

TRITOM™

Small animal whole-body photoacoustic and fluorescence tomography



Molecular and functional imaging with
high-resolution anatomical registration

Co-registered *in vivo* tomographic imaging

Fast, reproducible quantitative 3D data

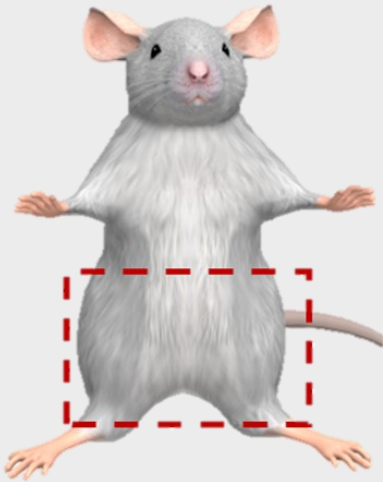


System Overview

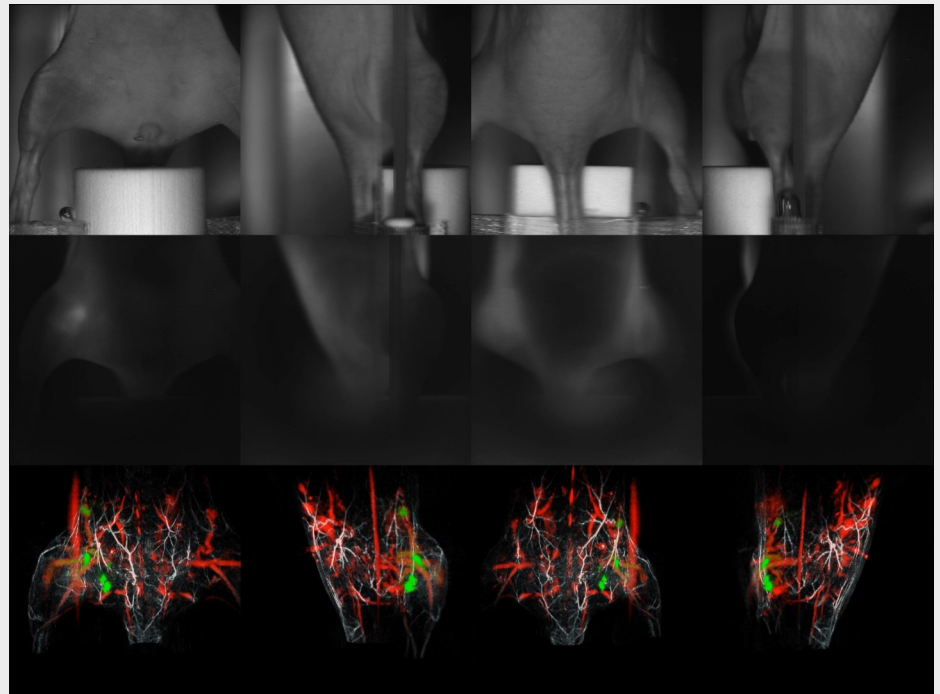


The TriTom platform is based on Photoacoustic Fluorescence Tomography (PAFT) technology that provides unparalleled capabilities for whole-body imaging and *in vivo* characterization of small animal models. Combining high-resolution photoacoustic imaging with fluorescence tomography allows deep tissue analysis, superior molecular sensitivity, and 3D localization of anatomical, functional, and molecular data. Utilizing an innovative and compact design, simultaneous spatially co-registered photoacoustic and fluorescent 3D images of live tissue can be acquired with high-resolution robust anatomical registration of optical biomarkers while maintaining high molecular sensitivity.

Perfectly co-registered 3D photoacoustic and fluorescence imaging



The TriTom enables true 3D photoacoustic and fluorescence tomographies with perfect co-registration within large volumes



Key Features:

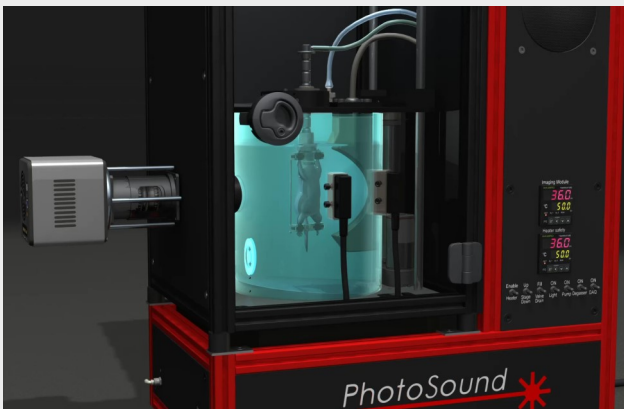
- Combined 3D photoacoustic and fluorescence tomographies enabling whole-body *in vivo* imaging with high spatial resolution (up to 150 μm) and superior molecular sensitivity.
- Fast imaging scans (36 s or less) with up to 360° rotational coverage for imaging large (30—60 cm^3) volumes and whole-body imaging.
- Nanosecond-pulsed laser tunable in a wide spectral range (460—1320 nm) for quantitative molecular imaging.
- Integrated gas anesthesia line and adjustable mouse holder designed for convenient operation and repeatable *in vivo* longitudinal studies.

TriTom Imaging Workflow

1. Subject preparation

Time Required: < 5 min

- Fast, accurate, and repeatable imaging.
- Sample holder (left) allows interrogation of up to 10 samples at a time and requires < 50 μ L sample volumes.
- Tissue holder (middle) enables imaging studies of ex vivo samples and other small objects.
- Adjustable mouse holder (right) with cushioned paw mounts and a bite bar to ensure animal well-being.
- Mouse holders are optimized for longitudinal *in vivo* whole body or brain imaging studies.



3. Image Acquisition

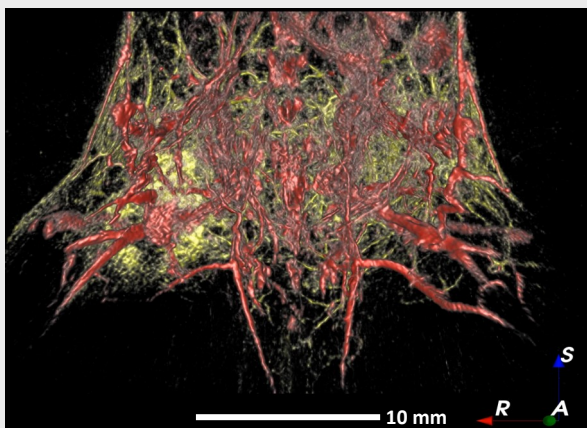
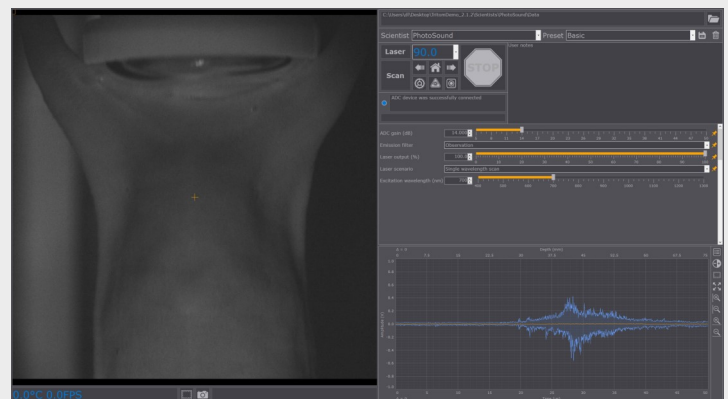
Time Required: 36 sec per scan

- Integrated software suite with user-friendly interface allows easy image acquisition.
- Quick-start and customizable scanning presets minimize image setup and data collection times.
- Open-data format enabling image reconstruction and data management with third-party software.

2. Secure subject in the TriTom

Time Required: < 2 min

- TriTom holders are designed for quick and easy mounting in the TriTom imaging chamber.
- Real-time camera feed allows users to optimize the sample position and monitor animal health during scans.
- Continuous anesthesia gas flow for *in vivo* imaging studies.
- Temperature control unit maintains the sample environment within $\pm 0.1^{\circ}\text{C}$ of the user-specified temperature.



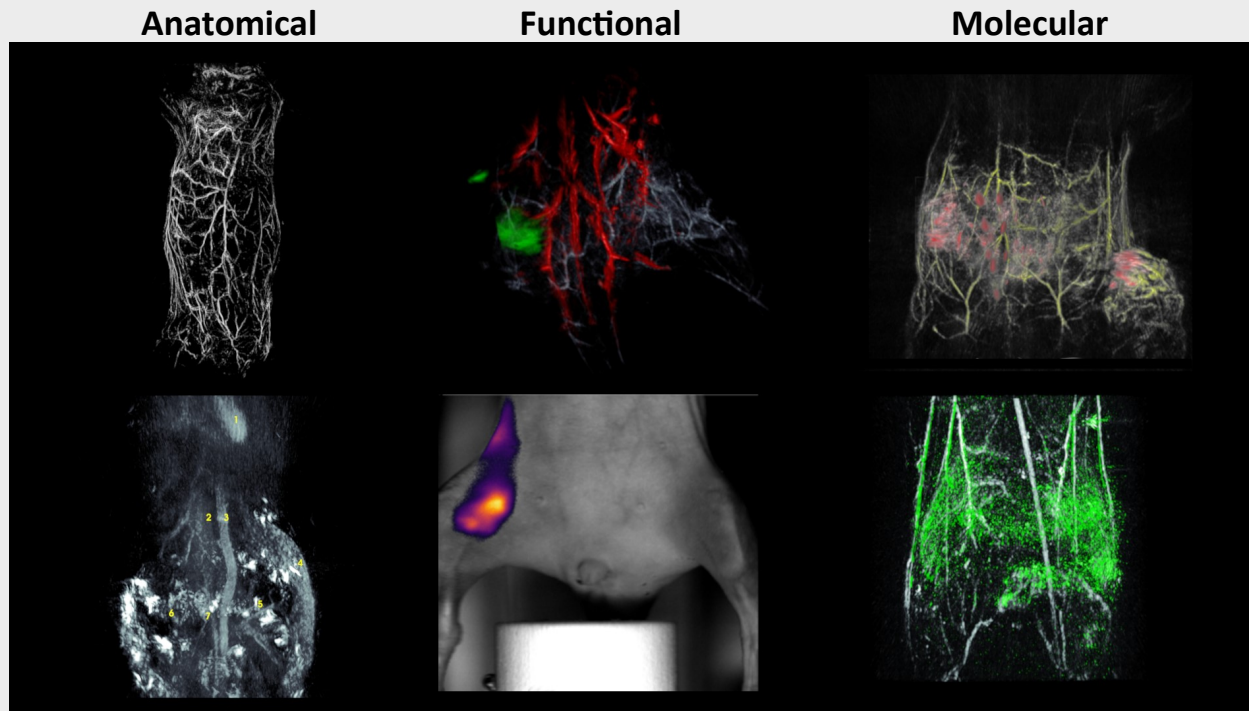
4. Reconstruct and visualize 3D data

Time Required: < 1 min

- Optimized reconstruction generates accurate large-scale volumes and molecular maps in seconds.
- Simplified data management allows photoacoustic and fluorescence data to be quickly selected for reconstruction and analysis.
- Reconstructed data is saved in standard volumetric data format for fast and easy visualization and analysis in third-party software.

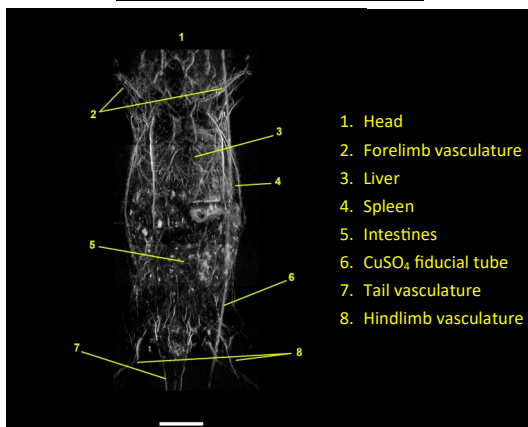
TriTom Applications

Triple analysis:



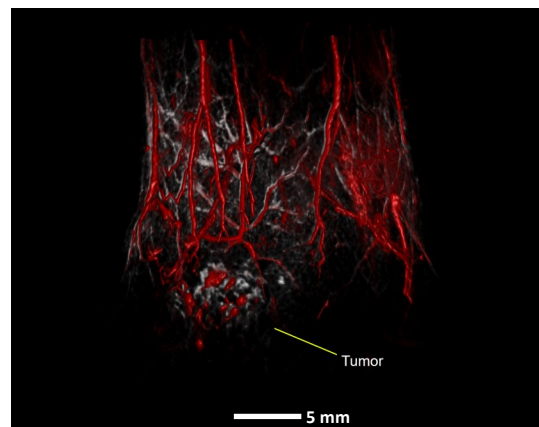
(Left) 3D photoacoustic images acquired with 532 nm (top) and 1064 nm (bottom) laser excitation showing high-resolution whole body images of superficial and deep tissue structures. (Middle) Functional PAT (top) and FL (bottom) imaging of regional lymphatic drainage. (Right) Molecular analysis of total tumor hemoglobin (top) and contrast agent biodistribution (bottom).

Anatomical Imaging



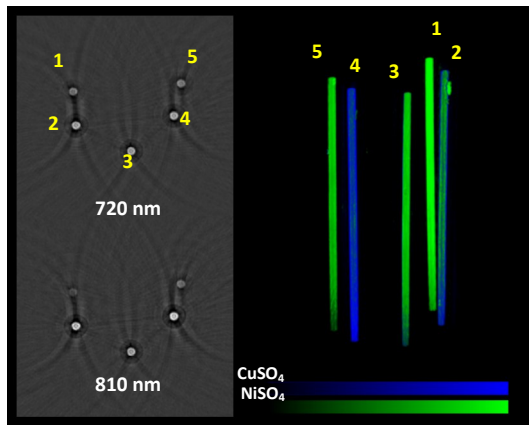
Whole-body reconstruction of a nu/nu nude mouse acquired with 800 nm laser excitation. The high-resolution volumetric imaging enables accurate anatomical registration and tissue characterization.

Cancer Research



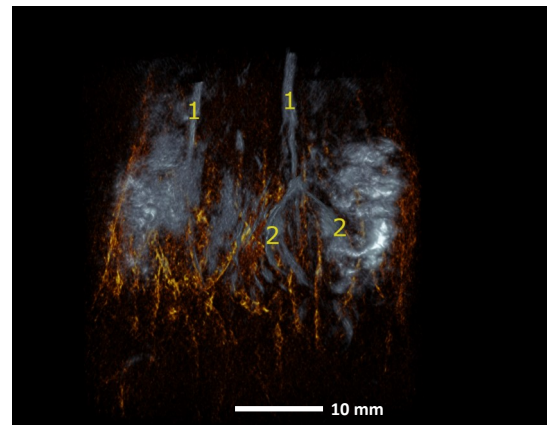
Composite 3D PAT reconstruction of 532 nm (white) and 890 nm (red) laser excitations provide high-resolution images of superficial and deep tissue structures surrounding the tumor. Tumor size = $10.6 \times 4.7 \times 11.6 \text{ mm}^3$.

Contrast Agent Development

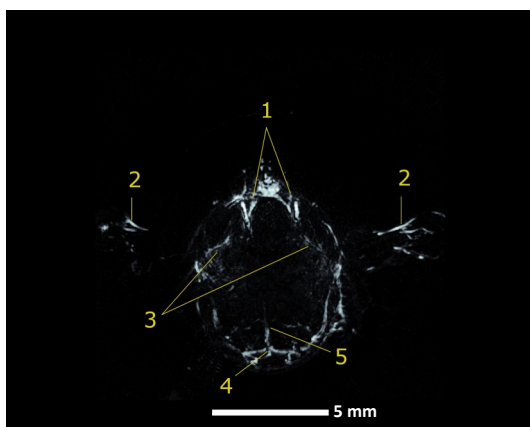


Microcuvettes loaded with contrast agent mixtures are hard to differentiate in single wavelength scans (left) compared to the spectrally unmixed volumes (right). The volumetric ratios of CuSO_4 to NiSO_4 are: [1] 0:1, [2] 1:3, [3] 1:1, [4] 1:0, [5] 3:1.

Developmental Biology

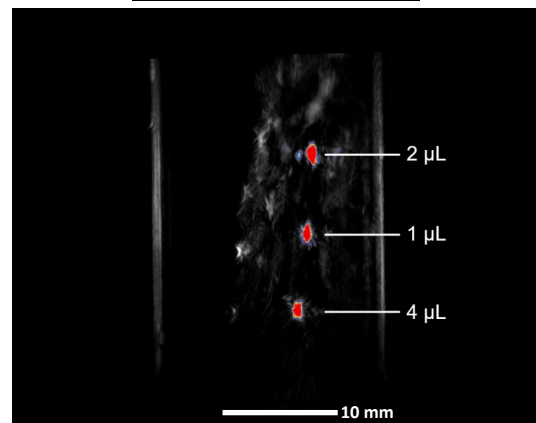


PAT imaging of a pregnant mouse at gestational day 12 acquired with 532 nm (red) and 800 nm (grey) laser excitation enables volumetric monitoring of maternal iliac arteries (1) supplying the placenta (2) and developing fetus.



10 mm thick transverse PAT reconstruction of an *ex vivo* mouse brain showing superficial and deep brain structures. (1) jugular vein, (2) brachial artery, (3) ophthalmic arteries, (4) confluence of sinus, (5) cerebral artery.

Tissue Engineering

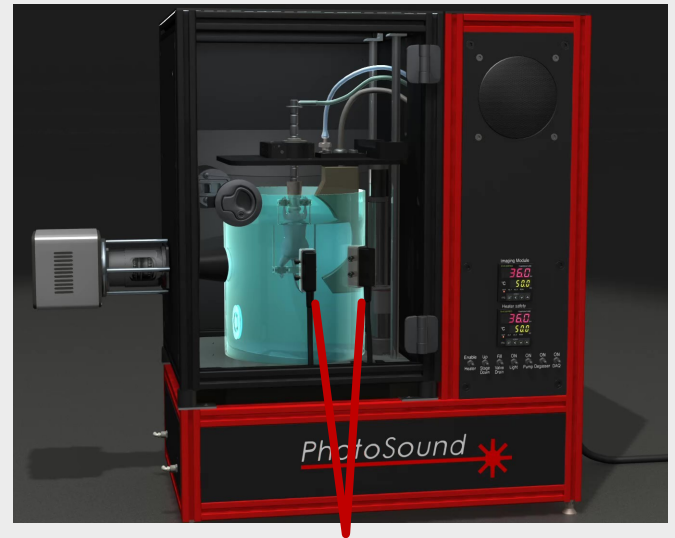


3D PAT reconstruction of stem cell tracking in an excised rat spinal cord. Contrast agent-loaded mesenchymal stem cells were injected at the indicated volumes.

TriTom Technology

High-resolution molecular imaging of large volumes

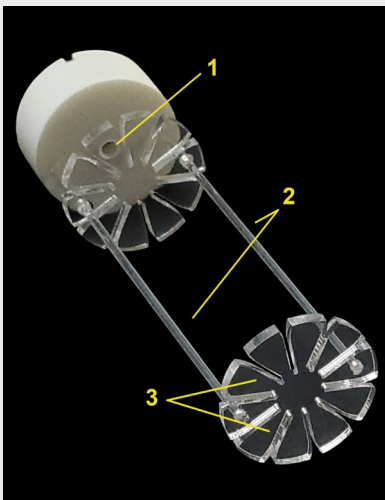
- Simultaneous photoacoustic and fluorescence excitation using tunable pulsed laser light.
- Fast wavelength switching enabling multi-excitation wavelength per scan.
- Wavelength tuning range covering all popular visible, NIR I, and NIR II fluorophores and nanoparticles:
 - 650—1320 nm (standard)
 - 460—649 nm (extended visible excitation)
- 20 Hz pulse repetition frequency with up to 160 mJ peak energy at 700 nm.
- Narrow excitation linewidth (< 0.5 nm) equivalent to 1,280 excitation filters.
- Integrated power meter for quantitative imaging.



High-efficiency optical fiber bundle with four excitation ports for safe and uniform light delivery

Accessories

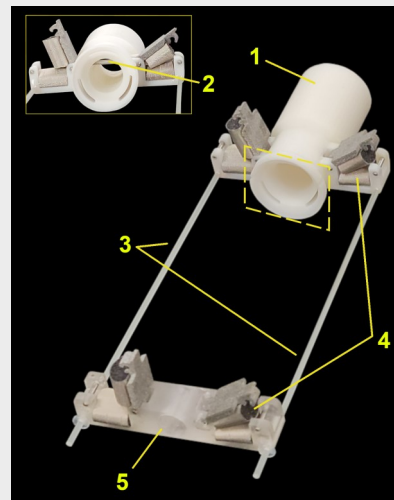
Sample Holder Contrast agent development



- **High throughput**
 - ◇ Interrogate up to 10 samples per scan
- **Small sample volume**
 - ◇ Tiny (< 50 μ L) volume saves valuable samples
- **Minimal preparation required**
 - ◇ Radially oriented slots consistently hold samples in place
 - ◇ Preparation time < 5 min

- (1) Port for administering liquid scattering background
- (2) Plastic support rods
- (3) Radial slots for quick setup and removal of microcuvettes

Mouse Holder *in vivo* imaging



- **Reliable anesthesia gas delivery**
 - ◇ Continuous gas flow via the hollow mounting shaft
- **Secure, repeatable positioning**
 - ◇ Cushioned paw mounts and bite bar ensure consistent mounting for longitudinal studies
 - ◇ Designed for minimal stress to the animal
- **Quick and convenient procedure**
 - ◇ Preparation time < 5 min

- (1) Hollow shaft for continuous anesthesia gas delivery
- (2) Bite bar for secure and repeatable positioning
- (3) Plastic support rods
- (4) Cushioned paw mounts
- (5) Adjustable hindlimb support block

Additional accessories included:

- Phantom kit
- Brain scan mouse holder
- Animal prep station
- Small animal anesthesia system
- Tissue sample holder

Key Specifications

TriTom Model	Basic Edition	Premium Edition	Features
PhotoAcoustic (PA) Imaging Channel			
PA excitation range	532 nm & 650 - 1320 nm	460 - 1320 nm	Covers visible, NIR I, and NIR II excitation
Spatial resolution	150 x 150 μ m 150 x 230 μ m		Transverse anatomical planes Sagittal and coronal anatomical planes
Molecular imaging sensitivity	100 nM ICG		In blood plasma, multispecies molecular unmixing, CNR = 2
Detector configuration	96-element curve-linear array		Wide-angle 3D imaging transducers; cylindrical focusing
Detection points per scan	> 69,000		Single scan, 360° azimuthal rotation
Detector central frequency	6 MHz $\pm$ 10%		T/R measurements, optimized sensitivity in receive mode
Detector bandwidth	$\geq$ 55% @ -6 dB		T/R measurements
PA signal digitizer	LEGION ADC: 12-bit, 256 parallel channels, up to 400 Hz frame rate, 40 MHz sampling rate, programmable amplifier 46-91 dB		

Fluorescence (FL) Imaging Channel			
Type	NA	3D Molecular imaging co-registered with PA and camera observation Real-time 2D in coronal, sagittal or any intermediate view at 20 fps	
Detector type	-	Back-illuminated sCMOS	High sensitivity cooled scientific camera
Spatial resolution	-	70 μ m x 125 μ m	At a skin level of a live test subject
Number of pixels	-	2048 x 2048	19.5 μ m pixel resolution
Max frame rate	-	40 fps	
Dynamic range	-	86 dB	
Quantum efficiency	-	95% @ 600 nm	30% - 95% in 400 – 900 nm spectral range
FL excitation range	-	460 - 800 nm	
Excitation linewidth	-	< 1 nm - Tuning step - 1 nm, equivalent to 150 - 340 extremely narrow-band excitation filters	
Emission filter set	-	8 filters: 510 - 995 nm emission range	Two additional filter slots available

Laser Excitation Unit			
Fast wavelength switching	650 - 1320 nm	460 - 649 nm & 650 - 1320 nm	Tunable to any wavelength in range allowing for multiple excitation wavelengths in a single scan
Pulse repetition frequency	20 Hz		
Pulse energy	> 160 mJ @ 700 nm > 20 mJ @ 532 nm	> 130 mJ @ 700 nm > 10 mJ @ 500 nm	Before fiber bundle transmission
Energy meter	Real-time automatic pulse energy measurements		
Excitation fiber bundle	> 70% transmission, 2 m length		
Dimensions (L x W x H)	68 cm x 44 cm x 89 cm mobile unit		

Image Acquisition Unit	
Single scan time	36 s; 360° azimuthal rotation, 720 data frames
Scan types	Continuous azimuthal rotation or reverse scans ($\leq$ 360°), time-limited by 10 min
Excitation sequence	Single wavelength; Linear or custom wavelength sweep; Popular spectral unmixing presets for molecular, physiological and anatomical imaging
Max single scan volume size	50 mm x 50 mm x 30 mm
Whole body imaging	Enabled as a stack of 3D volumes, manual axial positioning allows optimal subject placement for single scans covering the area of interest; 40 mm total positioning range with 10 mm steps, 70 mm total imaging range
In vivo imaging subjects	Mice, rats (< 200 g); fur covering the studied section should be shaved/depilated prior to imaging
Coupling medium	DI water; Temperature control range from 20 - 40 $\pm$ 0.5°C; degassing enabled
Laser safety	Light-tight imaging chamber, laser interlocks, no eye protection required
Dimensions (L x W x H)	78 cm x 35 cm x 70 cm benchtop instrument; 55 cm x 35 cm footprint

Full specifications for TriTom Basic and Premium editions available at www.photosound.com



About PhotoSound®

PhotoSound Technologies, Inc. (Houston, Texas USA) develops new imaging products and technologies for life sciences. A 3D imaging platform for in vivo preclinical research and drug discovery (TriTom™) is implemented on patented PhotoAcoustic Fluorescent Tomography (PAFT) technology, which utilizes simultaneous spectrally-selective optical and photoacoustic excitation and detection to create unparallel volumetric assessment of live organisms, organs, and tissues. A MoleculUS™ system is developed for clinical research that can benefit from co-registered ultrasound and molecular photoacoustic imaging. We also offer a variety of OEM electronic components for multi-channel parallel data acquisition.

All PhotoSound technology solutions are designed and built by experts in biomedical imaging systems, photoacoustics, ultrasound, optics, electronics and tunable lasers. Our employees are committed to provide every customer with the highest quality products and services, short delivery times and competitive pricing. Visit us at www.photosound.com to learn more about our products and proprietary technologies.

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All specifications are subject to change without notice.

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TriTom™ is designated for pre-clinical research only and is not suitable for human clinical trials.



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