Unsupervised Deep Learning for Microcopy Imaging Restoration

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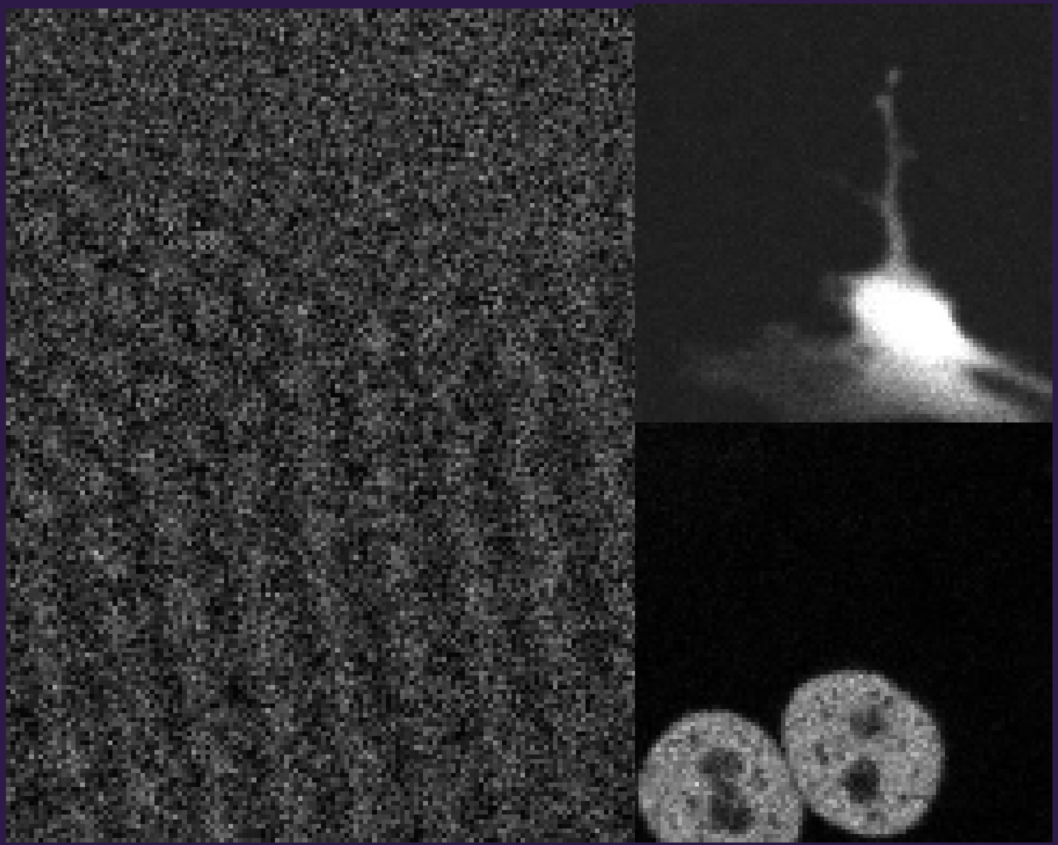
Motivation

Challenges with Microscopy Imaging

- Low signal-to-noise ratio
- Different noise types
- Multi-modality
- Motion Blur

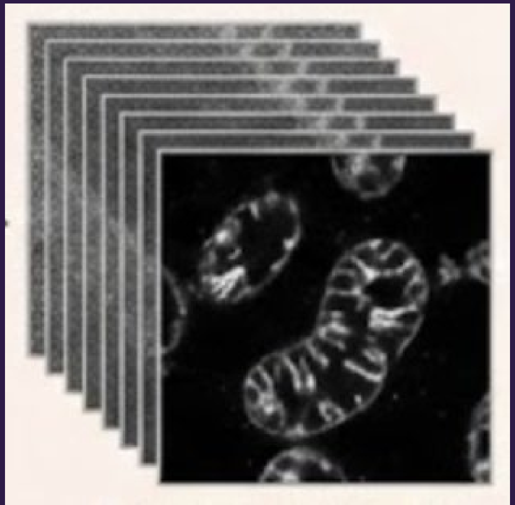


Motivation

Challenges with Microscopy Imaging

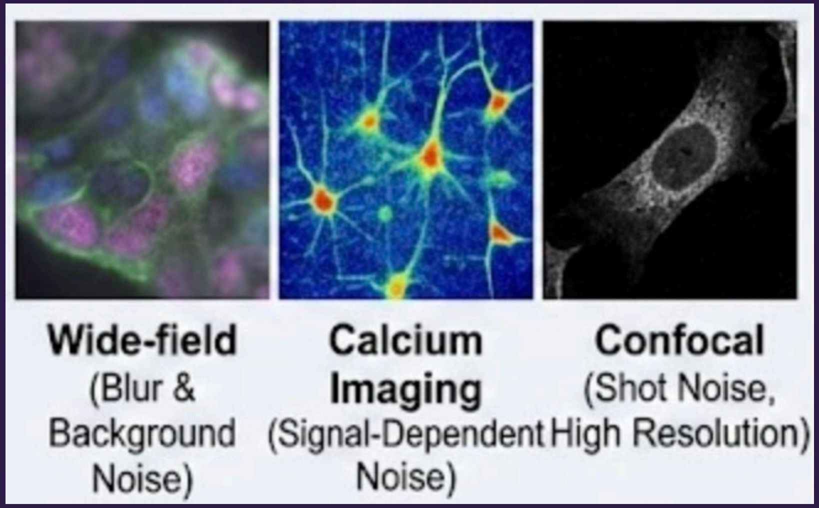
Small data problem:

Most denoising models require thousands of frames



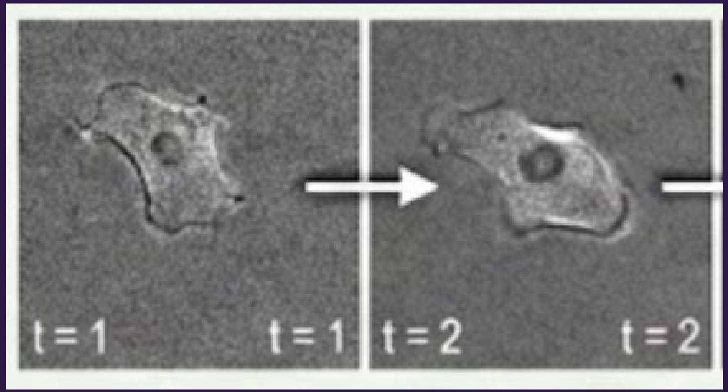
Modality diversity:

Noise varies across wide-field, calcium, and confocal imaging, limiting cross-modality generalization.



Temporal complexity:

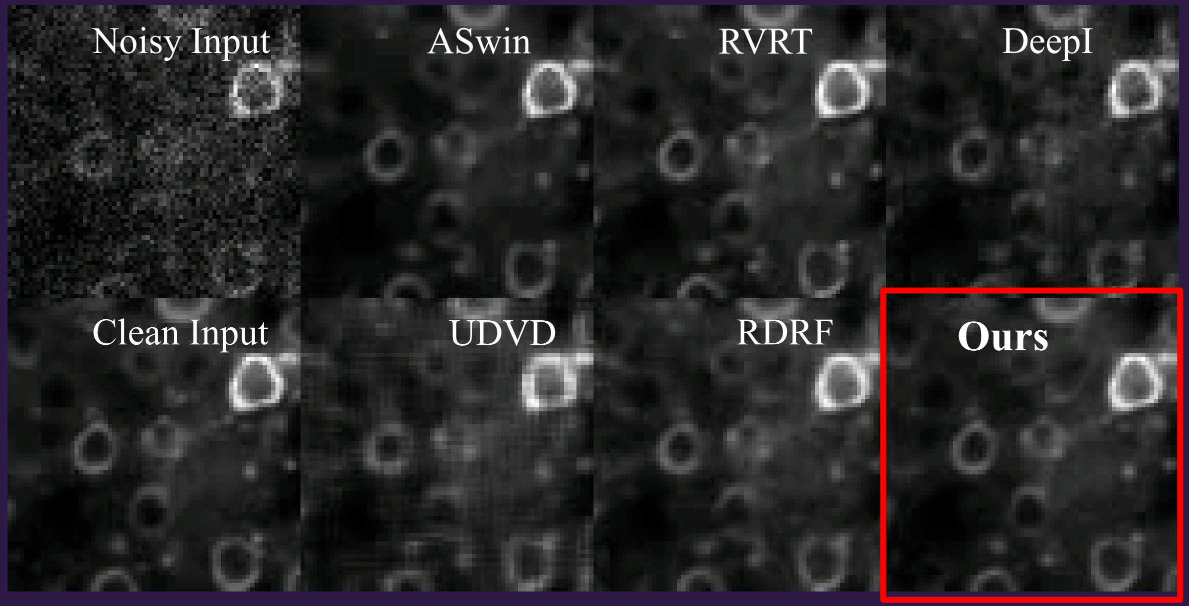
Balancing noise removal and motion preservation in complex or rapid motion scenarios.



Moving cells

Related Works

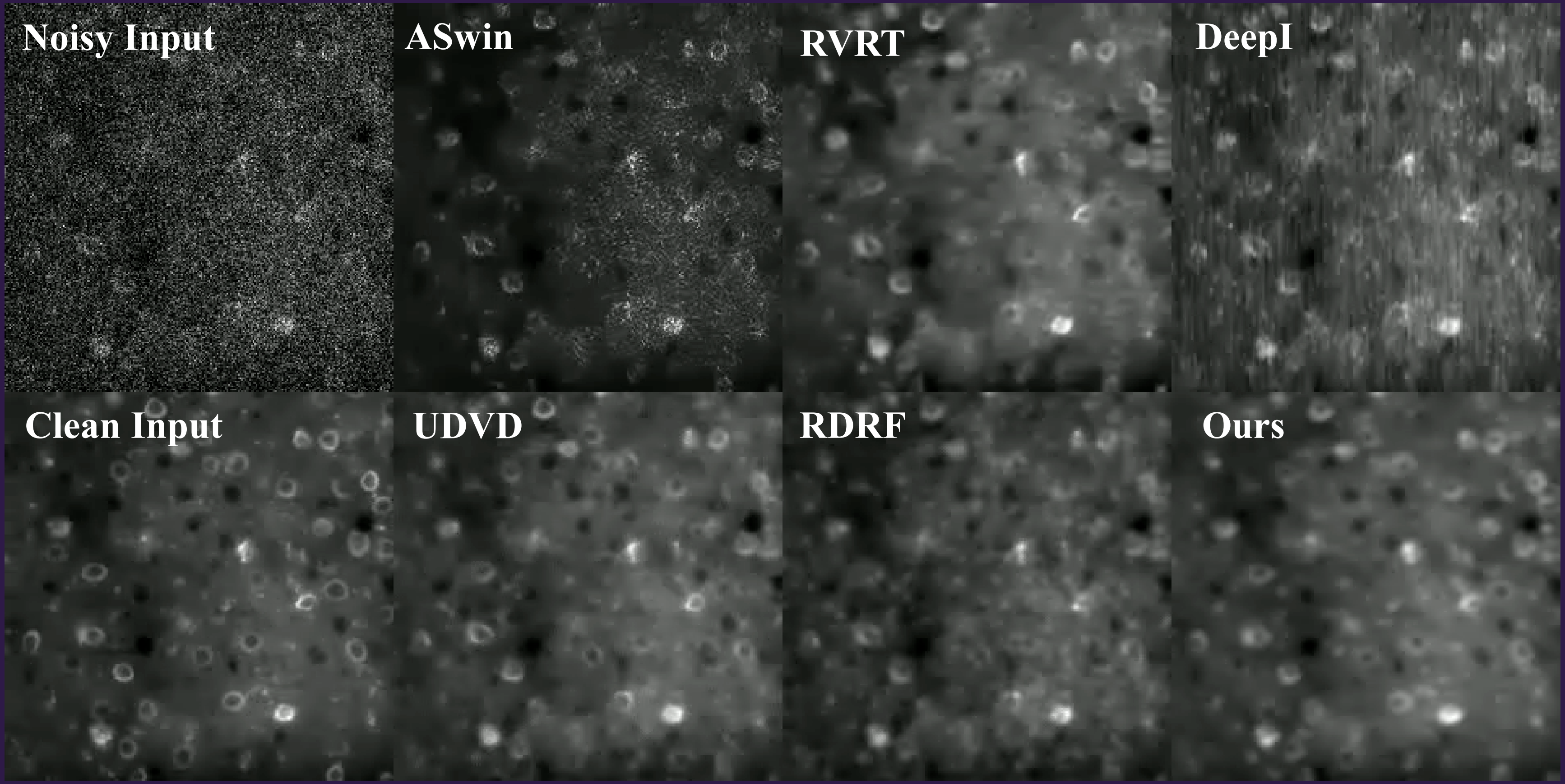
- Employs Blindspot mechanism to prevent identity noise mapping
- Extensively exploits neighboring pixels, which can lead to overfitting in microscopy images [1,2,3].
- Most existing denoising techniques are primarily designed for Gaussian noise [4,5,6].



[1] Lecoq et al., Removing independent noise in systems neuroscience data using deepinterpolation. Nature methods, 2021.
[2] Sheth et al., Unsupervised deep video denoising. ICCV, 2022
[3] Wang et al., Recurrent self-supervised video denoising with denser receptive field. In Proceedings of the 31st ACM ICM, 2023.
[4] Song et al., Neural anatomy and optical microscopy (naomi) simulation for evaluating calcium imaging methods. Journal of neuroscience methods 2
[5] Liang et al., Recurrent video restoration transformer with guided deformable attention. Neurips 2022
[6] Lindner et al., Lightweight video denoising using aggregated shifted window attention. ICCV, 2023.

Previous Solutions

Unsupervised/Self-Supervised Deep Learning solutions are not efficient and accurate



Outline

- Unsupervised Microscopy Video Denoising
- Unsupervised Denoising for Multi-Modal Microscopy Imaging for Small Data Scenarios

Unsupervised Microscopy Video Denoising

Our solution – Removing the Noise

- Calcium imaging is used for recording neural activity over a period
- Presence of noise can impact the detection of potential neural activities in the recordings
- Some learning-based approaches [1,2] for denoising require noisy/clean pair of images to train. However, ground truth clean images are usually unavailable in medical imaging
- We used an **unsupervised** learning-based approach trained on noisy data by learning the spatio-temporal relationship in the data

[1] HC. Burger, CJ. Schuler, and S. Harmeling (2012). Image denoising: Can plain neural networks compete with bm3d? CVPR
[2] Pascal V., Hugo L., Isabelle L., Y., and Pierre-Antoine M. (2010). Stacked denoising autoencoders: Learning useful representations in a deep network with a local denoising criterion. JMLR

Pre-processed Denoised Frame

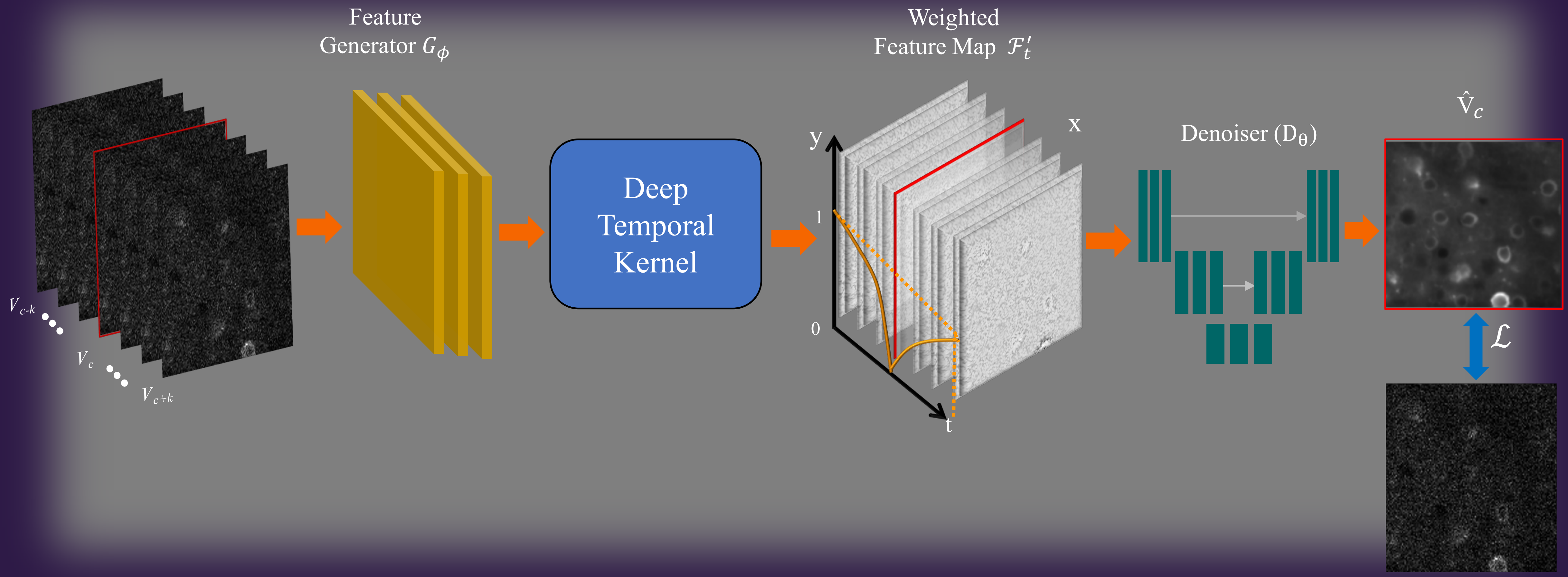
However, it will remove the subtle neuron signals and blur out the response area.

Our solution: Pre-process the input video to amplify the neuron response, then apply denoise algorithm

Pre-processed Denoised Frame

Our solution: Pre-process the input video to amplify the neuron response, then apply denoise algorithm

Our Method



Aiyetigbo et al. Unsupervised Microscopy Video Denoising. CVPRW 2024

Method

• DeepTemporal Filter (ω)

- Linear signal filter (ω) to adjust feature map weights based on temporal proximity to central frame

$$\omega(.) = \left[\left\{ \frac{k-i}{k} \right\}_{i=0}^{k-1}, \left\{ \frac{i}{k} \right\}_{i=1}^{k} \right]$$

$$\omega(.) = \{1, \dots, 0, \dots, 1\}$$

where

- $k = \lfloor N/2 \rfloor$ and N is the number of frames in the input
- $\{\omega_t | t = 1, \dots, N\}$

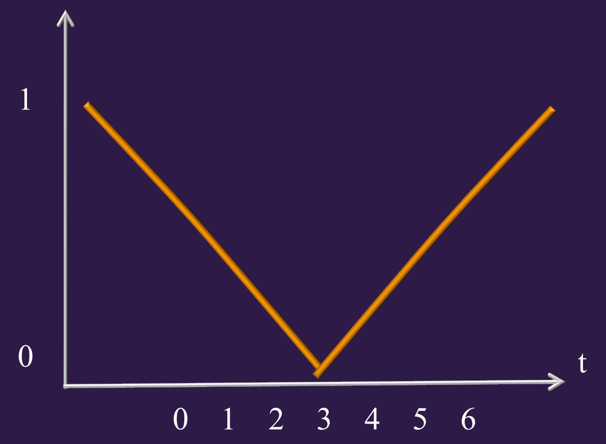


Fig 1. DeepTemporal filter shape

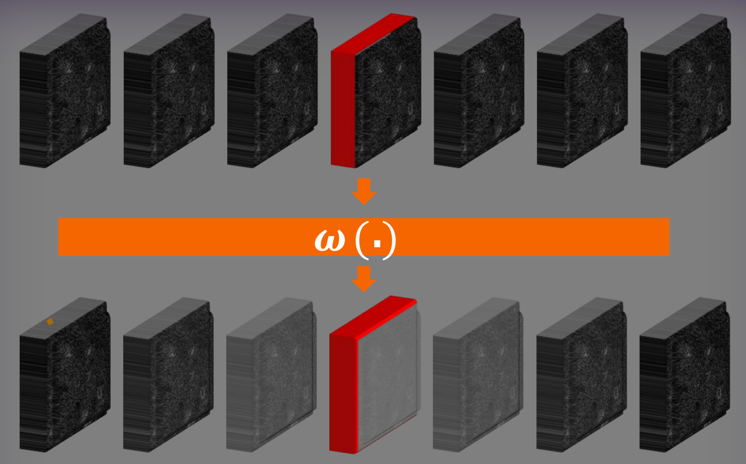


Fig 2. Illustration of our DeepTemporal filter

Method

- **Feature Generator**
  - Given a stack consecutive N input frame $\mathcal{V}_t \in \mathbb{R}^{N \times H \times W \times 1}$
  - Applies group convolution layers to extract features of each frame.

$$\mathcal{F}_t = \mathrm{G}_\phi(\mathcal{V}_t) \qquad \mathcal{F}_t \in \mathbb{R}^{N \times H \times W \times C}$$

- **DeepTemporal Filter**

$$\mathcal{F}_t' = \omega_t \odot \mathcal{F}_t \quad \forall t \in \{1, \dots, N\}$$

$$\{\mathcal{F}_t' | t = 1, \dots, c, \dots, N\}$$

$$\boldsymbol{\mathcal{L}} = \left\| \hat{\mathrm{V}}_{\boldsymbol{c}} - \boldsymbol{\mathcal{V}}_{\boldsymbol{c}} \right\|_{\mathbf{2}}$$

- **Denoiser**
  - UNet architecture. Comprises of 3 encoder layers and 2 decoder layers

$$\hat{\mathrm{v}}_c = \mathcal{D}_\theta([\mathcal{F}_1', \dots, \mathcal{F}_N'])$$

Experiment

Dataset

Simulated two-photon Calcium Imaging [1]

One-photon Calcium Imaging [2]

Fluorescence Microscopy [3]

Evaluation Metrics

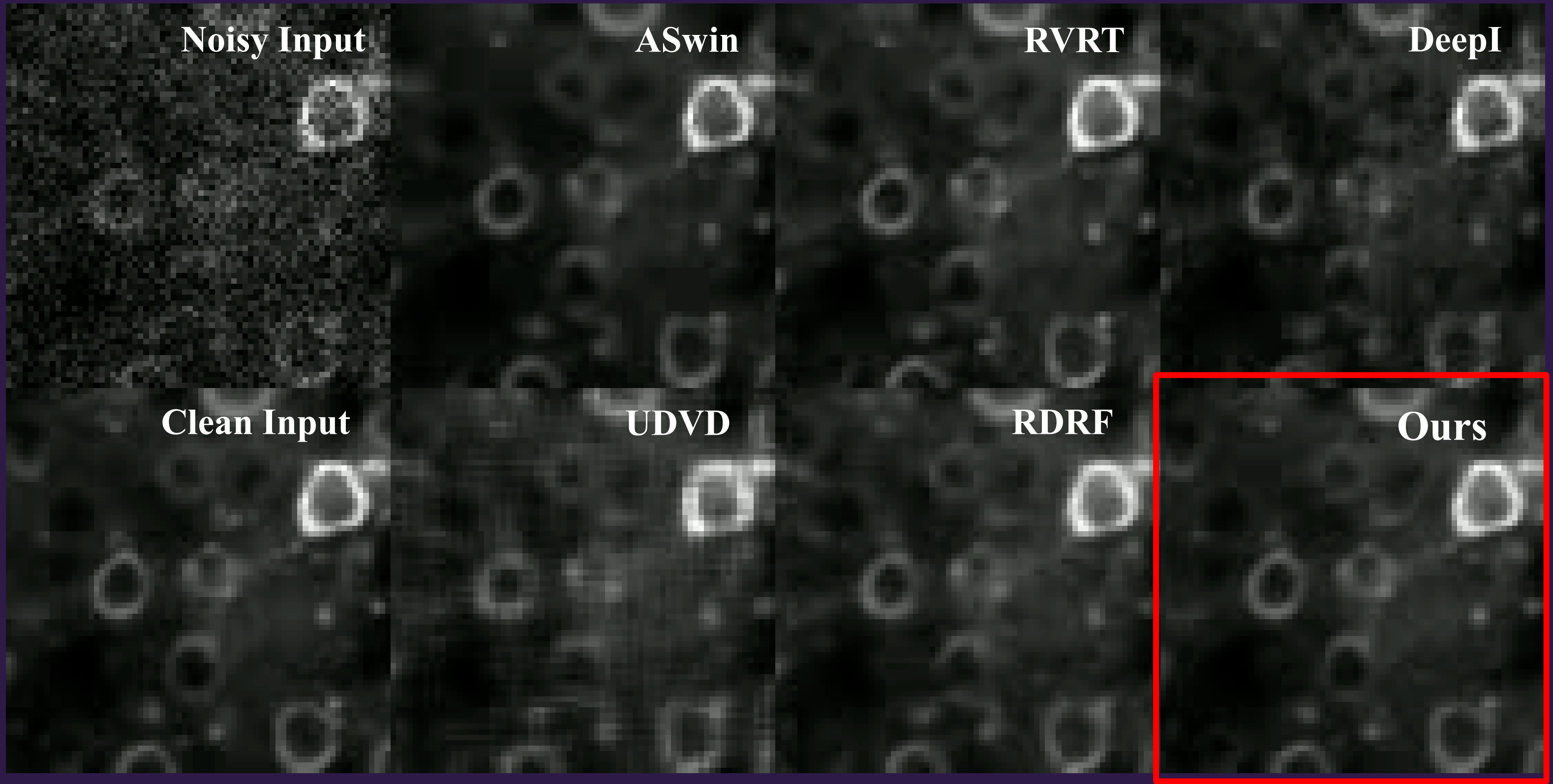
PSNR

SSIM

[1] Song et al., Neural anatomy and optical microscopy (naomi) simulation for evaluating calcium imaging methods. Journal of neuroscience methods 2021
[2] Ulman et al., An objective comparison of cell-tracking algorithms. Nature methods 2017.
[3] Kalivas Laboratory, The Medical University of South Carolina.

Results

Simulated two-photon calcium imaging



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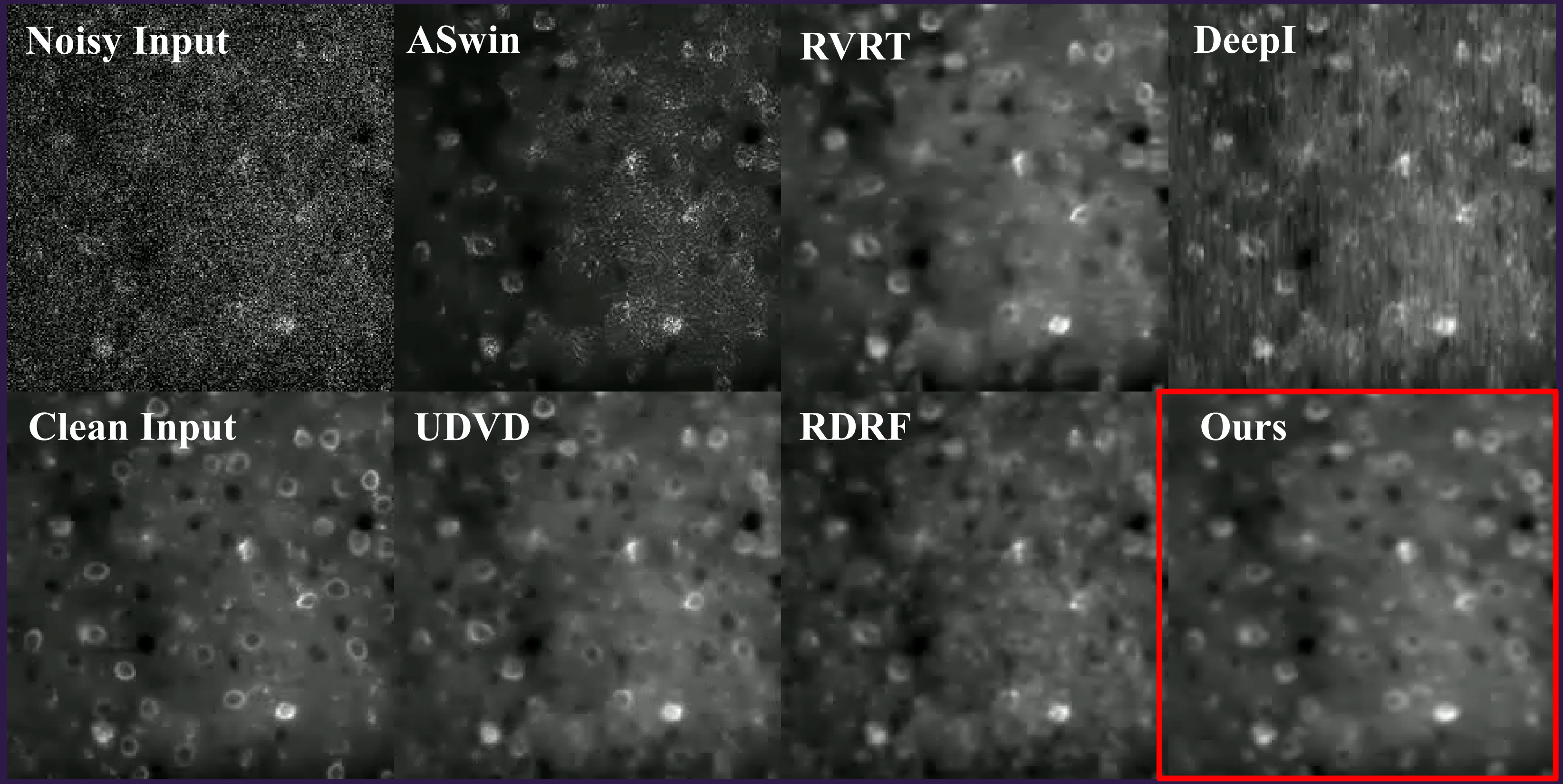
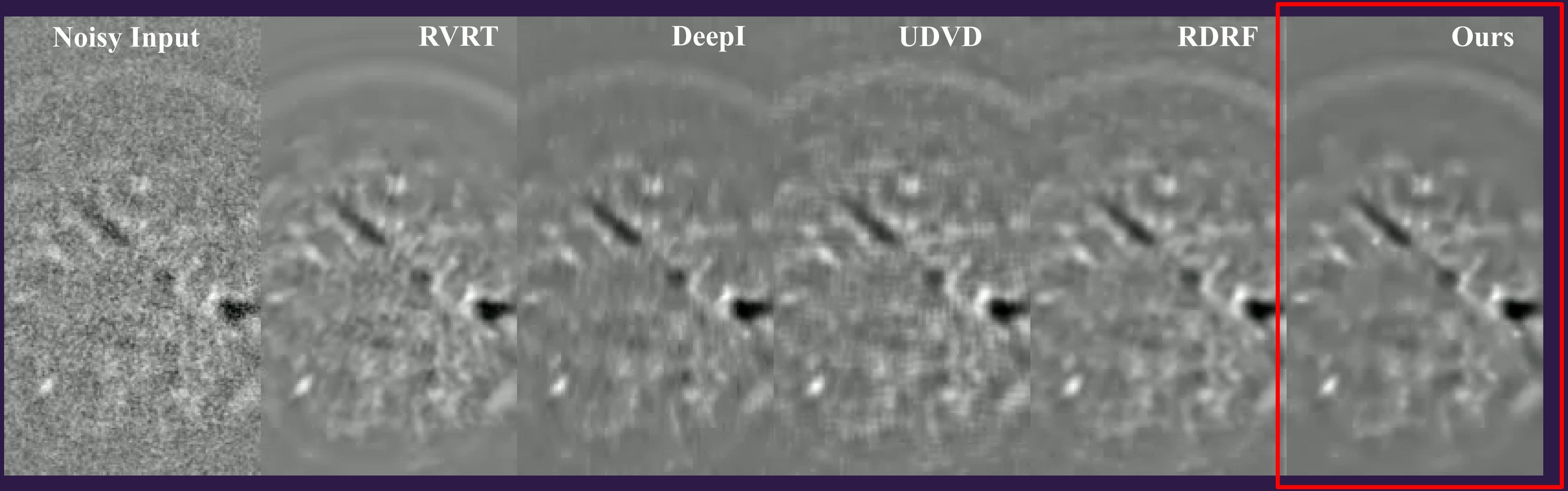
Simulated two-photon calcium imaging



Song et al., Neural anatomy and optical microscopy (naomi) simulation for evaluating calcium imaging methods. Journal of neuroscience methods 2021

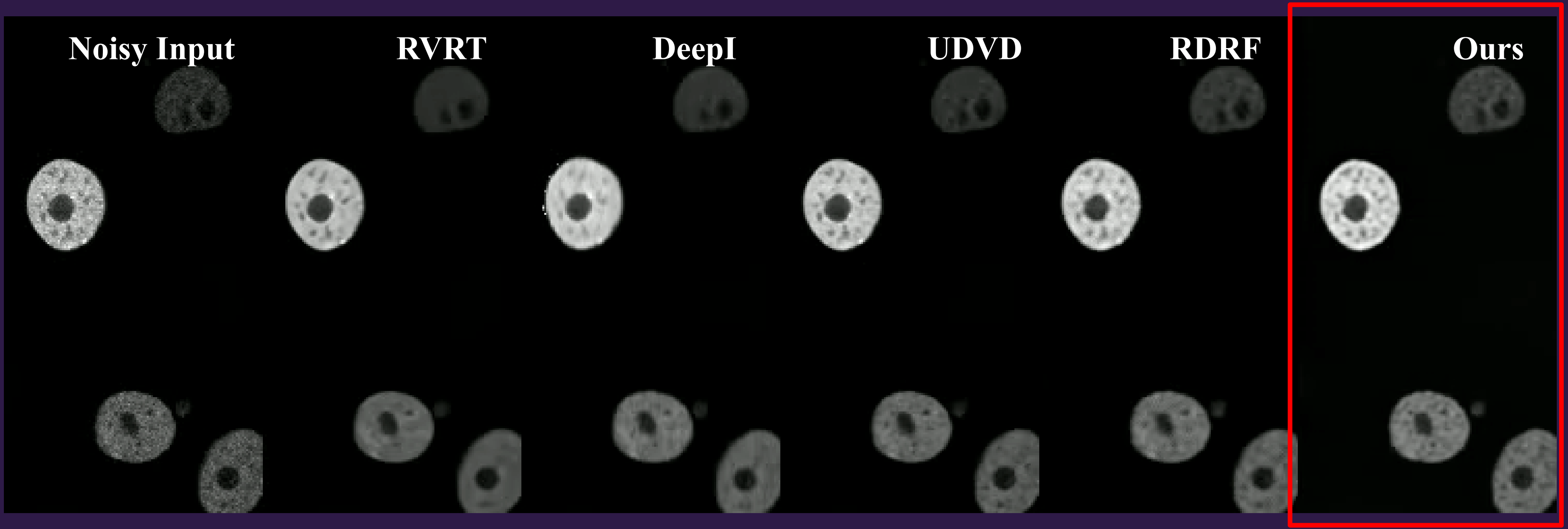
Results

One-photon calcium imaging



Results

Fluorescence Microscopy

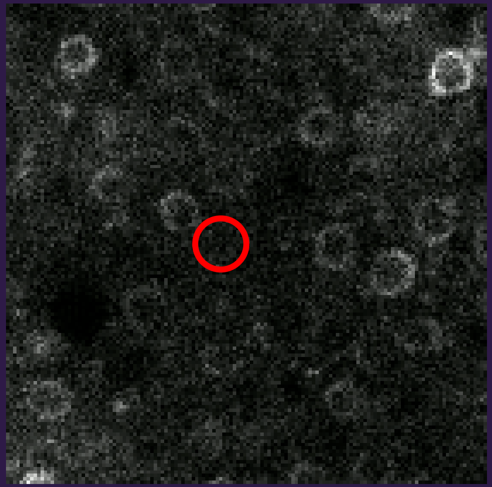


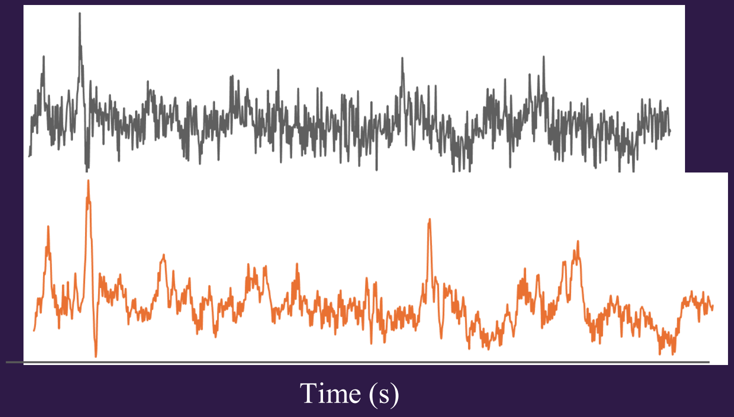
Results

- The result shows the average PSNR and SSIM values on the 2P imaging simulated with NAOMi[1]

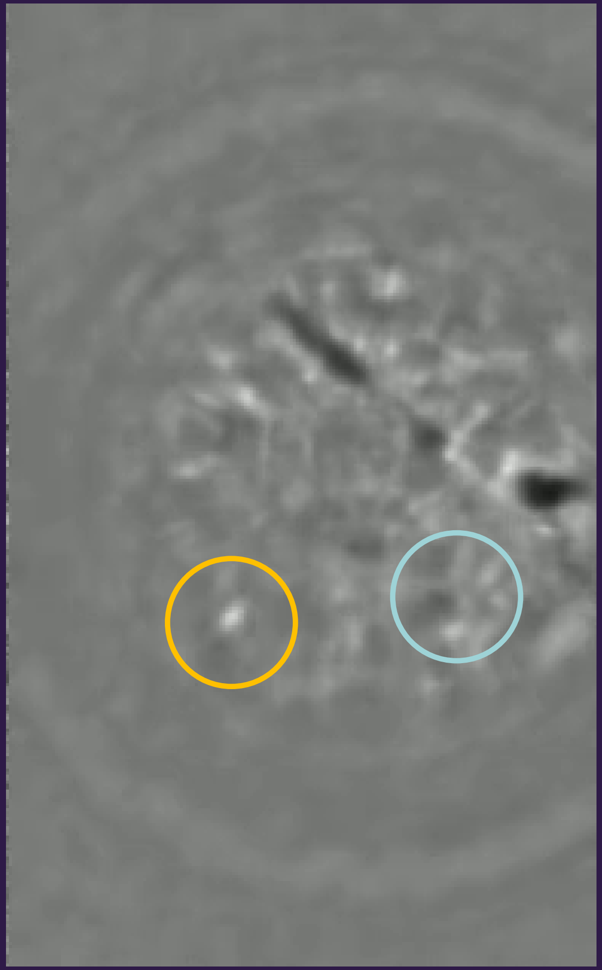
	RVRT	ASwin	DeepI	UDVD	RDRF	Ours
PSNR	<u>30.05</u>	25.94	28.64	25.75	28.31	**35.90**
SSIM	<u>0.92</u>	0.90	0.91	0.74	<u>0.92</u>	**0.95**

- Calcium traces of an example ROI from the two-photon calcium imaging



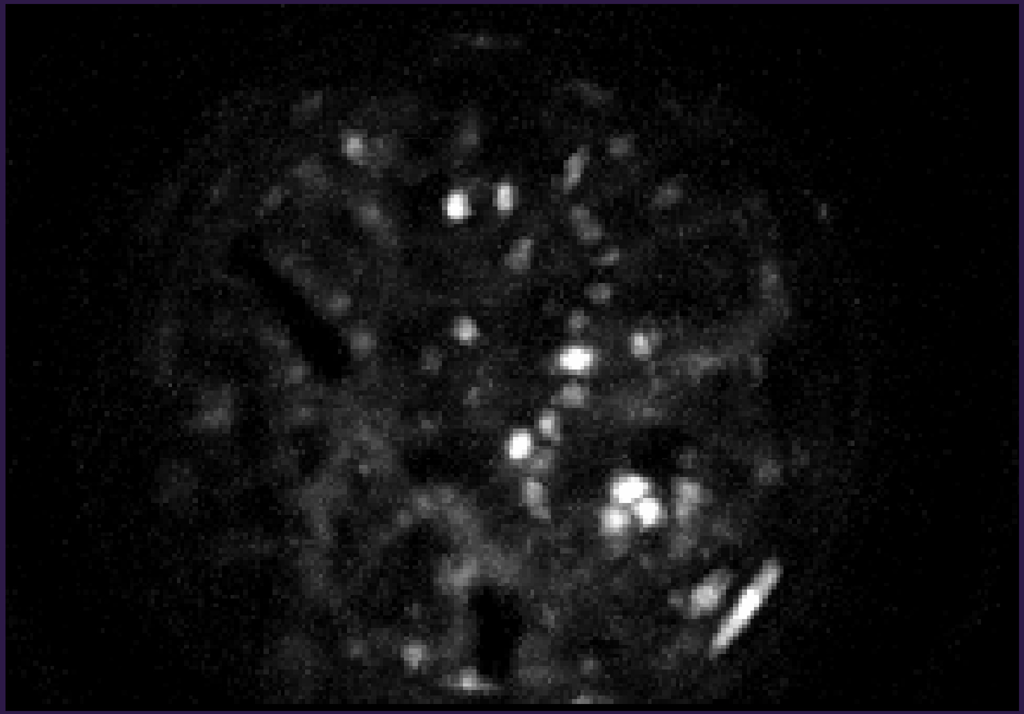


Results

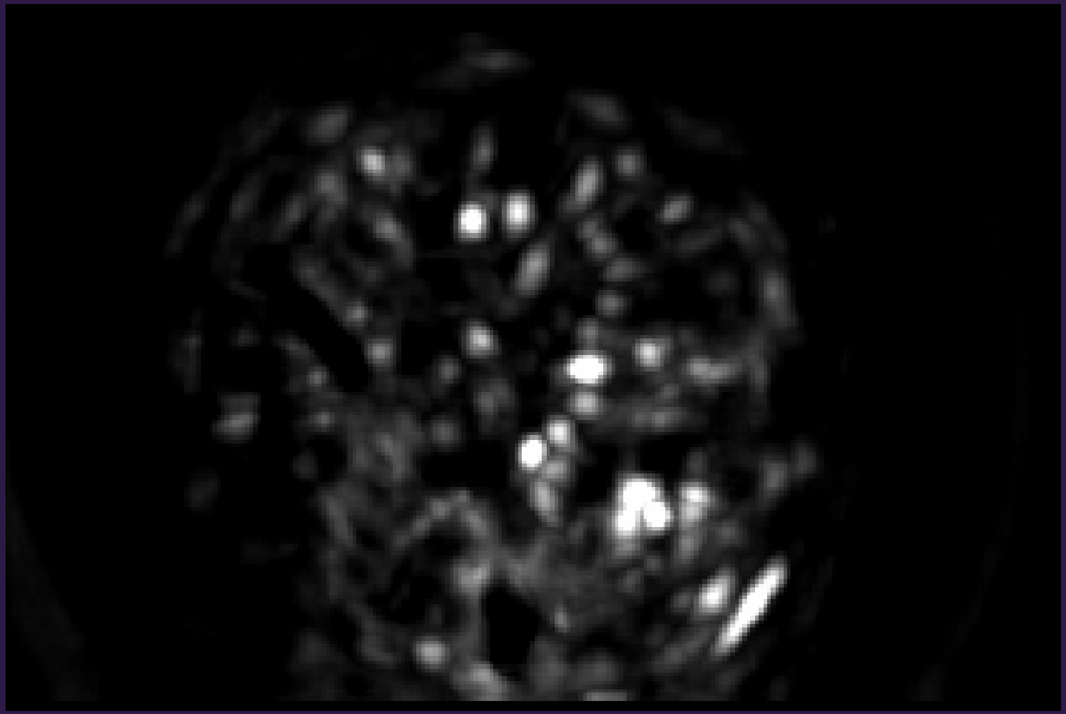


Effectively removed the noise while preserved the subtle neuron activity!

Maximum Projection

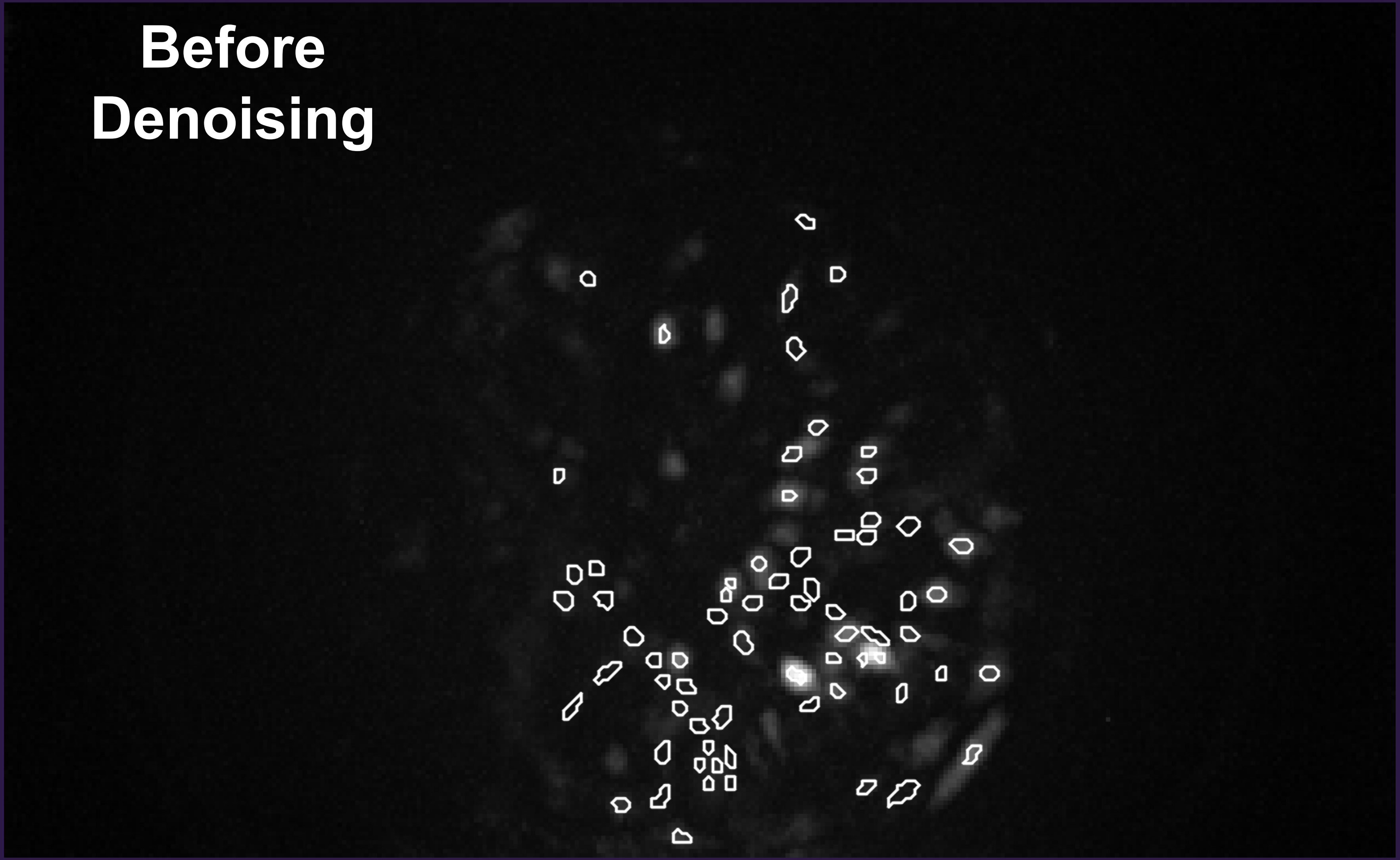


Original

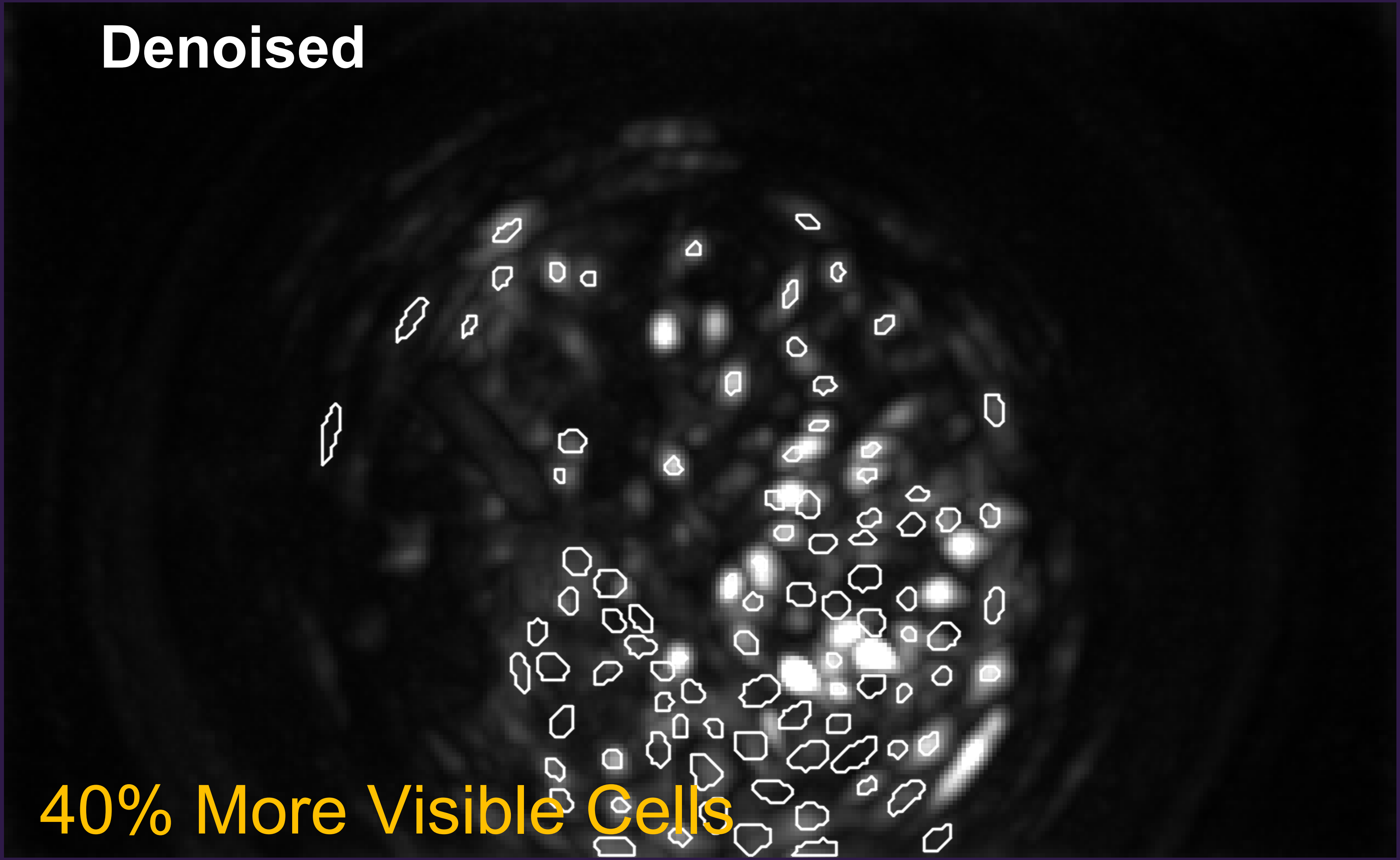


Ours

Results – Cell Tracking Rate



Results – Cell Tracking Rate

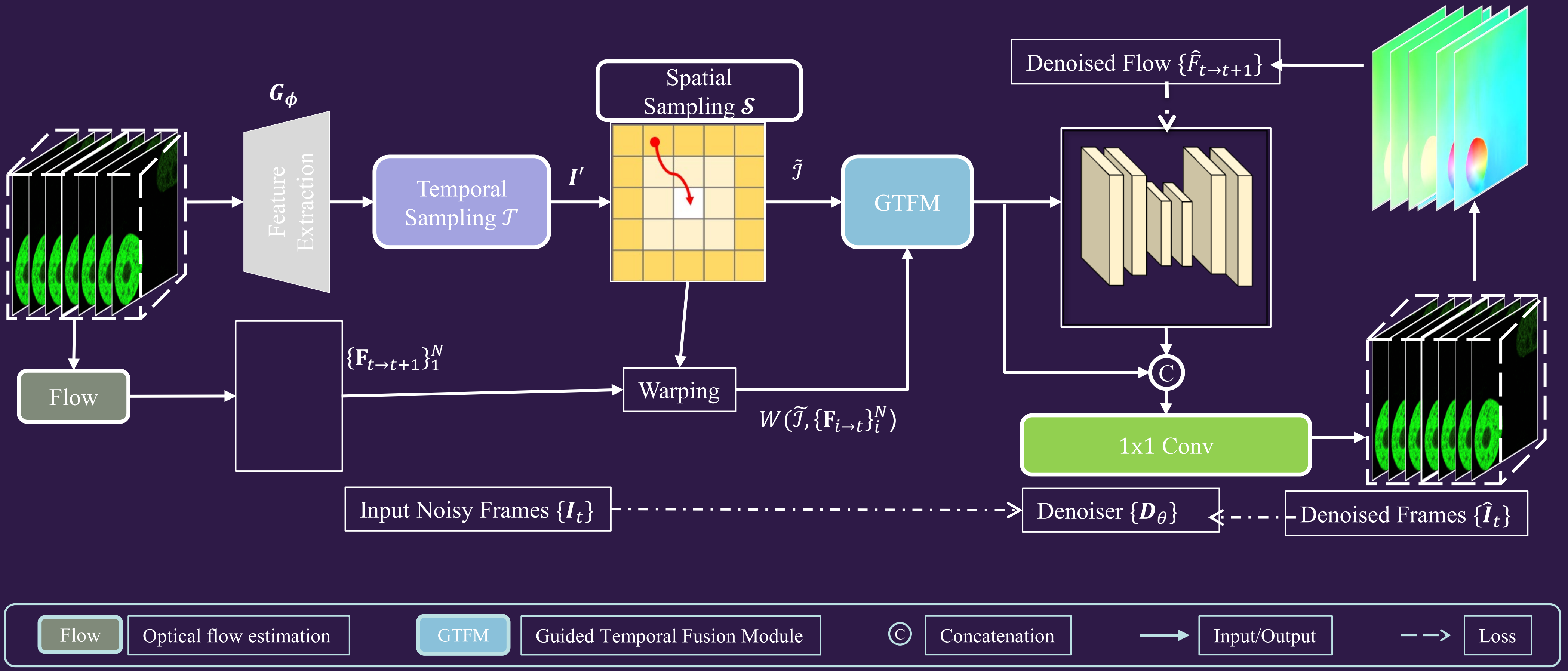


Conclusion

- We presented a novel unsupervised denoising method utilizing DeepTemporal Interpolation.
- Our approach demonstrates strong generalization across a variety of medical imaging and color datasets and shows robustness to different noise types and intensities.
- Limiting motion handling, due to the fixed temporal filter.

Unsupervised Denoising for Multi-Modal Microscopy Imaging for Small Data Scenarios

Method



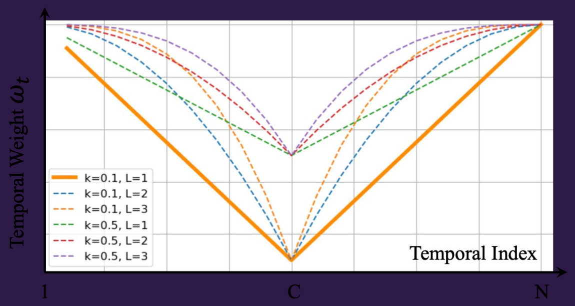
G_ϕ
Feature Extraction
Temporal Sampling $\mathcal{T}$
I'
Spatial Sampling $\mathcal{S}$
$\tilde{\mathcal{J}}$
GTFM
Denoised Flow $\{\hat{F}_{t\to t+1}\}$
Flow
$\{\mathbf{F}_{t\to t+1}\}_1^N$
Warping
$W(\tilde{\mathcal{J}}, \{\mathbf{F}_{i\to t}\}_i^N)$
C
1x1 Conv
Input Noisy Frames $\{I_t\}$
Denoiser $\{D_\theta\}$
Denoised Frames $\{\hat{I}_t\}$
Flow
Optical flow estimation
GTFM
Guided Temporal Fusion Module
C
Concatenation
Input/Output
Loss

Method

Weighted Temporal Sampling

1. Given the Magnitude $\bar{M}$ of optical flow, $\{F_{i\to t}\}_i^N$, we compute the initial minimum weight κ_0.

$$\kappa_0 = 0.2 \cdot \left(1 - e^{-\eta \times \bar{M}}\right) \qquad \kappa = \max\left(0, \kappa_0\left(1 - \frac{\text{epoch}}{\text{max\_epoch}}\right)\right)$$

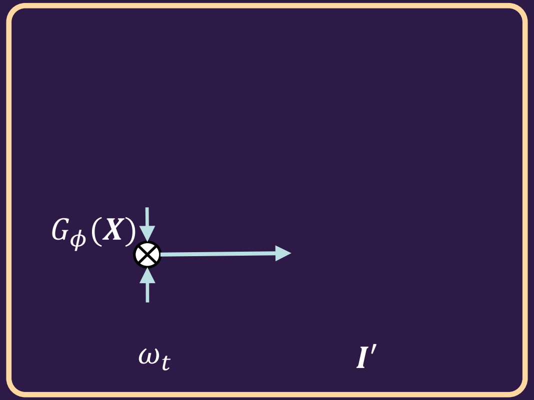


2. We compute the temporal weights $\{\omega_t | t = 1, \ldots, N\}$

$$\omega_t = \begin{cases} \kappa + (1-\kappa)\left(1 - \left(\frac{t}{C}\right)^L\right), & \text{if } t \le C \\ \kappa + (1-\kappa)\left(1 - \left(\frac{2C - t}{C}\right)^L\right), & \text{otherwise,} \end{cases}$$

3. We apply the temporal weights to the features

$$I'_t = \omega_t \odot G_\phi(X_t) \qquad \forall t \in 1, \ldots, N$$

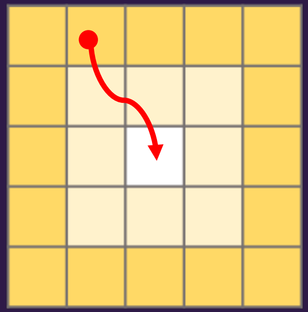


Method

Weighted Spatial Sampling

- **Replaces randomly selected pixels p with neighboring pixels q**

$$P(q|p) = \frac{e^{-\alpha \cdot d(p,q)}}{\sum_{q' \in \Omega_p} e^{-\alpha \cdot d(p,q')}}$$



where:
$P(q|p)$ is the probability of choosing a neighboring pixel
$d(p,q)$ is the l_1 distance between pixels **p** and **q**

Method

Weighted Spatial Sampling

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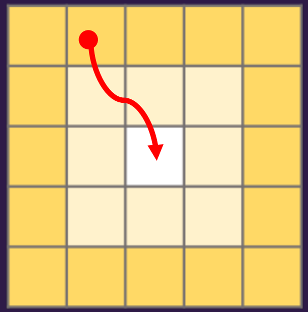

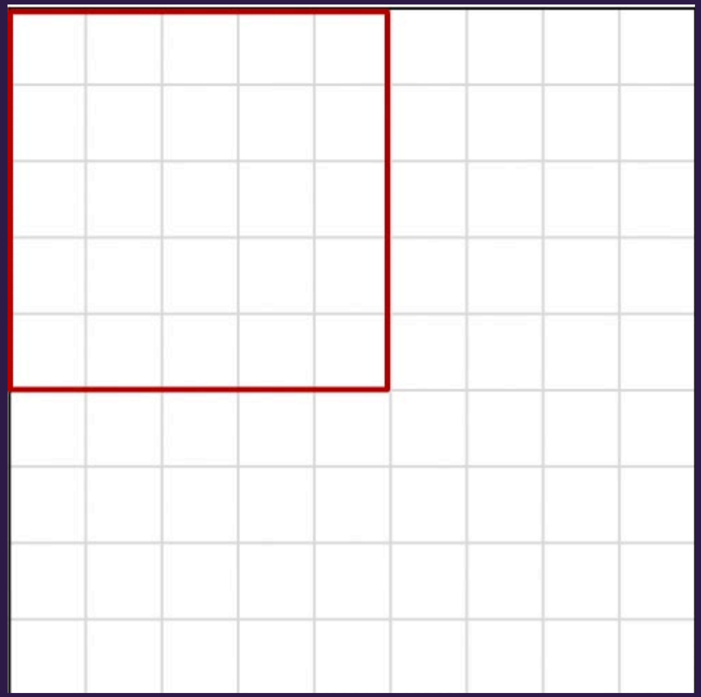

where:
$P(q|p)$ is the probability of choosing a neighboring pixel
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Method

Guided Temporal Fusion

1. Given spatiotemporal sampled features, we first warp the neighboring features to the central frame and the mean warped $\bar{W}$.

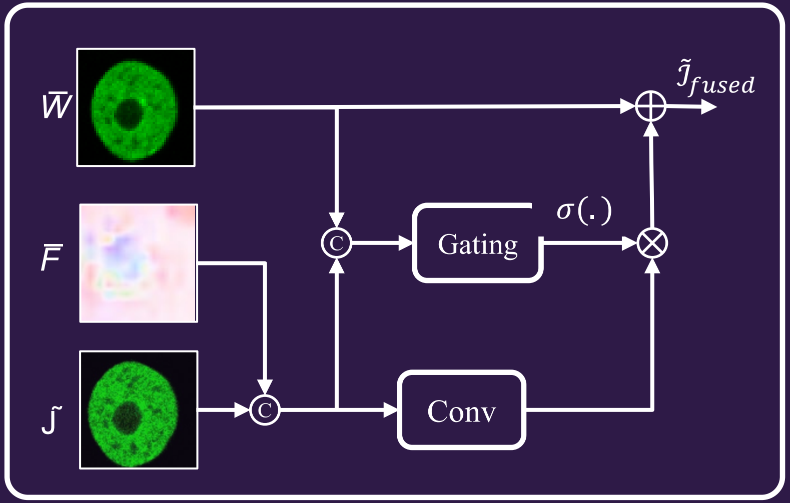
$$\bar{W} = \frac{1}{N\_1} \sum_{i \neq t}^{N} warp(\tilde{J}_i, \mathbf{F}_{i \to t})$$

2. We concatenate the mean flow $\bar{F}$, mean warped $\bar{W}$ and the central feature and pass to gating network to yield spatial attention map $\sigma(.)$

3. We concatenate the mean flow $\bar{F}$ and the central feature through convolution to get residual candidate.

4. Finally, we fused the temporal features using:

$$\tilde{J}_{fused} = \bar{W} + \sigma * \Delta\tilde{J}_t$$



Method

Recurrent Optimization

- To effectively guide the network during training, we employ two loss terms. One is the pixel-wise L1 loss between $\hat{I}_t$ and the reference frame I_t^{ref}, and the second loss term is a consistency loss based on the optical flow estimation of the denoised frames.

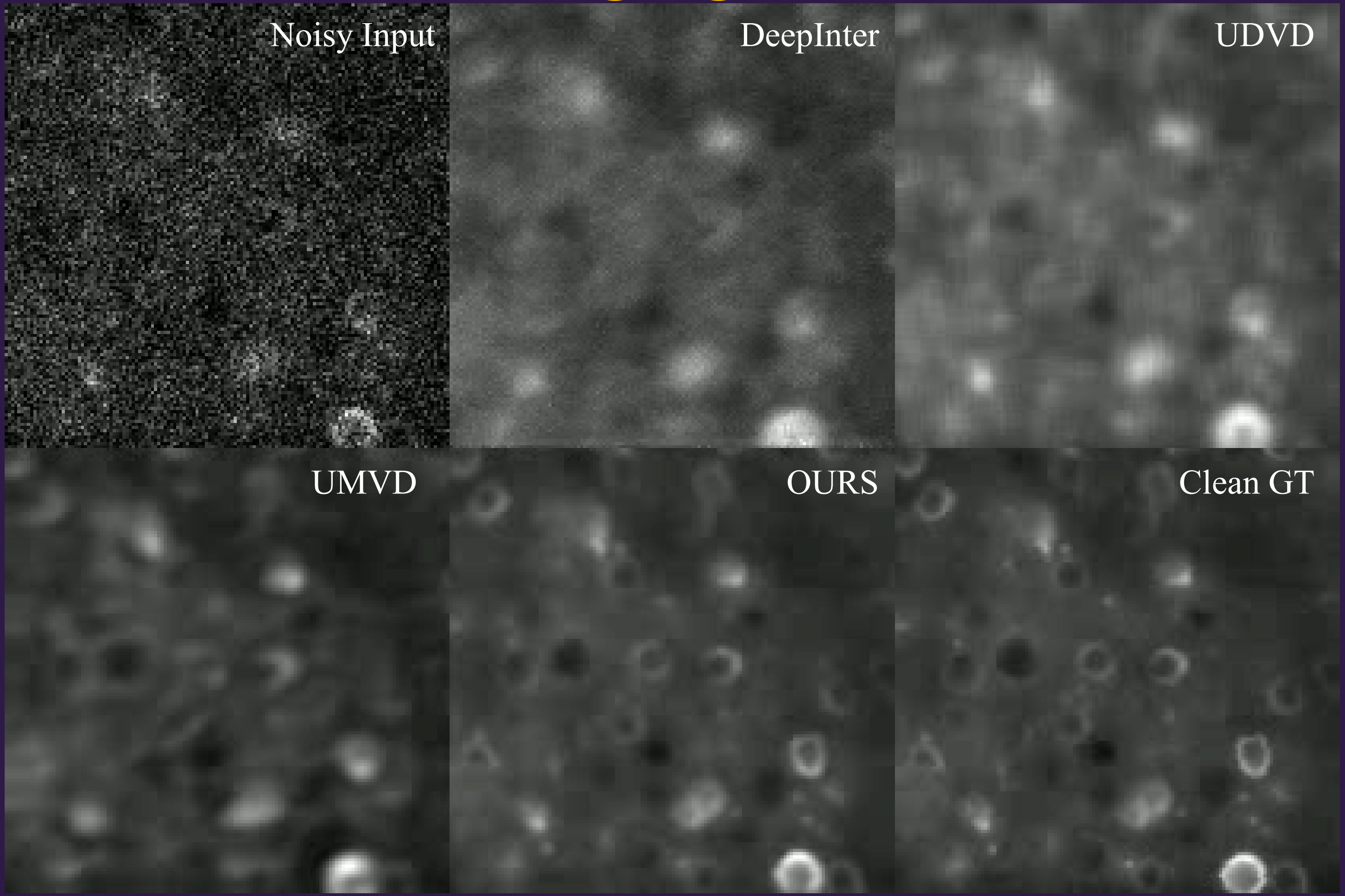
$$\mathcal{L}_1 = \sum_{p \in I_t} \left| \hat{I}_t(p) - I_t^{ref}(p) \right|$$

$$\mathcal{L}_2 = \frac{1}{N} \sum_{t=1}^{N-1} \left\| W(\hat{I}_{t+1}, \hat{F}_{t \to t+1}) - \hat{I}_t \right\|^2$$

$$\mathcal{L} = \mathcal{L}_1 + \mathcal{L}_2$$

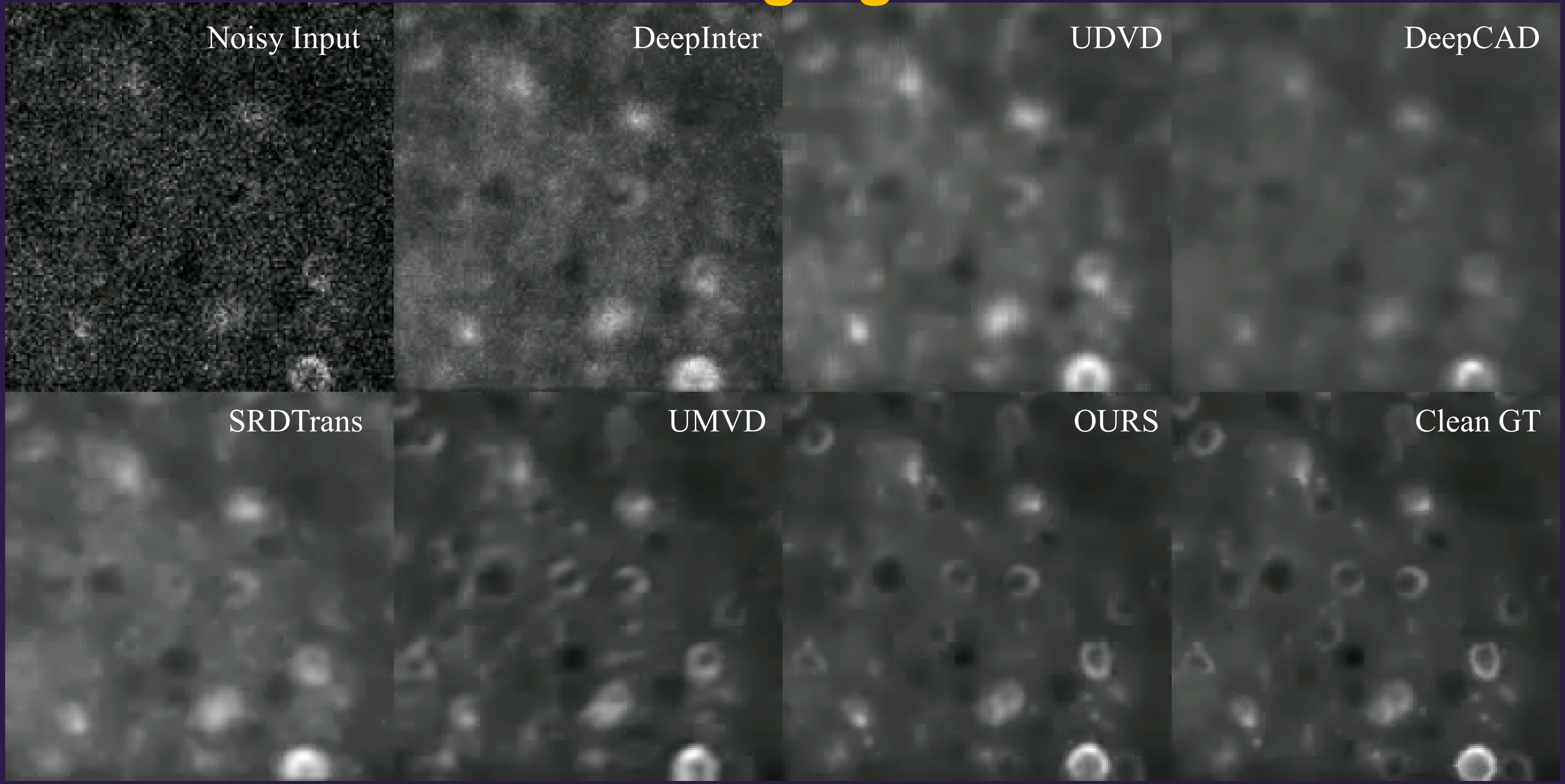
Result

Calcium Imaging – 10 Frames



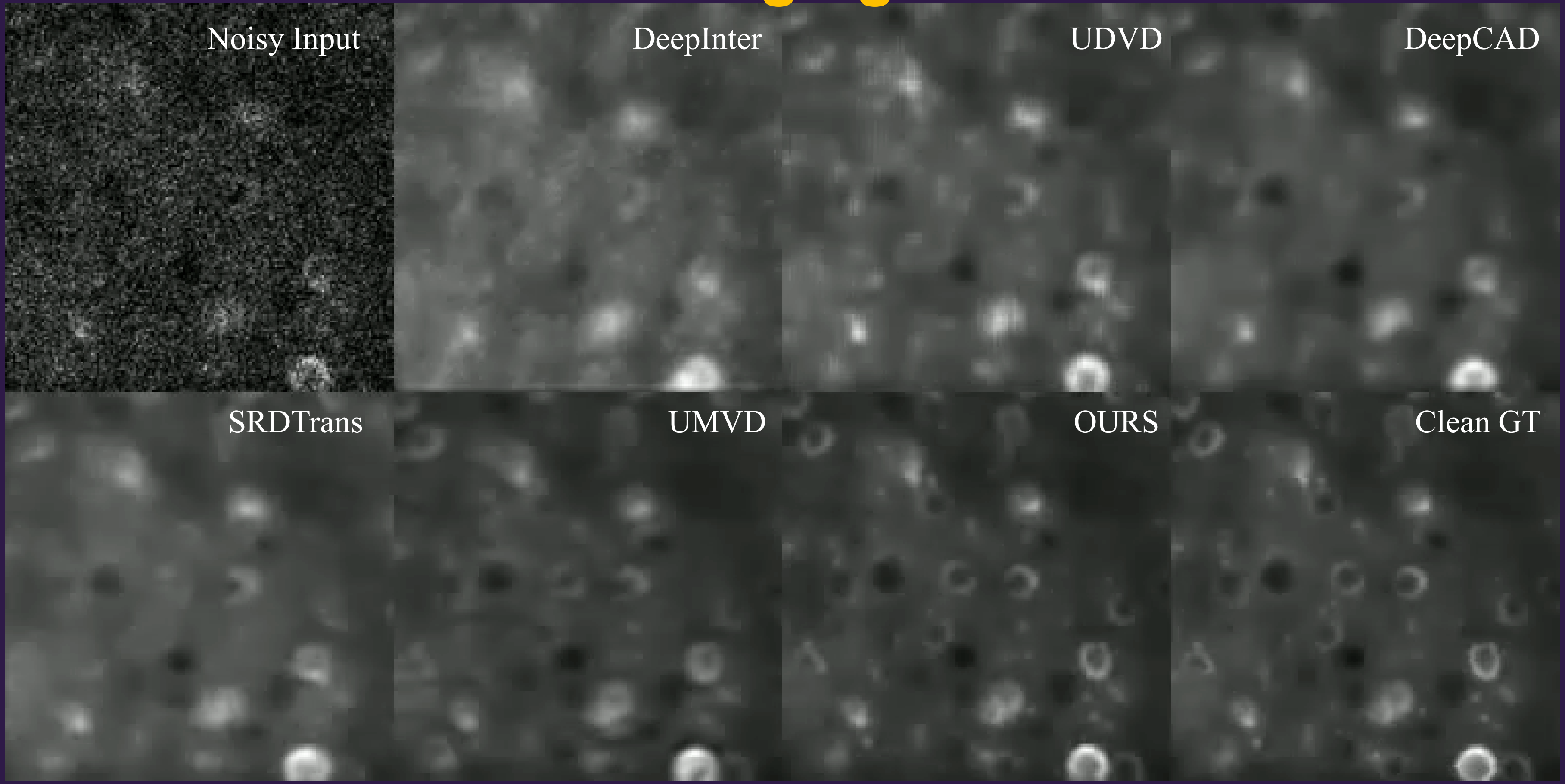
Result

Calcium Imaging – 20 Frames



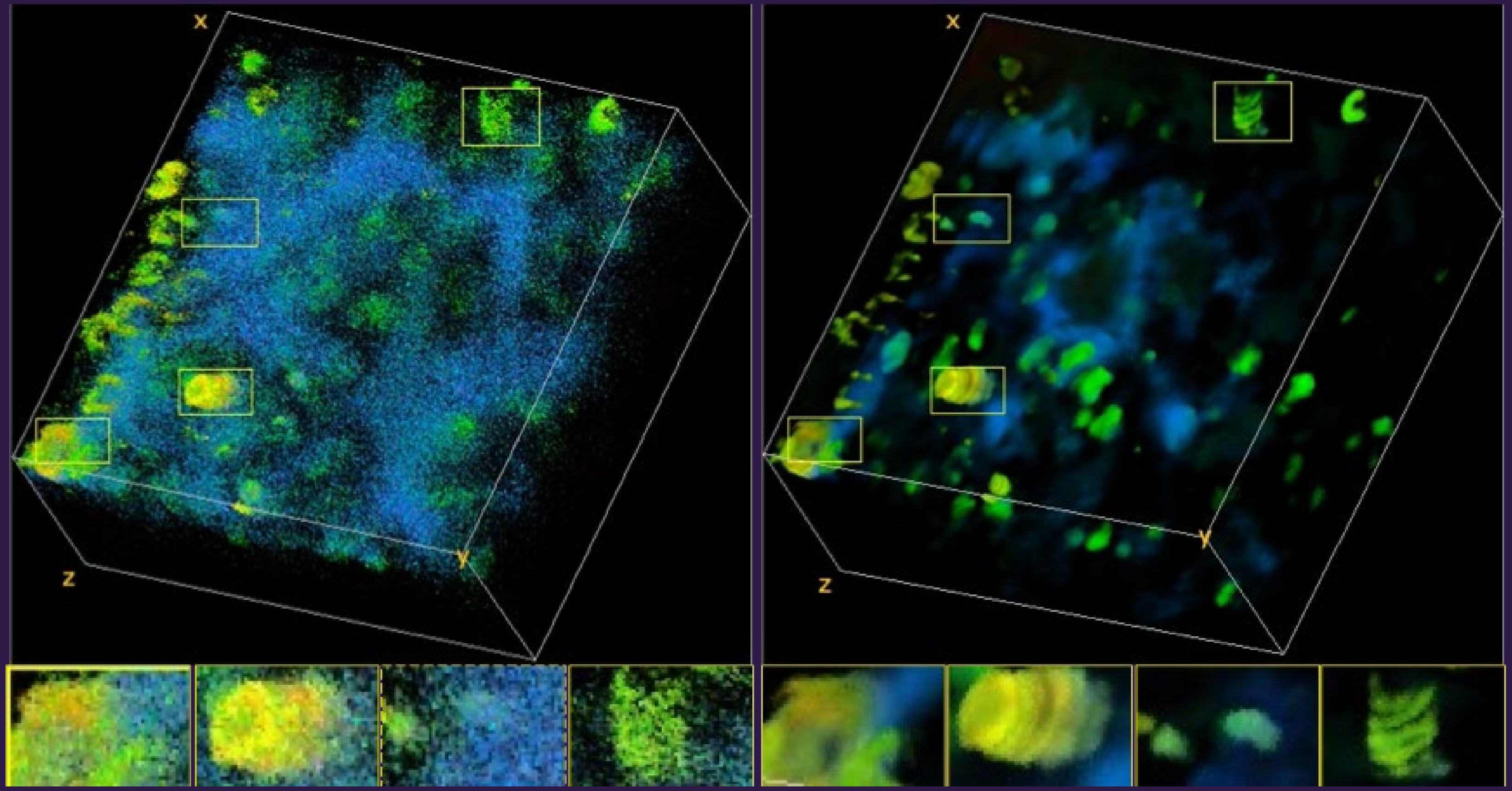
Result

Calcium Imaging – 50 Frames



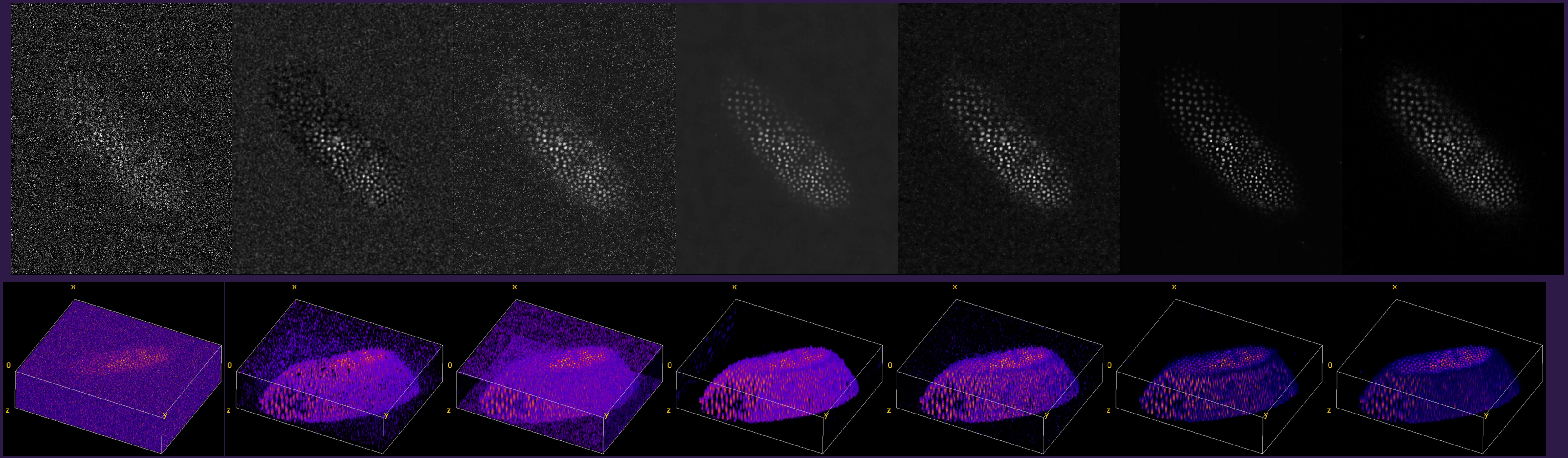
Result

Confocal Imaging



Results

Confocal Imaging



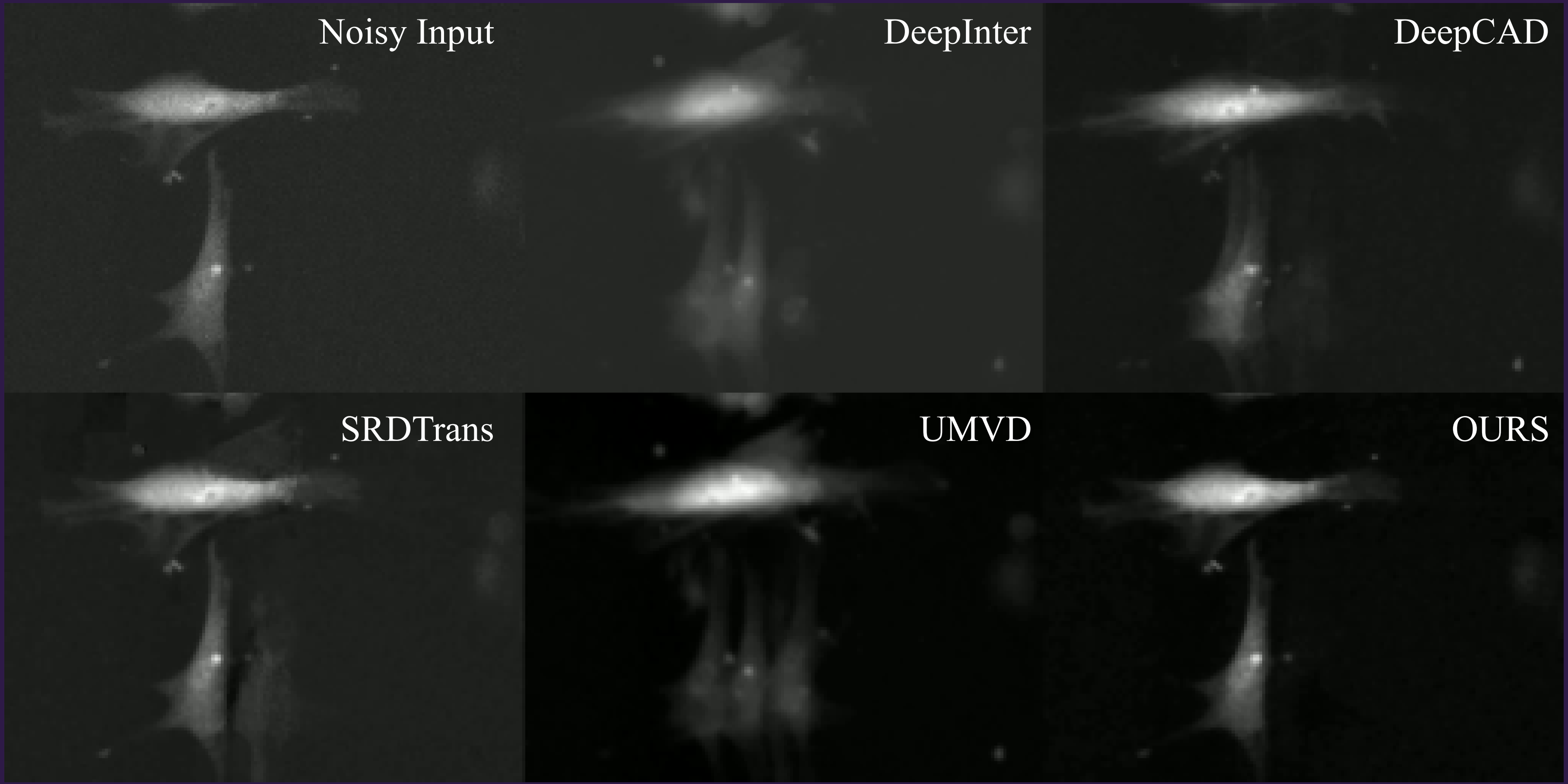
Noisy Input DeepCad DeepInterpolation UDVD UMVD SRDTrans Ours

Result

Fluorescence Microscopy

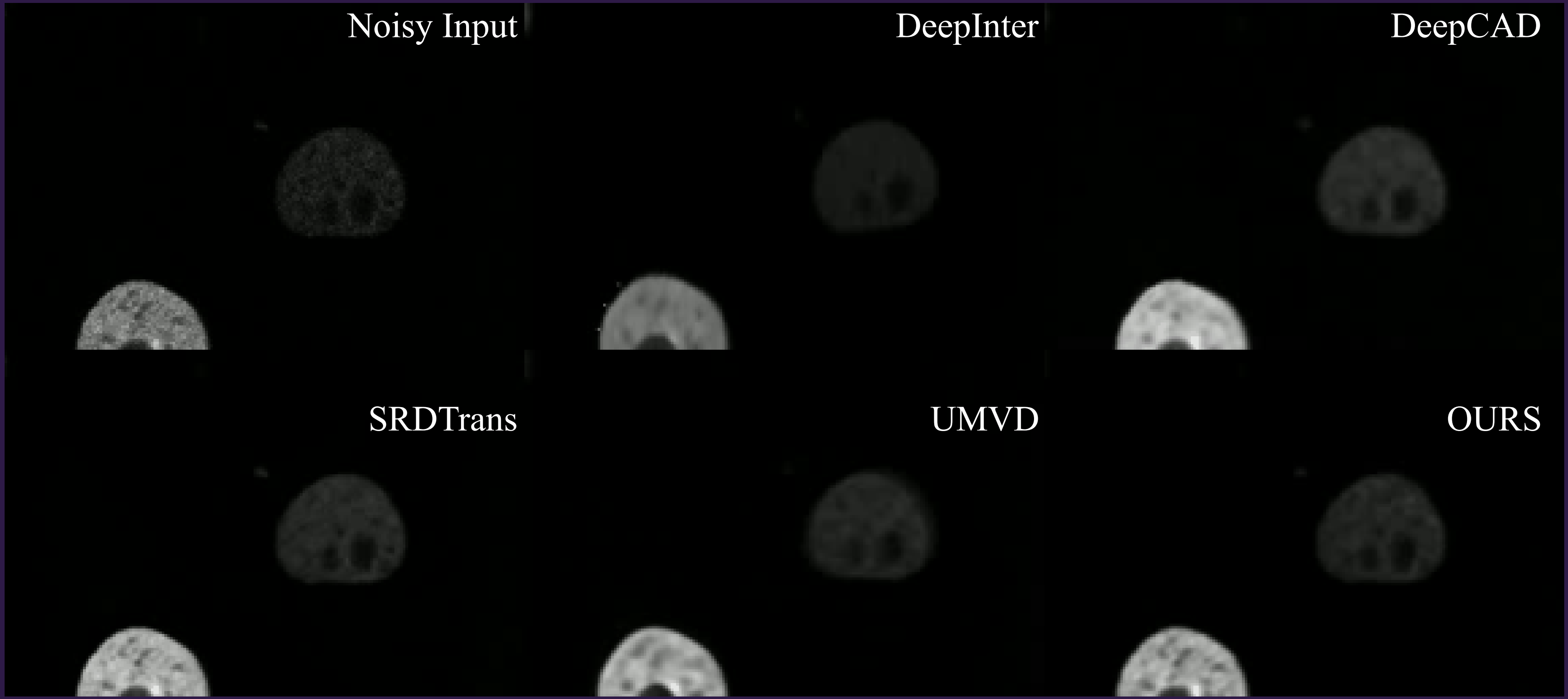
Result

Fluorescence Microscopy



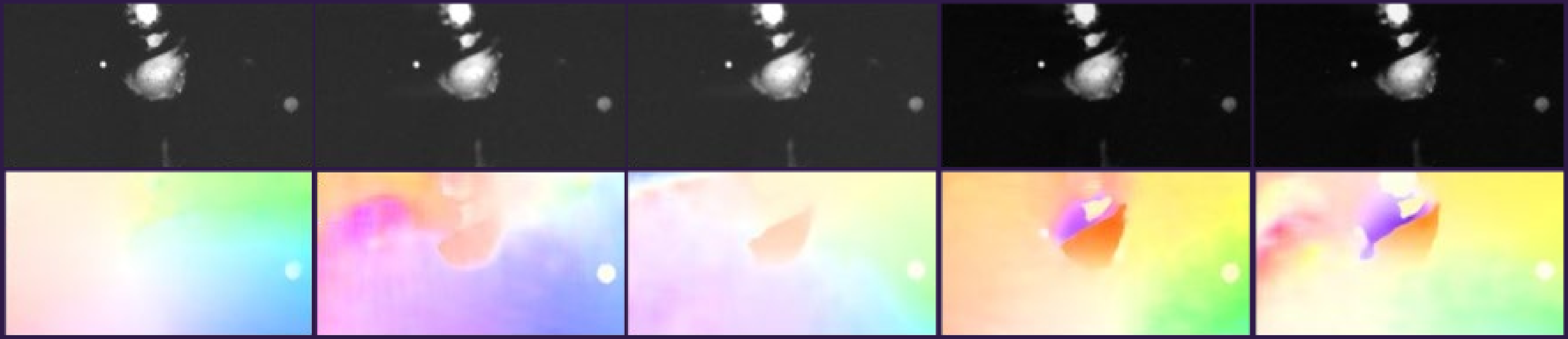
Result

Fluorescence Microscopy



Result

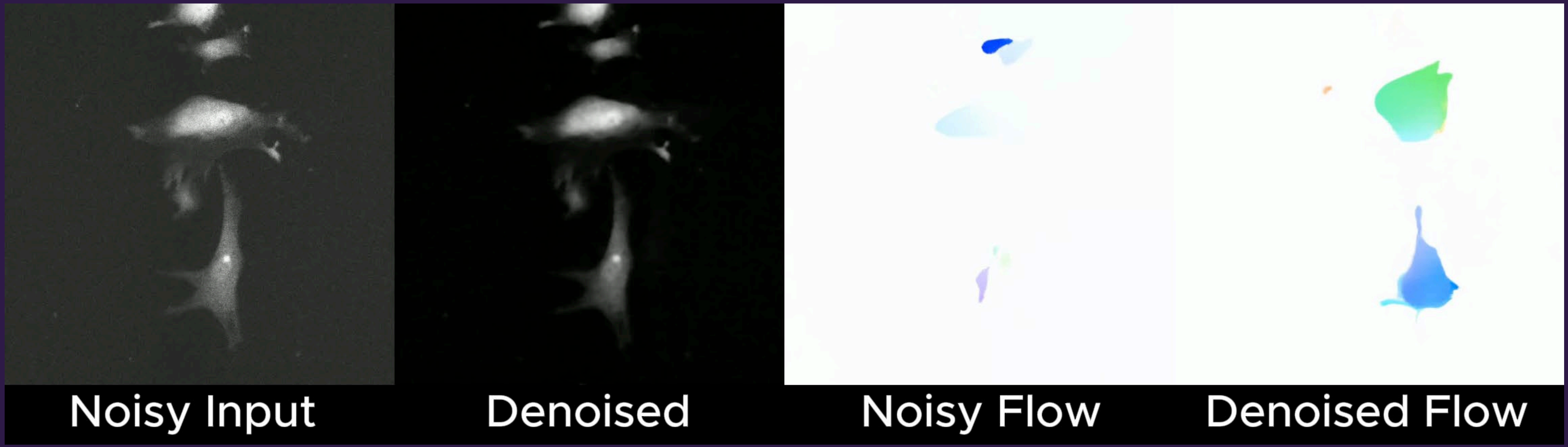
Refined Flow



Noisy Input | Epoch 1 | Epoch 2 | Epoch 4 | Denoised Output

Result

Optical Flow



Quantitative Results

- Quantitative comparison with SOTA video denoising techniques on simulated two-photon calcium imaging.

	DeepI		DeepCAD		UDVD		SRDTrans		UMVD		Ours	
	PSNR ↑	SSIM ↑	PSNR ↑	SSIM ↑	PSNR ↑	SSIM ↑	PSNR ↑	SSIM ↑	PSNR ↑	SSIM ↑	PSNR ↑	SSIM ↑
150 μm²	30.020	0.911	36.606	0.971	31.889	0.899	41.176	0.984	38.871	0.985	43.970	0.992
500 μm²	34.222	0.907	29.130	0.885	35.775	0.932	34.595	0.924	32.933	0.909	37.501	0.952
Average	32.121	0.909	32.868	0.928	33.832	0.916	<u>37.886</u>	<u>0.954</u>	35.902	0.947	**40.736**	**0.972**

Conclusion

- We presented a novel unsupervised denoising method which dynamically adapts to observed motion in videos by utilizing weighted SpatioTemporal sampling.
- Our approach demonstrates strong generalization across a variety of medical imaging datasets.
- To improve scalability to high-resolution or real-time settings, future work will explore lightweight optical flow models and adaptive sampling strategies.

Thanks to all Collaborators and Sponsors!

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Undergraduate
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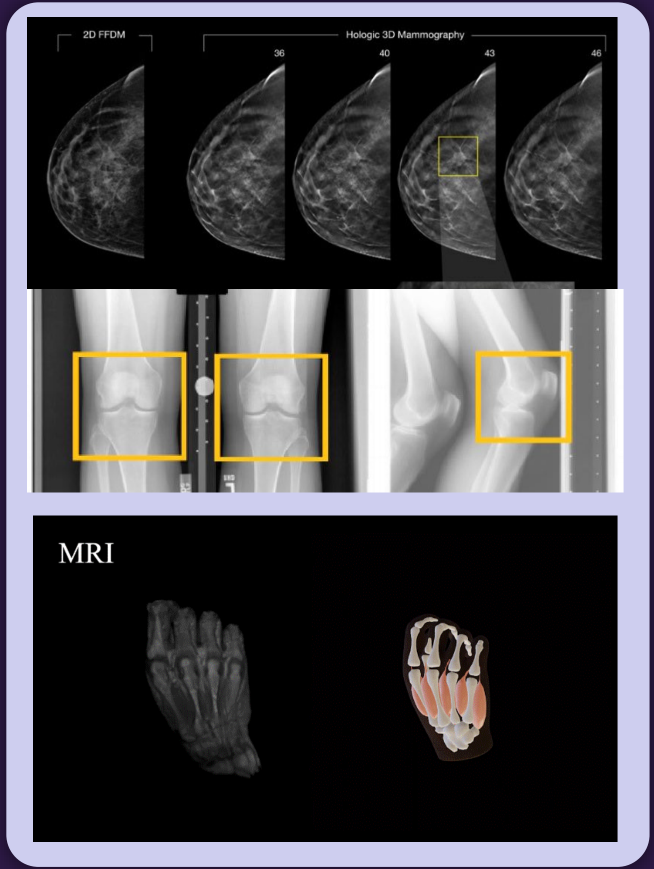
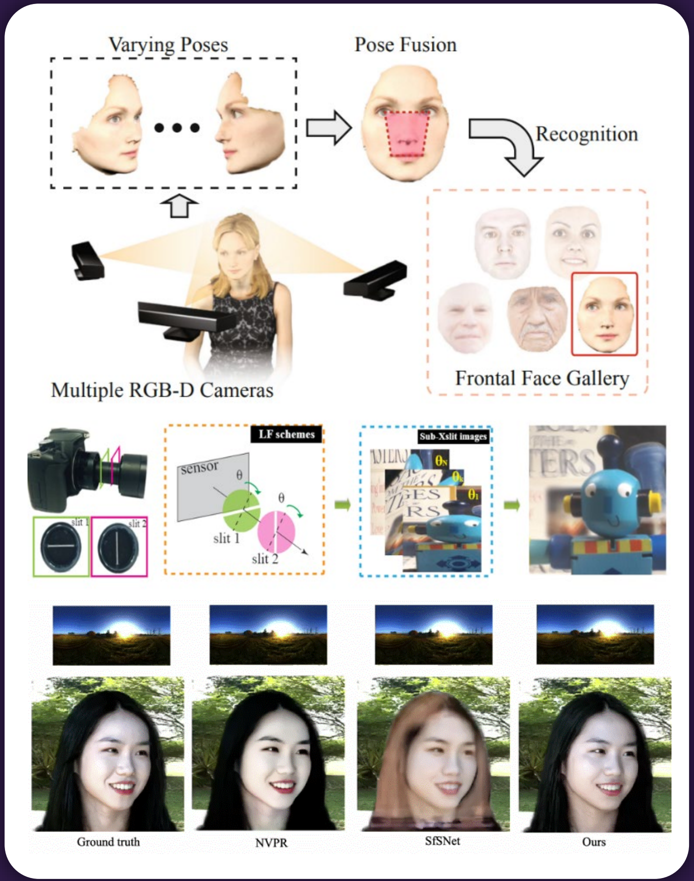
SC EPSCoR

Advanced Imaging Laboratory

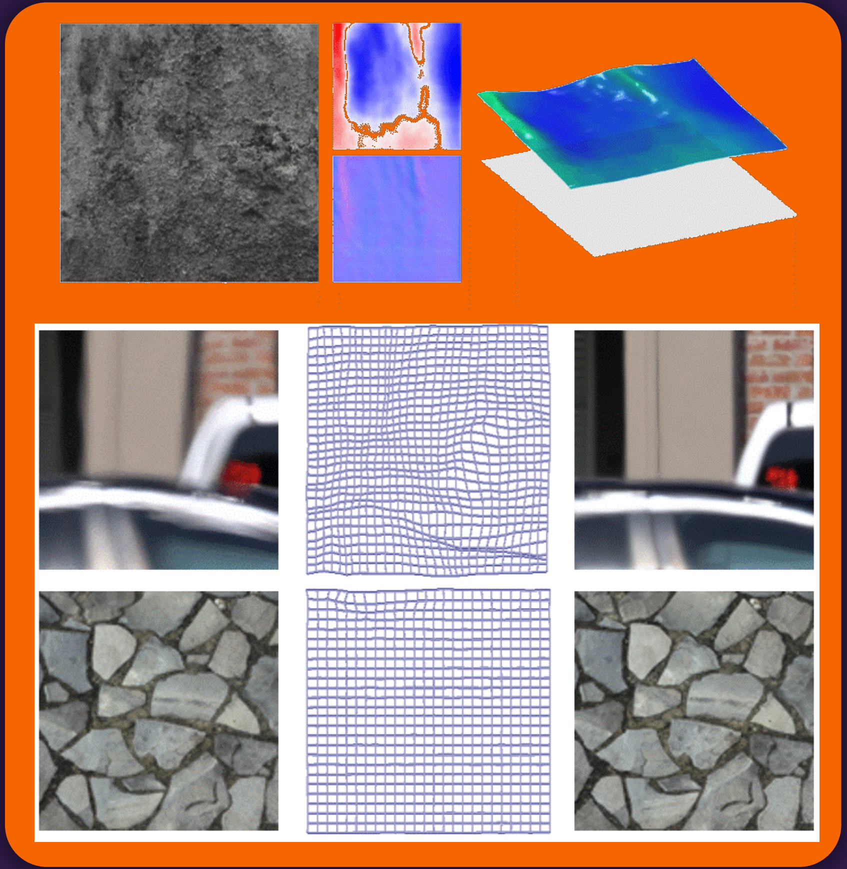
Medical Image Processing



Computer Vision



Physics-informed Machine Learning



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THANK YOU

Project Website

https://nianyil.people.clemson.edu/