Outline

• Principles/Light-Biomolecular Interactions
  
  \textit{optics/photonics: history}

  \textit{“tissue optics”: theoretical & experimental}

• Techniques & Applications

\begin{align*}
\text{diagnostic/analytic} & \quad \text{therapeutic} \\
\text{optical spectrosopies} & \quad \text{optical imaging} & \quad \text{light - based} & \quad \text{optically- guided}
\end{align*}

• Convergence with nanotechnology
1903 Nobel Laureate in Medicine
in recognition of his contribution to the treatment of diseases,
especially lupus vulgaris,
with concentrated light radiation,
whereby he has opened a new avenue for medical science
1960  **(Ruby) Laser** invented by Maiman

1961  **Use of Ruby Laser** in surgery

*Dr. Leon Goldman performs ruby laser treatment of melanoma*
Google “medical laser Images”

many instruments/systems   many applications
Google “light treatment images”

many other instruments/systems mostly skin applications
Laser surgery
PUVA therapy
Photodynamic therapy
Tissue Welding
Laserthermia
Laser-enhanced drug uptake
Photo-stimulation
Corneal refractive surgery
Retinal coagulation
Birth marks
Skin remodelling
Myocardial reperfusion

precise/minimally-invasive/selective
Basic law of photobiology

Any (photo)biological effect requires absorption of energy from the photon by a component of the biological system.
Any phototherapeutic effect depends strongly on:

- **wavelength:**
  - determines
  - a) chromophores that absorb the energy
  - b) volume (depth) within which the energy is deposited
e.g.

**CO₂ laser** ($\lambda \sim 10.6 \, \mu m$)

- 1/e penetration depth $\sim 10 \, \mu m$
- Water is the primary absorber

$\rightarrow$ precision, any hydrated tissue

**Nd:YAG laser** ($\lambda \sim 1.06 \, \mu m$) 1/e penetration depth $\sim 1 \, mm$

- Blood is the primary absorber

$\rightarrow$ bulk tissue, blood coagulation (hemostatic)
- tissue composition:
  - determines a) chromophores (endogenous or exogeneous
  
b) volume (depth) of effect
  
c) biological responses to photophysical/photochemical changes
    - structural
    - functional

e.g.
- optical “pulse length”:
- determines dominant biophysical interaction mechanism(s)
Photobiomodulation

Definition:
Use of very low-power (<~10 mW), usually red/near-infrared exposure over extended time (minutes, often repeated) to modify cell/tissue function

Fundamental mechanism:
Light triggering of intrinsic metabolic pathways

Applications:
Chronic conditions: e.g. pain, joint stiffness,.. Accelerated wound healing
Others (controversial) e.g. behavioral modification
Fig. 11-2. Action spectrum for the promotion and inhibition of germination of light-sensitive lettuce seed. Percentage of germination is plotted as a function of wavelength. A control level of 50% germination was established by exposing the seed to a constant amount of red light prior to the experiment. Subsequent exposures were given at the wavelength indicated and the effectiveness for promotion or inhibition was measured. (From reference 5.)
Existence of these effects are reasonably well established at the cellular level” altered metabolic and proliferation, cell signaling, ...

Figure 3.12. Action spectra of visible monochromatic light upon growth of Saccharomyces ludwigii expressed as amount of synthesized protein in irradiated and nonirradiated cultures after 18 h incubation.

→ Complex interdependence between wavelength & power/energy density
- Some evidence *in vivo* in animal models for specific endpoints

- Few well-controlled human trials published in highly cited journals

→ *Effects are subtle,*

→ *Challenging to sort out the various contributions to the effects observed*

Nevertheless......
- Widely used in both human and veterinary practice (largest market for medical lasers/devices): physiotherapy, chiropractic, chronic care

→ No properly-controlled, rigorous, randomized clinical trials
→ Some credible reports
→ Lot of dubious claims
Definition:
Use of sub-thermal power (<\sim 200\ mW/cm^2\), usually red/near-infrared (for deeper tissue penetration) over minutes or tens of minutes, sometimes repeated, primarily to kill pathological cells/tissues/microorganisms

Fundamental mechanism:
Light-induced chemical reactions, usually through generation of reactive (singlet-state) oxygen or radicals

Applications (approved):
PUVA, blue-light for hyperbilirubinemia
Photodynamic therapy (PDT) for:
- wet-form macular degeneration
- solid tumors/pre-malignant lesions
- Treatment of local bacterial infection
  (multiple others under development)
PUVA (psoralen-ultraviolet A) for skin conditions

Thymine in DNA Æ 8-Methoxypsoralen Æ Cycloadduct

Figure 9.4 Absorption spectra of psoralen derivatives in carbon tetrachloride: (a) psoralen, (b) 8-methoxypsoralen, (c) angelicin.

psoriasis    vitiligo
Blue-light treatment of hyperbilirubinemia (neonatal jaundice)

**FIGURE 17.1A** Structure of bilirubin and scheme of the photoconversion of bilirubin IX-α(2,2) to the EZ form. The photoisomerization affects the first ring on the left side of the figure. R1,R2,Me = methyl; R3,R4(CH=CH2) = vinyl.
Photodynamic therapy
(use of light-activated drugs to kill or modify cells)

(Potential) clinical applications

- solid tumors*
- dyslasias*
- papillomas
- rheumatoid arthritis
- age related macular degeneration*
- cosmesis
- actinic keratosis*
- psoriasis
- endometrial ablation
- localized infection*
- prophylaxis of arterial restenosis

*approved

In Vivo

Ex Vivo

von Tappeiner and Jesionek

1903
5% eosin solution topically
70 year old patient with Ulcus rodens.
Ed. P. Calandra-Pastré, R. M. Lissiansis and B. Opot (2005) in
Photodynamic Therapy and Fluorescence Diagnosis in Dermatology, Elsevier.

1905
5% eosin solution topically
50 year old man with Ulcus rodens.

Photodynamic therapy (use of light-activated drugs to kill or modify cells)

Ex Vivo

- extracorporeal photophoresis
- blood purging:
  HIV, Hepatitis B, protozoa
- bone marrow purging

• solid tumors*
• dyslasias*
• papillomas
• rheumatoid arthritis
• age related macular degeneration*
• cosmesis
• actinic keratosis*
• psoriasis
• endometrial ablation
• localized infection*
• prophylaxis of arterial restenosis

*approved
PDT Technologies

Drugs
- photosensitizers
- photosensitizer delivery systems

Devices
- light sources
- light delivery/control
- dosimetry and treatment monitoring
Photophysics, photochemistry of PDT

Singlet-state oxygen generation

Characteristic chemical interactions of singlet oxygen
Examples of clinical applications

- e.g. skin cancer
- age related macular degeneration
- Obstructive cancer
- Anti-infective PDT (in clinical trials)
Definition:

*Use of higher power (1-10^5 W/cm²) over minutes to microseconds, to selectively destroy or remove target cells/tissue or for hemostasis*

Fundamental mechanism:

*Light-energy induced thermal coagulation (destruction without direct tissue/dell removal from target site) or ablation (“explosive” removal from surface)*

Applications (approved):

*Surgical laser applications in multiple specialties* (ophthalmology, dermatology, Gynae, endoscopy, cardiology,...)
e.g. burn debridement
e.g. vocal chord surgery

e.g. disk surgery

Fig. 7. Fuzzy rat skin wound after pulsed CO$_2$ laser excision of a full-thickness burn.
Air Force devises Laser Medical Pen for the battlefield

KIRTLAND AFB, N.M. — Researchers at the Air Force’s Phillips Laboratory here have developed what they call the Laser Medical Pen, a compact and battery-operated laser that can cut like a scalpel and coagulate bleeding.

The device — a foot long, less than an inch in diameter and a pound in weight — is intended for coagulation and closure of wounds under battlefield conditions or during emergency medical evacuations.

The 5-W device, powered by 3-V lithium batteries, produces output wavelengths at 980 nm in the near-IR and is capable of both contact and free-beam lasing. Contact lasing is used for cutting and coagulation. Tissue welding and sterilization is done by free-beam lasing.

Phillips is currently transferring the system design to the private sector via a Cooperative Research and Development Agreement with hopes of expanding the development of this biomedical laser tool. One of many potential civilian uses might be to help stabilize highway accident victims until they can be brought to a hospital.

The Laser Medical Pen is intended for coagulation and closure of wounds on the battlefield.
Dermatological applications

PORT WINE STAIN

HAIR REMOVAL

ACNE

SPIDER VEINS

TATTOO REMOVAL

ANGIOMAS

REJUVENATION

ROSACEA
1983 CONCEPT OF PHOTOTHEMOLYSIS
BY ROX ANDERSON & JOHN PARISH

Selective Photothermolysis: Precise Microsurgery by
Selective Absorption of Pulsed Radiation

Science 220: 524-7, 1983

Fig. 2. Transmission electron micrograph
(×9,300) of cutaneous melanocyte after in
vivo irradiation with 351-nm excimer laser
pulse, showing specific disruption of melano-
somes (arrows) (inset, ×32,000). Nu, nucleus.

ns-μs PULSED LASERS OF OPIMAL λ
Biophysics of port wine stain laser treatment:

wavelength, pulse length critical
**Definition:**

Use of very high power density \((10^6-10^9 W/cm^2)\) over ns-\(\mu s\) per pulse to ablate target cells/tissues

**Fundamental mechanism:**

Direct molecular bond breaking and fragment ejection by short-\(\mu s\)e UV or id-IR light pulses

**Applications (approved):**

Laser corneal reshaping (PRK, LASIK), microsurgery
Micro-ablation of single human hair
Drilling of tooth enamel

Photo-thermal

Photo-MECHANICAL

Fig. 5. SEM view of enamel surface after CO$_2$ laser treatment of one pulse with 2 W. ×160.

Fig. 7. SEM view of enamel uncut surface after Er:YAG laser treatment of ten pulses with 200 mJ each pulse; ×80. The normal calcifying stripes (Retzius-stripes) are visible.
PRK

LASIK

Sophisticated Integrated systems

e.g. Corneal reshaping

Laser system

Eye tracker

Topography analyzer
Definition:
*Use of extremely high power density (>10⁹W/cm²) over sub-ns-µs per pulse* to ablate target cells/tissues

Fundamental mechanism:
*Multi-photon absorption → ionization & plasma formation → Plasma formation and collapse → shock wave → cavitation → Mechanical (micro) disruption of tissue*

Applications (approved):

*in situ* stone fragmentation, intravitreal microsurgery
stage | timescale
--- | ---
photon absorption, plasma formation | \(< 10^{-10} \text{ s}\)
plasma collapse | \(~ 10^{-10} \text{ s}\)
shock wave | \(~ 10^{-9} \text{ s}\)
cavitation | \(~ 10^{-8} \text{ s}\)
mechanical disruption of tissue | \(~ 10^{-6} \text{ s}\)

Fig. 2. Photographs of plasmas that were produced by ns- and ps-Nd:YAG laser pulses with various pulse energies. The laser beam is incident from the right. The arrows indicate the location of the laser focus. The length of the scale corresponds to 100 \(\mu\text{m}\).

Fig. 13. Sequence of images obtained by laser-flash photography in the ablation process of \(\text{K}_2\text{CO}_3\) solution by an excimer-laser pulse in the liquid (left column) and air (right column). The delay times \(t\) between the onset of laser irradiation and the image acquisition are marked in the figure. The excimer-laser fluence is \(F = 2.3 \text{ J/cm}^2\).
Photo-ELECTROMECHANICAL

e.g. kidney stone fragmentation

An optical fiber directs a laser pulse onto a kidney stone. The very fast temperature rise produces a plasma, and the first shock wave appears a few microseconds after the laser is fired. A cavitation (4 to 8 mm in diameter) forms after 300 to 500 µs, then implodes to produce a second shock wave that disintegrates the stone. The fragments are evacuated through natural channels.
Outline

• Principles/Light-Biomolecular Interactions
  
  *optics/photonics: history*
  
  "tissue optics": theoretical & experimental

• Techniques & Applications

  diagnostic/analytic
  
  *optical*  *optical*
  
  *spectroscopies*  *imaging*

  therapeutic
  
  *light-based*  *optically-guided*

• Convergence with nanotechnology
# Potential Roles for Optical Guidance of Therapeutic Interventions

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e.g. fluorescence image guidance

Indocyanine green (vascular)

Porphyrin (tumor specific)

e.g. Artemis system
- e.g. combining optical and non-optical

  e.g. merging real and virtual endoscopy
Optimizing target volume delineation: anatomical + tumor biomarker imaging at multiple times during fractionated course.
Monitoring efficacy of drug regimen

e.g. neoadjuvant treatment of breast cancer (prior to surgery) using diffuse optical tomography

**Courtesy Brian Pogue**
Guiding/assessing stenting

e.g. intravascular OCT

Courtesy Brett Bouma & Gary Tearney
Guiding infected wound cleaning & antibiotic choices

Overall Accuracy Comparison

- Wound Bed: Standard Practice 52.5% vs. PRODIGI 74.5%
- Wound Periphery: Standard Practice 17.6% vs. PRODIGI 82.4%
- Off-Site: Standard Practice 32.9% vs. PRODIGI 67.1%

“decontamination”

PRODIGY®
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  diagnostic/analytic
    optical
    spectroscopies
  optical
  imaging

  therapeutic
    light -
    based
  optically-
  guided

• Convergence with nanotechnology
  ▪ Nanomaterials
  ▪ Nanodevices
- Biomedical applications of nanoparticles
  *(incl. optically active)*

- As ‘reporters’ to probe biological functions
  *by spectroscopy, optical imaging*

  &/or

- As targeted ‘delivery vehicles’ to transport ‘payloads’ to specific sites (intracellular, organs/tissues)
  *drugs, genes, image contrast agents,..*

  &/or

- As *(photo)*active agents to elicit biological responses

Ranges from biodiscovery to clinical applications
Examples of optically-active nanoparticles

**Metal NPs**
- Ag Nanoprisms ~100 nm
- Au Sphere ~100 nm
- Au Sphere ~50 nm
- Ag Sphere ~120 nm
- Ag Spheres ~80 nm
- Ag Spheres ~40 nm

**Iron Oxide NPs**

**liposomes**

**Quantum Dots**
Why use NPs rather than other optical “reporters” for biological studies?

✓ Strong signals → bright optical images

✓ Low photobleaching

✓ Uncouple photophysics from biological behavior
  (e.g. fluorescence)    (e.g. biomarker targeting)

✓ Multiplexing (report several biomarkers simultaneously)

✓ Multimodal (2 or more optical &/or non-optical imaging methods)
Multimodal NP imaging

e.g. MR+optical

Receptor Targeted MR and Optic Imaging Probe for Imaging of Breast Cancer

Pre contrast
ATF-IO contrast enhanced
NIR imaging

Cy5.5-ATF-IO

e.g. CT+optical
NP-based nanosensors

Selective Parameters:

- NP class/material
- “indicator”
- Optical read out:
  - e.g. fluorescence, FRET, quenching, scattering,…
- Biofunction probed (“reportee”):
  - e.g. pH, glucose, phosphate, ions, oxygen, metals,…

Optical nanosensors—smart tools in bioanalytics

Analyst 2008

Sergey M. Borisov* and Ingo Klimant
e.g. Polyacrylamide nanosensor incorporating pH-sensitive dye

Advantage: Increase selectivity by blocking interfering reactants/effects of (e.g. protein) binding on the fluorescence properties

Ray & Kopelman, Nanomed 2013
e.g. optical nanodevice: intracellular probes

- Interrogate cells
- Transport fluid
- Optical/electrochemical sensing
  ..spatial resolution ~ 100 nm

Multifunctional carbon-nanotube cellular endoscopes

Singhal et al, Nature Nano 2010