Lasers for Biophotonics & Bioanalytics
Diode and Femtosecond Fiber Lasers

A Passion for Precision.
The term “Biophotonics” is used for a plethora of applications in life sciences — all of them using light in a very controlled manner. In order to overcome the challenges encountered, researchers need to understand the underlying biological processes and the light microscope is their most important tool. Invented in the 17th century, the microscope literally changed the view of the living world.

Today, light is more than just a means of transport of information from the living world magnifying our two-dimensional field of view. We can generate pictures that provide much more information than we would ever be able to capture with our eye alone. Light, in particular the precise control of laser light, allows us to look precisely at the various layers of a sample generating three-dimensional images. We can study living species and even manipulate the sample using controlled light. The close partnership between photonic technologies and the life sciences is reflected in the word biophotonics. Their common quest aims at improved means of contrast, with better spatial and temporal resolution.

TOPTICA Photonics has been the technology and market leader for diode lasers and ultrafast fiber lasers for science and industry for more than a decade. Our laser sources are found in all areas of biophotonics ranging from OEM lasers hidden in the interior of other complex systems, to lasers on the optical tables of academic laboratories. Having been the first company to develop tunable diode lasers and tunable ultrafast fiber lasers, TOPTICA’s lasers cover the widest wavelength range on the market. Fastest temporal control, highest spatial beam quality, highest power and lowest noise are further characteristics of our well-established diode lasers. TOPTICA has made the step into the world of femtoseconds and picoseconds much less daunting for its customers than it had been in the past. In fact, with our FemtoFiber® series, we have entered an era in which ultrafast lasers have become fit for hands-off OEM products.

In the last few years, TOPTICA’s laser innovations have been instrumental in supporting rapidly emerging new applications and we are continuously looking for more challenges in this field.
# Lasers for All Applications

Highest Power, Best Beam Quality, Broarest Wavelength Coverage

<table>
<thead>
<tr>
<th>Laser</th>
<th>Properties</th>
<th>Biophotonics / Microscopy</th>
<th>Bioanalytics</th>
</tr>
</thead>
<tbody>
<tr>
<td>iBeam / iPulse</td>
<td>CW or pulsed diode laser (375, 405, 445, 473, 640, 643, 660, 785 nm; up to 120 mW)</td>
<td>Confocal: TIRF, FLIM, FRET, FRAP ... Nonlinear: Multiphoton SHG, CARS, STED ... Raman: μ-Raman, Process-Raman</td>
<td>Cytology, Cytometry, HCS / HTS, OCT Terahertz Biinstrumentation</td>
</tr>
<tr>
<td>XTRA</td>
<td>Single frequency diode laser (785 nm, 300 mW)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>XTL</td>
<td>High power diode laser (650 nm, 250 mW)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>iWave</td>
<td>Coherent blue diode laser (405 nm, 50 mW, 50 GHz linewidth)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>dfTune</td>
<td>Tunable diode laser (760 – 2800 nm, ± 700 GHz tuning range)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>FFS</td>
<td>Femtosecond fiber laser (485 – 2100 nm, 100 fs – 2 ps, 40 to 100 MHz repetition rate)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Modern microscopy is attacking three main issues from several directions:

- Confocal microscopy suppresses out-of-focus signals, achieving sub micron spatial resolution. The use of non-linear processes like Multiphoton microscopy improves the resolution and increases signal discrimination and depth penetration.

- Spectroscopic resolution such as fluorescence and the Raman effect add chemically specific information on the composition of the biological sample.

- Other techniques assist in revealing the time sequence of biological processes. These "in situ" or "in vivo" studies often involve pulsed excitation to improve the temporal resolution.

Lasers are an essential part of these techniques. Besides being used passively as a light source for observation, there is also a strong trend towards using lasers actively for manipulation of the microscopic samples. Examples of such manipulation include excision of samples, moving tissue sections, triggering fluorescence capabilities or perforating cell membranes for transgenetic experiments.

The most widespread laser method today is confocal microscopy, allowing a spatial resolution down to a diffraction limited focus of 200 nm. This approach has been refined using various subtechniques (like TIRF, FLIM, FRET, FRAP, etc.) that all need specialized laser parameters, in order to achieve best contrast and add chemical sensitivity or temporal resolution. TOPTICA’s diode laser solutions for this field, like the iBeam, have been deployed with the single source microscopists but also in volume by OEM integrators around the world, defining the standard of wavefront control.
Novel Applications

Lasers are not only used for excitation of fluorescence but can also be used to exert mechanical forces on biological probes. Microdissection by high peak power lasers is a standard technique today—the prepared biological probes are then held and manipulated by laser traps or optical tweezers.

A new field has just started as an extension of the well-known photodynamic medical treatments to use photoactivation or so-called uncaging techniques on a cellular or molecular level. Recently nanosurgery using femtosecond lasers has emerged as a promising new technique.

Matching lasers to the microscope

TOPTICA has a long tradition of customizing OEM laser systems for microscopes. It is our experience that optimum interfacing of the laser to the microscope is as essential as the right laser specifications. This concerns the delivery of the laser beam, for example via a single-mode fiber, as well as intelligent laser driver concepts which enable the microscope software to control the laser operation.

Microscope objectives are known to introduce significant dispersion when using an ultrashort pulse laser. For TOPTICA an active dispersion control adjustment is a crucial building block of our femtosecond lasers. Ample output power for our diode laser solutions allows us to offer fiber-based alternatives to the free-space approaches in the past.

Laser speckle can be a big nuisance in standard light microscopes causing ghost images or other unwanted artefacts. We therefore have all means in place to tailor the coherence of the laser just to what is needed, from an ultra narrow-band emitter to a white light laser source.

FFS — Ultrafast fiber lasers, modern technology

- State-of-the-art ultrafast laser technology
- Largest wavelength range
- Highest reliability
- Smallest size
- Lowest cost

Spectral coverage with tunable visible quasi-continuous light generated from an FFS fiber laser, providing a few mW of optical power for fluorescence microscopy.

Covering 485 to 700 nm with one single laser source is a revolution in fluorescence microscopy. The proprietary TOPTICA approach to generate visible tunable light with best power stability will change the way we look at microscopes in the future.
Using all properties of laser light

The high peak powers of ultrashort laser pulses enable the use of nonlinear processes for generating additional contrast in microscopy. Two-photon or SHG microscopy, for example, achieves high spatial resolution with deep penetration into biological samples. The four-photon processes used in CARS add valuable chemical selectivity.

TOPTICA has developed fiber-based femtosecond lasers with significant advantages in time resolution, polarization and noise properties. Virtually no excess heat is generated from the fiber laser, limiting secondary problems with the microscope.

Hitherto, nonlinear microscopy has mainly relied on the Titanium-Sapphire laser. Initially pumped by Argon-ion lasers, this laser is nowadays equipped with a diode-based green pump laser and a closed-cycle chiller—a mature solution half the size of an optical table. However, it can be clearly seen that this will forever stay a laboratory solution, due to its boundaries in size, cost and efficiency.

Only very recently has a new laser solution entered the scene, quickly claiming ground: the fiber laser. TOPTICA was one of the first companies to commercially introduce ultrafast fiber lasers in 2004. Fiber technology has developed at a stunning rate for telecom applications. Now it allows us to build compact, passively cooled ultrafast lasers with a reliability unprecedented in femtosecond laser technology.

Being easily integrated, fiber-based femtosecond laser technology is most probably going to change the look of modern microscopes in the years to come.

Fiber-based frequency conversion processes explain the enthusiasm of microscopists, allowing them to follow “the one laser for all” approach.

The evolution of ultrafast lasers in terms of reliability and cost.

**Pulse Domain Microscopy — Multiphoton Techniques**

**TiSa vs. fiber lasers**

**FFS / FFI — Next generation ultrafast fiber lasers**

- Modern fiber-based femtosecond technology with the reliability of the telecom world
- No external pump source or water-cooling needed
- Self-starting mode-locking
- Compact footprint
- Largest spectral coverage in the industry
Lasers in Microscopy — The White Light Revolution

The FemtoFiber Scientific (FFS) and FemtoFiber Industrial (FFI) laser sources provide an unmatched modular approach to microscopy, combining e.g. a narrow-band visible ps-source with a NIR multiphoton source. The powerful output at 780 nm can be used to generate white light, offering all colors simultaneously when dispersed by either a grating or an acousto-optic tunable filter (AOTF) for rapid wavelength selection. On top of that, the time structure of the 40 or 80 MHz FFS enables time-resolved techniques with the same laser.

FFS — The modular family of ultrafast fiber lasers for biophotonics
- Visible and infrared supercontinuums (white light lasers)
- Tunable narrow-band visible light for fluorescence microscopy, (485 – 700 nm)
- High power 775 nm, 970 – 1400 nm ultrafast beams for multiphoton microscopy
- Two-color emission with sub-femtosecond precision of synchronization
- High-end synchronization to other laser sources

White light lasers for biophotonics

One of the big hype words of these days is “white light laser”. At first glance this looks like a contradiction in terms, since one of the distinctive features of lasers is their one-color emission. This is of course strictly true only for continuous wave lasers with a high finesse cavity. With pulsed or even ultrashort pulsed operation, the optical laser pulses contain a broader range of wavelengths. The very high intensities of these short pulses are exploited in nonlinear optics and offer several methods for color conversion. Second harmonic generation with nonlinear crystals is the simplest and best known form of this effect. Very recently nonlinear fibers were developed, which allow the generation of supercontinua, i.e. broadband “white” laser spectra with widths previously only known for incoherent lamps. Femtosecond fiber lasers are the ideal pump source for these supercontinua. Pulsed lasers with repetition rates of multiple 10 MHz can be used as quasi continuous sources for “classical” microscopy, but also as pulsed sources for modern schemes like FLIM or for multi-color flow cytometry setups.

The spatial properties of the laser remain untouched in the case of a laser-based white light generator. This still allows diffraction-free beam delivery with highest intensity in a tiny spot under a microscope objective, or transport of the beam over larger distances. Increasingly important for the future: the white light can be delivered in single (spatial) mode fibers.


Excited at 530 nm with an FFS laser system: lifetime image of H14 cells stained with CTB-Alexa594. (Gerritsen, unpublished)
Analysis of chemical composition by light

Researchers need to detect and distinguish the composition of samples in a fast, accurate, and non-destructive fashion. Raman microscopy supports these requirements: laser light probes the molecular or band structure of gases, liquids and solids. According to the structure and chemical composition, a small portion of the laser light is shifted in wavelength and thus provides insight into the chemical identification of the sample under test. No time-consuming preparation or labeling of the sample is required. Applications of Raman techniques in biophotonics include:
- Localization of chemical substances within living cells.
- Food analysis for quality control.
- Screening of drugs in high-throughput processes.
- Cancer cell detection and discrimination from residual tissue.

For many reasons, single-mode diode lasers are the first choice for Raman microscopy. They provide high power levels to compensate for the low efficiency of the Raman effect, achieve best beam quality for high spatial resolution, consume little energy and are very compact plus easy to operate.

In the past, gas lasers were used frequently, but they never met the diode lasers advantages mentioned above. For high-resolution Raman spectra, gas lasers benefited from an intrinsical narrow linewidth, a feature now achieved by DFB diodes or external cavity diode lasers.

Latest diode technology opens up new wavelength ranges (375 – 473 nm) which are used for Raman microscopy of non-fluorescent samples. Comparing lifetime values, diode modules exceed 10,000 hours, much more than common gas lasers.

Diode lasers strengthen Raman microscopy

XTRA —
Premium Raman microscopy laser
- External cavity diode laser
- Best laser beam quality (< 1 micron resolution)
- 785 nm emission
- Highest laser power (300 mW)
- Single-mode fiber coupling (optional)

ddfBeam —
Economic Raman microscopy laser
- DFB diode technology
- Best laser beam quality (< 1 micron resolution)
- 785 nm emission
- High laser power (120 mW)

iWave —
First blue Raman diode laser
- Best laser beam quality (< 1 micron resolution)
- Enhanced linewidth, typ. 2 cm⁻¹
- Wavelengths 375, 405 nm
- Power up to 50 mW

XTRA system stability:
frequency drift < 1 GHz within 10 h,
power drift < 5 % within 10 h.
Cytometry embraces all aspects of characterization and measurement of cells and cellular constituents for biological, diagnostic, and therapeutic reasons. In this context, lasers play a pivotal role. For instance, flow cytometry exploits light scattering, light excitation, and emission of fluorochrome molecules to generate specific multi-parameter data from tiny particles and cells. TOPTICA provides laser systems exactly tailored to these requirements and supports OEM customers.

The fully automated study of the entire genome of an organism (genomics) is not possible without dedicated light sources. Investigation of single genes in high throughput sequencers, their functions and roles, are pursued with TOPTICA lasers. Our portfolio offers a unique wavelength coverage with highest available output power.

**Deep UV:** The native fluorescence of biological tissue is in the UV range. Exploiting this as a marker-free technique seems a natural choice. TOPTICA has developed a family of deep UV cw lasers using nonlinear conversion of high power red/IR diode lasers. The unique wavelength coverage from 205 to 400 nm with diffraction limited focusability and attractive power regions provides access to a series of new investigations.

**Diode lasers — the future of gas lasers**

In the past, gas lasers were the predominant light source for biophotonic instruments, like flow cytometers, high throughput sequencers or confocal microscopes. Today single-mode diode lasers have taken over the pole position, for several reasons:

- Diode lifetime values exceed 10,000 hours whilst most gas lasers require retubing after approx. 5,000 hours.

- Low heat generation and power consumption are intrinsic for diode lasers.

- High power gas lasers need external water cooling, diodes don’t.

- As to size, diode modules outperform their gas laser counterparts by a factor of 5 to 10.

**XTL — Strongest red TEM\(_{00}\) diode laser**

- 650 nm, 250 mW, single-mode
- High beam quality (M\(^2\) < 1.5)
- Excellent power stability (< 0.5 %, 8 h)

**UV diode laser — Deep UV under control**

- Wavelengths 205 to 400 nm
- TEM\(_{00}\) laser beam, high beam quality
- Industrial versions coming up (e.g. 325 nm, 100 mW, CW)

Infrared to Ultra Violet — High Power Diode Lasers for Challenging Applications

Diode lasers are easier to operate as they do not require long warm-up times.

Both the UV/blue and the red/NIR spectral ranges are readily accessed with diode modules. Lines in the green-yellow gap are covered by frequency-converted diode modules also provided by TOPTICA.

Wavelength coverage of TOPTICA’s single-mode and single-frequency diode lasers.
Using the complete optical spectrum from UV to THz

Modern lasers have been stimulating new fields of applications. TOPTICA’s sources are well suited for applications like Optical Coherence Tomography (OCT), a method that benefits from the customized spectral bandwidth of our pulsed fiber lasers.

Terahertz radiation with wavelengths beyond the infrared is intensively investigated at present. Application include biomedical imaging or the analysis of pharmaceuticals. TOPTICA’s DFB diode lasers are the best choice for generating tunable (0 - 3 THz), high resolution (up to < 10^{-6} THz) continuous-wave terahertz radiation by means of difference-frequency mixing.

TOPTICA’s femtosecond fiber lasers, on the other hand, are the preferred choice for generating broadband (> 2 THz), ultrafast terahertz pulses for time-domain spectroscopy. Further details can be found in our leaflet “Lasers for Terahertz”.

The detection of water, ammonia or oxygen in biological, pharmaceutical or chemical applications benefits greatly from our unique grating-stabilized diode laser solutions. TOPTICA has the world’s largest stock of spectroscopically relevant diode wavelengths, and draws from this advantage for your latest application. Laser diode experience for more than 10 years, combined with in-house know-how of ultralow noise electronic driver design, is available for your application in analytics.

dfTune —
For gas analysis or THz generation
- Single-frequency operation, fixed or tunable
- Wavelength agile lasers allow to match the wavelength to your application, fine and coarse detuning
- Diffraction limited focusing: < 1 μm
- Output power up to 120 mW
- Spectral coverage 760 to 3000 nm
- RS 232 computer interface
- Ready for system integration
Lasers for Biophotonics and Bioanalytics — Specifications

### Diode Lasers

<table>
<thead>
<tr>
<th>System</th>
<th>Wavelength</th>
<th>Power</th>
<th>Operation mode</th>
<th>Single-frequency</th>
<th>Applications</th>
</tr>
</thead>
<tbody>
<tr>
<td>iBeam</td>
<td>375 – 785 nm</td>
<td>Up to 120 mW</td>
<td>CW</td>
<td>No</td>
<td>Confocal microscopy, Flow cytometry, HCS / HTS</td>
</tr>
<tr>
<td>iPulse</td>
<td>375 – 785 nm</td>
<td>Up to 120 mW</td>
<td>Pulsed CW</td>
<td>No</td>
<td>Confocal microscopy, Flow cytometry, FLIM, FRAP</td>
</tr>
<tr>
<td>XTL</td>
<td>650 nm</td>
<td>250 mW</td>
<td>CW</td>
<td>No</td>
<td>Cytometry, HCS / HTS</td>
</tr>
<tr>
<td>iWave</td>
<td>405 nm</td>
<td>50 mW</td>
<td>CW</td>
<td>No (2 cm⁻¹)</td>
<td>Raman microscopy</td>
</tr>
<tr>
<td>dfBeam</td>
<td>785 nm</td>
<td>120 mW</td>
<td>CW</td>
<td>&lt; 10 MHz</td>
<td>Raman microscopy</td>
</tr>
<tr>
<td>XTRA</td>
<td>785 nm</td>
<td>300 mW</td>
<td>CW</td>
<td>&lt; 10 MHz</td>
<td>Raman microscopy</td>
</tr>
<tr>
<td>dfTune</td>
<td>760 – 2800 nm</td>
<td>Up to 120 mW</td>
<td>CW</td>
<td>&lt; 4 MHz</td>
<td>THz spectroscopy, Gas sensing</td>
</tr>
</tbody>
</table>

### Femtosecond Fiber Lasers

<table>
<thead>
<tr>
<th>System</th>
<th>Wavelength</th>
<th>Pulse length</th>
<th>Average power</th>
<th>Applications</th>
</tr>
</thead>
<tbody>
<tr>
<td>FFS.BU</td>
<td>1550 nm</td>
<td>&lt; 150 fs</td>
<td>10 mW</td>
<td>Reference / master oscillator</td>
</tr>
<tr>
<td>FFS.SYS</td>
<td>1550 nm</td>
<td>&lt; 100 fs</td>
<td>&gt; 250 mW</td>
<td>Multiphoton THz spectroscopy</td>
</tr>
<tr>
<td>FFS.SYS.HP</td>
<td>1550 nm</td>
<td>100 fs</td>
<td>&gt; 350 mW</td>
<td>Multiphoton</td>
</tr>
<tr>
<td>FFS.SYS.2B</td>
<td>1550 nm</td>
<td>&lt; 100 fs</td>
<td>2 x &gt; 250 mW</td>
<td>CARS</td>
</tr>
<tr>
<td>FFS.SYS.SHG</td>
<td>775 nm</td>
<td>&lt; 100 fs</td>
<td>&gt; 60 mW</td>
<td>Multiphoton THz spectroscopy</td>
</tr>
<tr>
<td>FFS.SYS.HP-SHG</td>
<td>775 nm</td>
<td>120 fs, 250 fs</td>
<td>140 mW, 170 mW</td>
<td>Multiphoton</td>
</tr>
<tr>
<td>FFS.SYS-CONT</td>
<td>970 – 2100 nm</td>
<td>&lt; 100 fs @ 1700 – 2100 nm</td>
<td>5 – 40 mW</td>
<td>Survey spectroscopy, Seeding</td>
</tr>
<tr>
<td>FFS.SYS-CONT-COMP</td>
<td>970 – 1400 nm</td>
<td>&lt; 40 fs</td>
<td>20 – 30 mW</td>
<td>Multiphoton</td>
</tr>
<tr>
<td>FFS.SYS-CONT-COMP-TSHG</td>
<td>485 – 700 nm</td>
<td>&lt; 1 ps</td>
<td>1 – 10 mW (&gt; 1 mW / nm)</td>
<td>Confocal microscopy, FLIM</td>
</tr>
<tr>
<td>FFS / FFI Custom Design</td>
<td>Please inquire</td>
<td></td>
<td></td>
<td>CARS, STED</td>
</tr>
</tbody>
</table>
Distributors

**Australia & New Zealand**
Lastek Pty. Ltd.
Mr. Alex Stanco
Thebarton Campus
University of Adelaide
10 Reid Street
5031 Thebarton, SA
Australia
Phone: +61 8 8443 8668
Fax: +61 8 8443 8427
sales@lastek.com.au
www.lastek.com.au

**Israel**
GR-VAM Optronics Ltd.
Mr. Shmuel Kaufman
80 Harav Yitzhak Nissim street
Hemat Shmuel
93125 Jerusalem, Israel
Phone: +972 6 622 1212
Fax: +972 2 676 8538
or-ym@013.net

**Korea**
JINSUNG LASER
Mr. Ha-Won Lee
#535-5, Bongmyung-Dong
Yusung-Gu
Hanjin Officetel Rmd #1016
Daeeun, 365-301, South Korea
Phone: +82 42 823 5300
Fax: +82 42 823 7447
sales@jinsunglaser.com
www.jinsunglaser.com

**Taiwan**
SLEO Photonics Co. Ltd.
Mr. Jimmy Chao
6F, No. 2, Lane 74
An-der Street
Hsin Tien City, Taipei County
Taiwan 231, P.O.C.
Phone: +886 2 2211 5408
Fax: +886 2 2211 5401
sleo.jimmy@msa.hinet.net

**USA & Canada**
TOPTICA Photonics, Inc.
1286 Blossom Drive
Victor / Rochester, NY 14564
U.S.A.
Phone: +1 585 657 6663
Fax: +1 877 277 9897
sales@toptica.com
www.toptica.com

---

**Every other country not listed above:**

TOPTICA Photonics AG
Lochhamer Schlag 19
D-82166 Graefelfing / Munich
Germany
Phone: +49 89 85837-0
Fax: +49 89 85837-200
sales@toptica.com
www.toptica.com

---

**Japan**

**ALTECH ADS Co. Ltd.**
Digital Storage Media Div.
Mr. Toshinori Matsuura
3F YM Shinjuku Building
4-1 Yotsuya 4-Chome
Shinjuku-Ku
160-0004 Tokyo, Japan
Phone: +81 3 3563 0945
Fax: +81 3 5363 0945
info@altech.co.jp
www.altech.co.jp

**United Kingdom & Ireland**

Mr. Howard Potter
Unit 4H Lansbury Estate
Woking, Surr, UK, GU21 2EP
Great Britain
Phone: +44 1483 799 030
Fax: +44 1483 799 076
howard.potter@toptica.com
www.toptica.com

**USA & Canada**

TOPTICA Photonics, Inc.
1286 Blossom Drive
Victor / Rochester, NY 14564
U.S.A.
Phone: +1 585 657 6663
Fax: +1 877 277 9897
sales@toptica.com
www.toptica.com

---

**Distributors only for Optical Disc Testing:**

**Taiwan**

Omega Scientific Taiwan Ltd.
Mr. James Ting
3F-3, No.415, Sec. 4
Sinyi Road
110 Taipei City
Taiwan R.O.C.
Phone: +886 2 8780 5228
Fax: +886 2 8780 5225
omega001@ms3.hinet.net

---

**Every other country not listed above:**

TOPTICA Photonics AG
Lochhamer Schlag 19
D-82166 Graefelfing / Munich
Germany
Phone: +49 89 85837-0
Fax: +49 89 85837-200
sales@toptica.com
www.toptica.com

---

**TOPTICA Photonics AG**
Lochhamer Schlag 19
D-82166 Graefelfing / Munich
Germany
Phone: +49 89 85837-0
Fax: +49 89 85837-200
sales@toptica.com
www.toptica.com

---

**TOPTICA Photonics, Inc.**
1286 Blossom Drive
Victor / Rochester, NY 14564
U.S.A.
Phone: +1 585 657 6663
Fax: +1 877 277 9897
sales@toptica.com
www.toptica.com

---

**TOPTICA Photonics, Inc.**
1286 Blossom Drive
Victor / Rochester, NY 14564
U.S.A.
Phone: +1 585 657 6663
Fax: +1 877 277 9897
sales@toptica.com
www.toptica.com

---

www.toptica.com