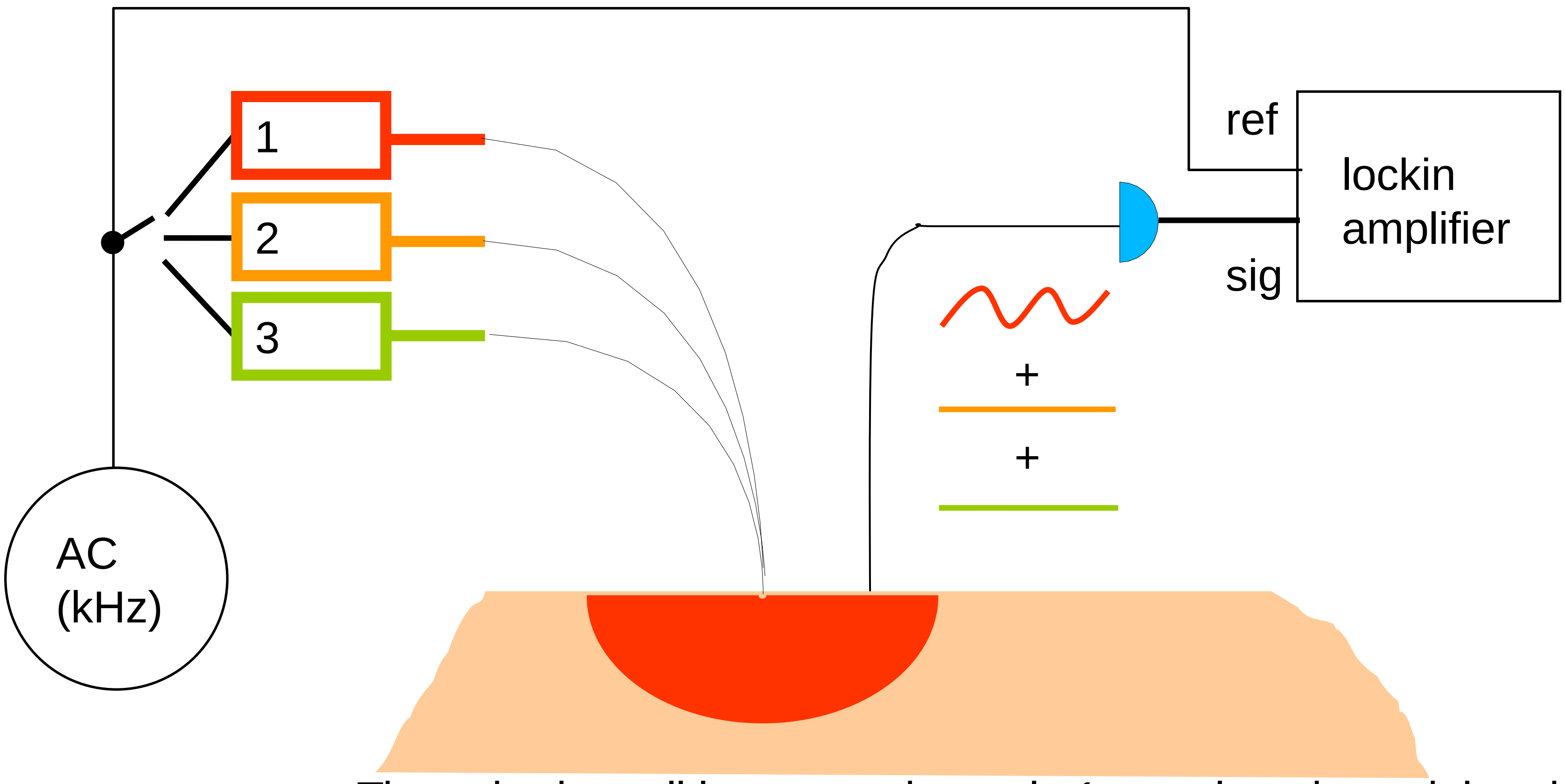
- much brighter: deeper penetration, or faster acquisition
- fewer wavelengths: less chemical information

Resolving wavelengths: time-sharing



Time-sharing: tissue sees only 1 wavelength at a time
- uses full dynamic range of detector(s)

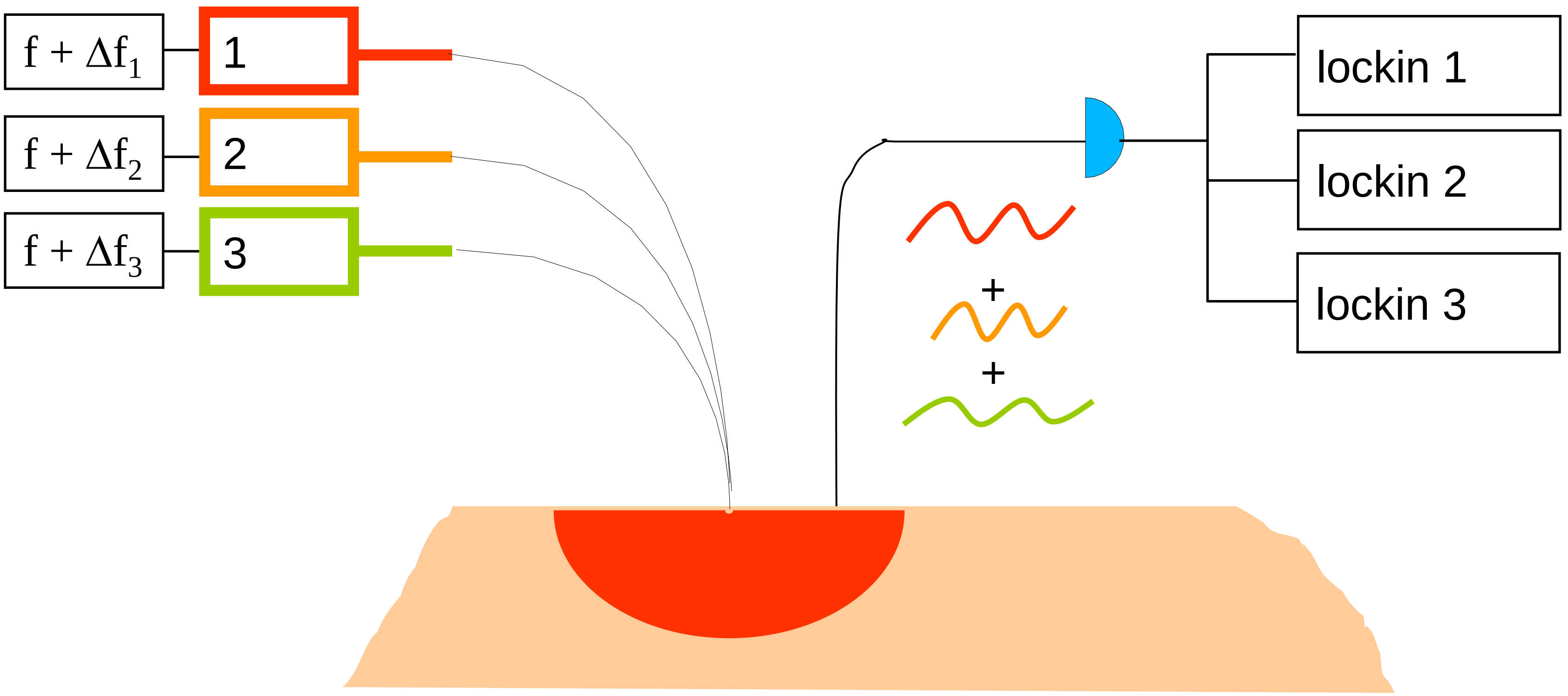
Resolving wavelengths: serial modulation



Time-sharing: all lasers on, but only 1 wavelength modulated

- reduced dynamic range
- robust against background light
- higher throughput (multimode fiber switches usually lossy)

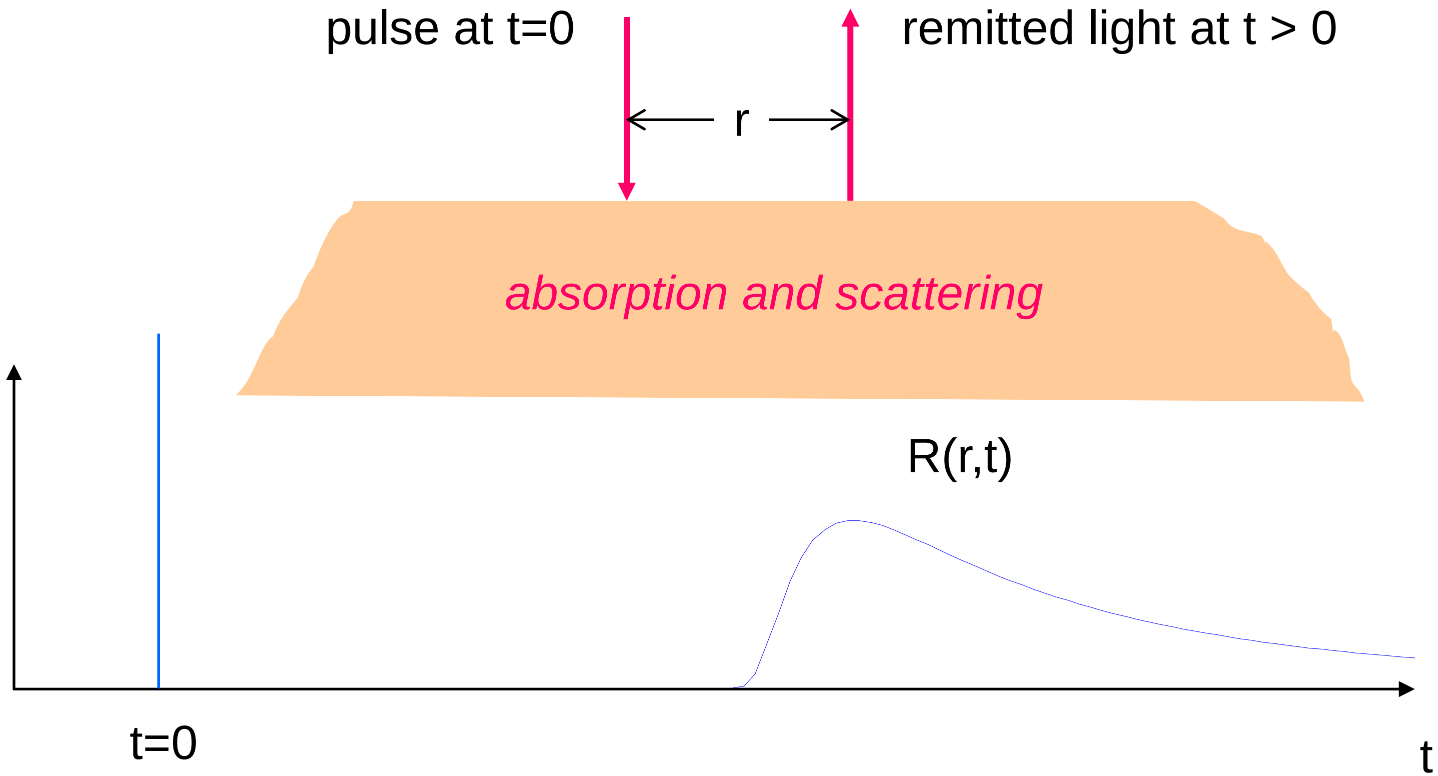
Resolving wavelengths: parallel modulation



Frequency-encoding: multiple wavelengths detected simultaneously

- good for higher time-resolution
- greater demand on instrumentation or signal processing

Time-resolved measurements



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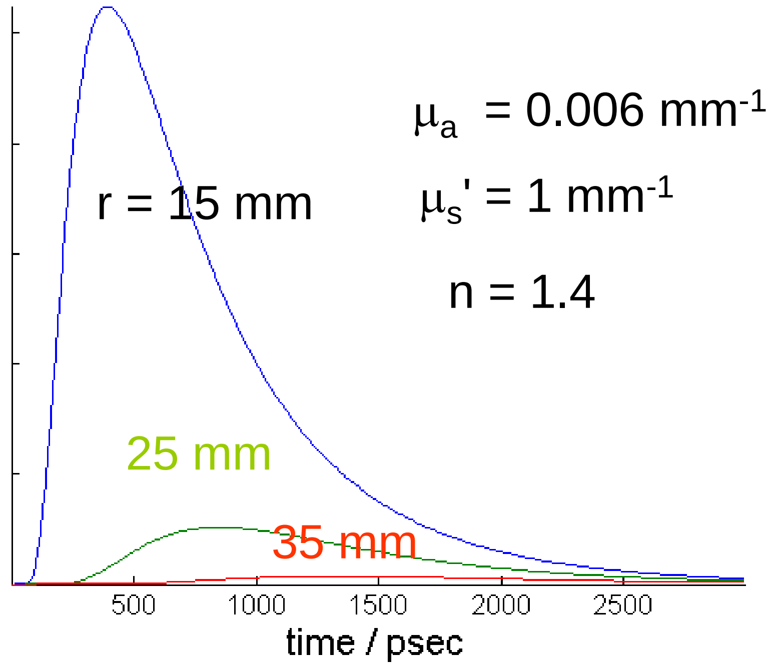

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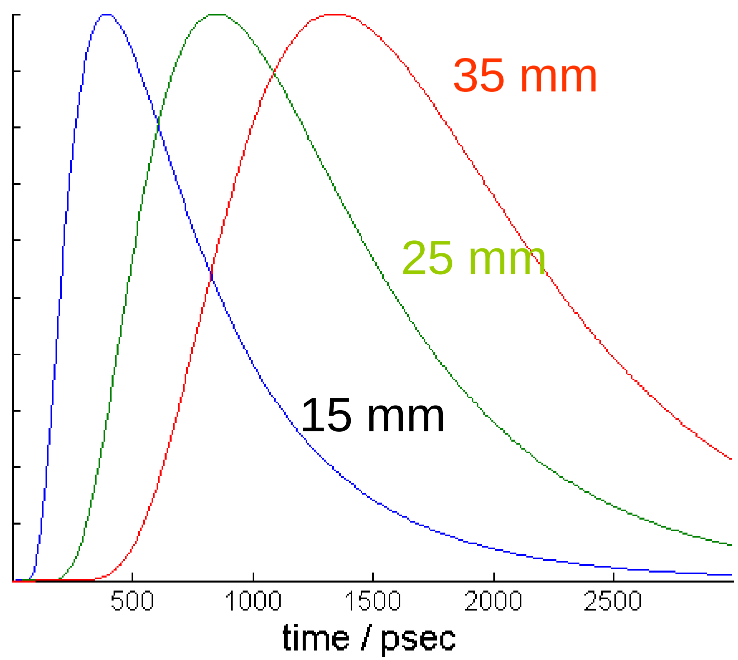
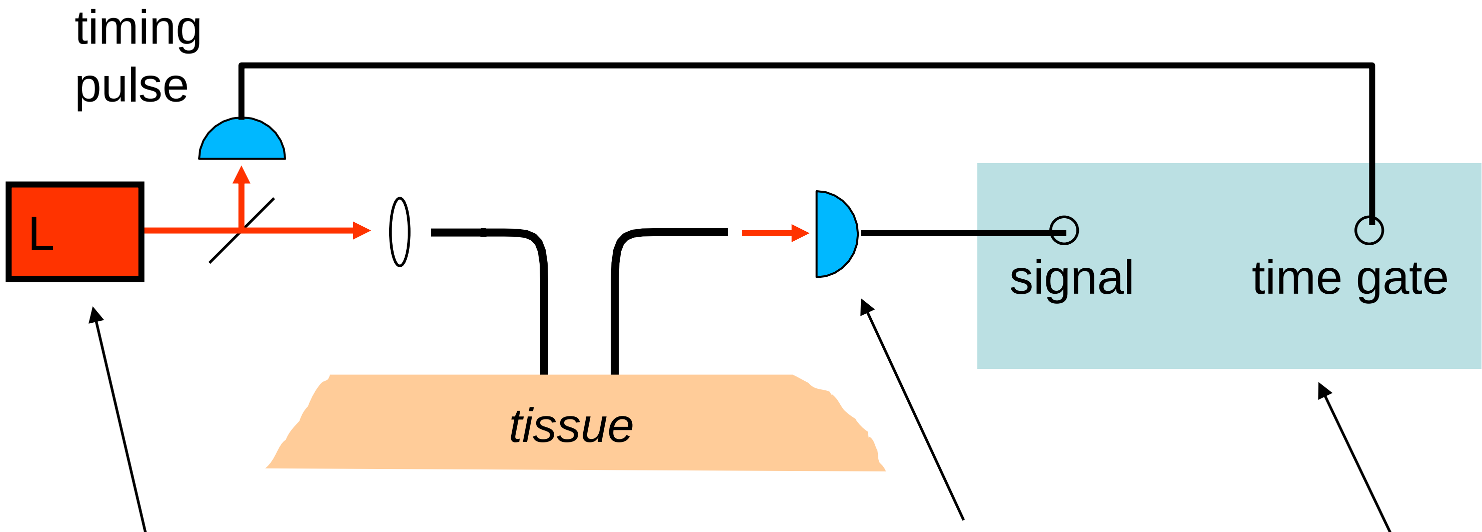


time resolution needed: psec scale; integration time needed: 100's-1000's of psec

Time-resolved measurements

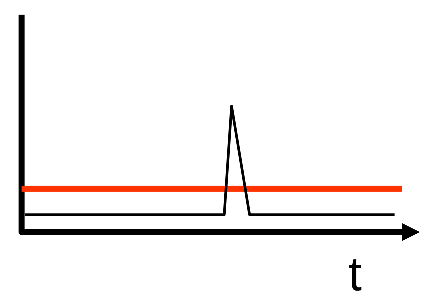


ps- or fs-pulse laser
(mode-locked dye, Ti:sapph, or fiber; pulsed diode)

Fast detector
- streak camera
- microchannel plate photomultiplier tube

time-correlated single-photon-counting (TCSPC) electronics

- many pulses, builds up a histogram
- typical integration time: ~ 0.1-1.0 sec



Multi-wavelength, time-domain data

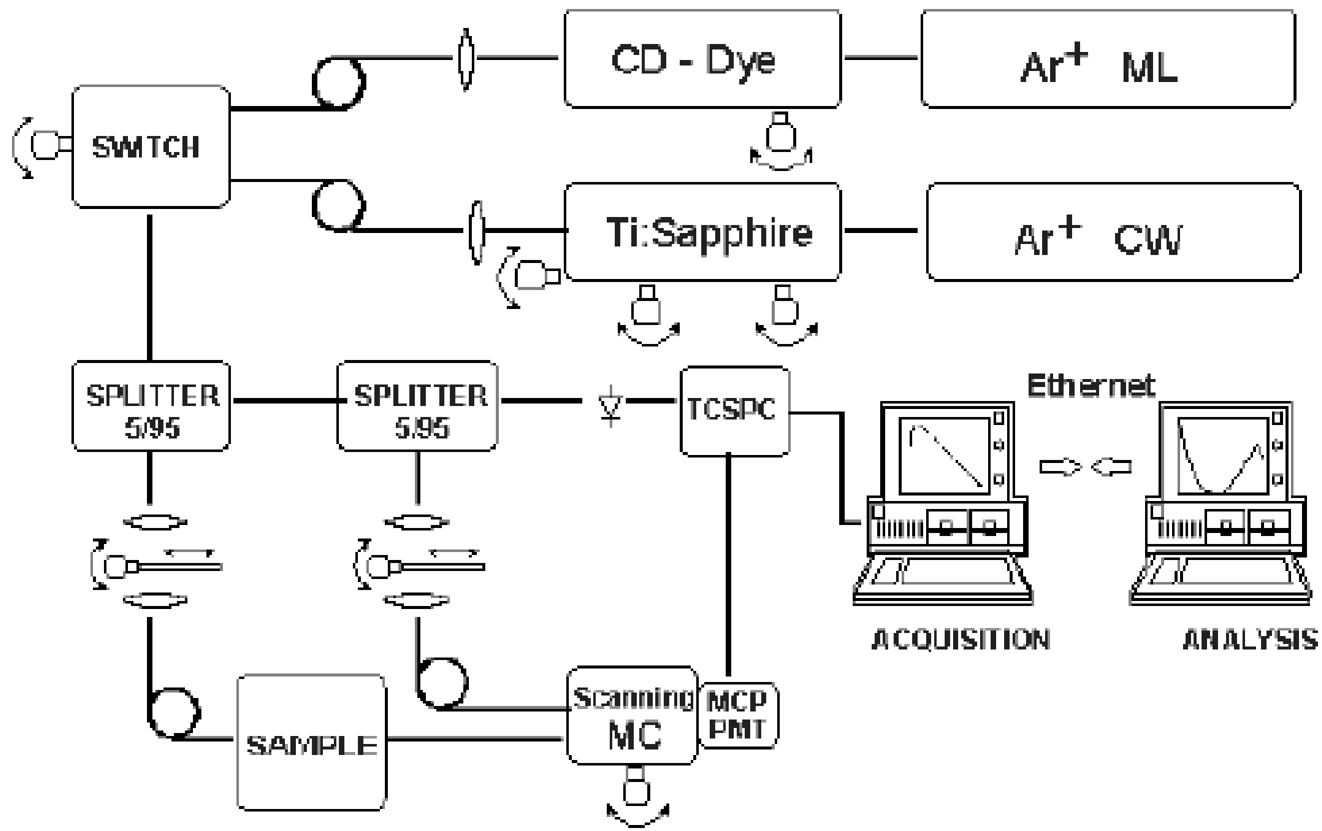


Figure 1. Schematic diagram of the system set-up. CD, cavity-dumped; ML, mode-locked; CW, continuous wave; TCSPC, time-correlated single-photon counting; MC, monochromator; MCP-PMT, microchannel plate photomultiplier.

***In vivo* optical characterization of human tissues from 610 to 1010 nm by time-resolved reflectance spectroscopy**

Alessandro Torricelli, Antonio Pifferi, Paola Taroni, Eleonora Giambattistelli and Rinaldo Cubeddu

INSTITUTE OF PHYSICS PUBLISHING

Phys. Med. Biol. **46** (2001) 2227–2237

Experimental time-resolved photon diffusion measurements on biological tissue

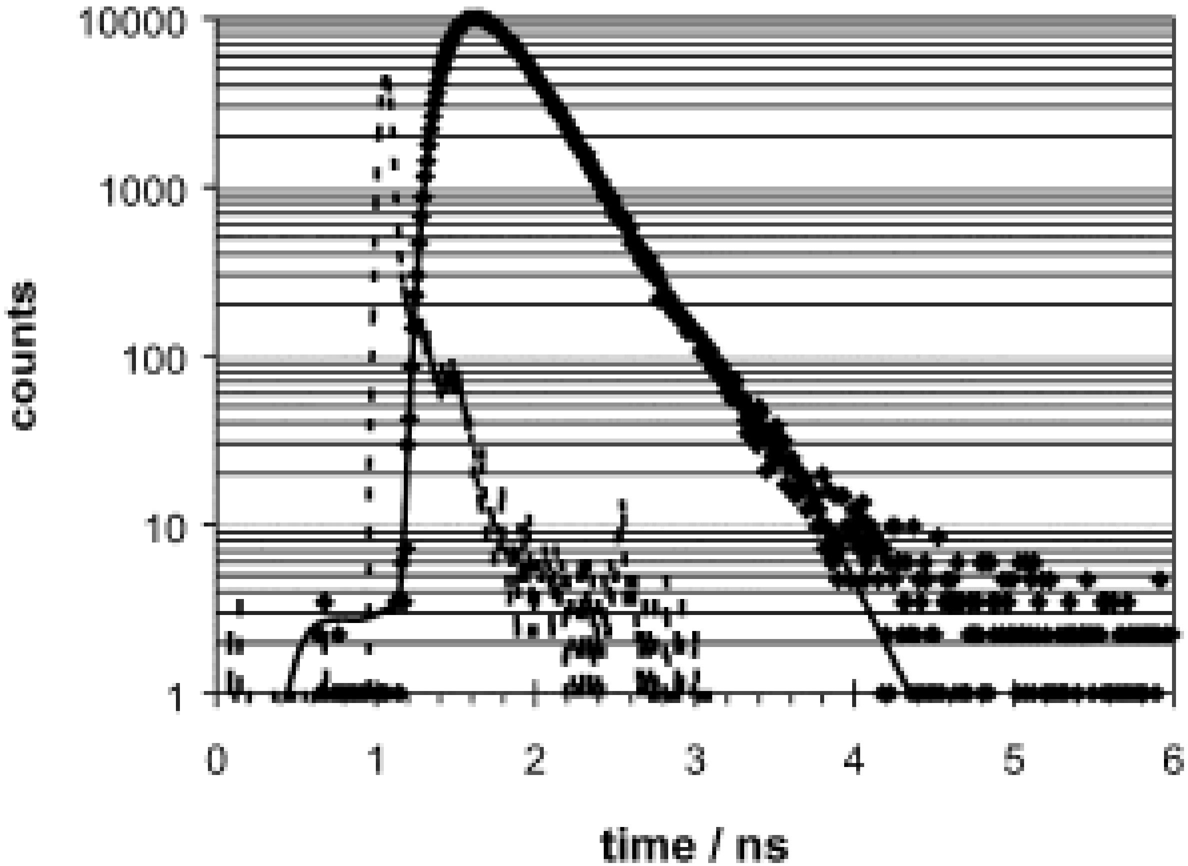
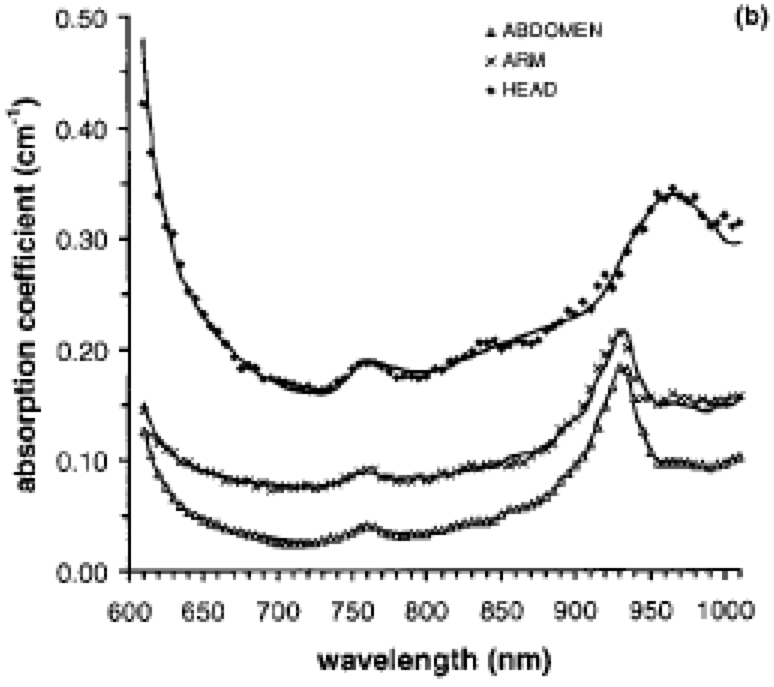
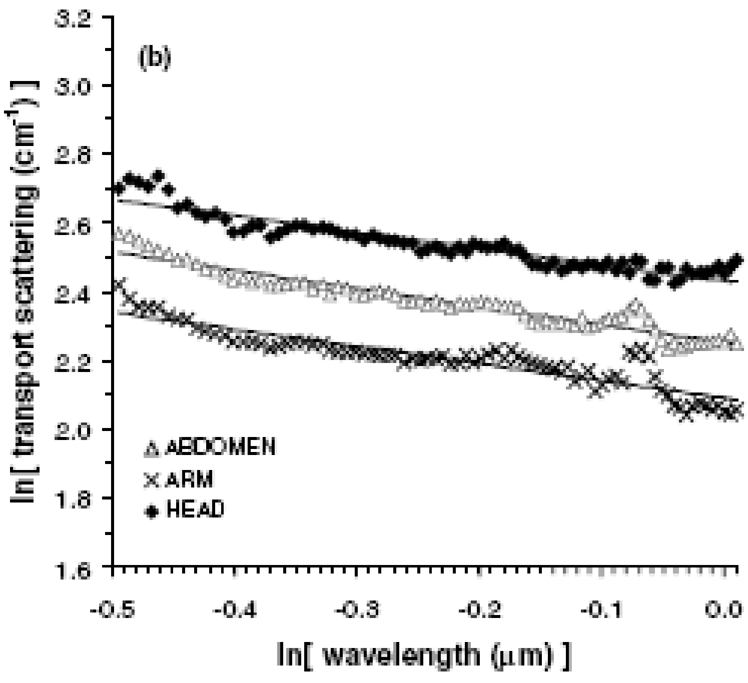


Fig. 1 Typical best fit of time-resolved reflectance data. The experimental data (◆) are fitted with the convolution (——) of the instrumental transfer function (- - -) with the theoretical curve (not shown).

Absorption and scattering spectra extracted from fitting time-resolved data to theory

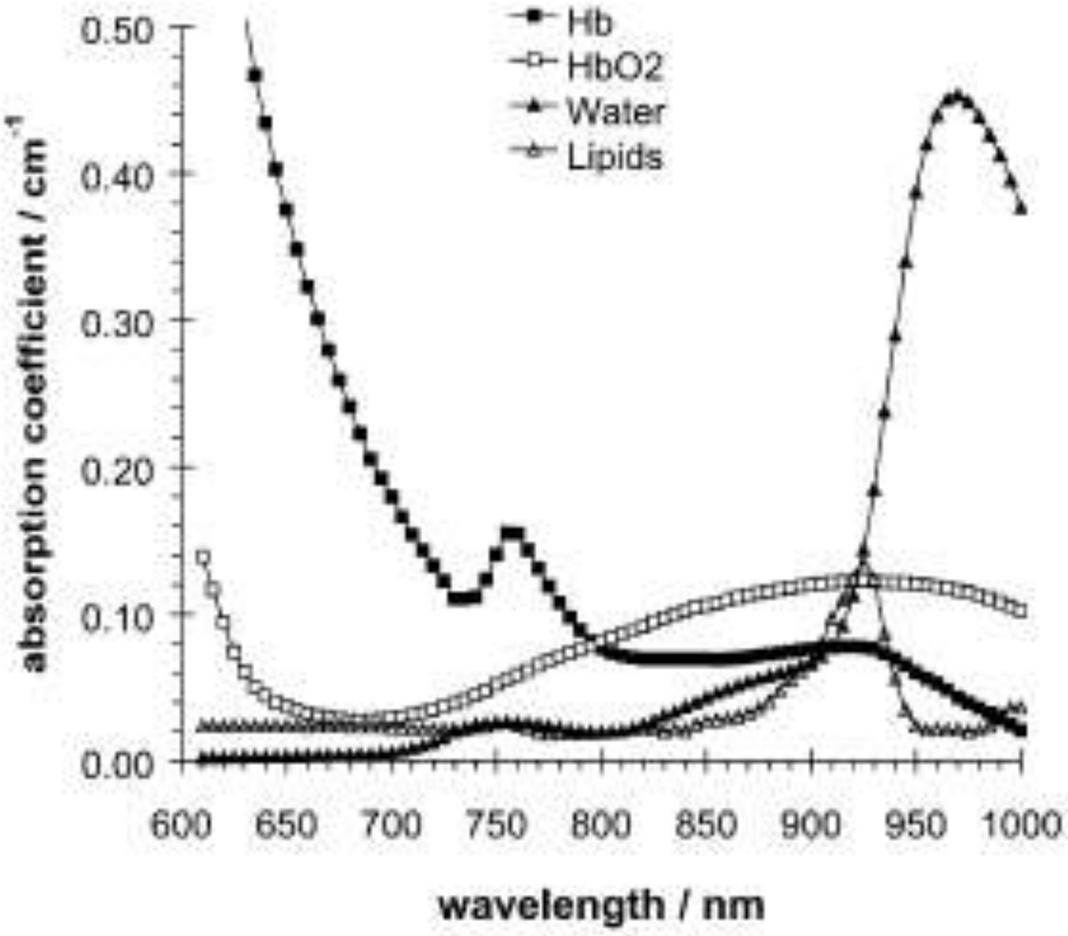


absorption



scattering

Important chemical absorbers in tissue



Calculating chemical concentrations

Table 1 Concentration of absorbers in the female breast

Subject	Hb + HbO_2/µM	Y (%)	Water (%)	Lipid (%)
1 (44 years)	37	78	13	86
2 (24 years)	71	75	46	29

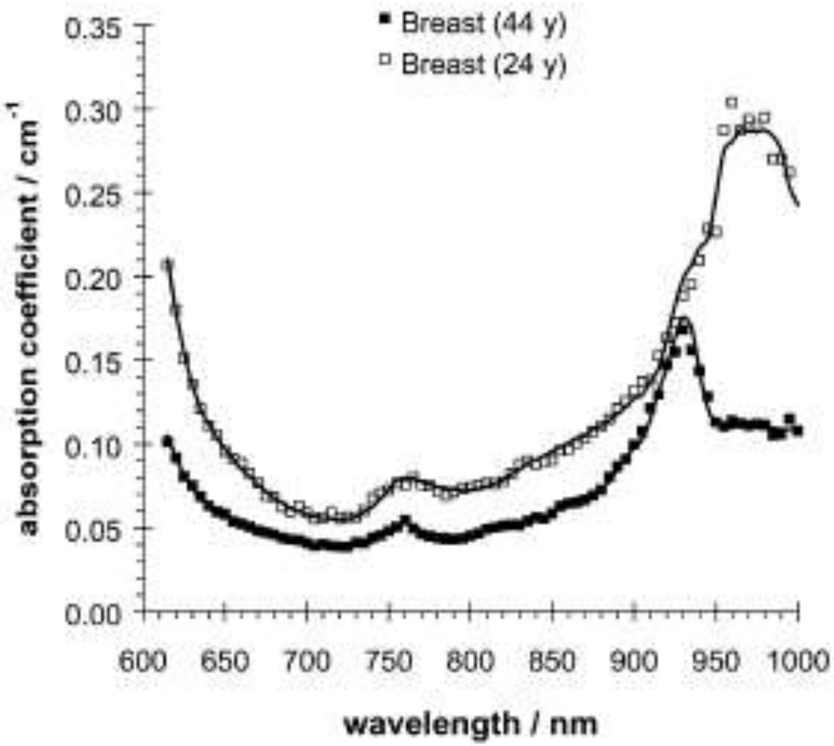
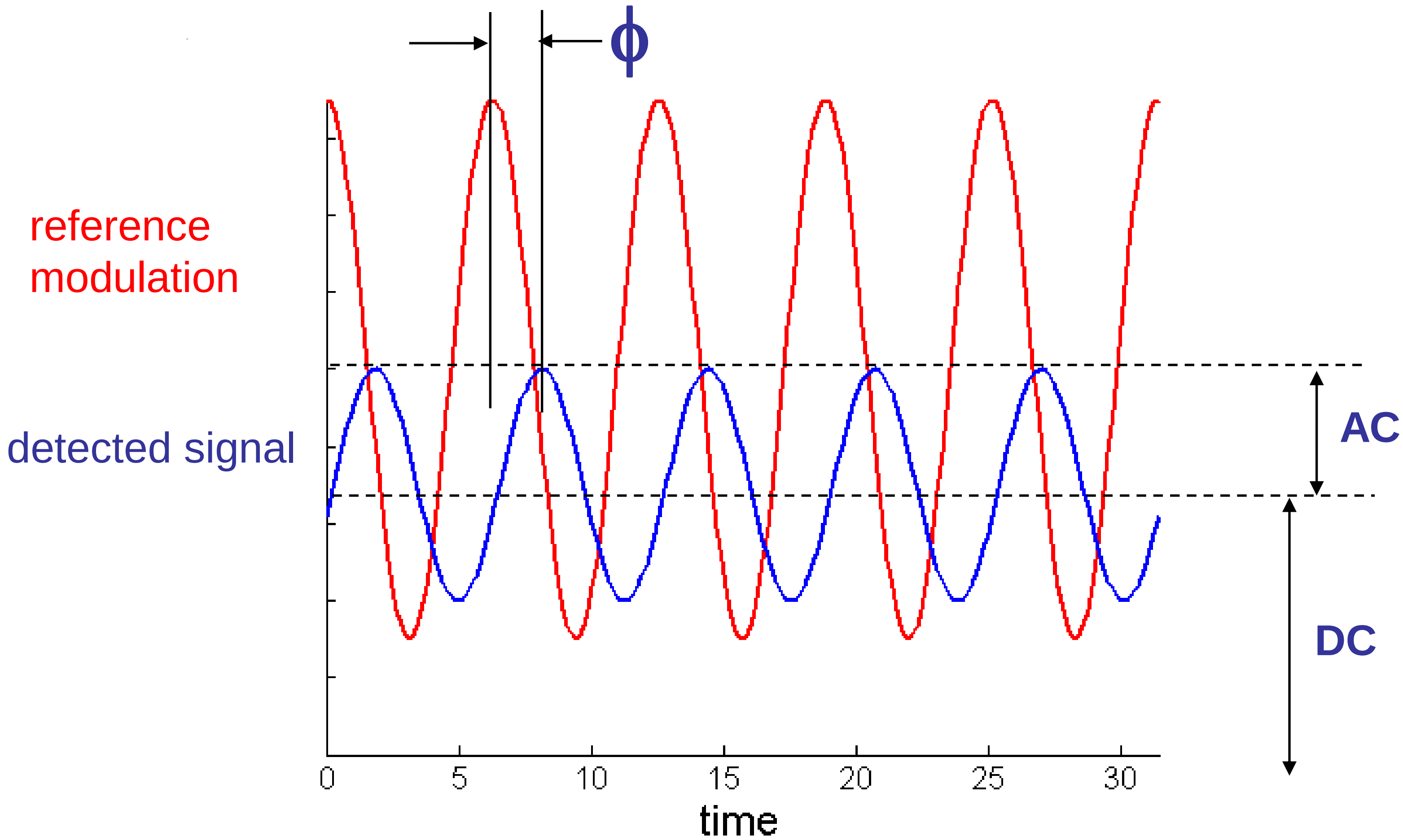


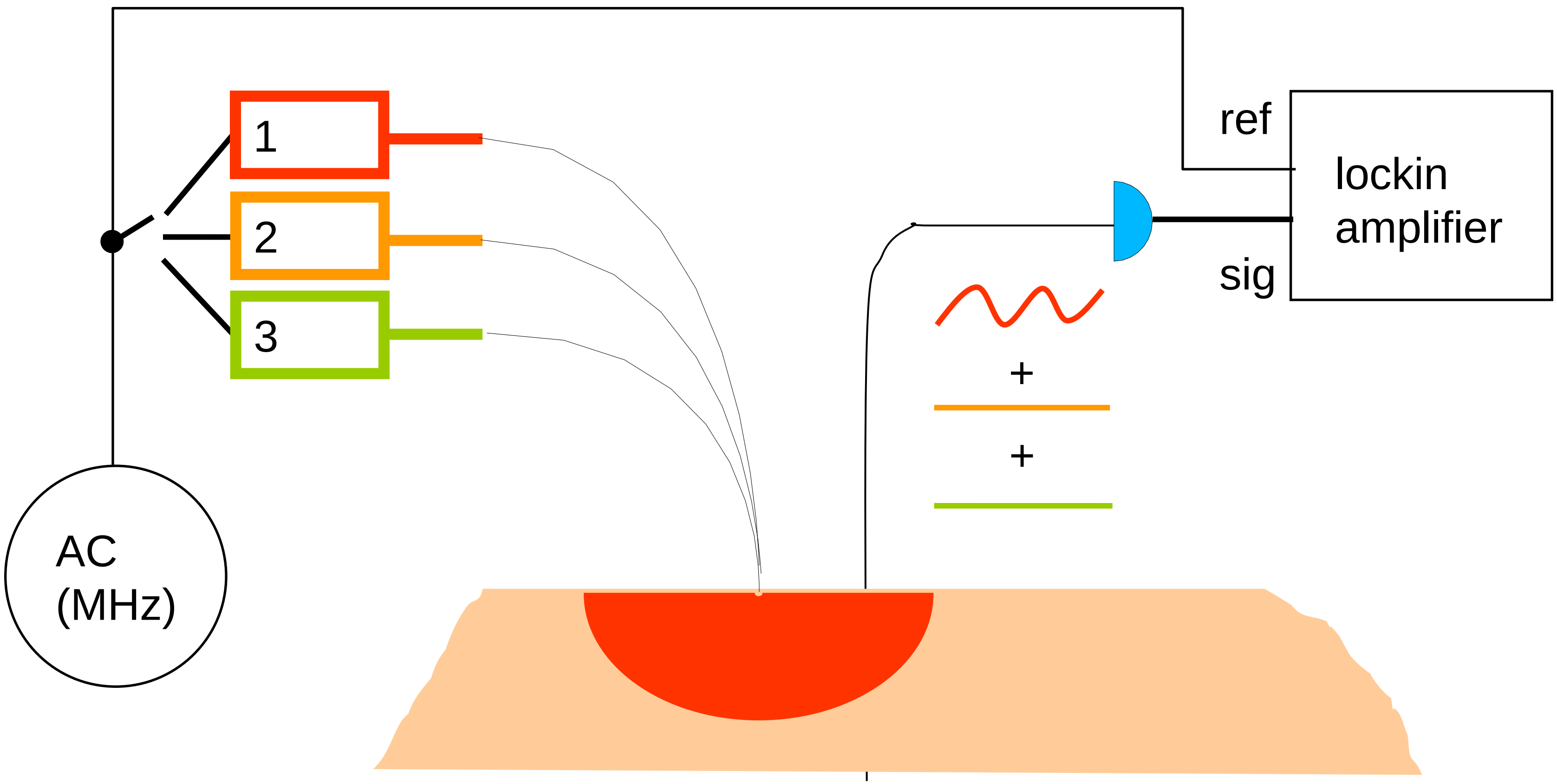
Fig. 3 Absorption spectra of the female breast on 44 year (■) and 24 year (□) old volunteers. The water and lipid content vary appreciably with age.

Frequency-resolved method

Reminder of the observables:

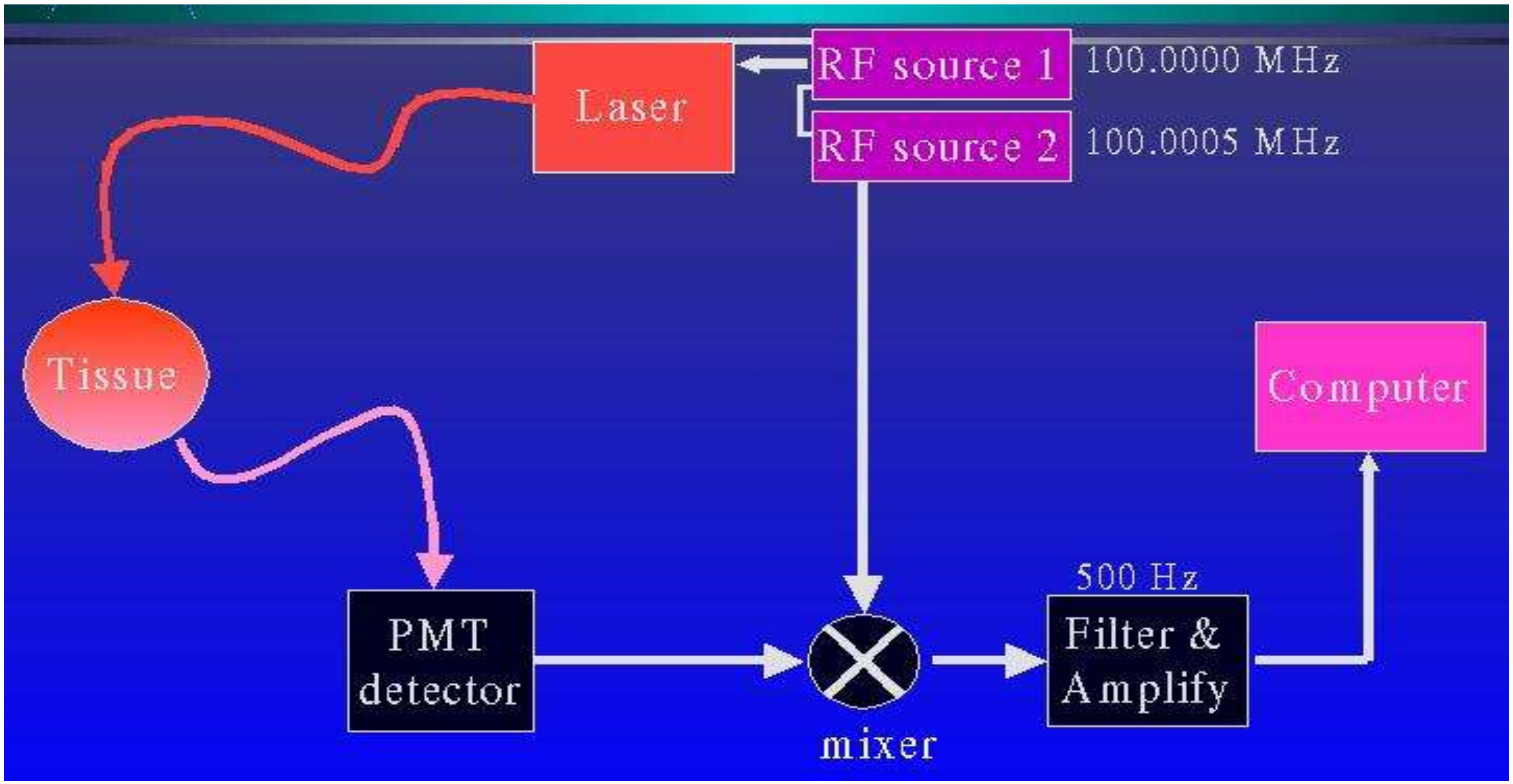


Frequency-resolved instrumentation



Frequency-resolved instrumentation: heterodyning

takes RF frequency → low frequency (better detection electronics)

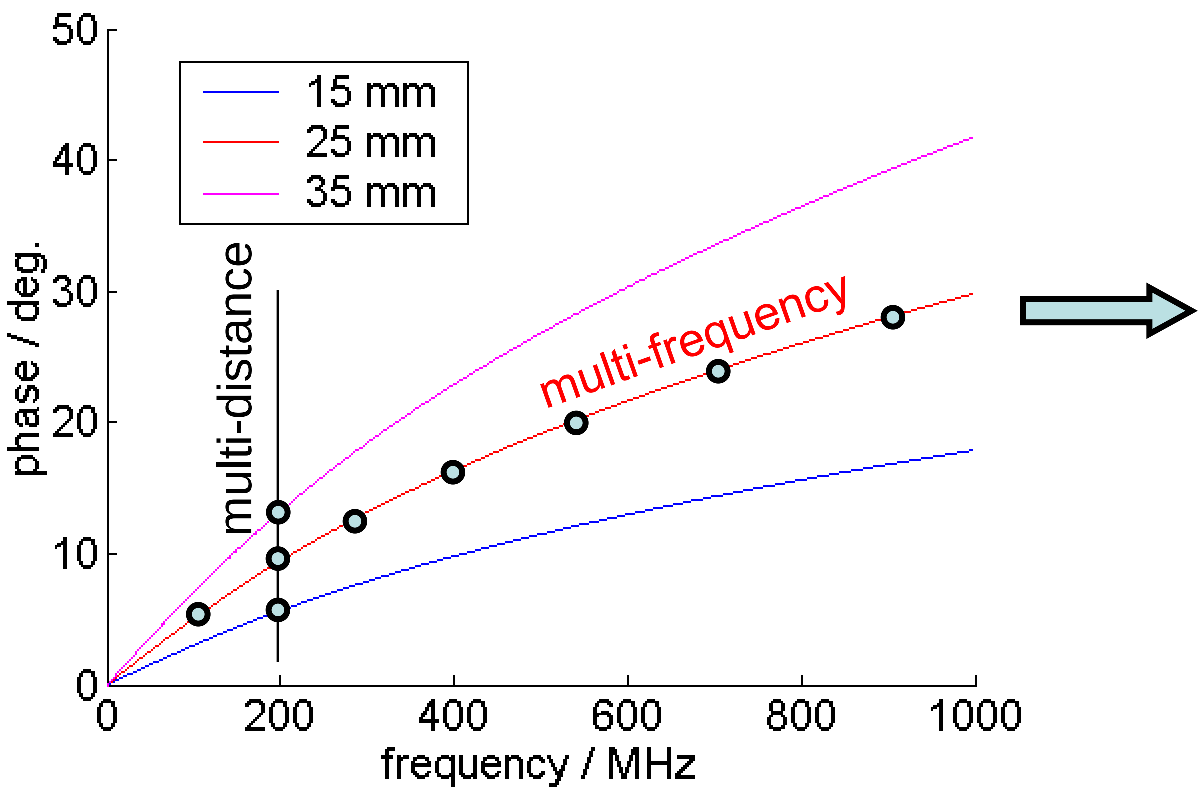


http://www-nml.dartmouth.edu/nir/index.html

Frequency domain: optical geometries

Ways to measure:

1) phase and/or amplitude vs. distance
2) phase and/or amplitude vs. frequency

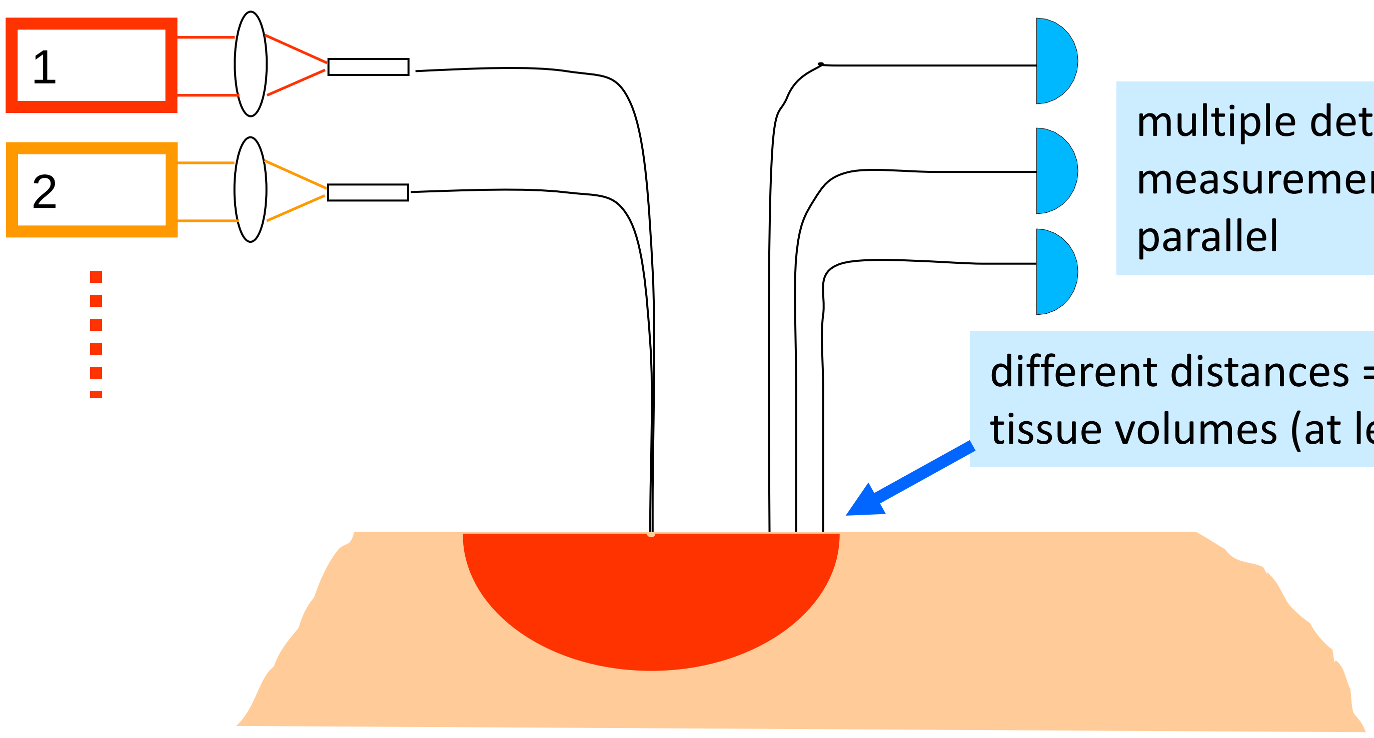


fit to a model
to get absorption
and scattering
coefficients

Multidistance vs. multifrequency

Multidistance

single modulation frequency = optimized impedance matching, simpler design



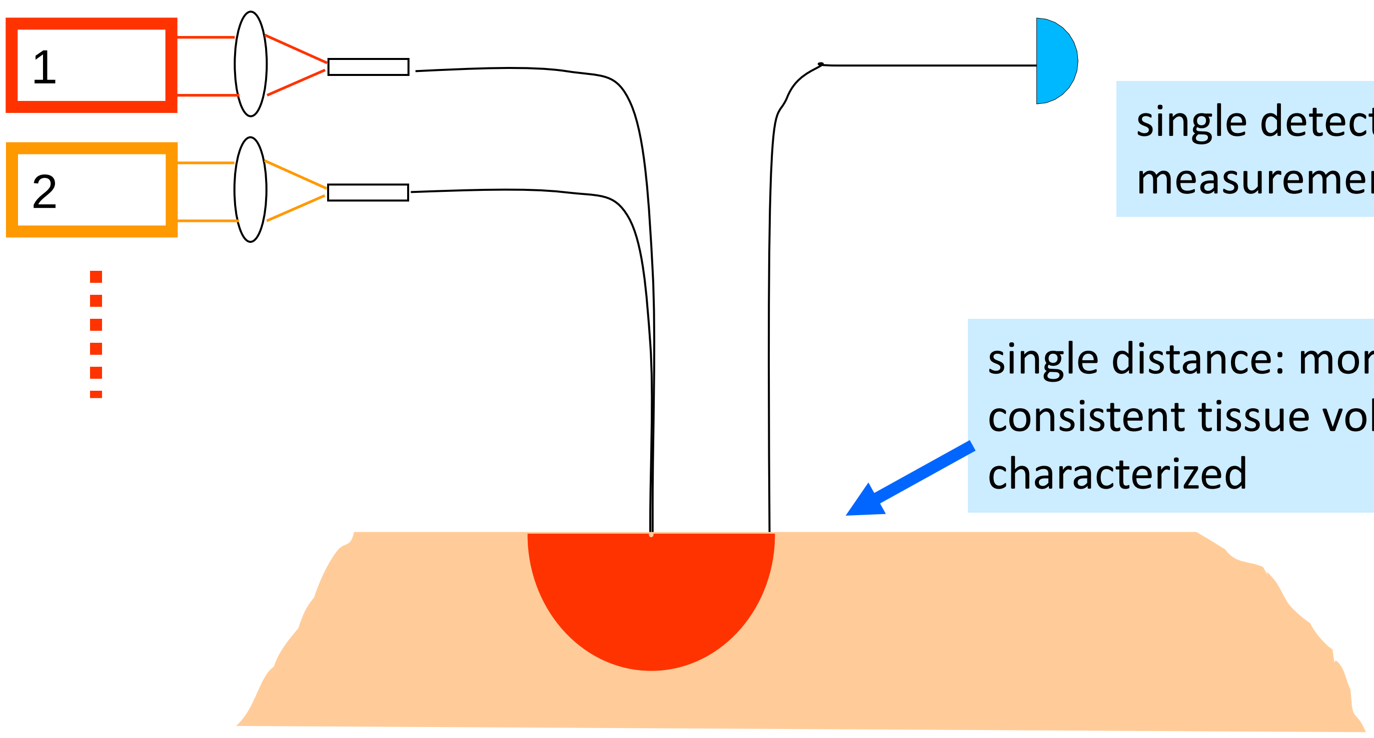
multiple detectors = measurements in parallel

different distances = different tissue volumes (at least slightly)

Multidistance vs. multifrequency

Multifrequency

multiple modulation frequencies
= lossier, more complex design



single detector:
measurements in series

single distance: more
consistent tissue volume
characterized

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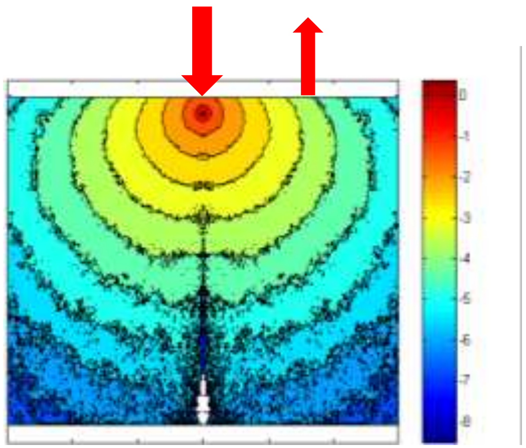

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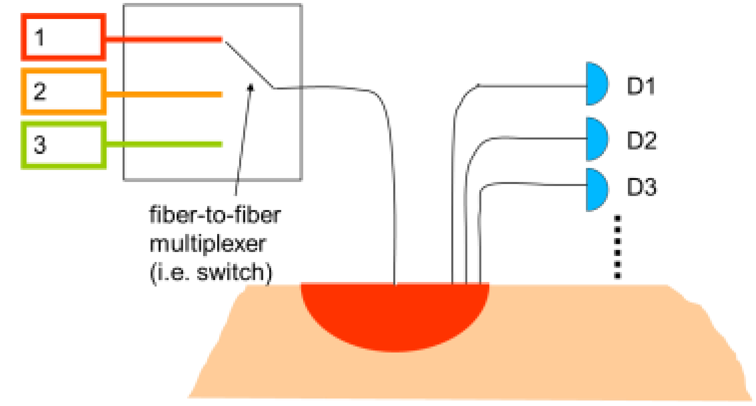
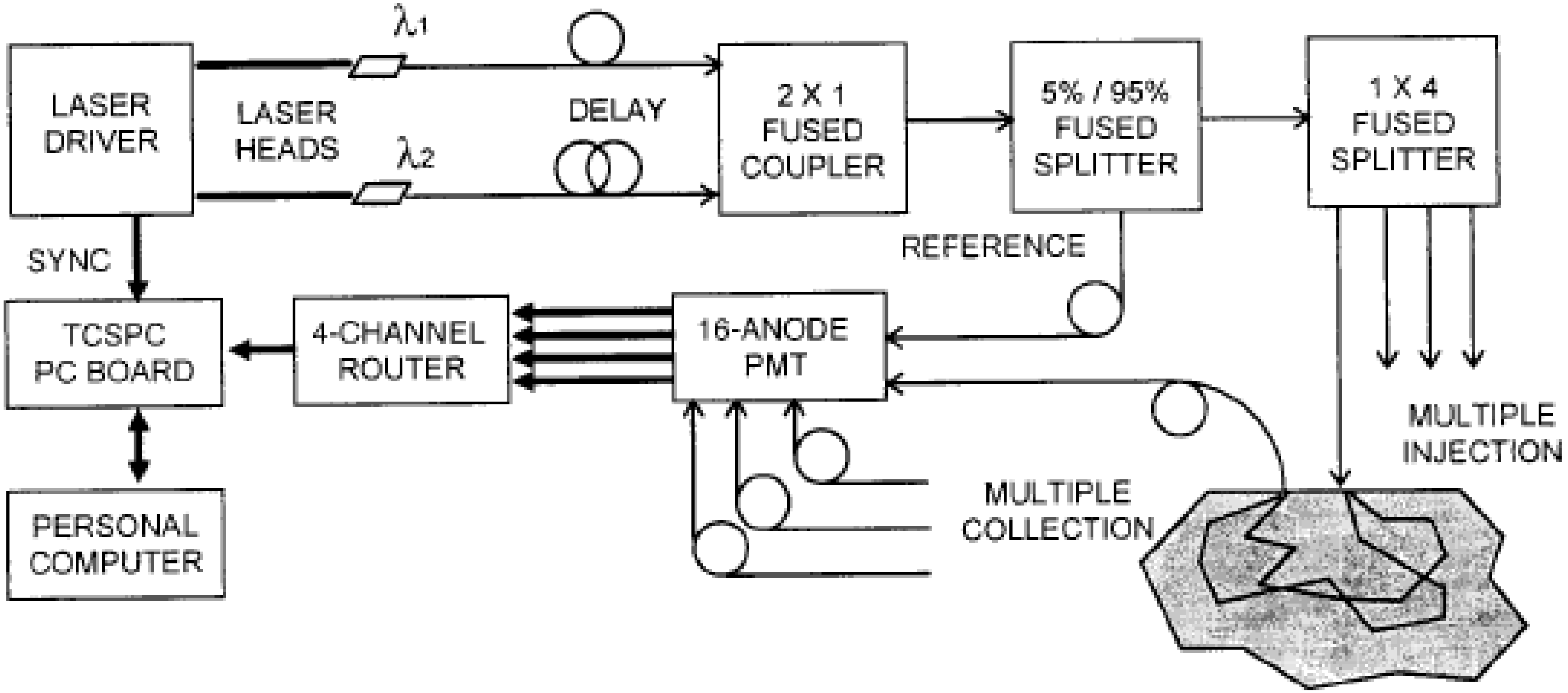


various applications

Application #1: time-domain tissue oximetry

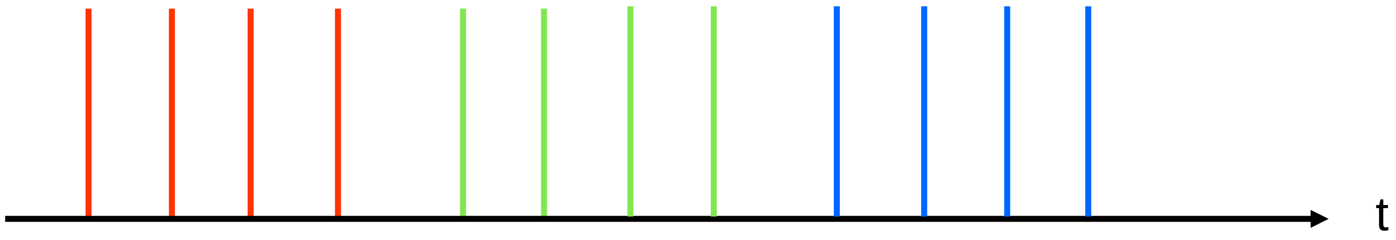


R. Cubeddu et al., "Compact tissue oximeter based on dual-wavelength multichannel time-resolved reflectance," Appl. Opt. 38(16), 3670-3680 (1999).

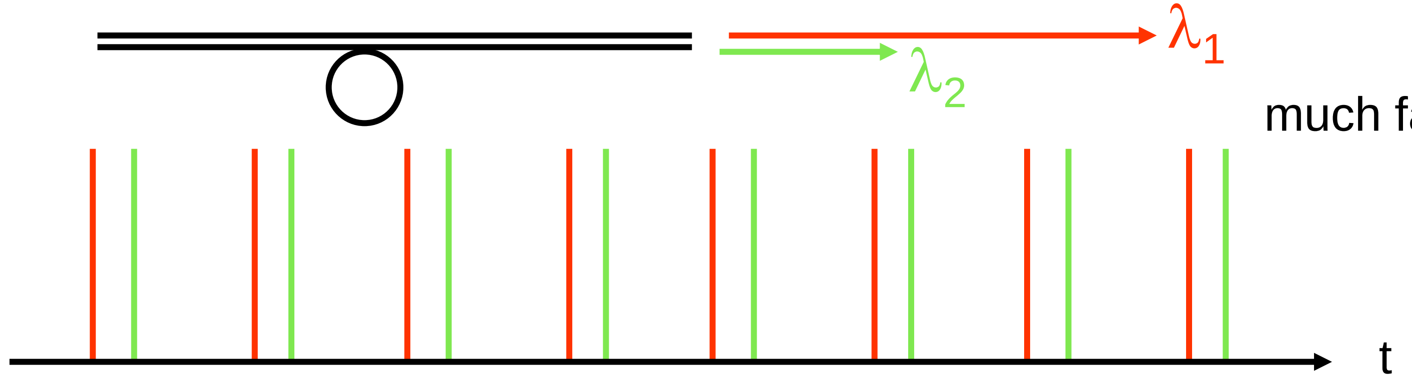
Time-resolved measurements

Resolving wavelengths

Tunable mode-locked laser (many wavelengths):



Simultaneous pulses with delay line (2 wavelengths):



much faster results

2-wavelength tissue oximeter

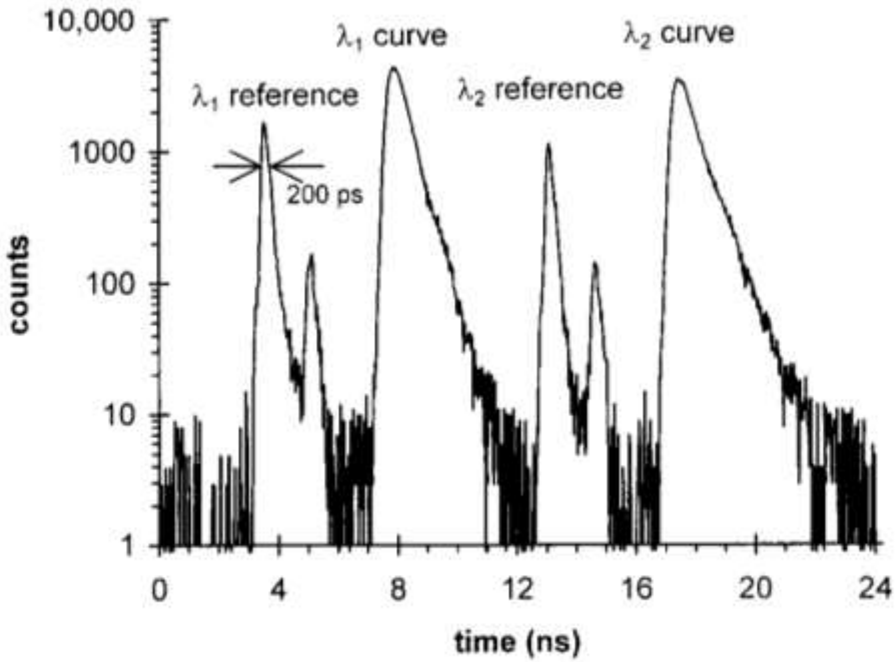


Fig. 2. Typical time-resolved reflectance curves and reference pulses. Note, in each reference pulse, the presence of the characteristic afterpulse of the PMT. The arrows mark a FWHM of 200 ps.

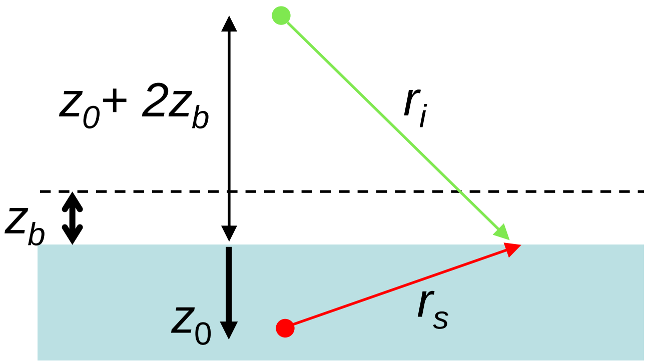
- multianode photomultiplier
- 672 & 818 nm diode lasers, 100 ps pulses
- 1 mW average power
- TCSPC board, 80 MHz acquisition
- integation time >= 100 msec

R. Cubeddu et al., "Compact tissue oximeter based on dual-wavelength multichannel time-resolved reflectance," Appl. Opt. 38(16), 3670-3680 (1999).

2-wavelength tissue oximeter

$$R_f = \frac{1}{2}(4\pi Dc)^{-3/2} t^{-5/2} \exp(-\mu_a ct) \left[z_o \exp\left(-\frac{{r_s}^2}{4Dct}\right) + (z_o + 2z_b) \exp\left(-\frac{{r_i}^2}{4Dct}\right)\right]$$

from previous lecture



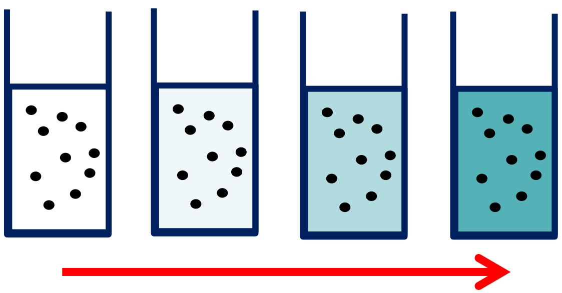
$$R(\rho, t) = \frac{1}{2}(4\pi vD)^{-3/2} t^{-5/2} \exp(-\mu_a vt) \exp\left(-\frac{\rho^2}{4Dvt}\right) \times \left\{ z_0 \exp\left(-\frac{{z_0}^2}{4Dvt}\right) - (z_0 + 2z_e) \times \exp\left[-\frac{(z_0 + 2z_e)^2}{4Dvt}\right]\right\}, \quad (2)$$

where v is the speed of light in the medium, z_0 [$=(\mu_s')^{-1}$] is the effective mean-free path, D ($=z_0/3$) is the diffusion coefficient, z_e [$=2D(1 + r_d)/(1 - r_d)$] is the extrapolated distance, and r_d can be approximated by $r_d = -1.440n^{-2} + 0.710n^{-1} + 0.668 + 0.0636n$, with indices of refraction of $n = 1.33$ for the experiments on phantoms and $n = 1.4$ for the *in vivo* measurements.

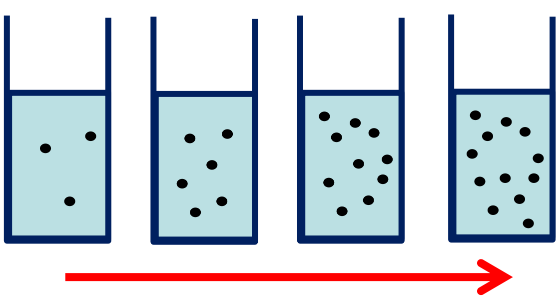
R. Cubeddu et al., "Compact tissue oximeter based on dual-wavelength multichannel time-resolved reflectance," Appl. Opt. 38(16), 3670-3680 (1999).

from oximeter paper

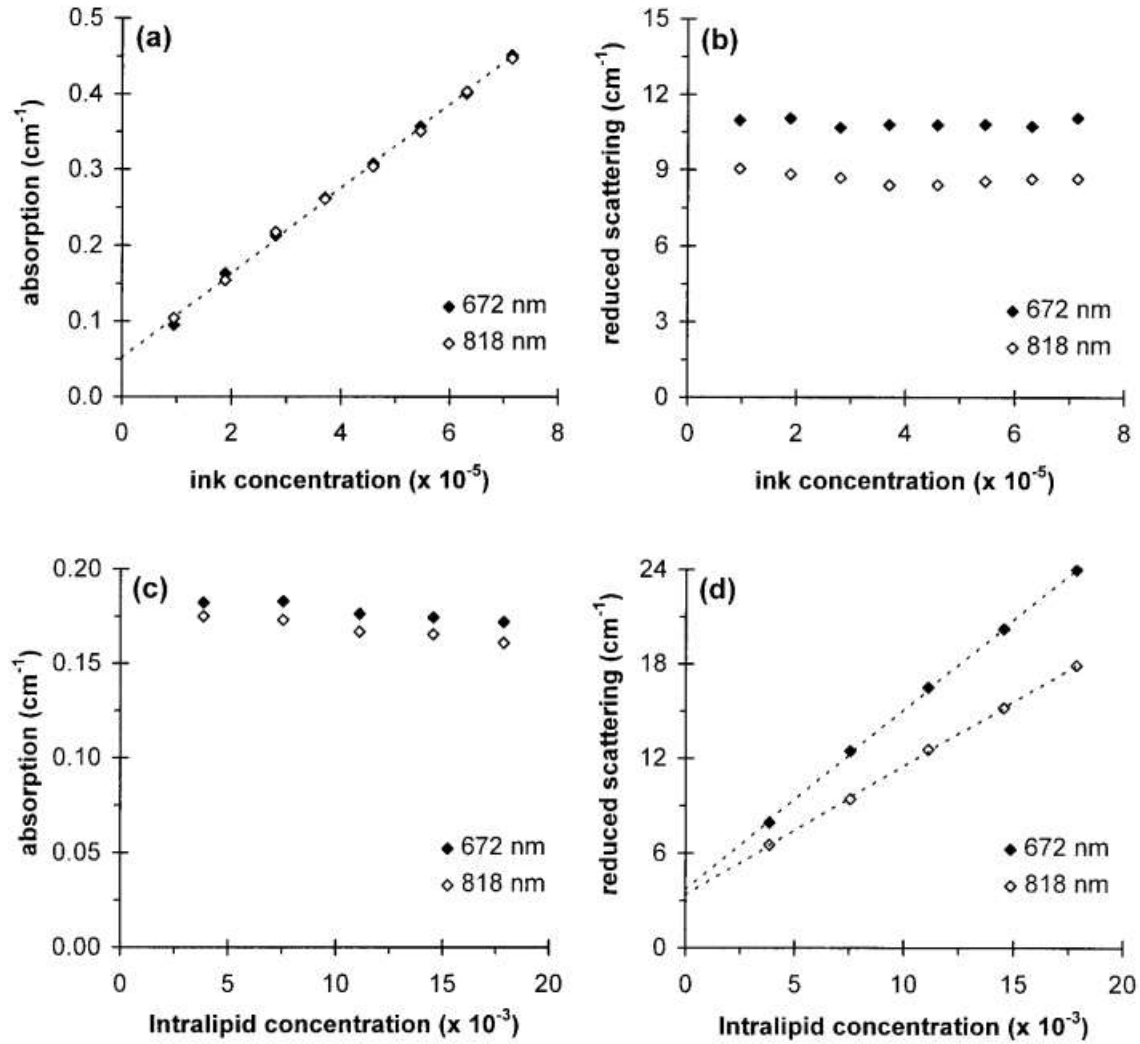
Validation: unmixing absorption & scattering



absorption



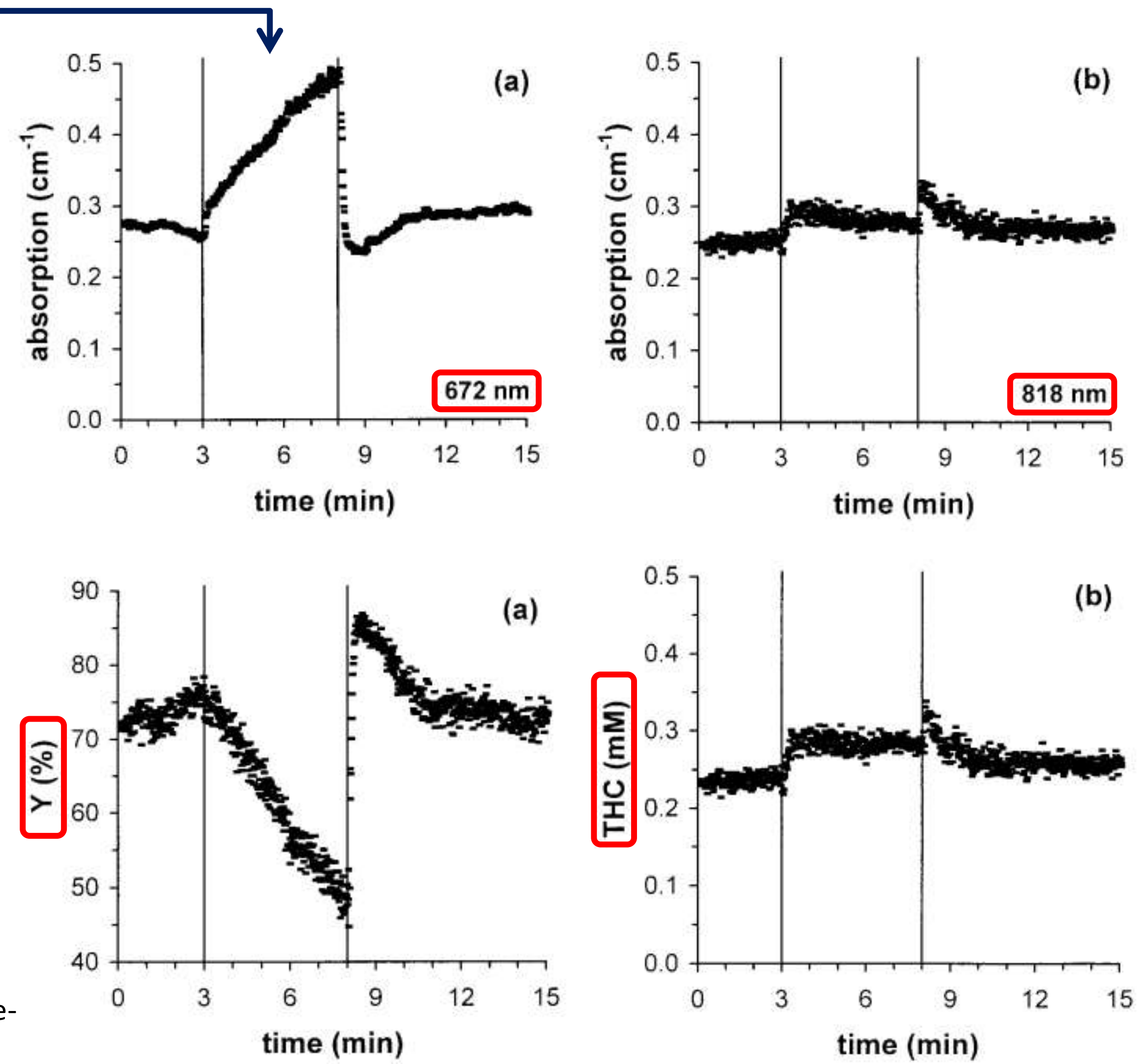
scattering



R. Cubeddu et al., "Compact tissue oximeter based on dual-wavelength multichannel time-resolved reflectance," Appl. Opt. 38(16), 3670-3680 (1999).

Time domain *in vivo* blood measurements

pressure cuff on



R. Cubeddu et al., “Compact tissue oximeter based on dual-wavelength multichannel time-resolved reflectance,” Appl. Opt. 38(16), 3670-3680 (1999).

Application #2: Breast tissue

Breast tissue analysis

Clinical goals:

- Flag abnormal tissue (i.e. tumors)
- Coregister with higher-resolution imaging modalities (e.g. mammographic x-ray)

Instrumentation goals:

- Absorption spectroscopy of breast
- Tomographic reconstructions in breast with tumors

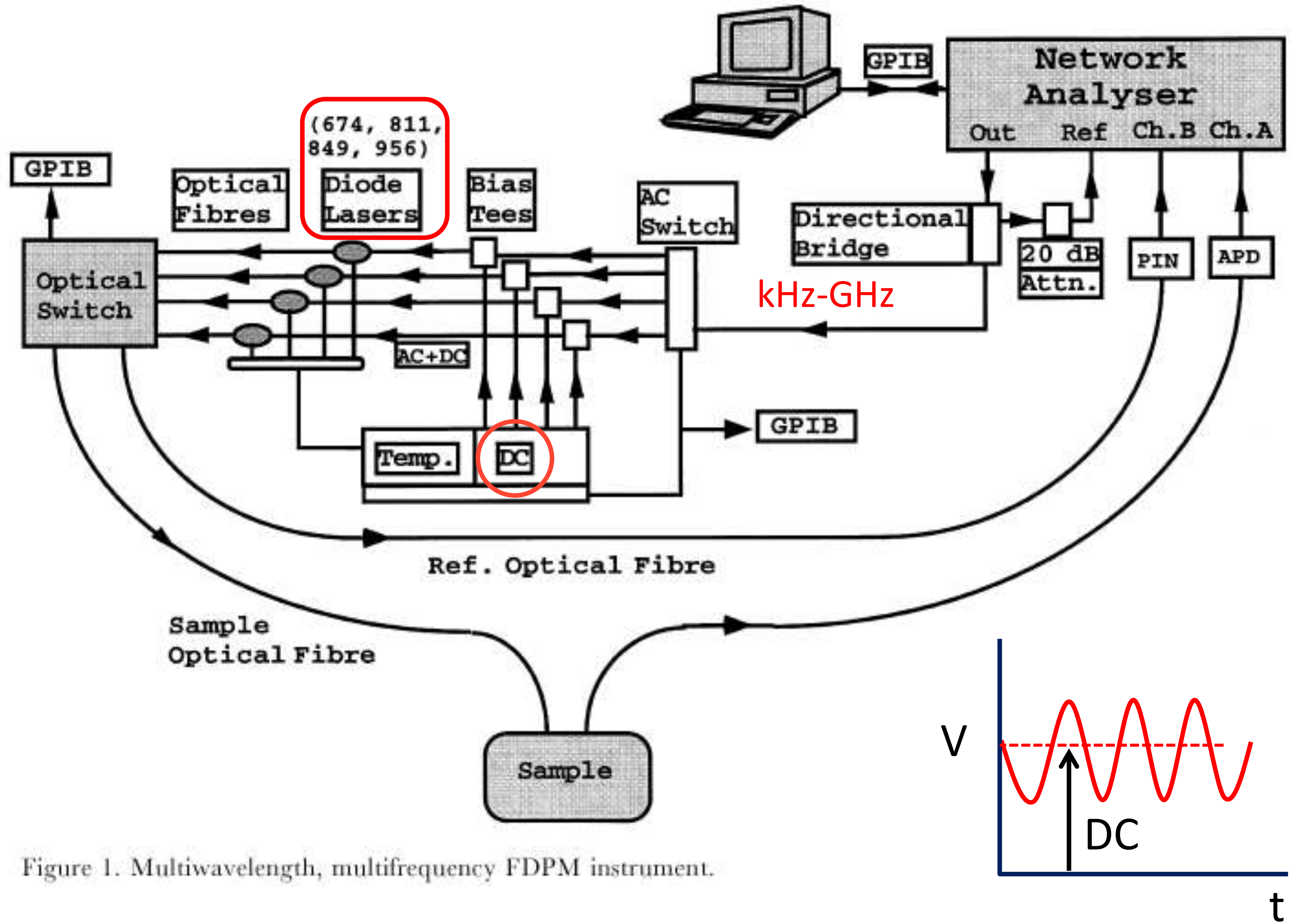


Figure 1. Multiwavelength, multifrequency FDPM instrument.

Calculations

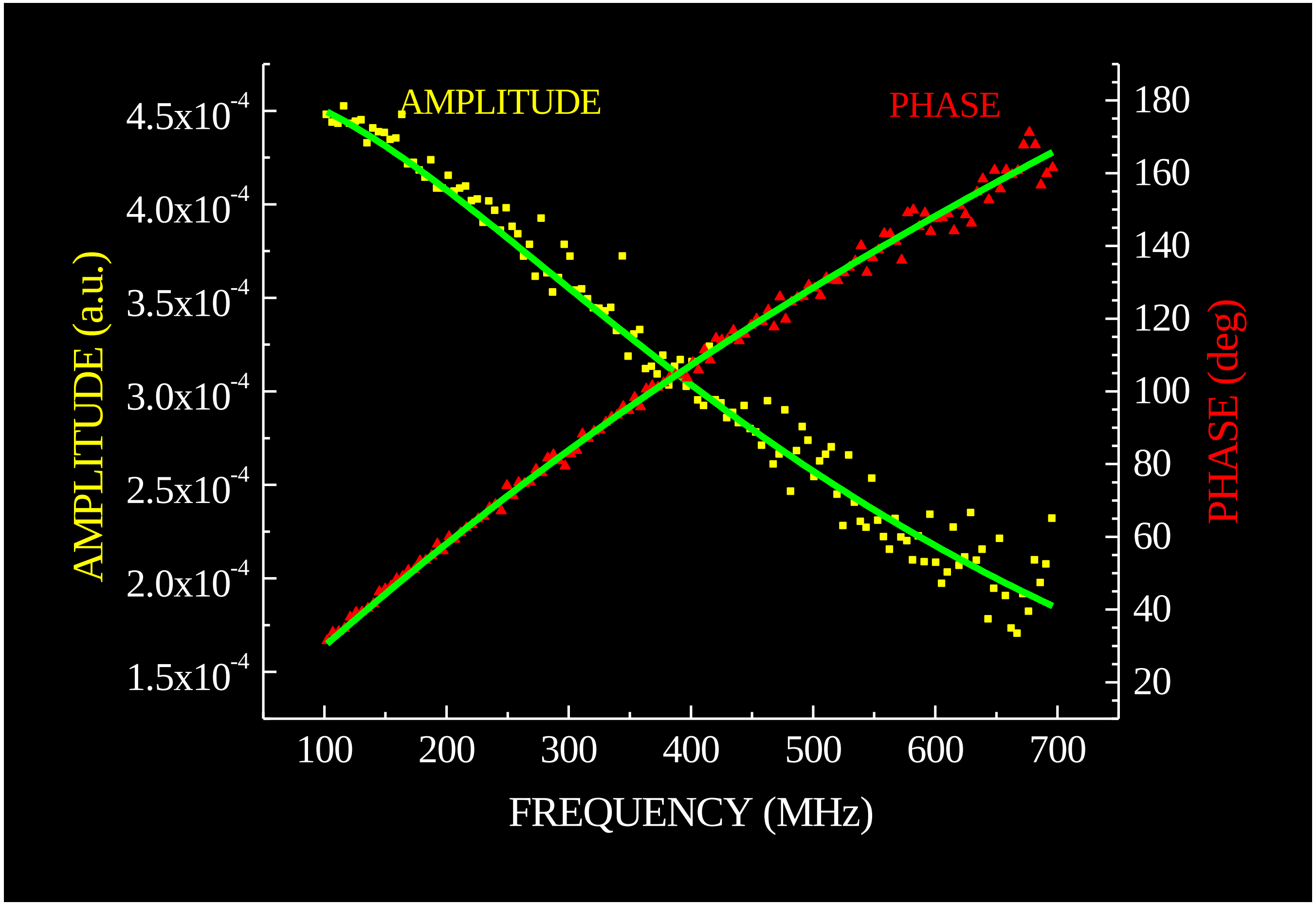
Data

DIFFUSION FIT

absorption and reduced scattering coefficients

concentrations of major chromophores

simultaneous fit

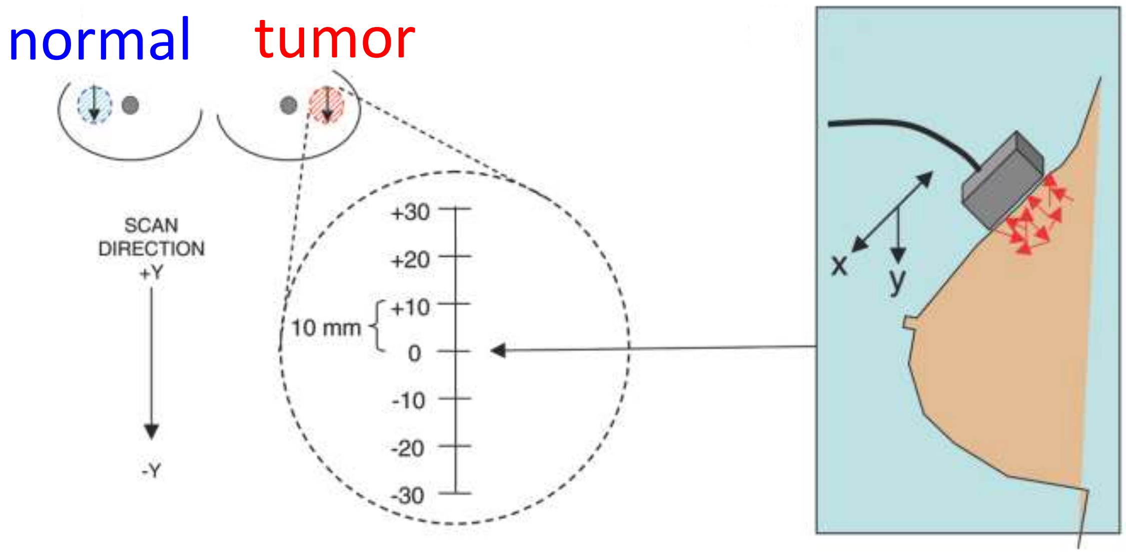


(courtesy F. Bevilacqua)

Optical scanning of breast tissue

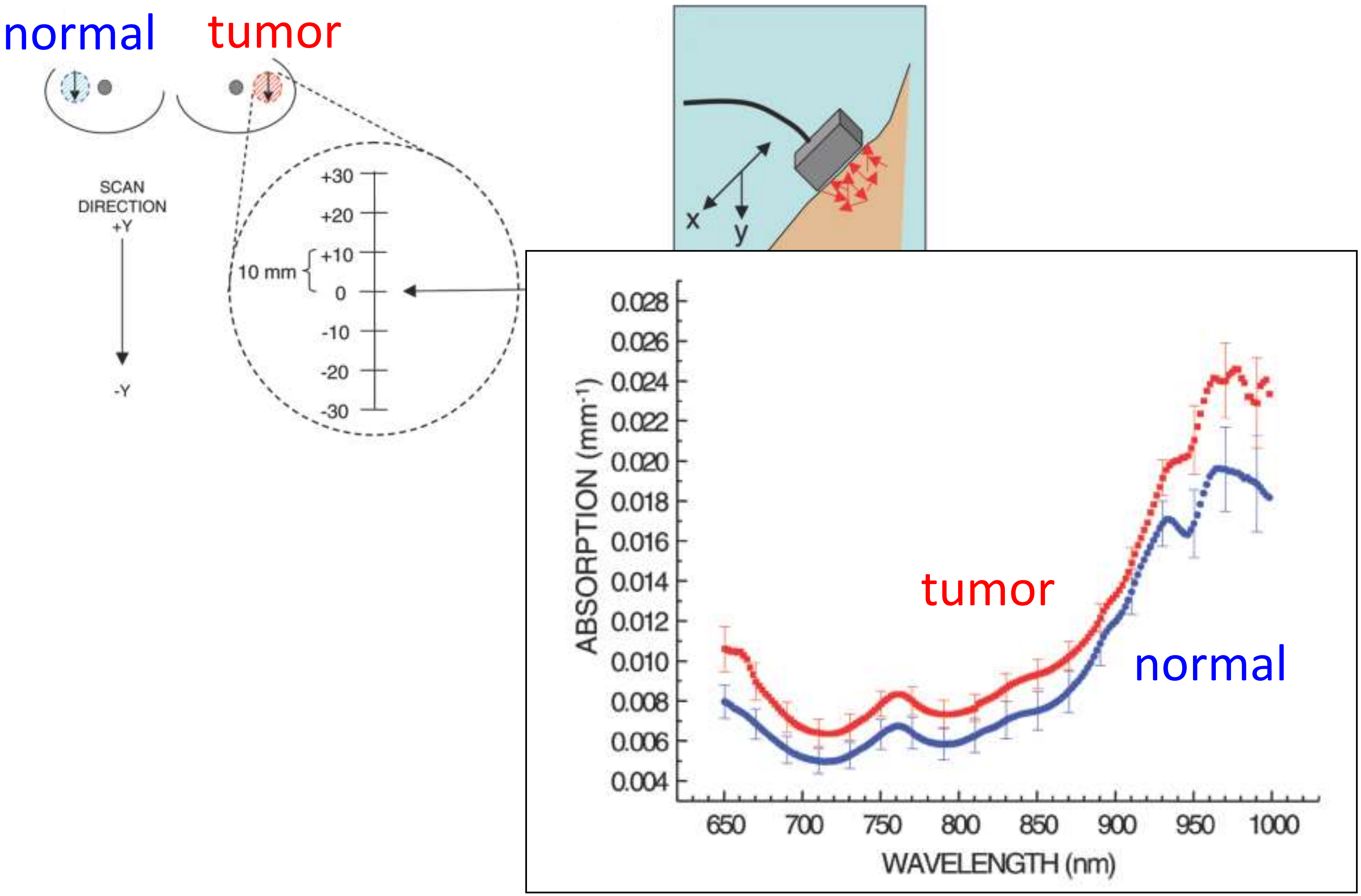
Pham, TH., et al. Review of Scientific Instruments, 71 , 1 – 14, (2000).
Bevilacqua, F., et al. Applied Optics, 39, 6498-6507, (2000).
Jakobowski et al., J. Biomed. Opt., 9(1), 230-238 (2004).

Diffuse optical scanning of tumor vs. normal



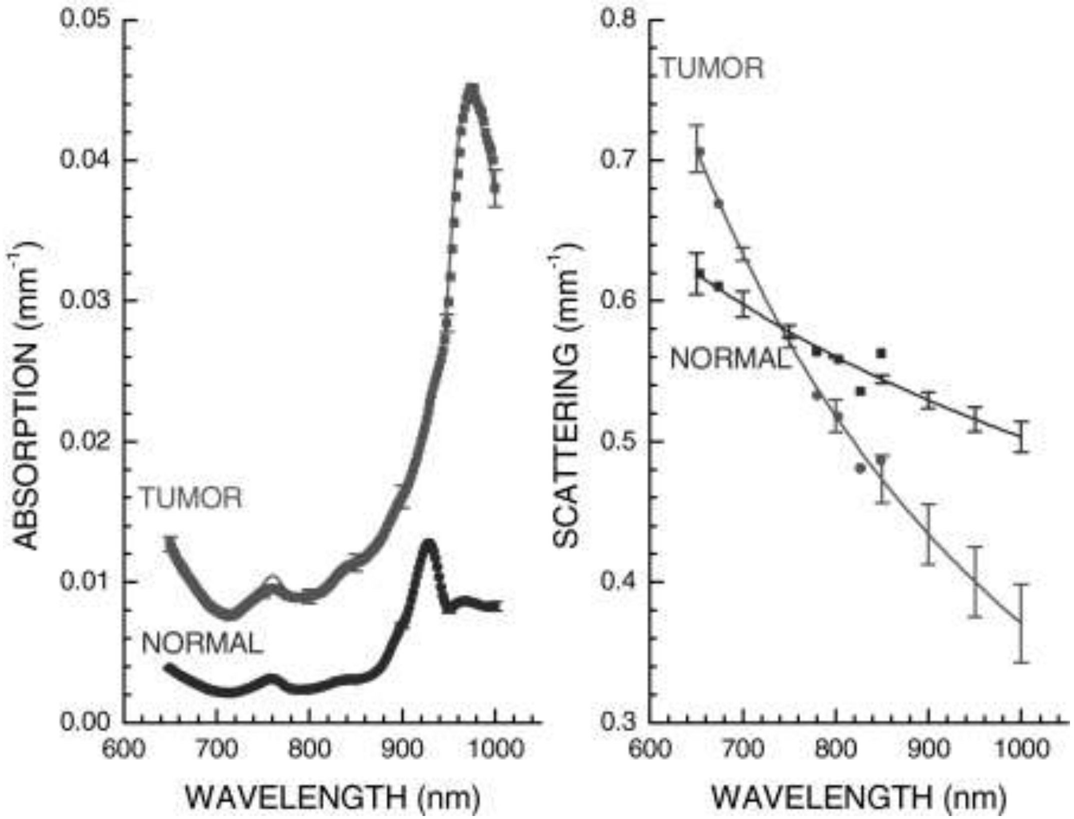
Tromberg et al., *Breast Cancer Research* 2005, 7:279-285 (DOI 10.1186/bcr1358)

Diffuse optical scanning of tumor vs. normal



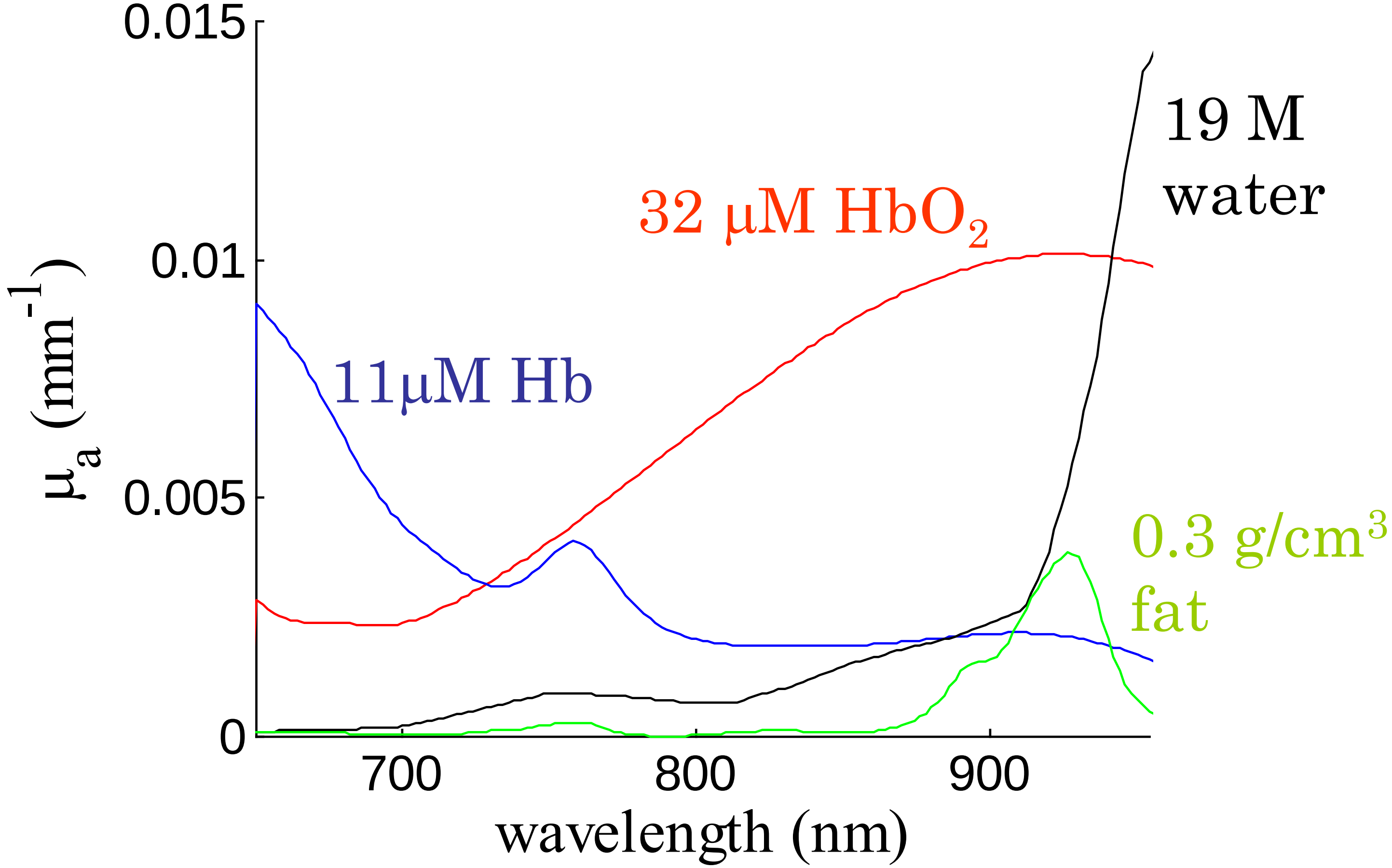
Tromberg et al., *Breast Cancer Research* 2005, 7:279-285 (DOI 10.1186/bcr1358)

Tumor vs. normal: contrast in both modalities



Cerussi et al., *Proc. Natl. Acad. Sci.* U S A. 2007 March 6; 104(10): 4014–4019.

Physiological components of fit



Choosing the most diagnostic parameters

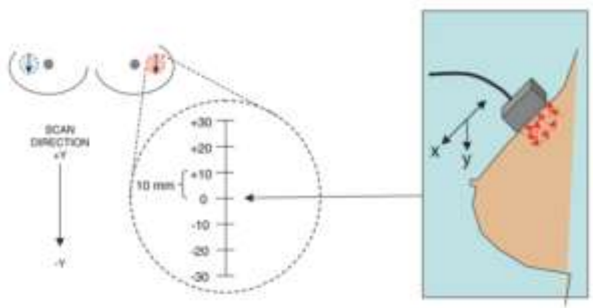


Table 1

Physiological properties of normal breast and malignant tumors (12 subjects, aged 30–39 years)

	Normal		T_{PEAK}		*P*
Parameter	Mean	Median	Mean	Median	Wilcoxin
ctHHb	6.73 ± 2.08	6.57	15.3 ± 8.16	12.6	0.005
ctO_2Hb	18.6 ± 6.9	18.9	33.3 ± 12.0	32.9	0.002
%Lipid	55.5 ± 8.7	54.9	30.6 ± 13.7	24.2	0.0003
$\%H_2O$	27.5 ± 12.1	25.4	49.9 ± 25.4	44.2	0.014
Scatter power	0.800 ± 0.362	0.830	1.17 ± 0.503	1.22	0.065

ctHHb, deoxygenated hemoglobin concentration; ctO_2Hb, oxygenated hemoglobin concentration; T_{PEAK}, peak tumor values.

Tromberg et al., *Breast Cancer Research* 2005, 7:279-285 (DOI 10.1186/bcr1358)

Pre and post-menopausal breast examples

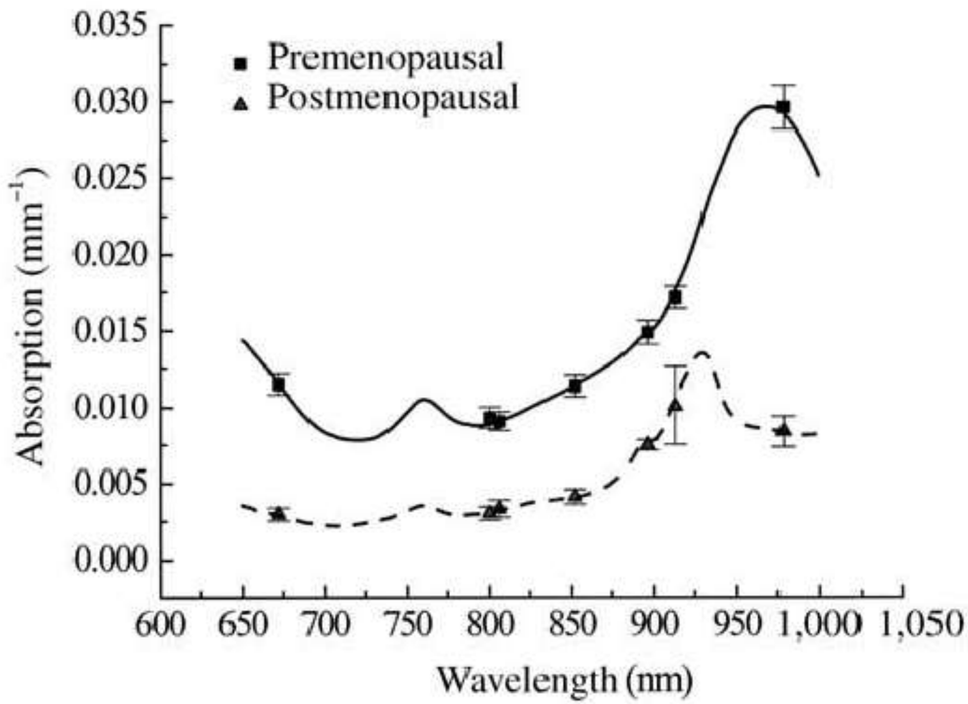
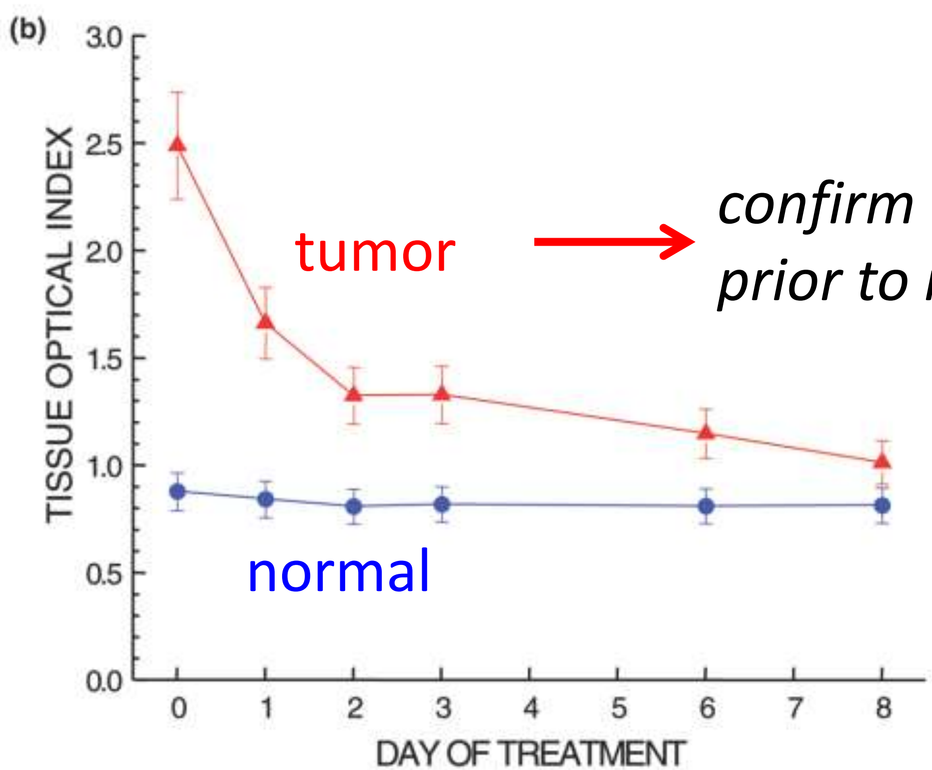


Figure 2. Measured absorption spectra for a 32-year-old premenopausal subject (■, solid line) and a 54-year-old postmenopausal subject (▲, dashed line). Points represent the average of several measurements, and lines represent a least-squares fit (extrapolated to all wavelengths) based on the assumption that breast absorption is due only to Hb-R, Hb-O_2, H_2O, and lipids.

A. E. Cerussi, D. Jakubowski, N. Shah, F. Bevilacqua, R. Lanning, A. J. Berger, D. Hsiang, J. Butler, R. F. Holcombe, and B. J. Tromberg, Spectroscopy enhances the information content of optical mammography, **Journal of Biomedical Optics**, 7(1), 60-71 (Jan. 2002).

Tumor response to neoadjuvant therapy

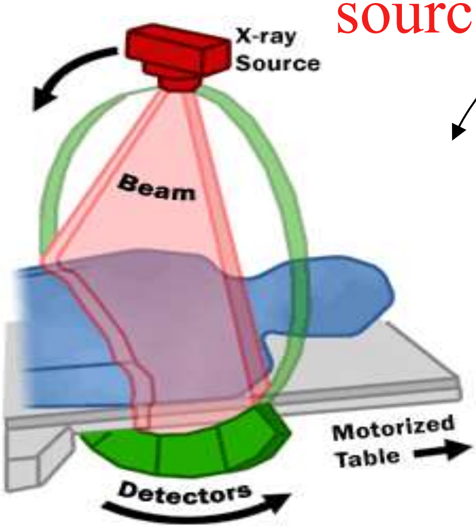


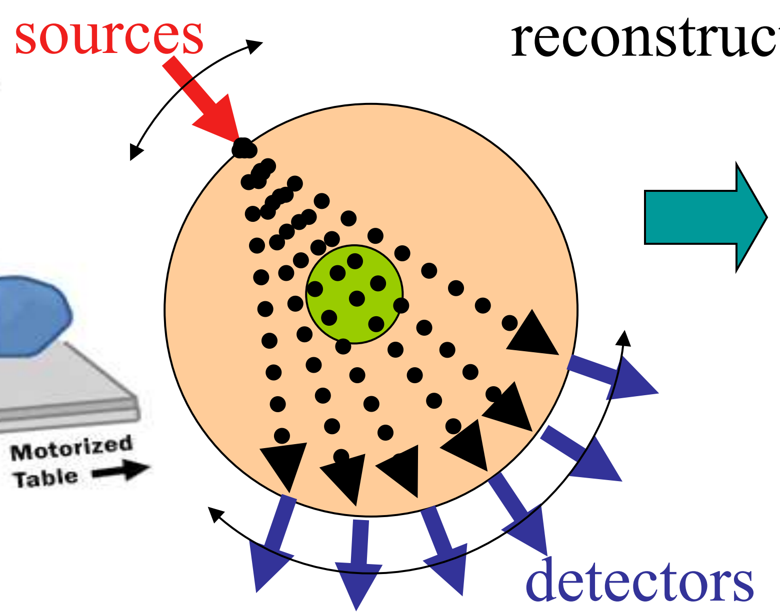
confirm reduction in tumor prior to radiation therapy

Tromberg et al., *Breast Cancer Research* 2005, 7:279-285 (DOI 10.1186/bcr1358)

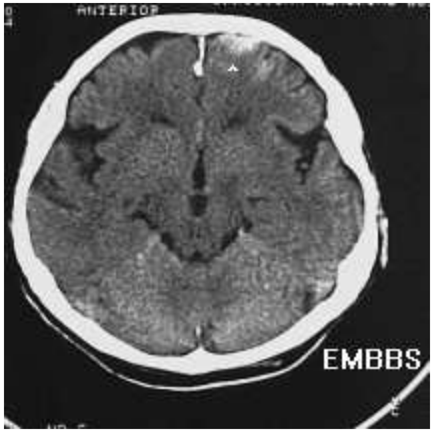
Breast imaging: Computed Tomography

CT-scan (x-ray)





numerical reconstruction

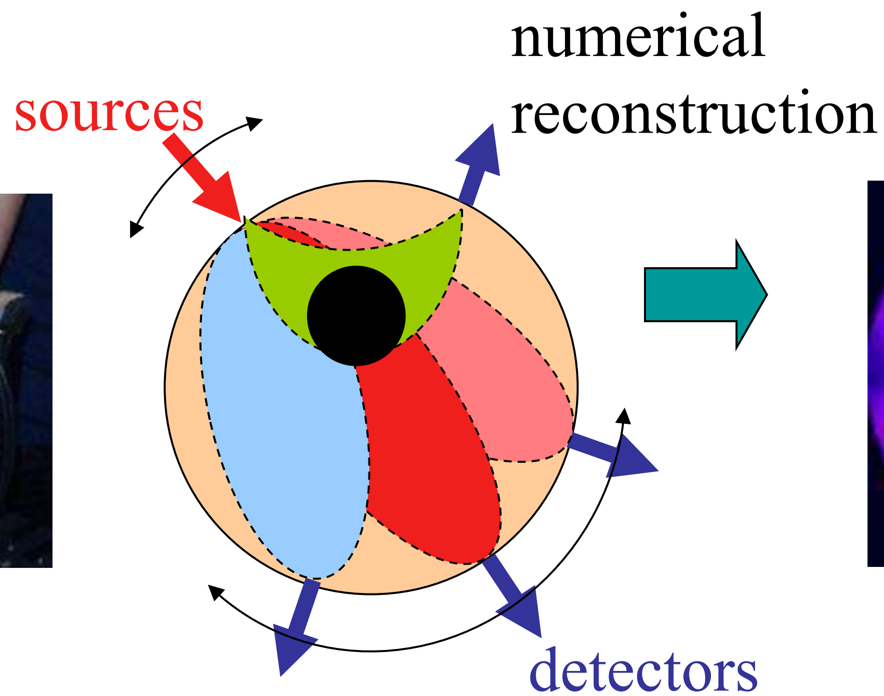


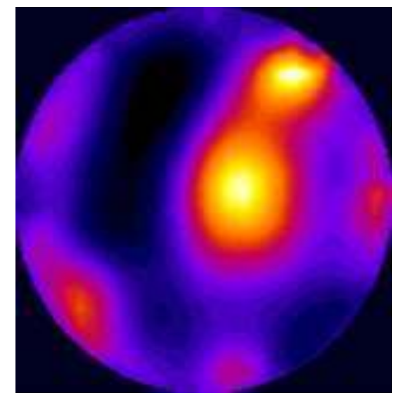
scattering << absorption ⇒ paths = straight lines

(courtesy F. Bevilacqua)

Breast imaging: *Optical* Computed Tomography

near-infrared light





http://www.medphys.ucl.ac.uk/research/borg/index.htm

scattering >> absorption ⇒ broad probability of paths
⇒ challenges: ill-posed problems (non-unique solution)
poor resolution

(courtesy F. Bevilacqua)

Tomography in the frequency domain

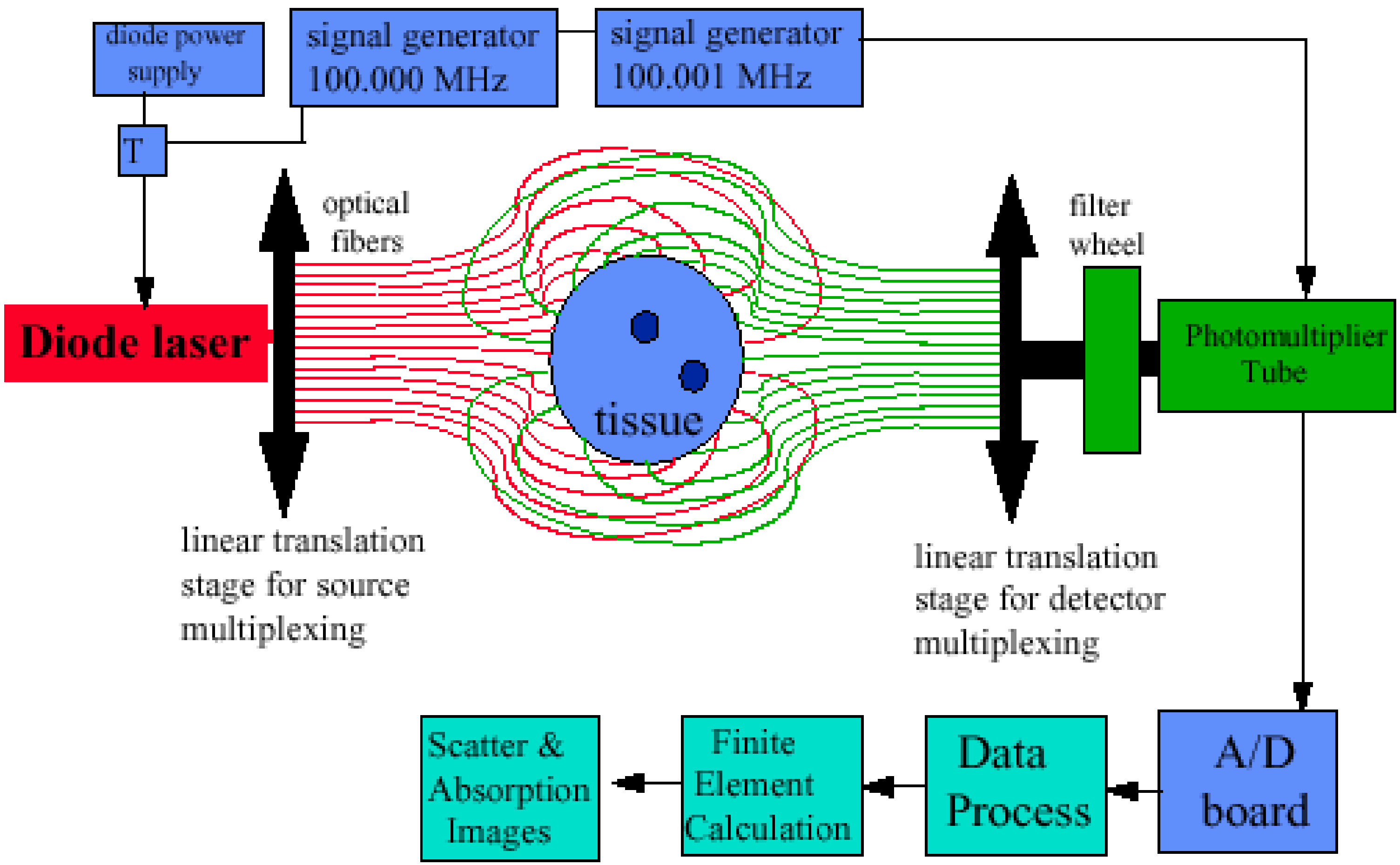
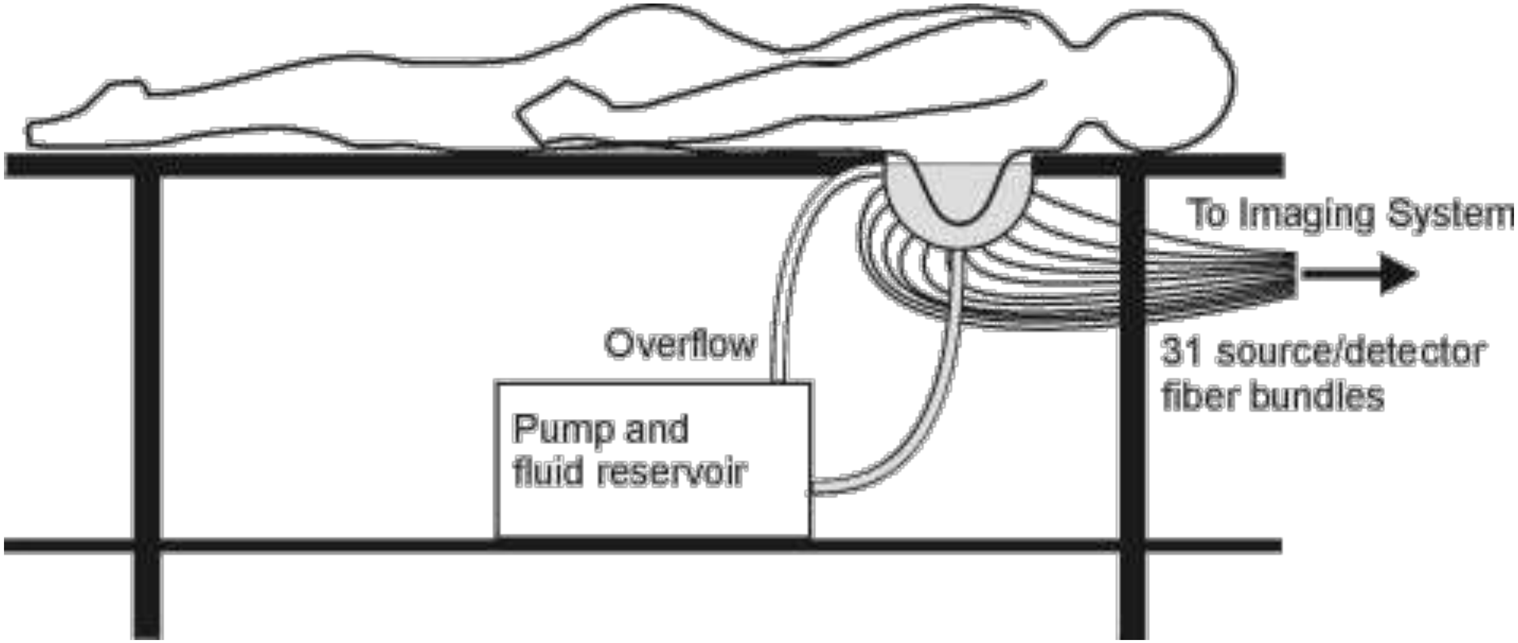


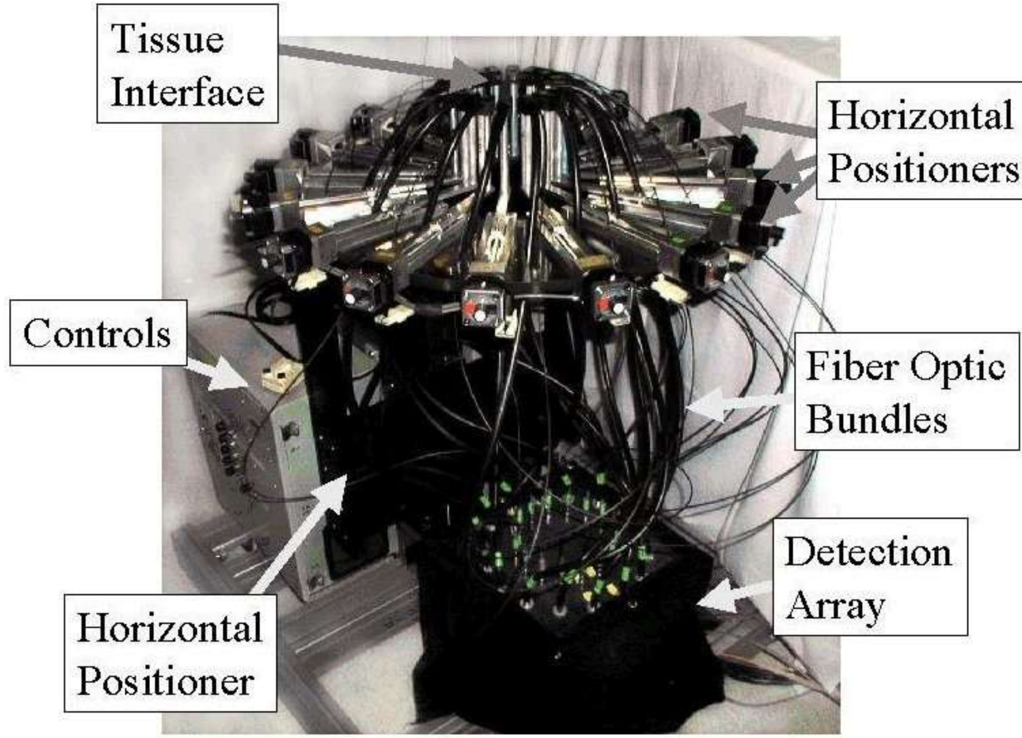
Fig. 1. Schematic of the automated imaging instrument including hardware and software processing. Source optical fibers are indicated in red and detector optical fibers in green.

B. W. Pogueet al, Opt. Express **1**, 391-403 (1997), http://www.opticsexpress.org/abstract.cfm?URI=OPEX-1-13-391

Optical mammography: diffuse imaging

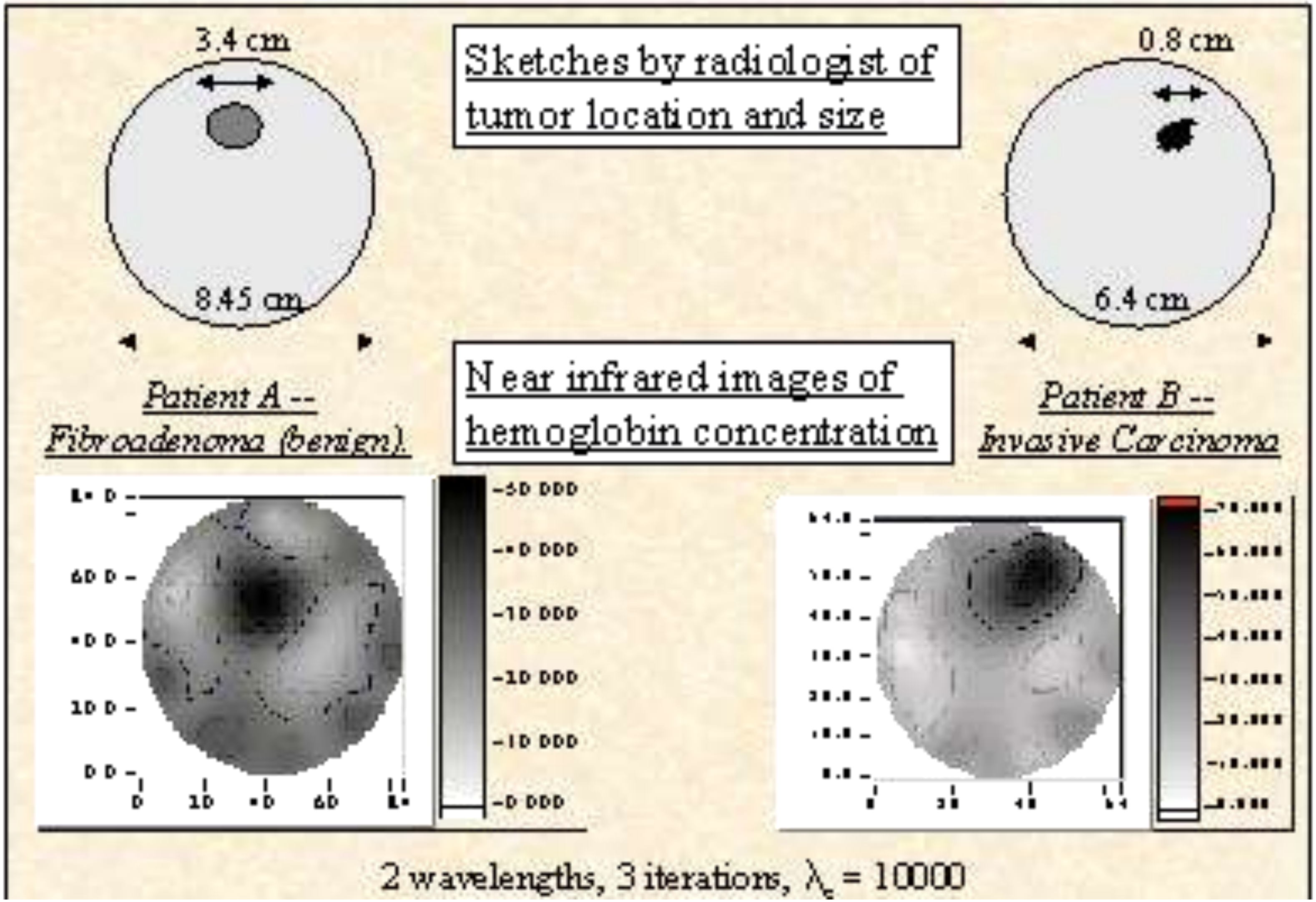


http://www.ucl.ac.uk/medphys/research/borl/imaging/monstir/breast

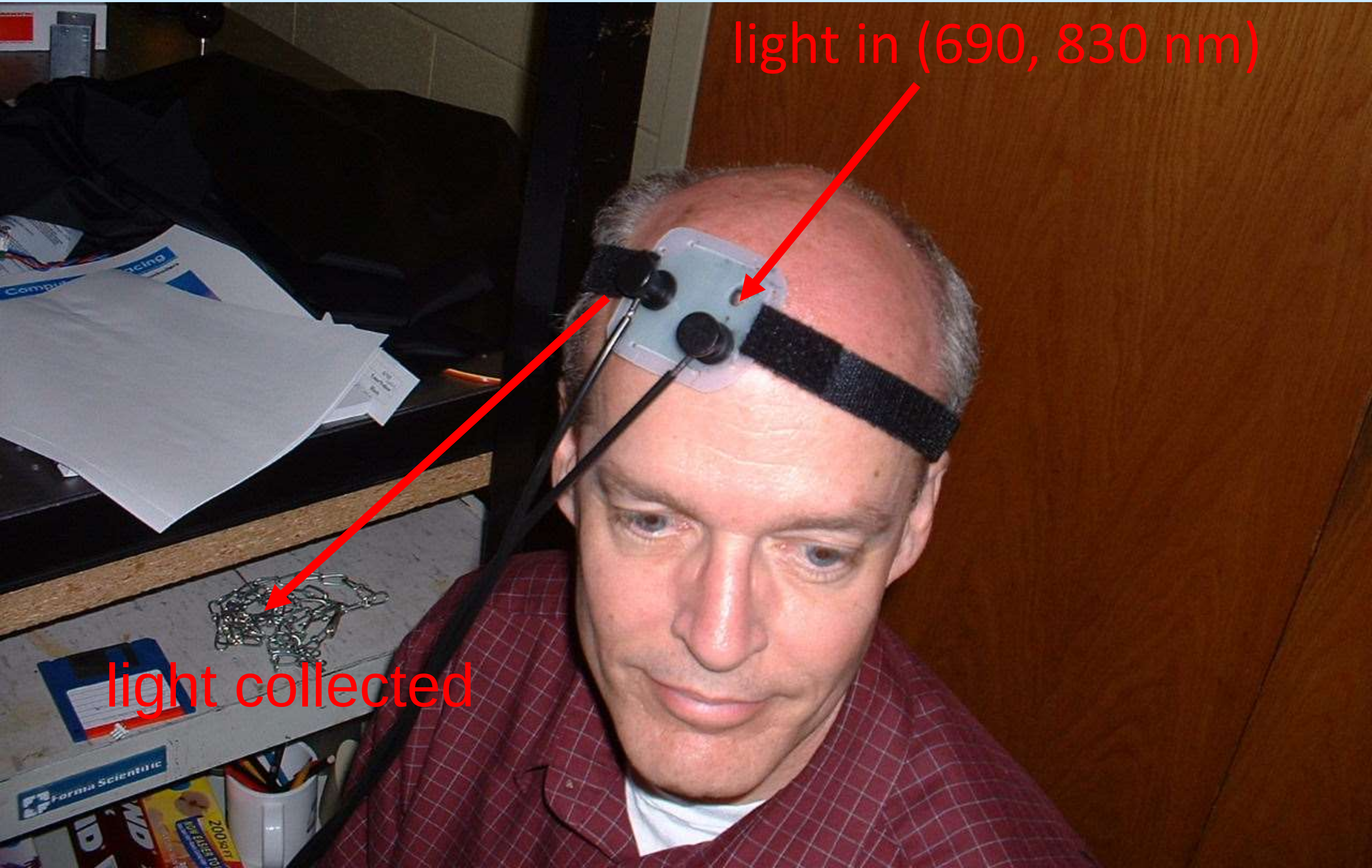


http://www-nml.dartmouth.edu/nir/instrumentation.html http://www-nml.dartmouth.edu/biomedprg/NIR/sld013.htm

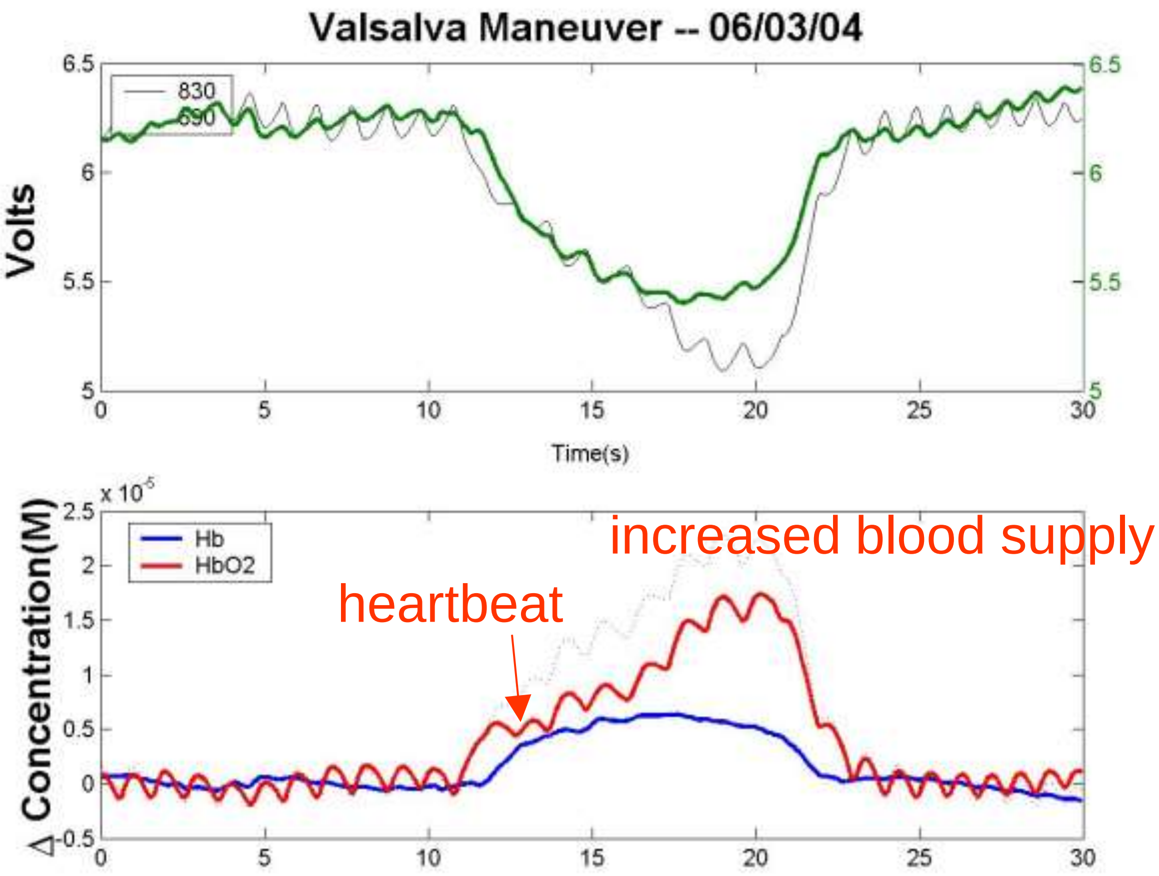
NIR Images of Volunteers with tumors



Application #3: brain hemodynamics

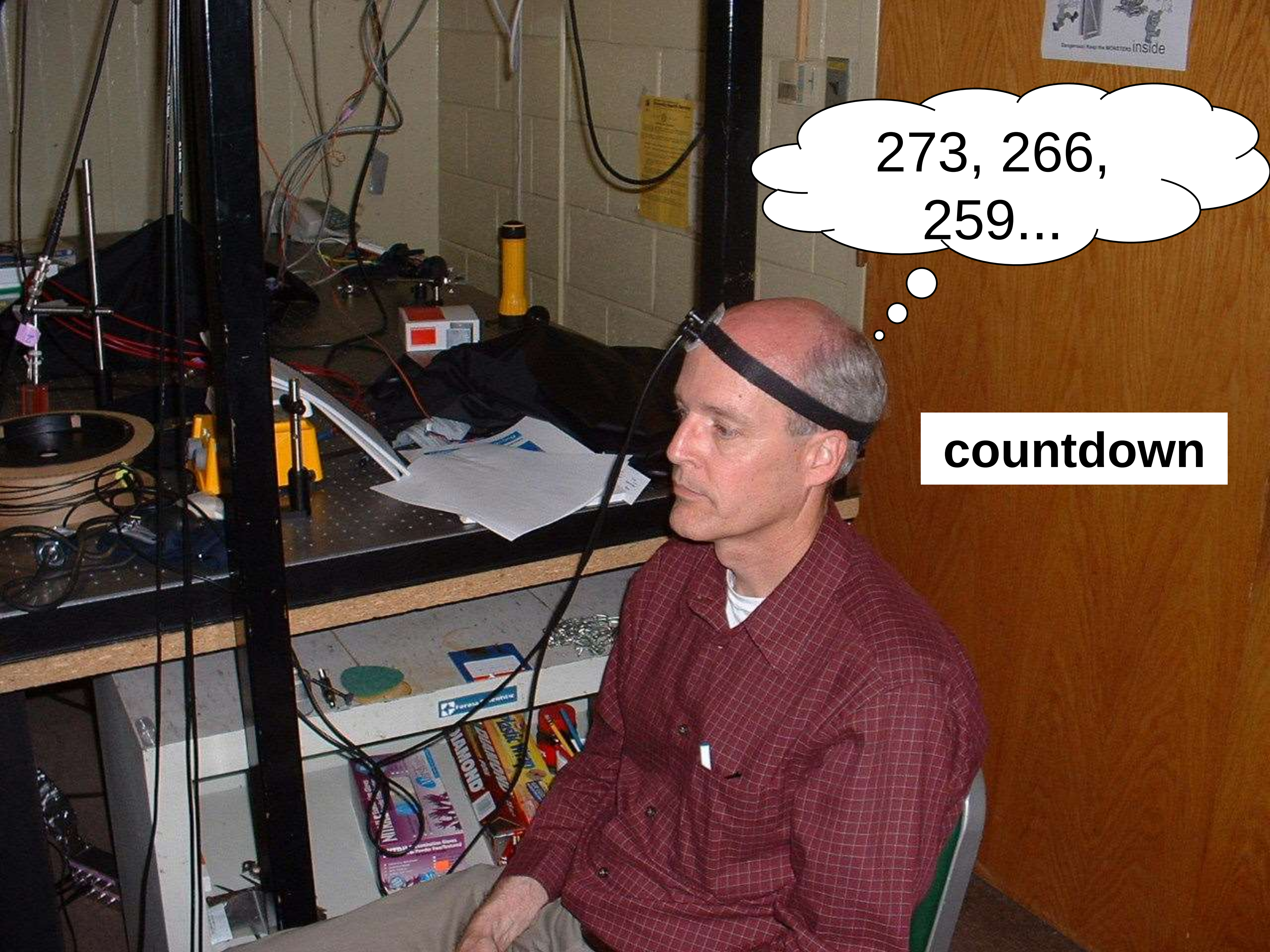


Noninvasive monitoring of hemodynamics

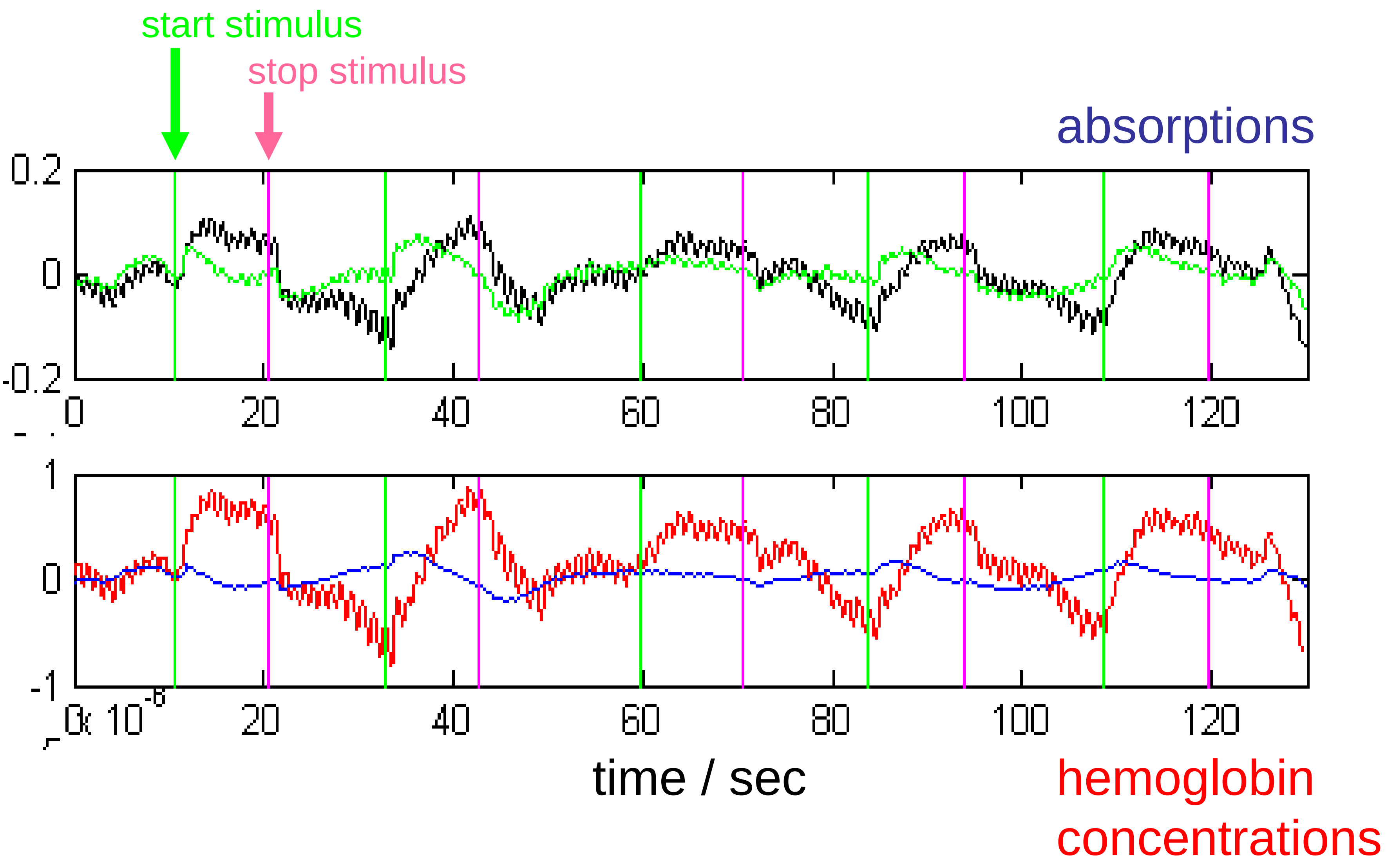


optical power measurements

oxy- and deoxy-hemoglobin concentration *changes*


273, 266,
259...
countdown

Single subject countdown timecourse



Typical headpiece for adults

optical fiber bundles

First Nearest Neighbors 1.3 cm

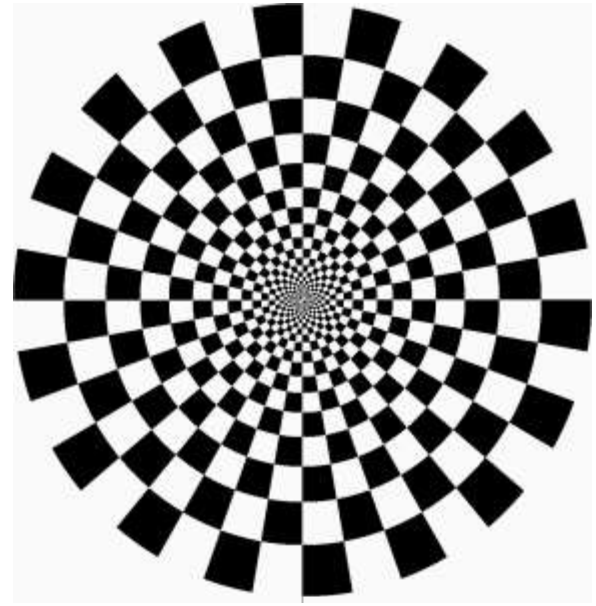
Second Nearest Neighbors 3 cm

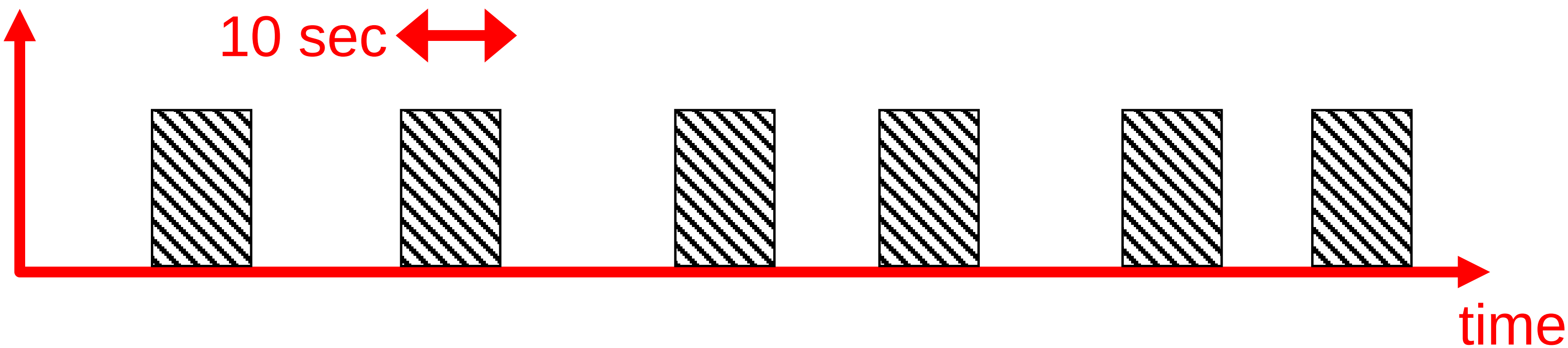
Third Nearest Neighbors 3.9 cm

Visual Stimulation Protocol

- 6 stimulus periods of pattern reversal at 10 Hz

  based upon code by Brian White and Joseph Culver, Washington University (St. Louis)



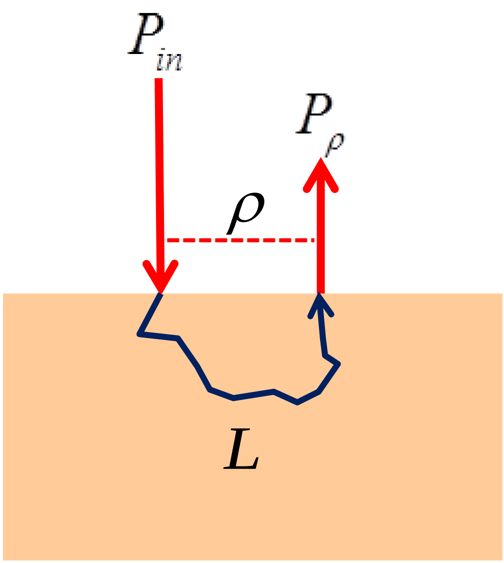


Simplified model of reflectance vs. time

$$\frac{P_{out}}{P_{in}} = e^{-\mu_a L}$$

Simplified model of reflectance vs. time

$$\frac{P_{out}}{P_{in}} = e^{-\mu_a L}$$



$$\frac{P_{\rho}}{P_{in}} = G(\mu_a, \mu_s', \rho) e^{-\mu_a \langle L \rangle}$$

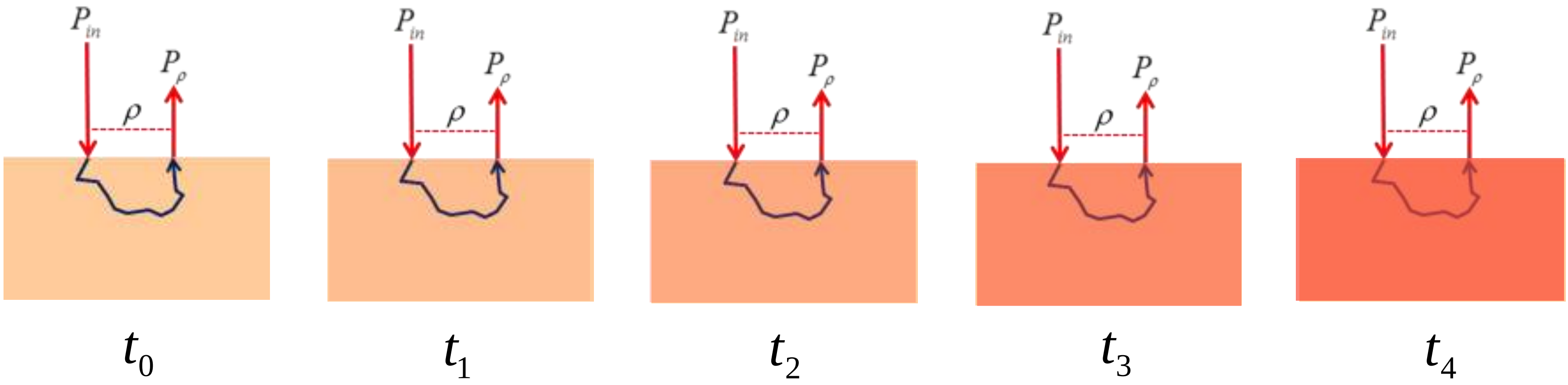
$$= G(\mu_a, \mu_s', \rho) e^{-\mu_a [DPF \cdot \rho]}$$

"geometry factor"

$$\langle L \rangle \approx DPF \cdot \rho$$

e.g. *DPF = 6*

Simplified model of reflectance vs. time

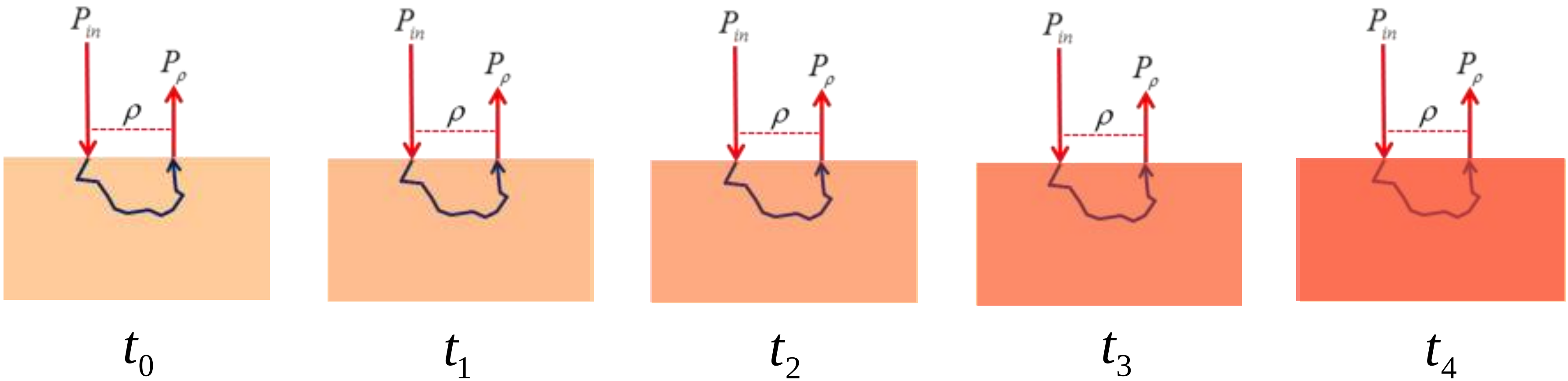


sequential measurements: $$\frac{P_\rho(t)}{P_{in}} = G(\mu_a, \mu_s', \rho)e^{-\mu_a(t)[DPF\cdot\rho]}$$

changes between measurements: $$\frac{P_\rho(t)}{P_\rho(t_0)} = e^{-[\mu_a(t)-\mu_a(t_0)][DPF\cdot\rho]}$$

$$= e^{-\boxed{\Delta\mu_a(t)}[DPF\cdot\rho]}$$

Simplified model of reflectance vs. time



$$\frac{P_\rho(t)}{P_\rho(t_0)} = e^{-\Delta\mu_a(t)\cdot[DPF\cdot\rho]}$$

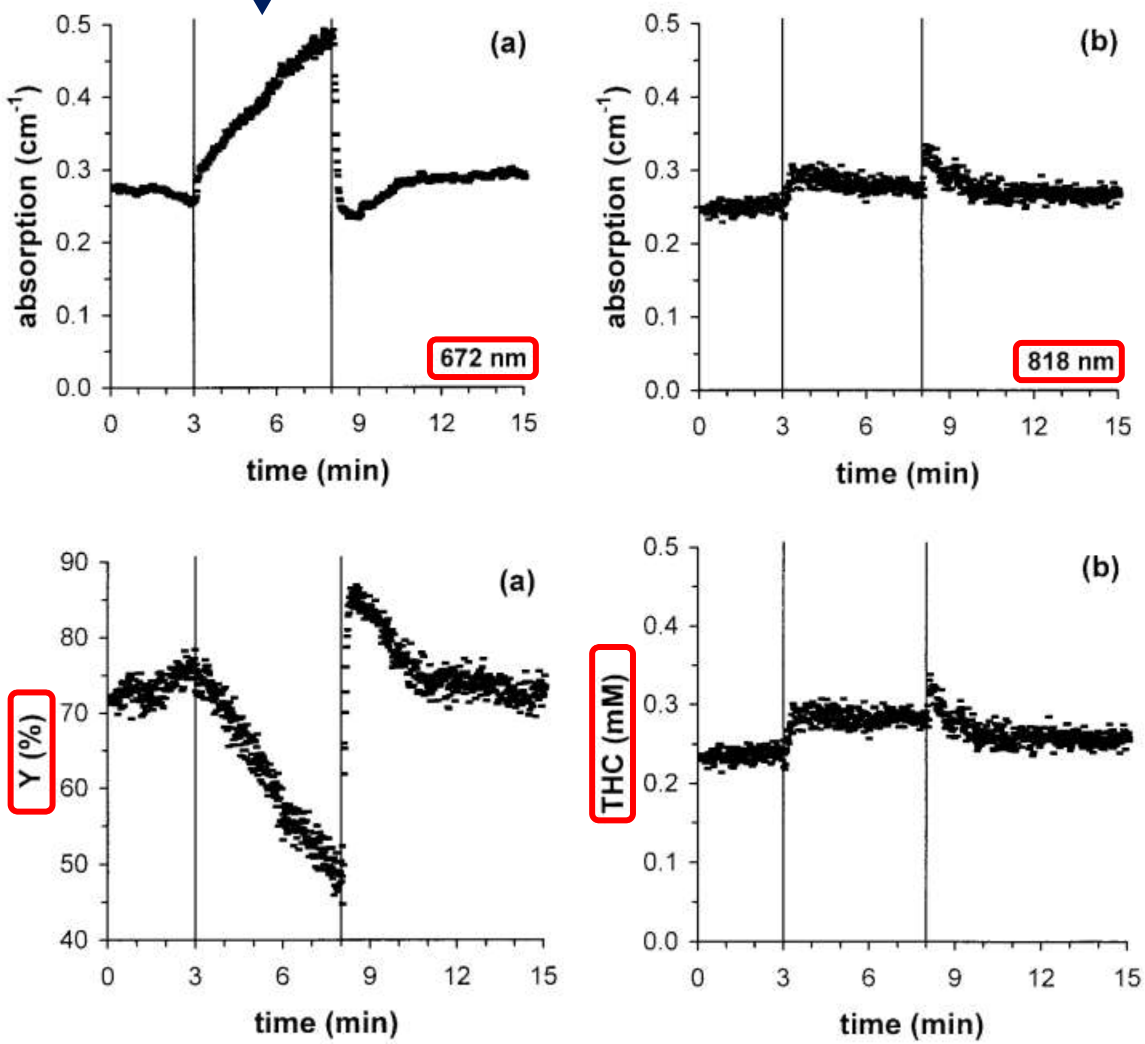
"Modified Beer-Lambert Law (MBLL)"

$$\frac{P_{out}}{P_{in}} = e^{-\mu_a L}$$

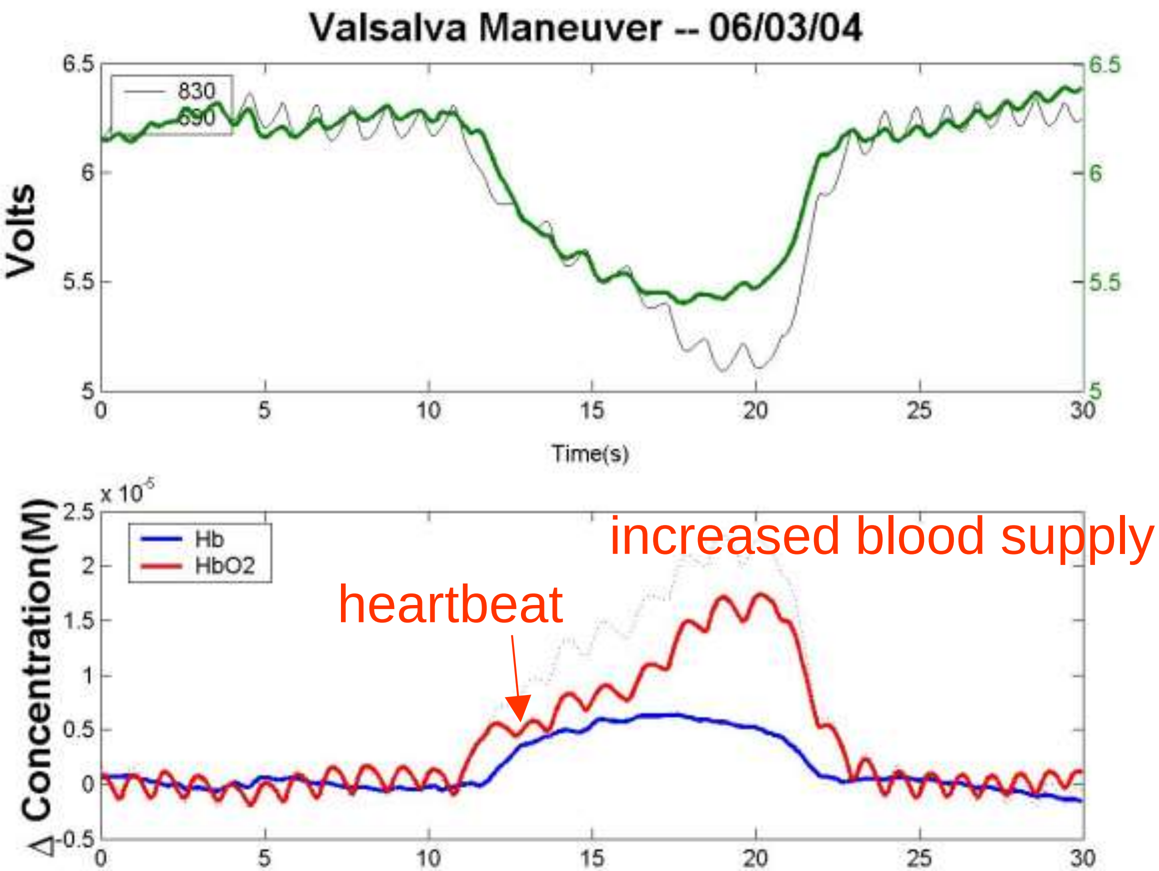
Reminder: time domain *in vivo* blood measurements

pressure cuff on

absolute
measurements



Reminder: CW *in vivo* measurements

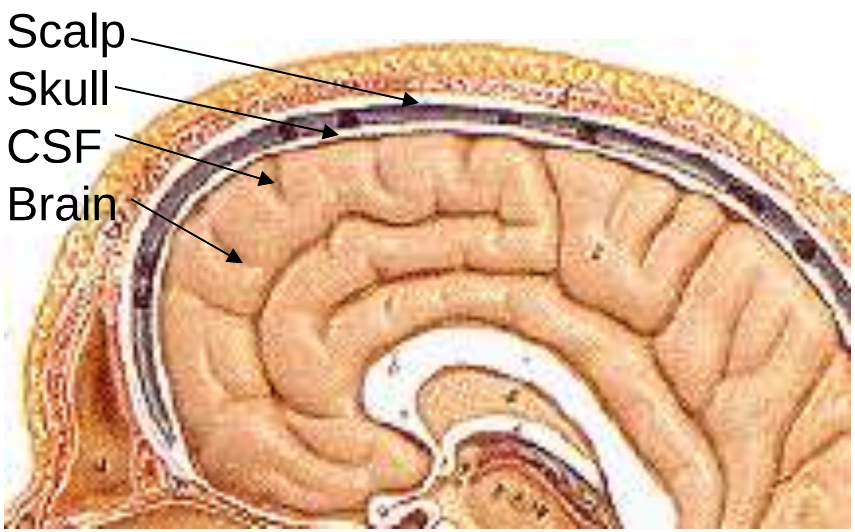


optical power measurements

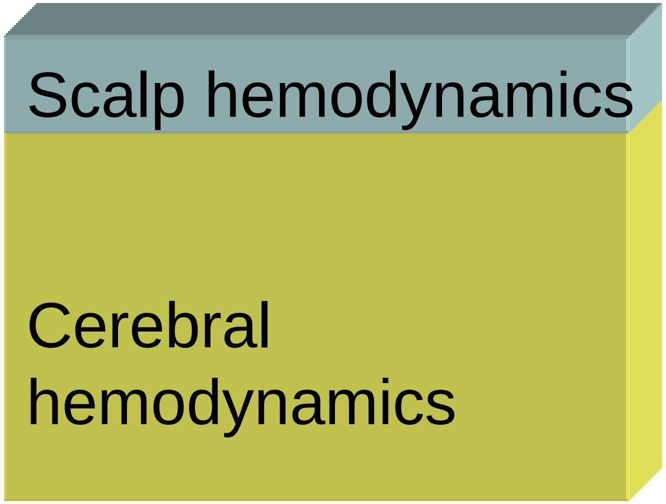
oxy- and deoxy-hemoglobin concentration *changes*

The optical geometry

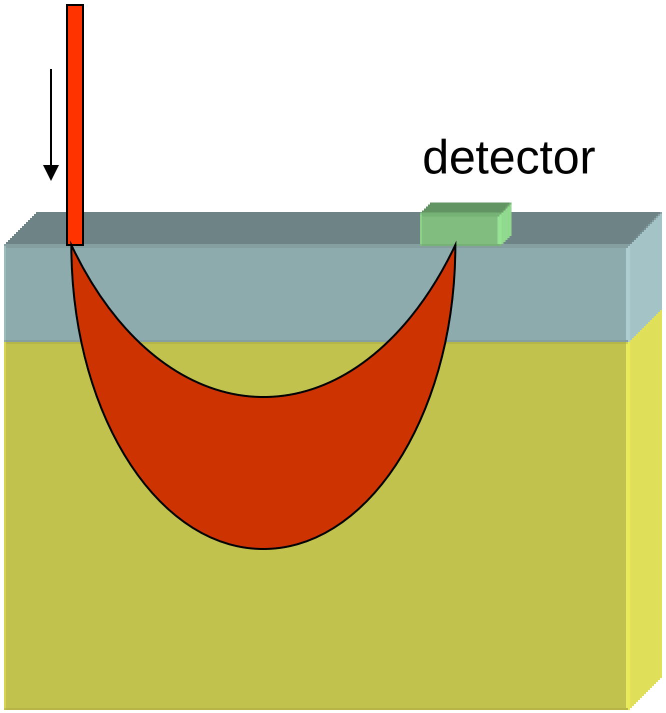
A real head



A physicist's head



Problem: not all blood is in the brain!

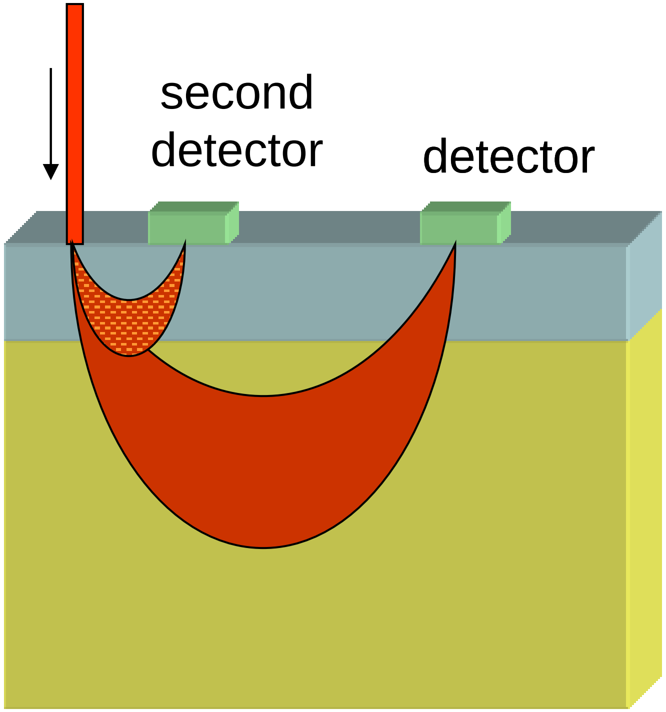


Measurement sensitive to both scalp and brain hemodynamics

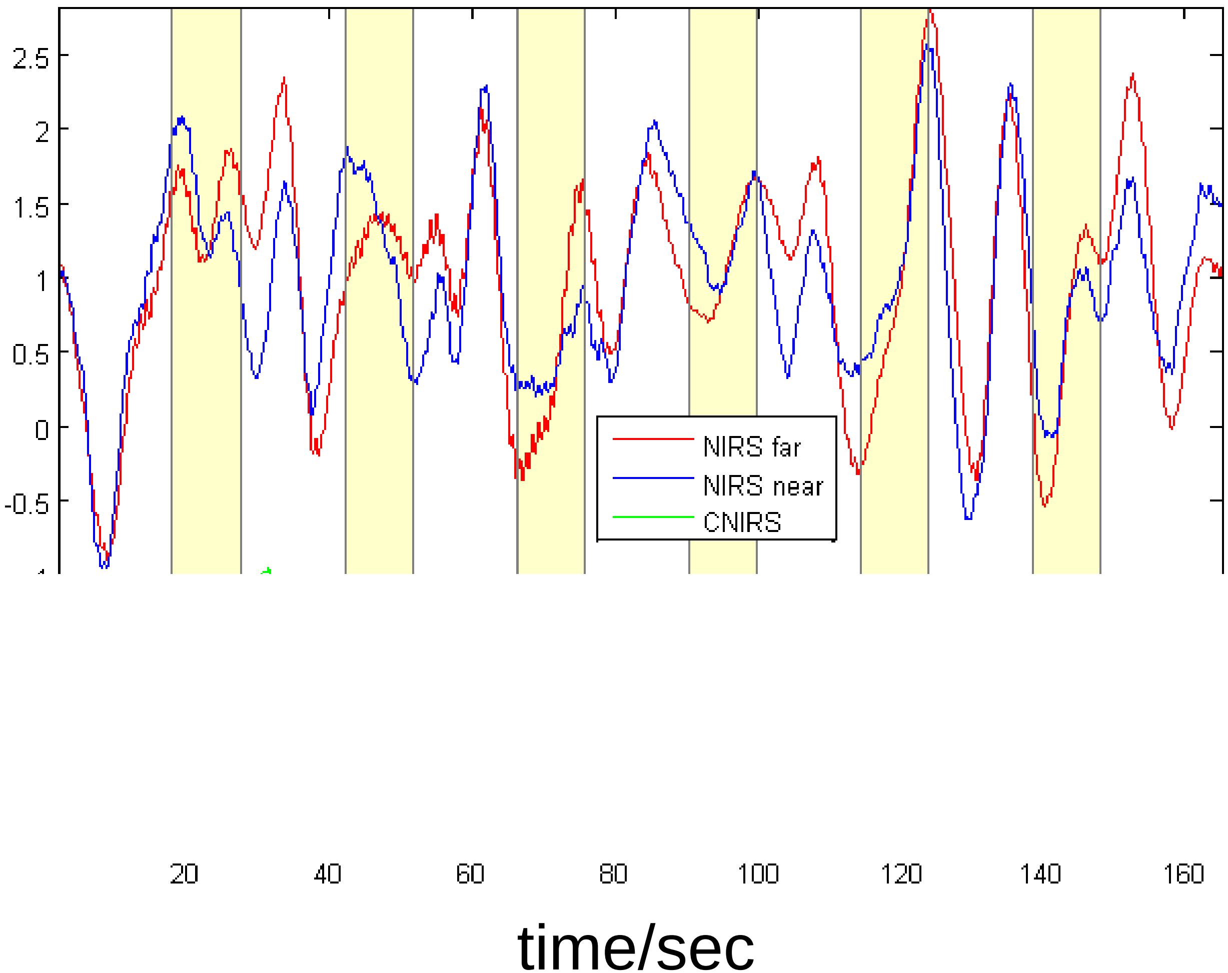
Want to isolate brain-specific trends

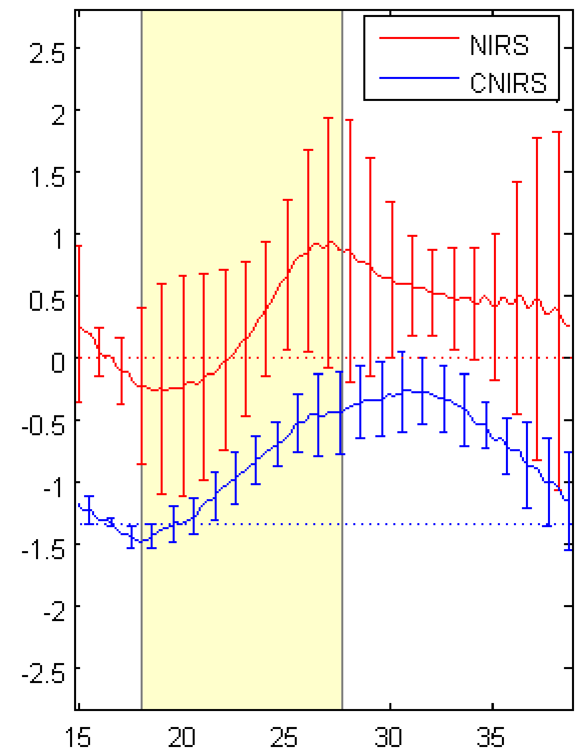
Saager et al., NeuroImage 55(4), 1679--1685 (April 2011)

2 detectors, 2 different depths probed!

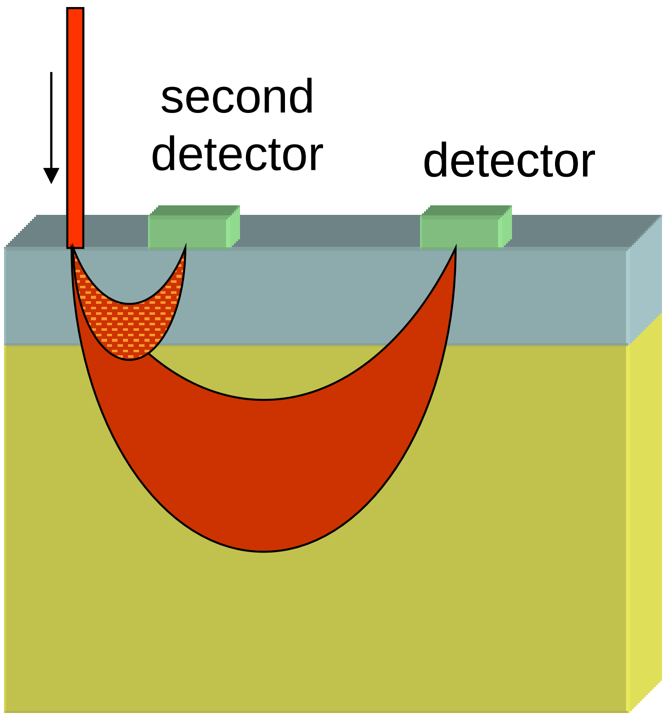


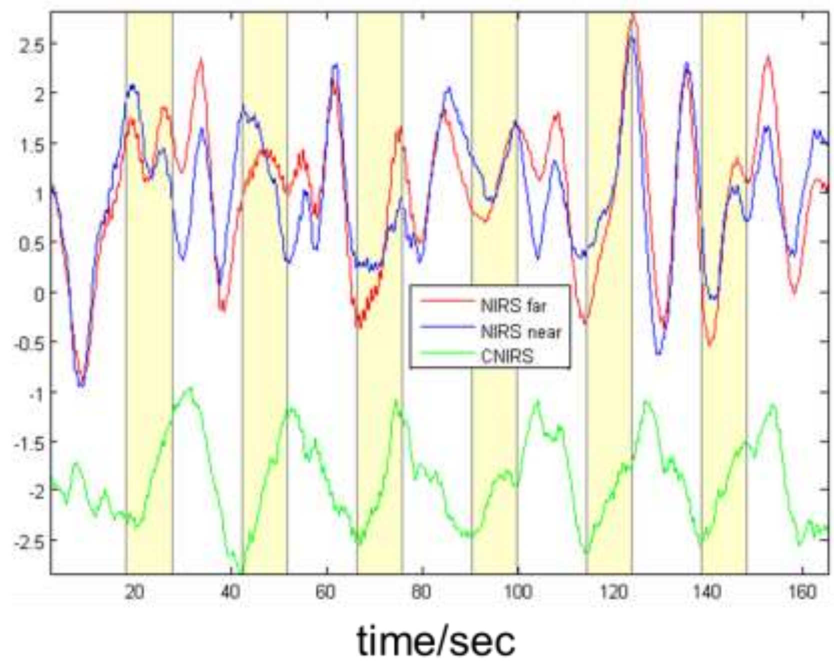
Improving signal-to-noise by subtracting “scalp” signal





Good summary case for diffuse spectroscopy

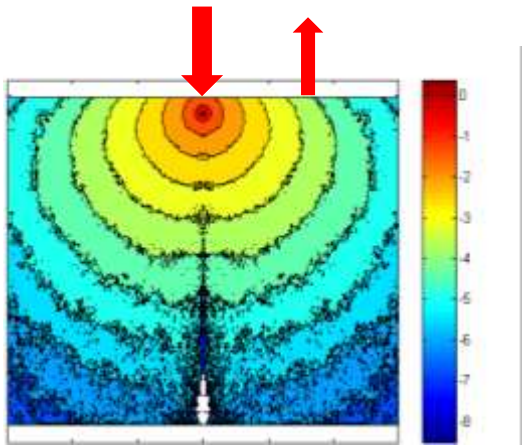




Reviewing the roadmap

review of basic concepts from last time

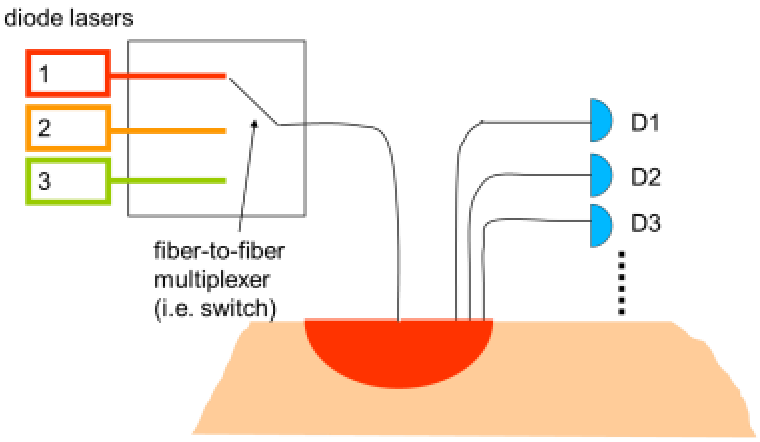
the Virtual Tissue Simulator



reflectance measurements: three types

- steady-state
- pulsed
- sinusoidally-modulated *("frequency domain")*

instrument design considerations



various applications

To learn more

Tuesdays in January (7.1, 14.1, 21.1, 28.1), 2:00 pm, IPHT Sitzungssaal

Lecture 2 - Turbid tissue optics I: *Introduction*

Lecture 3 - Turbid tissue optics II: *Instrumentation and measurements*

Lecture 4 - Turbid tissue optics III: *(More) Applications*

Lecture 5 - A different view of turbidity: *elastic scattering analysis*