



# Utilization of Deep Learning Methods for Automatic Reconstruction of Quantitative Phase Images in Non-telecentric Digital Holographic Microscopy

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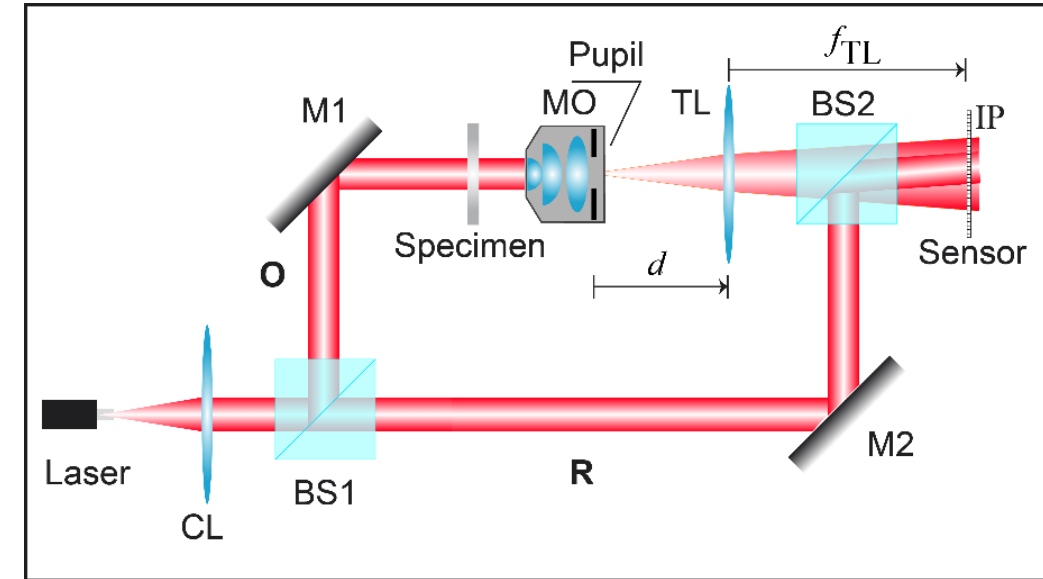
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# Digital Holographic Microscopy (DHM)

- ❑ Principle: DHM offers advantages of recording and reconstructing complex object distributions (e.g., both amplitude and phase information)
- ❑ Advantages of DHM
  - ❑ Capable of 3-D reconstructions of samples in a single shot.
  - ❑ Label-free imaging technique
  - ❑ Perfect candidate for live-cell imaging.
- ❑ Recording: interferometry (e.g., interference between two wavefronts – object and reference waves)



**FIGURE.** Optical configuration of an off-axis DHM system based on a modified Mach-Zehnder interferometer. In a general case, the microscopic objective (MO) lens and the tube lens (TL) are arranged in non-telecentric mode ( $d \neq f_{TL}$ ). The remaining components of the system are denoted as: CL, converging lens; BS, beamsplitter; IP, image plane; M, mirror; O, object wave; R, reference wave.



# Non-telecentric DHM systems provide phase images distorted by a spherical phase term introduced by the optical imaging system

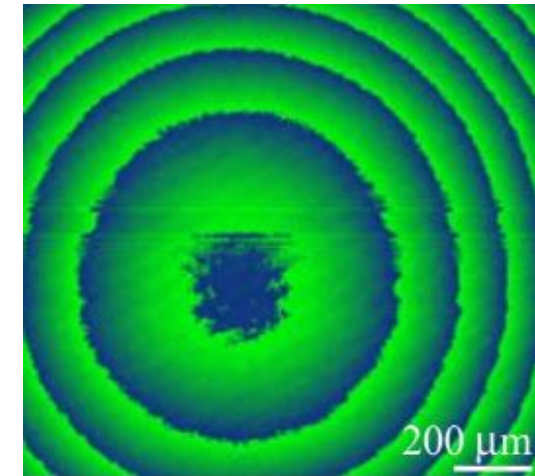
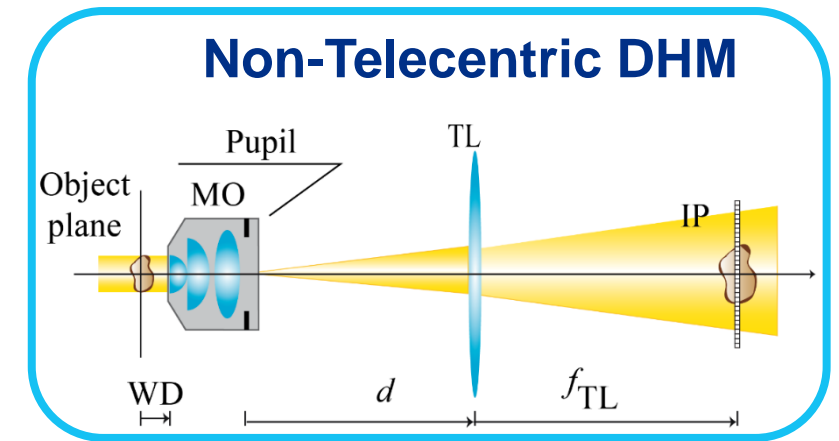
Complex amplitude distribution at the image plane (IP)

$$U_{IP}(\mathbf{x}) \propto \frac{1}{M^2} \exp\left(i \frac{k}{2C} |\mathbf{x}|^2\right) \times \left\{ O\left(\frac{\mathbf{x}}{M}\right) \otimes_2 \tilde{p}\left(\frac{\mathbf{x}}{\lambda f_{TL}}\right) \right\}$$

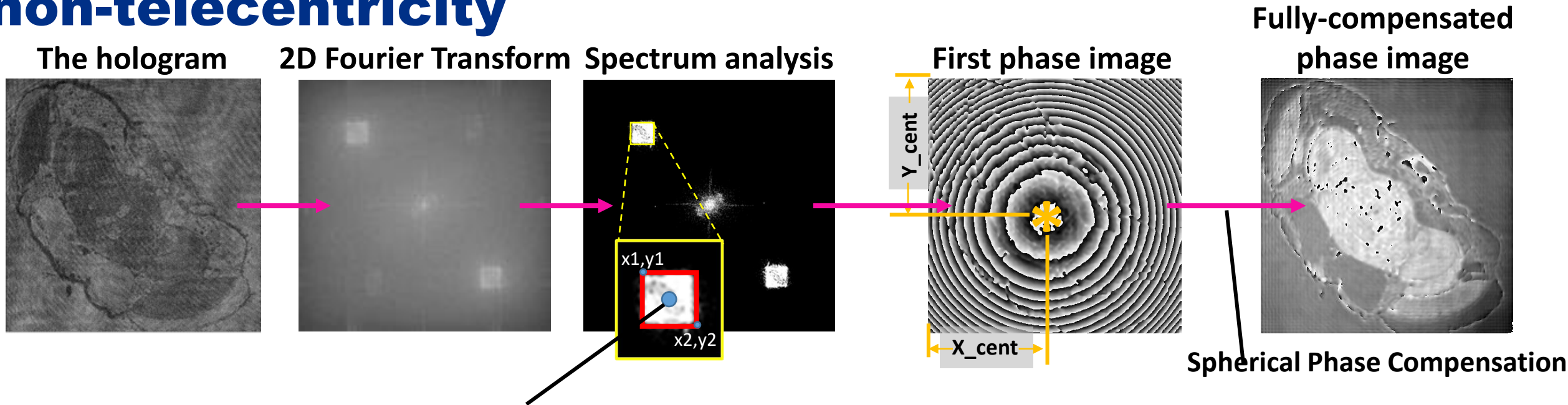
$$M = -f_{TL} / f_{MO}$$

$$C = f_{TL}^2 / (f_{TL} - d)$$

- ❑ The non-telecentric DHM systems are **shift-variant**.
- ❑ The phase measurement depends on the object position within the whole field of view.



# Tedious algorithm to compensate the spherical phase term introduced by the non-telecentricity



Reference wave

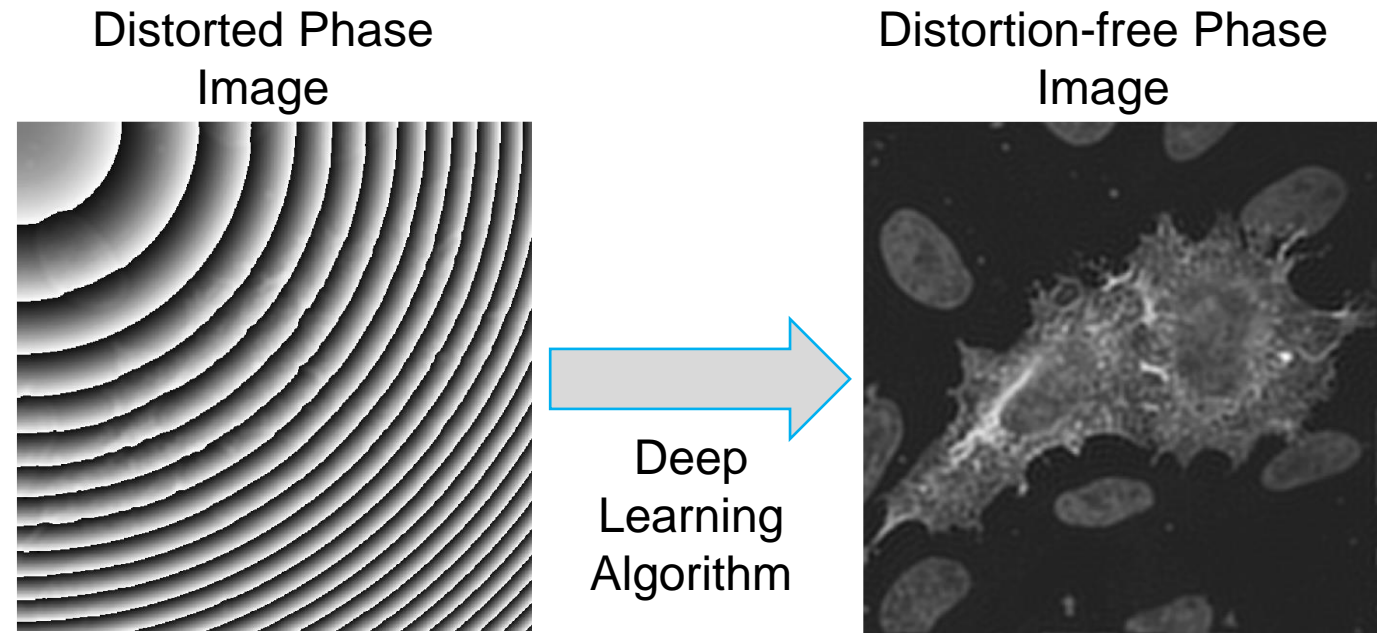
Steps:

1. Spectrum filtering with a rectangular filter from  $(x1, y1)$  to  $(x2, y2)$
2. Inverse Fourier Transform
3. Compensation of the tilt with a digital reference wave with position is the centroid of the rectangular filter



# Use of Deep Learning to correct the spherical distortions

- ❑ We propose the application of deep learning artificial intelligence algorithms for automatically correcting and reconstructing images from holograms obtained with non-telecentric Mach-Zehnder DHM systems.
- ❑ Convolutional neural network model: the **pix2pix** conditional Generative Adversarial Network (cGAN).
- ❑ The model that was trained on a simulated, and then evaluated on the accuracy of its reconstruction ability.

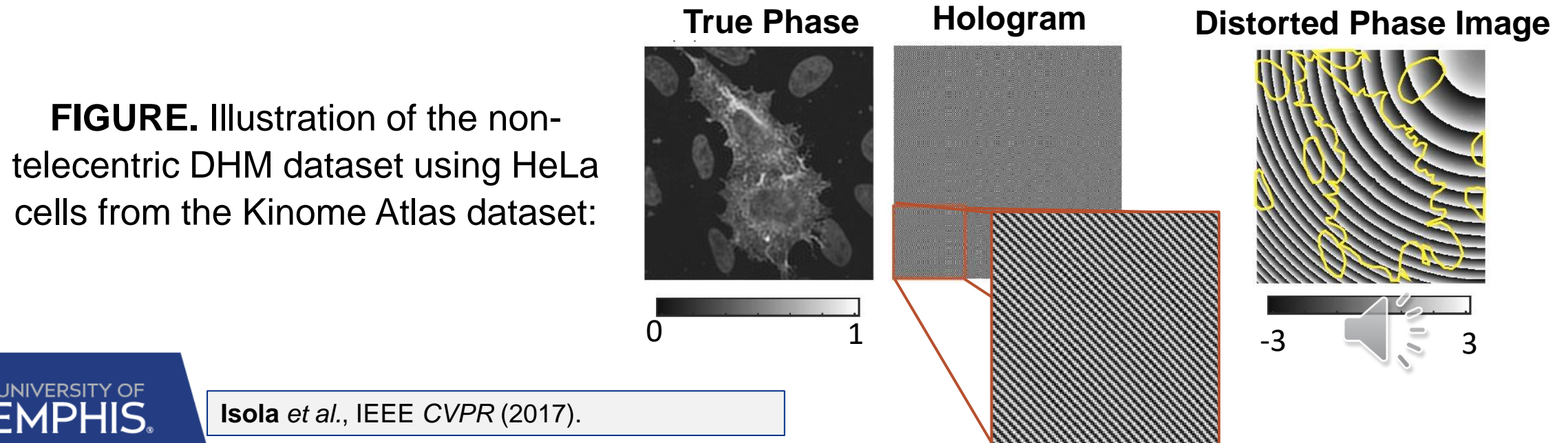


**FIGURE.** Description the proposed system and its intended functionality. Left image portrays the distorted image resulting from spherical aberrations and right image is the desired computationally corrected image.



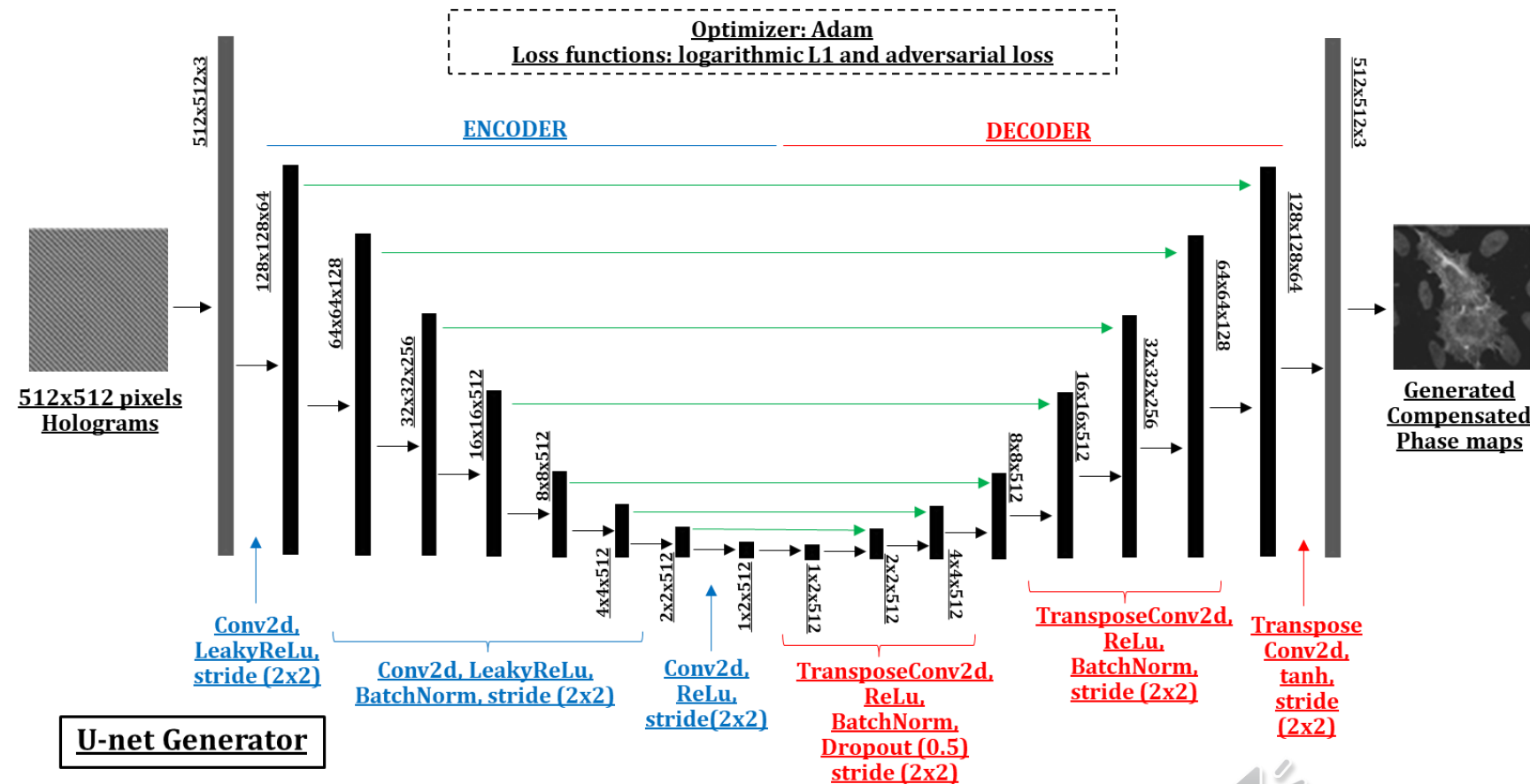
# Generation of the simulated dataset

- Kinome Atlas dataset from the Cell Image Library [8]: with 907 total images, of size 512x512 pixels
- Dual-path Mach-Zehnder DHM
- Hologram:  $h(x, y; z = 0) = |u(x, y)|^2 + |r(x, y)|^2 + u(x, y)r^*(x, y) + u^*(x, y)r(x, y)$
- Object beam:  $u(x, y) = \frac{1}{M^2} \exp\left[j \frac{k}{2C} (x^2 + y^2)\right] \times \left[o\left(\frac{x}{M}, \frac{y}{M}\right) \otimes_2 P\left(\frac{x}{\lambda f_{TL}}, \frac{y}{\lambda f_{TL}}\right)\right]$
- 3 simulated aberrations where  $C = 3,500, 4000, 5000$



# The PIX2PIX Conditional Generative Adversarial Network (cGAN)

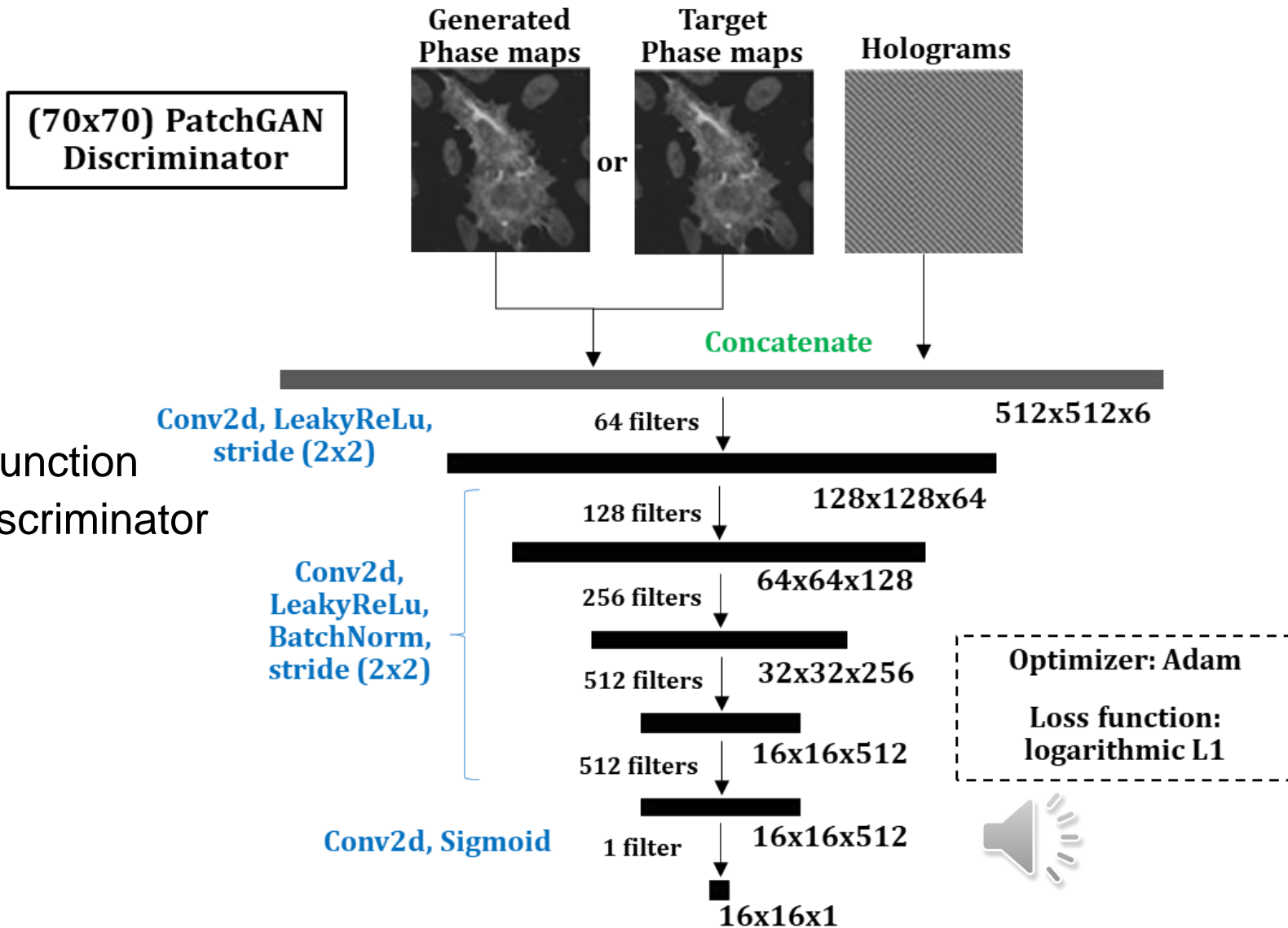
- pix2pix cGAN
- cGAN are a U-Net & CNN
- U-Net: Generator
- Encoding
  - Convolution 4x4 filters
  - Batch normalization
  - LeakyReLU activation
  - Final layer: tanh
- Decoding
  - ReLU



U-Net Architecture for the Generator of that cGAN.

# The PIX2PIX Conditional Generative Adversarial Network (cGAN)

- pix2pix cGAN
- cGAN are a U-Net & CNN
- CNN: Discriminator
  - 4x4 filters
  - LeakyReLU
  - Last layer uses a sigmoid function
  - Generator needs to trick discriminator

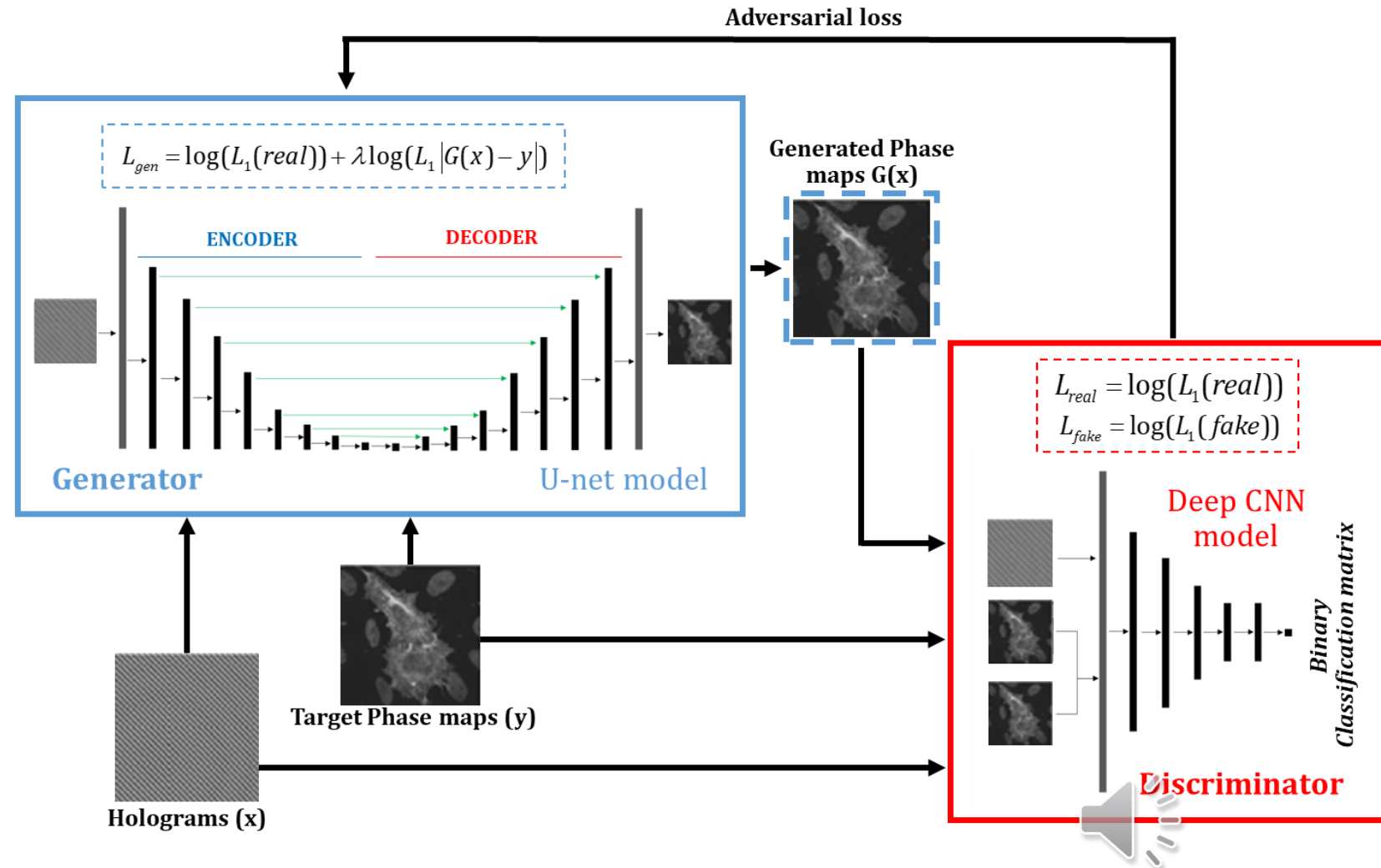


PatchGAN CNN Architecture for the Discriminator of that cGAN.



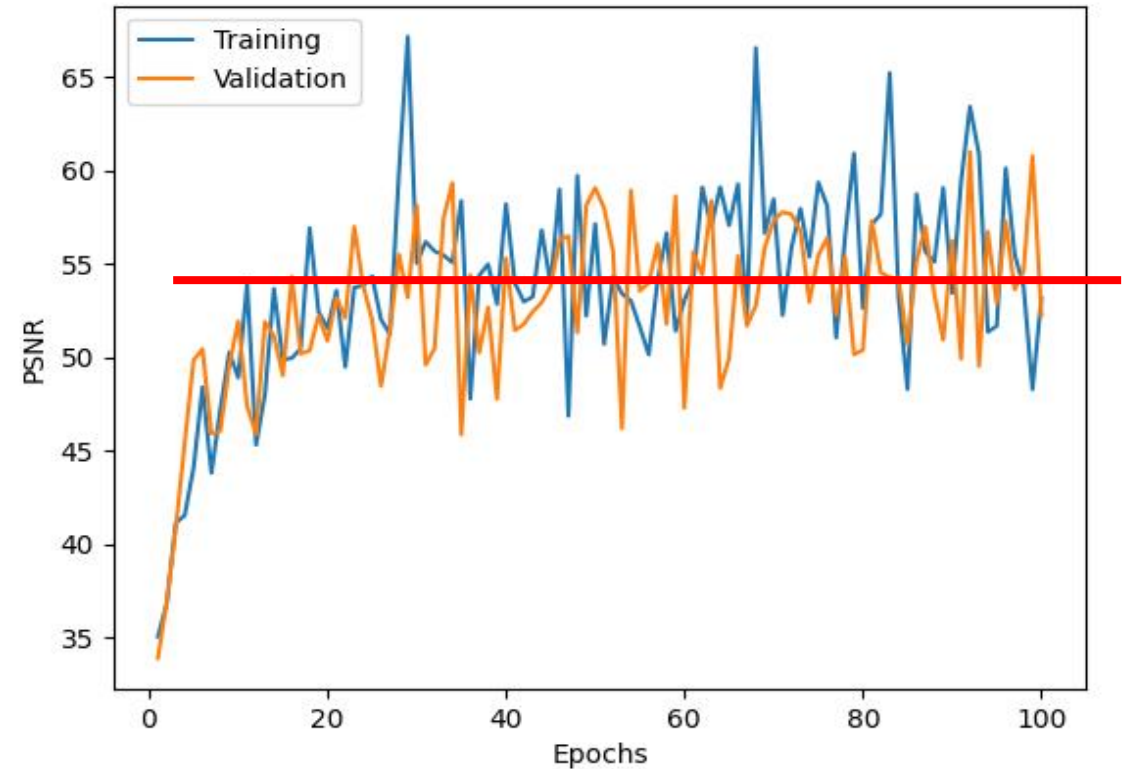
# Metrics and Hyperparameters used to train the cGAN

- Mean Absolute Error (MAE) by the generator
- Binary cross-entropy used by the discriminator
- Adam optimizer
  - Learning rate: 0.0002
  - beta-1 value of 0.5
  - beta-2 value of 0.999
- Training Procedure:
  - 100 epochs
  - batch size of 1



# Results – average Peak Signal-to-Noise Ratio (PSNR)

- Training/testing split of 80/20.
- Peak signal-to-noise ratio (PSNR)
- For Each epoch, the average PSNR of 15 random images in the training and test dataset is computed.
- The behavior of PSNR values is quite noisy.
- Trend of the PSNR fluctuates within a mean value after epoch 30.

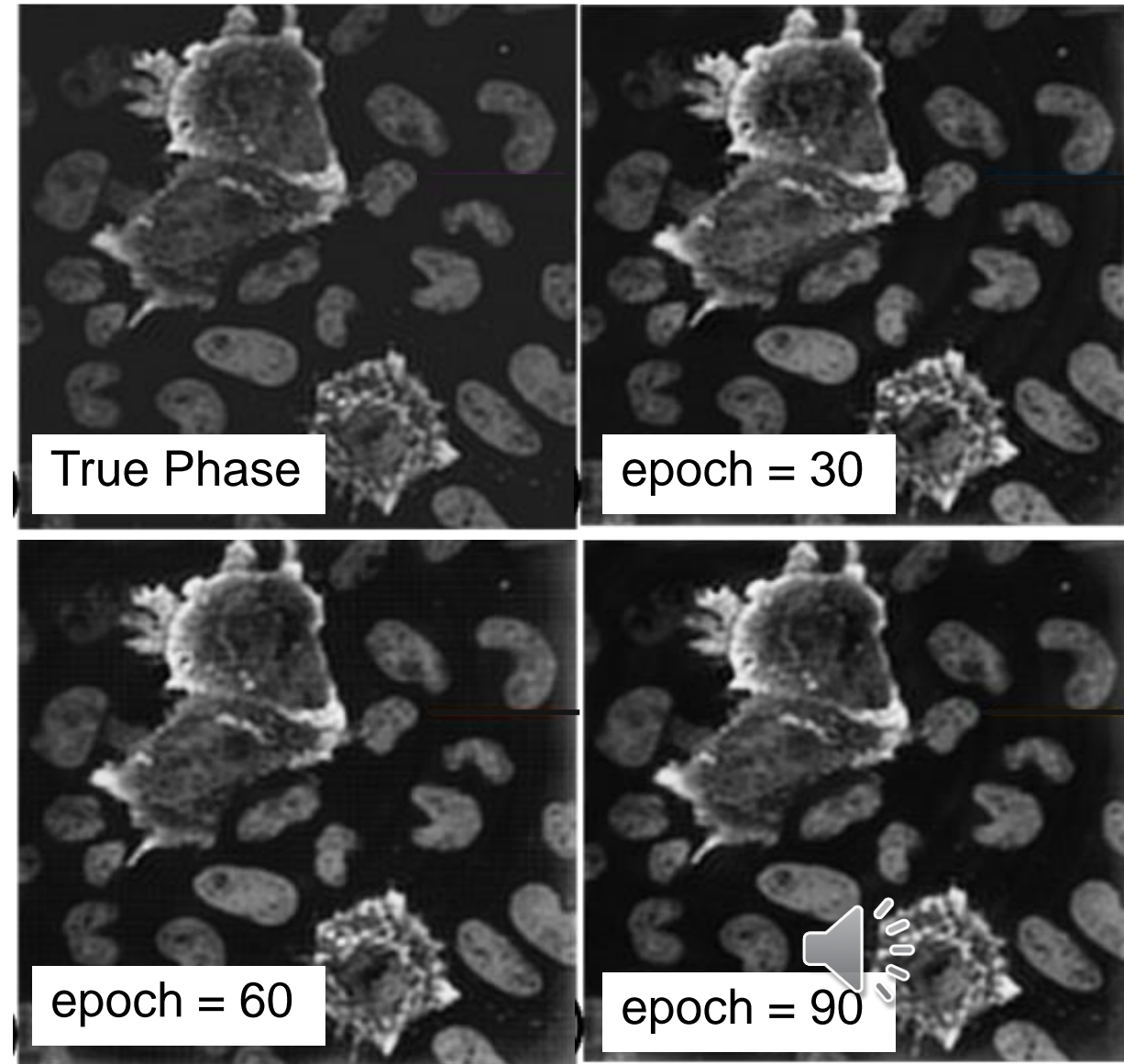


**FIGURE.** Peak Signal-to-Noise Ratio (PSNR) vs Number of Epochs of the Training and Testing datasets.



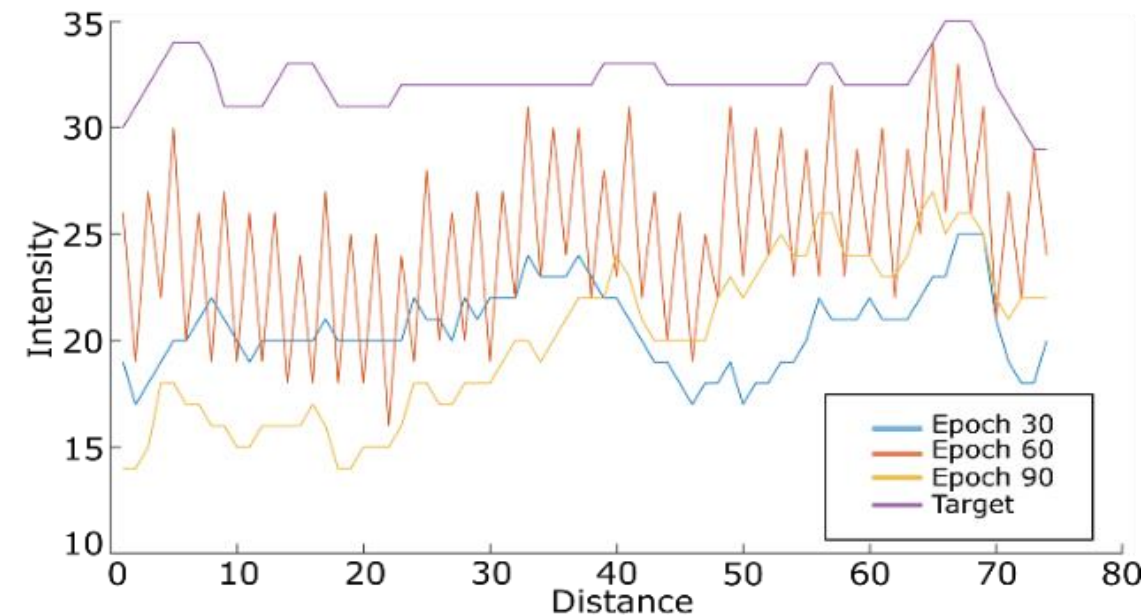
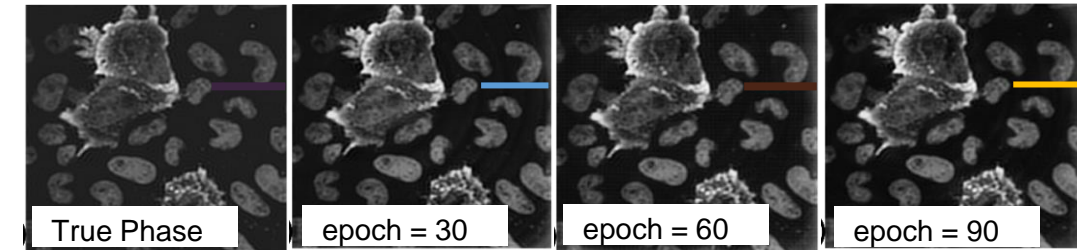
# Results – true versus predicted phase images

- Qualitatively, the agreement between the true and predicted phase images is high.



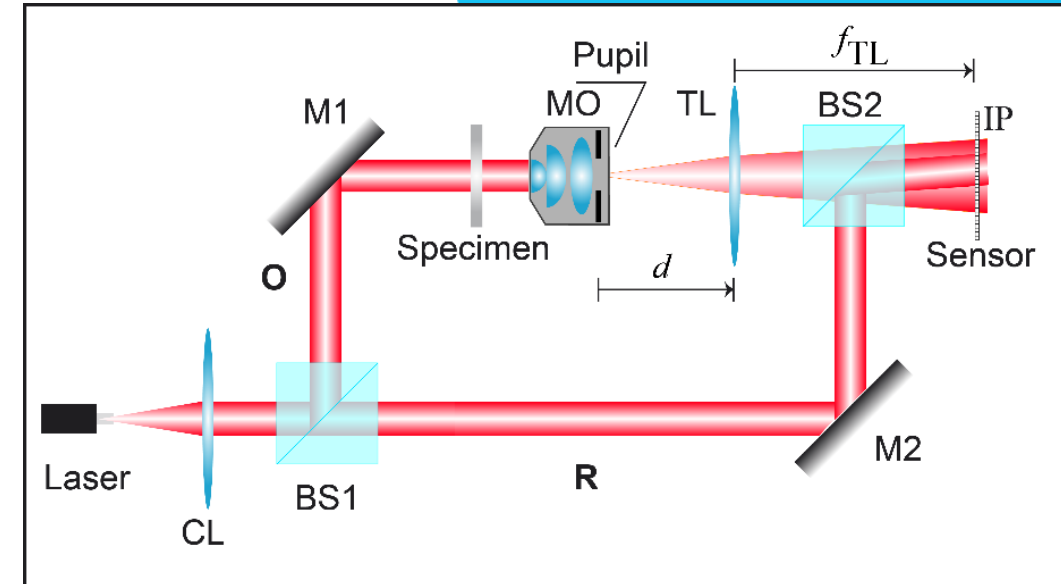
# Results – true versus predicted phase images

- Qualitatively, the agreement between the true and predicted phase images is high.
- The cGAN network introduces some undesired artifacts
  - *In all three epochs, there is a very dim low frequency ringing effect that is not present in the actual phase image.*
- In epoch 60, we observed a high frequency grid effect as well .



# Conclusions

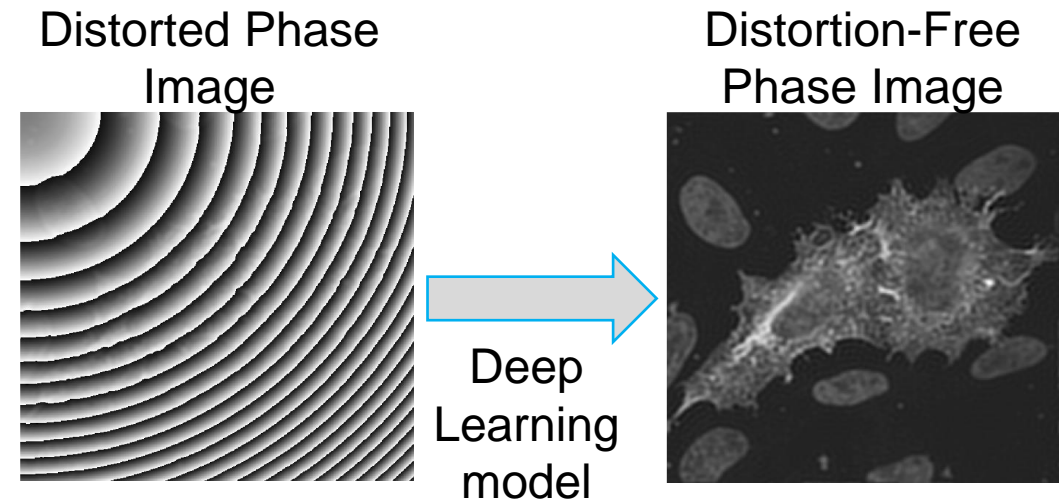
- ❑ DHM is widely used by the biomedical community for imaging of thin label-free samples and analyze the 3D phase images from a single recorded image.





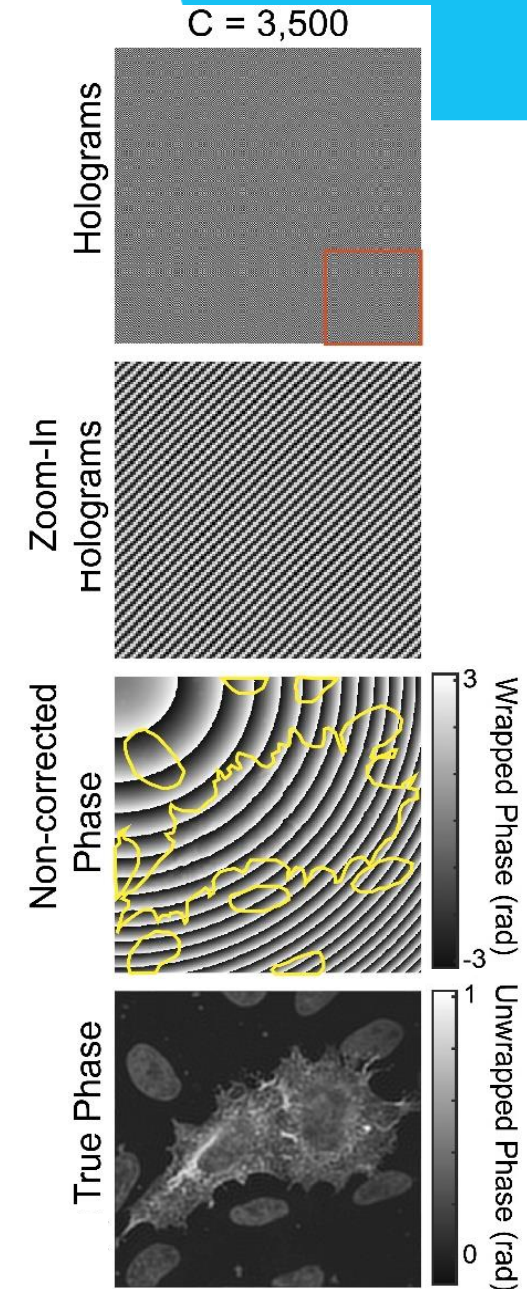
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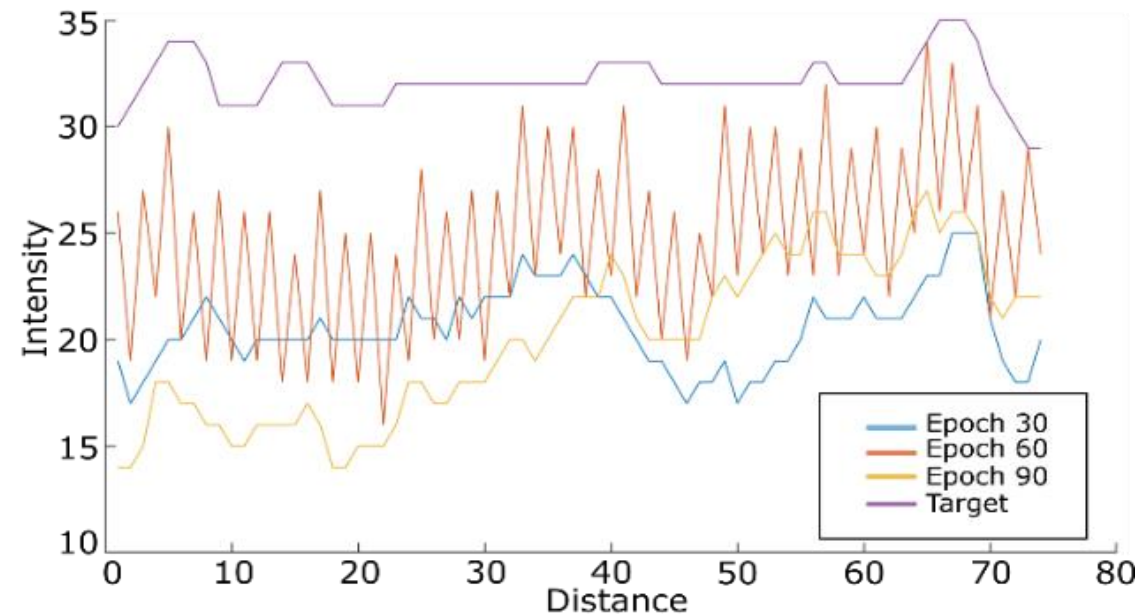
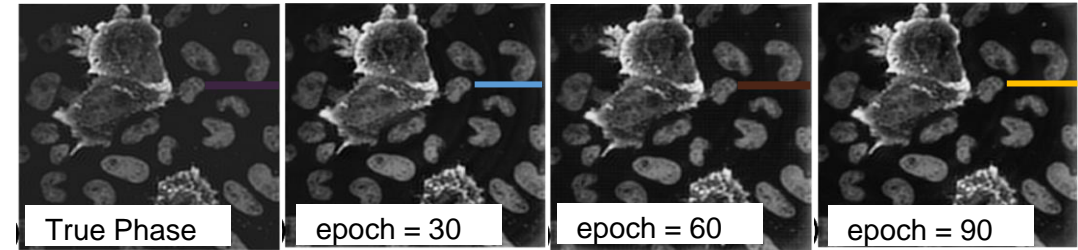
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- ❑ Non-telecentric DHM systems are shift variant systems, distorting the phase images with a spherical phase term.
- ❑ This study investigate the use of a cGAN learning-based model to automatically reconstruct holograms captured by non-telecentric DHM systems, including correction of the spherical aberrations.
- ❑ The trained cGAN model reconstructs phase images with a high similarity (e.g., minor artifacts with respect to the true phase).



Thanks...

# Q&A

