Photodisruption in biological samples using femtosecond laser pulses

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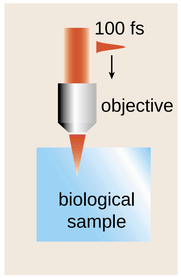
Introduction



Femtosecond laser pulses are tightly focused inside biological samples using high numerical aperture (NA) objectives.

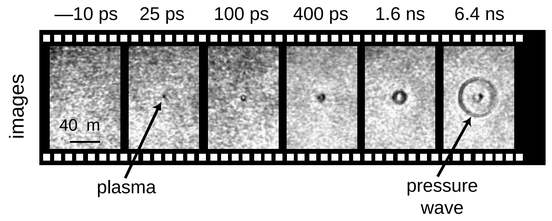
The short pulse and tight focusing produce high laser intensity at the focal volume (10^{17} W/m^2).

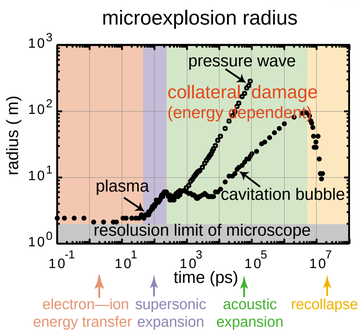
The laser ionizes the material at the focus through nonlinear processes, producing an expanding highly-excited plasma that produces a structural change.

- Compared to longer laser pulses, femtosecond pulses produce higher intensity under the same focusing condition.
- Because ionization occurs only at the focus, we can position the focus inside the bulk of the sample and produce subsurface microstructures.

Dynamics

Time-resolved imaging allows us to observe the photodisruption dynamics.

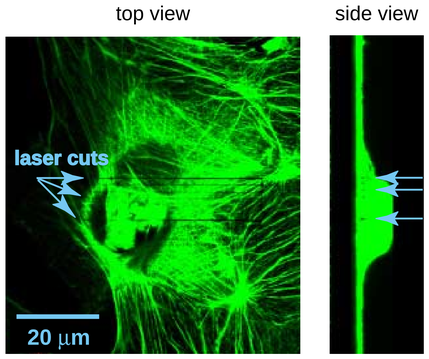


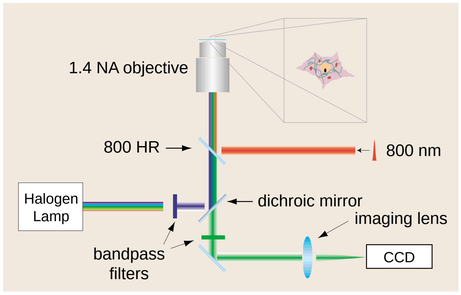


- Femtosecond pulses minimize collateral damage in biological samples.

Single cell photodisruption

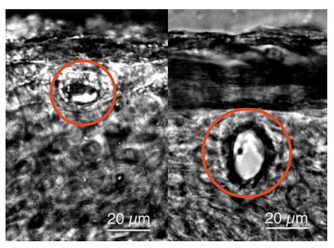
An epi-fluorescence microscope arranged colinearly with the photodisruption apparatus allows us to visualize fluorescently tagged cell structures. Femtosecond pulses are then brought in to target specific regions of interest within





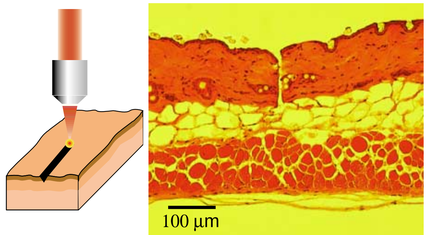
After photodisruption, we image the cell in 3-D using a confocal microscope; one slice of the image of a photodisrupted cell is shown above at left. Microfilaments in the cytoskeleton are tagged with fluorophore (green). Using 1.5 to 3 nJ of energy we made cuts and holes in the cell. As the side view at right shows, all cuts are entirely confined **inside** the cell.

Photodisruption in turbid tissue



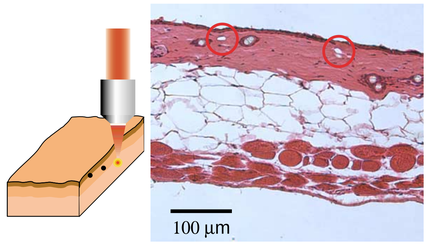
fs pulse ps pulse
subsurface microstructures
at 20μJ

Size of subsurface structures is comparable to that of a single mouse-skin cell.



100 fs, 20 μJ, 0.65 NA

Femtosecond lasers pulses produce smaller structures and less collateral damage in human dermis tissue culture.

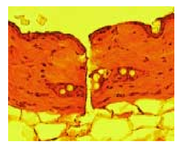


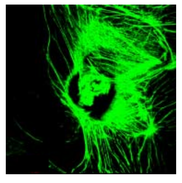
100 fs, 20 μJ, 0.65 NA

Incisions only a few micrometers wide in mouse skin tissue.

Applications

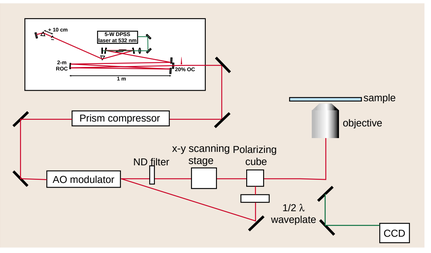
Femtosecond laser pulses can be used as scalpel in micro-surgery. Structures such as the channel shown at right, may provide a new way for transdermal drug delivery.





Photodisruption with subcellular precision has many applications in biological research. For example, by selectively disrupting organelles or receptors and watching the cell response, one can study specific cell functions, such as the relation of the cytoskeleton network to mechanical signal transduction.

Future work



- Incorporate a multiphoton laser scanning microscope into the femtosecond laser photodisruption set up for real-time high-resolution 3-D imaging.
- Use live cells that are genetically labeled with green fluorescent protein (GFP) and study specific organelle functions.