# SUPERN VA-100 Miniature Two-Photon Microscope

Exploring the brain, Lighting up the future



# SUPERN VA-100 Miniature Two-Photon Microscope

Complete Solutions for in vivo Imaging, Revolutionising Neuroscience Research!

Imaging neurons and synapses in the brain of free-moving animals with the resolution of a benchtop two-photon microscope, providing neuroscientists with a revolutionary

The easy-to-wear headpiece of SUPERNOVA-100 makes real of *in vivo* imaging in free-moving animals. SUPERNOVA-100 has already been used in cognition, attention, sensory motor integration researches and a variety of studies in neural circuitry and neurological diseases.



#### Small: Wearable microscope

- 2.6g miniature headpiece, easy for small animals to wear - All-in-one design and compact system

#### Superior: Excellent imaging performance

- Imaging single dendritic spine at 0.65 µm resolution - Recording over 1,000 neurons simultaneously at 1 mm×0.87 mm FOV - Accessing all layers of mouse cortex as deep as 800 µm

#### Smart: Flexible and user-friendly

- Compatible with femtosecond lasers from various manufacturers - Compatible with EEG, EMG and DBS - Standardized procedure to locate FOV and mount the headpiece



# Small

# 2.6g Headpiece

The headpiece is designed to provide distortion-free conduction of femtosecond laser pulses, high-speed scanning, high-efficiency fluorescence excitation and collection, empowering high-resolution imaging of brain neurons and synapses in freely behaving animals.



	FHIRM-HR	FHIRM-U	FHIRM-LF
Lateral Resolution@920 nm	0.65 µm	0.85 µm	1.38 µm
Axial Resolution@920 nm	3.9 μm	7.1 μm	-
FOV Diagonal	418 µm	640 μm	1.33 mm
Working Distance		1.08 mm	
Frame Rate	9 Hz@600×500 18 Hz@300×250		
Weight		2.6g	

# **Miniature Objectives**

#### **TVS-C Series**

With a diameter of only 3.6 mm, TVS-C Series offer high resolution, large field of view, long working distance, chromatic aberation correction, and imaging optimization for deep scattering tissue.



4.65X	3X	1.6X
0.65 μm	0.85 μm	1.38 µm
3.9 µm	7.1 μm	-
418 µm	640 µm	1.33 mm
Water/Silicon oil	Water/Silicon oil/Glycerol/Oil	Water/Silicon oil/Glycerol/Oil
	400~1100 nm	
1.08 mm		
3.6 mm		
11.7 mm		
	0.65 µm 3.9 µm 418 µm	0.65 μm       0.85 μm         3.9 μm       7.1 μm         418 μm       640 μm         Water/Silicon oil       Water/Silicon oil/Glycerol/Oil         400~1100 nm       1.08 mm         3.6 mm       3.6 mm

# **MEMS Scanning Mirror**

#### **TVS-SMM Series**

TVS-SMM Series scanning mirrors are monolithically fabricated as an integrated part of the gimbal-less actuator device structure. The package size is 8.89 mm×8.89 mm×1.65 mm, and a series of optional mirror size from 0.8 to 2.0 mm are available. TVS-SMM Series provide selectable resonant frequency from 1200 to 4500 Hz.





# Superior Visualize deep into the brain



# Free-moving animal imaging

Dendrites and spines imaging, 418 µm FOV (diagonal)



#### Single spine visualization, 640 µm FOV (diagonal)



Thy1-YFPH transgenic mouse (Left) Wild type mice cortex injected with AAV-hSyn-GCaMP6s (Right) Miniature headpiece: FHIRM-U Depth: 0~60 µm Projection (Left) 200~260 µm Projection (Right) Excitation wavelength: 920 nm Freely behaving mouse

## Visualizing subcellular structures and axons, 1.33 mm FOV (diagonal)



Thy1-YFPH transgenic mouse Miniature headpiece: FHIRM-HR Depth: 60 µm Excitation wavelength: 920 nm Freely behaving mouse



Mouse cortex injected with AAV-hSyn-GCaMP6s (Left) Thy1-YFPH transgenic mouse (Right) Miniature headpiece: FHIRM-LF Depth: 450 µm (Left) 300 µm (Right) Excitation wavelength: 920 nm Freely behaving mouse

# **Superior** Multiple operating modes

#### Visualize spines under GRIN Lens

Dendrites and spines were imaged through GRIN Lens using optimized FHIRM-U headpiece.





Thy1-YFPH mice cortex, imaging through GRIN Lens

mice (hSyn-GCaMP6s virus labeling), imaging in the hippocampus

#### Non-stopping continuous imaging

Non-stopping continuous imaging at 5 Hz lasts up to 24 hours.



mice (hSyn-GCaMP6s virus injection)

#### Long-term imaging

Long-term imaging enables tracking the same population of neuron up to 30 days.



mice (hSyn-GCaMP6s virus injection)

## **Dual-emission channel imaging**

Dual-channel images are acquired simultaneously by using FHIRM-U equipped with 920 nm excitation lasers.



zebrafish (neurons were labeled with GFP and blood vessels were labeled with mCherry)

#### Volumetric imaging

The miniature three-dimensional varifocal unit enables image acquisition at different focal plane.



#### **Multi-FOV** imaging

Multiple FOVs can be acquired simultaneously in different brain regions in one animal or in different animals by using FHIRM-HR equipped with multiple headpieces.



mice (hSyn-GCaMP6s virus injection) Neuronal activities in prefrontal cortex during social behavior

mice (hSyn-GCaMP6s virus injection)

# Smart All-in-One system

With a volume of 0.6 m<sup>3</sup>, the compact SUPERNOVA-100 is designed for easy set-up and move.



Wide field fluorescence unit

- Target ROI easily with the 3 mm×3 mm FOV



#### • Fiber coupling unit

Maximize the efficiency of two-photon excitation via dispersion compensation
 Modulate the laser power with AOM
 Auto-adjustment of fiber coupling to maintain constant laser power

• Mode-switching module

Switch between wide field and two-photon imaging modes
Find ROI and mount headpiece by the standard procedure

1. Easy to put head-fixed mouse onto the stage

2. Locate the ROI through wide field imaging

3. Switch to two-photon imaging mode

4. Release mouse after imaging Animal holder with treadmill
 Reduce stress for experiment animal
 Easy to hold experimental animals without tools

• XYZ-Stage - Micrometer step accuracy

Microscope touch pad
 /
 Display system status in detail



# **Smart** Stable and compatible

-

• Fluorescence detection unit

- High sensitivity detection by GaAsP PMT - Optimized optical design that effectively captures the scattered fluorescence photons

• FPGA based real-time control and acquisition

- FPGA to control real-time acquisition - Scan control at 16-bit precision - High speed acquisition at 120 Msps - Synchronization at nanosecond resolution



• Connector fiber

- Flexible connection of the host and coupling adapter

Sama Sama

SUPERN(JVA-100

0



#### • Built-in laser beam adjustment device

- Real-time monitoring of laser pointing
- Closed-loop locking design to minimize optical path drift



#### • Laser coupling adapter

- Integrated with devices for beam adjustment and shaping
- Compatible with femtosecond lasers from different commercial brands

# **Smart**

# Easy-to-use

## SUPERGIN System Control and Image Acquisition Software

SUPERGIN is designed for easy use with a short learning curve. The software platform includes modules for image collecting, data processing and analysis.



## -1- SUPERANALY Data Processing and Analysis Software

SUPERANALY provides functions of imaging preprocessing, automatic neuron identification, proofreading, trace extraction and trace generation. It also supports various correlation analysis between neuronal activities and behavioral events. The software supports different file export formats that are compatible with a variety of external image processing and data analysis software.



Coupling the image data with animal behavior

#### Features

- 1. Al algorithm: Effective denoising without degrading sharpness, automatic neuron segmentation with increased accuracy.
- 2. Algorithm pool: Powerful and extensive algorithm pool, supporting a variety of applications.
- 3. Powerful functions: accurate event calibration, multi-dimensional correlation analysis, flexible file adaptability and compatibility. 4. Easy operation: User-friendly through single-button preprocessing and stepwise preprocessing.

# Applications

The large-field two-color miniature two-photon microscope combined with the optogenetic module were used to observe overall network activity of dorsomedial prefrontal cortex (dmPFC) after inhibiting PV neurons or VIP neurons. By optogenetic manipulating and calcium imaging of cell-type specific neurons, the neuronal mechanism behind the "winner effect" was revealed.



Chaoyi Zhang et al. | Neuron | February 2, 2022

Study of the itch perception in freely behaving animals requires minimal input of stimulus perception. The miniature two-photon microscopy enables neuronal calcium imaging for itch study. Itch was induced by manipulation of GRPR neurons in spinal cord, and activity of S1Tr neurons was recorded while mouse scratching.



Xiao-Jun Chen et al. | National Science Review | June, 2022

Rest (5 s) Run (47 s) Bun (36 s) AAV9-hsyr -ACh3.0 Miniature two-photon microscope Bun (59 s) Rest (85 s) Average in run 0 r 0.2 0.5 0.5 -0.5 0 1 -0.5 1 -0.5 0.5 0 Time relative to stimulus onset (min)

Miao Jing et al. | Nature Methods | September 28, 2020

## perception and disorders in freely behaving mice was studied using a miniature two-photon microscope.



Furong Ju et al. | bioRxiv preprint | January 11, 2022

30

Social behavior research intrinsically requires animals in a state of freely behaving. To study the neural mechanism behind social behavior at the single-cell level, miniature two-photon microscopy was used here and revealed the neural coding mechanism of social behavior deficits in autistic mice.



Zhe Zhao et al. | Science Advances | August 31, 2022

To study the seizure propagation in free-behaving animals, miniature two-photon microscopy was used to visualize brain network hyper-excitation coupled with behavioral assessment in freely-moving mice.



12s Zhuoran Zhang et al. | Neurosci. Bull | May 11, 2022

The miniature two-photon microscopy was utilized to verify a flexible multimodal transparent electrophysiological hydrogel electrode (MTEHy), and also demonstrate its good biocompatibility and reduction of neuroinflammatory response and cortical tissue damage.



Fluorescence of genetically encoded fluorescent acetylcholine indicator (ACh3.0) was recorded using a miniature two-photon microscope while mouse running, to image the neurotransmitters in real-time in freely behaving animals.

Long-term imaging of spinal cord in freely behaving mice is been proved to be practical. The function of spinal cord on sensory





Dio-GCaMP6s



Wei Wei et al. | Acta Biomaterialia| August 28, 2022

# Specifications

Optical Headpiece	FHIRM-HR	FHIRM-U	FHIRM-LF		
Resolution	0.65 µm	0.85 µm	1.38 µm		
FOV Diagonal	418 μm	640 μm	1.33 mm		
Working Distance		1.08 mm			
Frame Rate		9 Hz@600×500 18 Hz@300×250			
Weight		2.6g			
luorescence collection nodule	High sensitivity GaAsP PMT Collection range: 300~720 nm Green fluorescent channel: 520+/-25 nm Red fluorescent channel: 625+/-25 nm (				
Controller	Sample Rate: ≥120 Msps	Analog input resolution: ≥14 bit	Analog bandwith: ≥60 MHz		
iber coupling unit	Built-in AOM (acoustic optical modulate	or) , response time<250 ns; with laser shutte	r protection		
ield of view searching nodule	XYZ Stage, Bidirectional Repeatability, T Be used for searching field of views and				
Viled field fluorescence	Excitation wavelength 470 nm         CCD Camera, Resolution1920×1200 pixels, full field of view imaging speed ≥40 Hz				
Init					
Software	SUPERGIN: System Control and Image A SUPERANALY: Processing and Analysis	Acquisition of Neuronal and Animal Behavioural Data			
System overall size	595×400×668 mm <sup>3</sup>				
Ainiature three- limensional varifocal Init ( Option )	~50 µm	~150 µm	~500 µm		
emtosecond pulsed aser ( Option )	920 nm femtosecond pulsed laser Compatible with all brands of femtosecond lasers				
Vork Station Option )	Imaging workstation Recommend Specification: OS-Win10, F	RAM-32G, HDD-512 SSD and 2T HDD			
nimal Behavior nstrument ( Option )	This imaging system is suitable for mos	st mice behavior experiment			
Antivibration table Option )	Recommended size: 1200×750×750 mm	13			
nstallation conditions	Temperature: 20~30°C, humidity<60%				

# **Dimensional Diagram**





Headpiece Size

Laser Coupling Adapter Size





Aunion Tech Co.,Ltd1850-166-2513021-510-83793info@auniontech.comwww.auniontech.com