

Nutzungsregeln

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Two-Photon Laser Scanning Microscope

Two-Photon Laser Scanning Microscope(TPLSM) imaging allows:

- ▶ Deep tissue imaging of live and fixed specimens with low phototoxicity and photobleaching
- ▶ In vivo (mouse) and in vitro (cultured cells, explanted organs) microscopy
- ▶ Confocal microscopy
- ▶ 3D volume scanning, time lapse and automated multi-region imaging

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TPLSM Specifications:

Microscope

- ▶ Upright DM6000 stand
- ▶ Motorized stage
- ▶ Nosepiece piezoelectric actuator and Motorized Moveable Base Plate stage adapter for a precise control of Z positioning

Scanner

- ▶ Tandem (conventional and resonant) galvanometer scanhead.
- Scanning speeds in bidirectional mode (conventional scanning): 1- 1,800 Hz/lines
Scanning speed in bidirectional mode (resonant scanning): 16,000 Hz/lines per sec
TCS (true confocal scanner)

Excitation sources

- ▶ Infrared laser Ti-Sapphire Chameleon Vision II OPO MPX
- ▶ Visible laser - Laser Blue (488 nm)

Objectives

- ▶ 10x/0.30 HCPL FLUOTAR, Distance 11 mm
- ▶ 25x/1.00 IMM HC FLUOTAR motCORR VISIR, Distance 6 mm
- ▶ 25x/1.00 W IIRAPO L motCORR VISIR, Distance 2.6 mm
- ▶ 40x/1.10 W HCPL APO W CORR CS1, Distance 0.65 mm

Detectors

- ▶ Non-descanned detectors: HyD-RDLs 2ch and PMT-NDDs

Acquisition software

- ▶ LAS X SP8 Control Software, X 3D Visualization, X Dye Finder, X Electrophysiology (Leica Application Suite Advanced Fluorescence)
- ▶ Experiment/image file format: LIF (export options to TIFF, JPEG, AVI, MOV)

Work station

- ▶ PC, Windows 7 Professional, 64-bit, Intel 8-Core Xeon 2.6 GHz, RAM 16GB, 32-inch LCD monitor

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User regulations and operation procedures

The Two-Photon Laser Scanning Microscope (TPLSM) is equipped with both expensive and potentially dangerous optical equipment. Moreover, the microscope is vulnerable for malfunctioning due to wrong usage. To maintain high quality standards, the following user regulations and procedures apply to all users.

Access to the TPLSM:

- ▶ A basic training of at least 2 hours is mandatory for all users (regardless of prior TPLSM experience) before being granted access to the online booking system and independent use of the TPLSM.
- ▶ Access to the laboratory is only permitted after instructions about S1 laboratory guidelines and laser safety rules. Copies of the S1 security/laser safety statement form will be handed over during the introduction on S1 lab security/laser safety. User will be informed initially and then annually about laser safety in a laser safety instruction by the laser safety officer Dr. Heinze along the guidelines of the Stabsstelle Arbeitssicherheit der Universität Würzburg. Adherence to these rules will need to be confirmed by signature of the user.

Access to the TPLSM:

- ▶ A basic training of at least 2 hours is mandatory for all users (regardless of prior TPLSM experience) before being granted access to the online booking system and independent use of the TPLSM.

Usage of the TPLSM:

- ▶ **Do not change the TPLSM settings** if you are not sure what the consequences are! It should always be possible to reach somebody with the appropriate knowledge to help out.
- ▶ **After usage, all equipment must be cleaned**, fully functional, and brought back to its original state, i.e. ready to be used by the next user.
- ▶ Any problems, malfunctioning equipment or software should be reported immediately to the Technical Support Team.
- ▶ The facility is not responsible for storage of acquired data. All users should **take care** of their **own data** by transferring it to an external storage device directly after usage of the facility.

Time rules:

- ▶ Time between the hours of **9:00 am to 6:00 pm Monday to Friday** is deemed „prime time“. Use of the instrument is **limited to 4 consecutive „prime time“ hours** per user per day and not more than 8 hours per week in order to ensure equal access to the microscope for all potential users. More consecutive hours can be added if outside prime time. Reservations exceeding user's allowance of hours are allowed only if the microscope is available, and can be made no earlier than 1 week in advance.
- ▶ Users can only book for himself/herself. Please book by your full name, phone number, and name of your group leader.
- ▶ Reservations more than 30 minutes late for their appointments may lose the slot. The instrument is then available for use by anyone else.
- ▶ Please call the next (or previous) user when finishing your slot early (or starting late)!

Reservations made in violation of this policy may be cancelled without notice. If a user does not show for his/her booked slot the group leader will be warned by email or phone.

Costs

A basic training of at least 2 hours, mandatory for all users, is free of charge.

- ▶ Usage of the TPLSM for external users: 25 €/ hour
- ▶ Usage of the TPLSM with support for performing measurements and/or data processing: 50 €/ hour

Declaration regarding S1 laboratory guidelines

The TPLSM microscope room is classified as a S1 laboratory and is subject to the official security regulations. I hereby state that I will follow the S1 laboratory guidelines and working regulations during the use of the TPLSM.

Declaration regarding laser safety

I hereby declare that I was informed about the potential risks of working with class 4 lasers and that I am familiar with and will follow the rules concerning working with 4 laser systems embedded in TPLSM system.

Date City Name

Signature

N.N. (Technical leader/Laser safety officer)