Generative Spatiotemporal Modeling of Neutrophil Behavior

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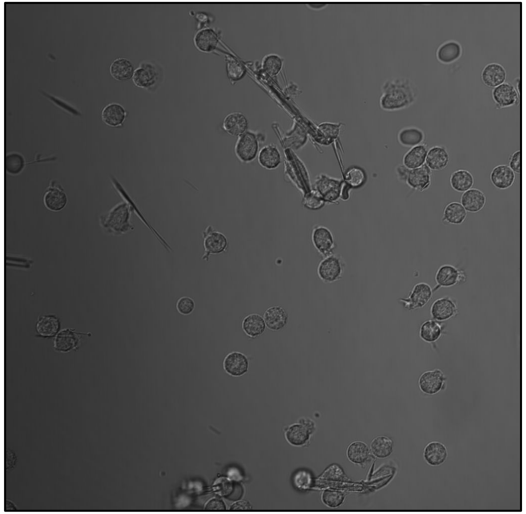
AGENDA

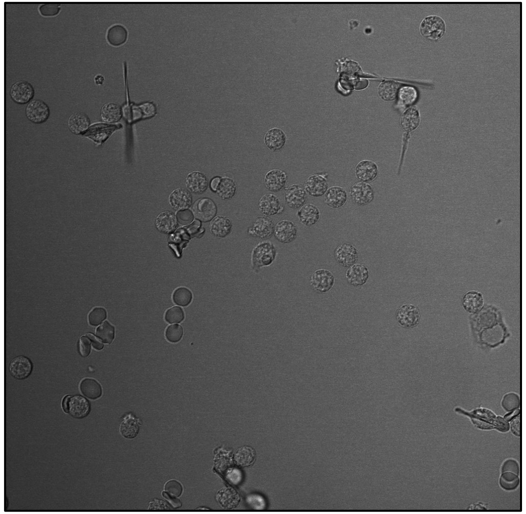
- Introduction
  - Goal
  - Previous Work
  - Proposed Approach
  - Dataset
- Data Preparation
  - Segmentation
  - Tracking
- Generative Modeling
  - Content Synthesis: Generative Adversarial Networks
  - Motion Synthesis: Autoregression
  - Content + Motion Synthesis
- Conclusion and Future Work
- References

INTRODUCTION

INTRODUCTION

- Neutrophils = Polymorphonuclear* granulocytes = Lobed White Blood Cells
- Most abundant
- Professional phagocytes
  - First line of defense of the immune system [1]
  - Highly Motile





GOAL

What?

- Develop a model of appearance/shape and motion of neutrophils.

Why?

- Studying neutrophils’ shape + motion → insights → behavior as a function of specific stimulus
- Scarce data
- Simulation → loopback for our understanding

How?

- Generative Adversarial Networks (GANs) + Autoregressive (AR) Models

Research Question:
Can we use Generative Adversarial Networks (GANs) features + weak motion model to realistically simulate a biological system?

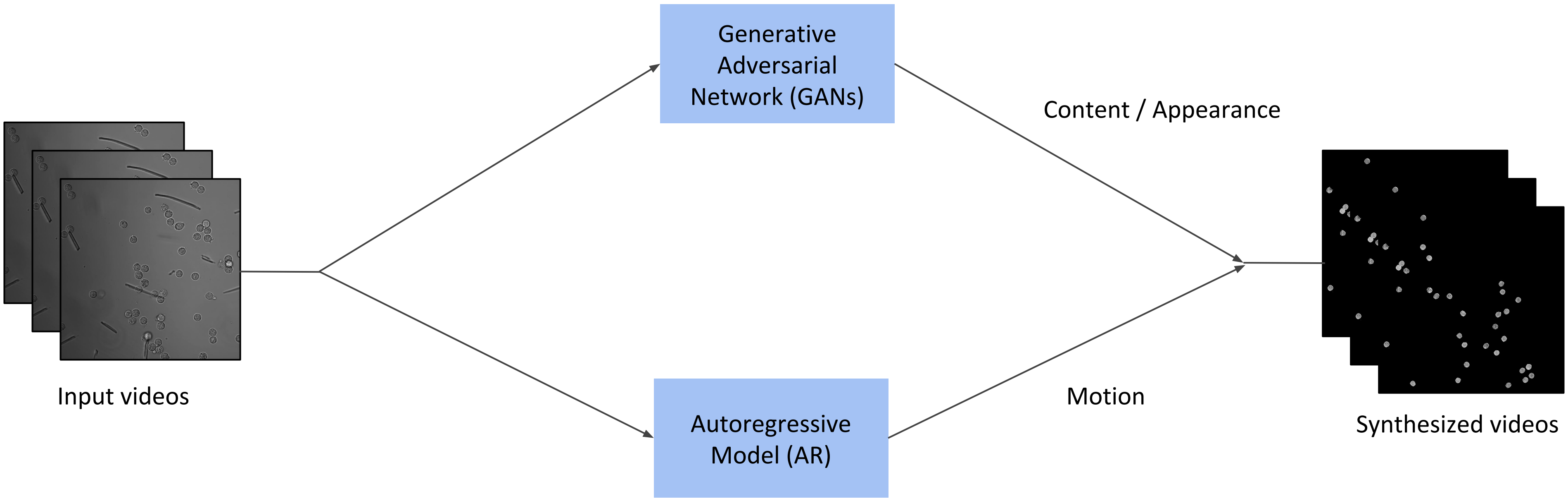
PREVIOUS WORK

- Active Shape Models (ASM)
  - Medial-axis + Texture model [9]
  - Medial-axis: (Cell shape | Nucleus shape) [10]

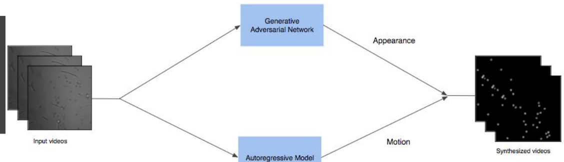
- For temporal evolution:
  - Random Walk, Autoregression [10]
  - Motion synthesis based on annotations [15]

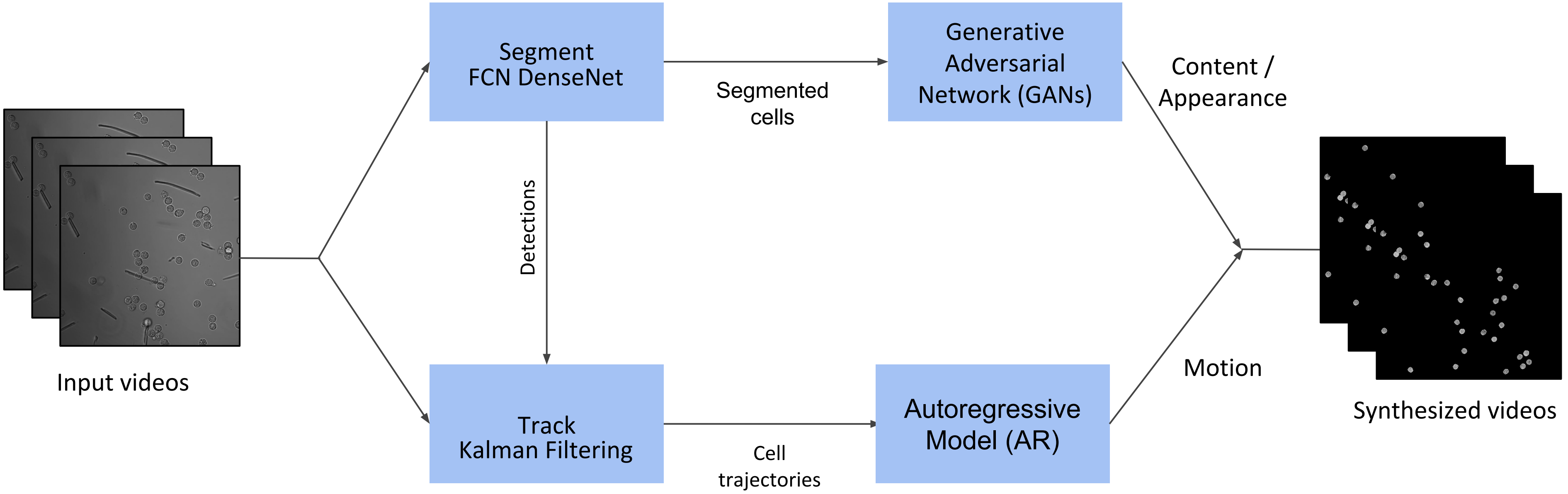
- GANs -> fluorescence microscopy images [11]

PROPOSED APPROACH: TWO-STREAM



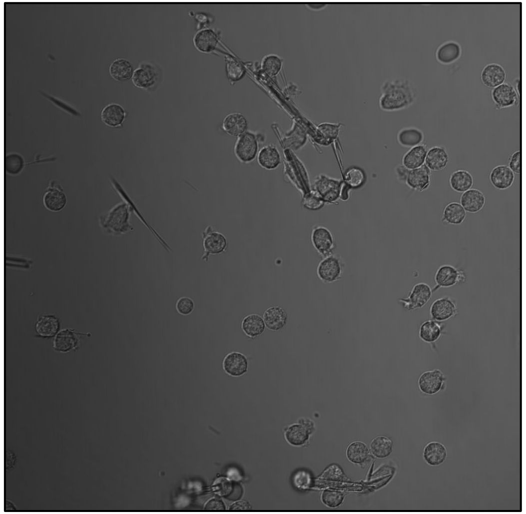
TWO-STREAM APPROACH

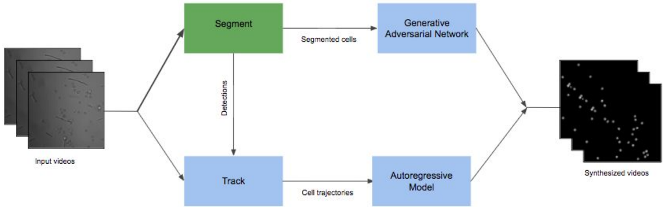




DATASET

- 11 Videos:
  - 3 of normal neutrophils
  - 8 of neutrophils treated with MRS2578 inhibitor
  - Differential Interference Contrast (DIC): Enhances contrast in unstained, transparent samples
- Length of most of the videos: 3.0 secs
- Extracted frames: 20fps
- Images of size: 1024 x 1024



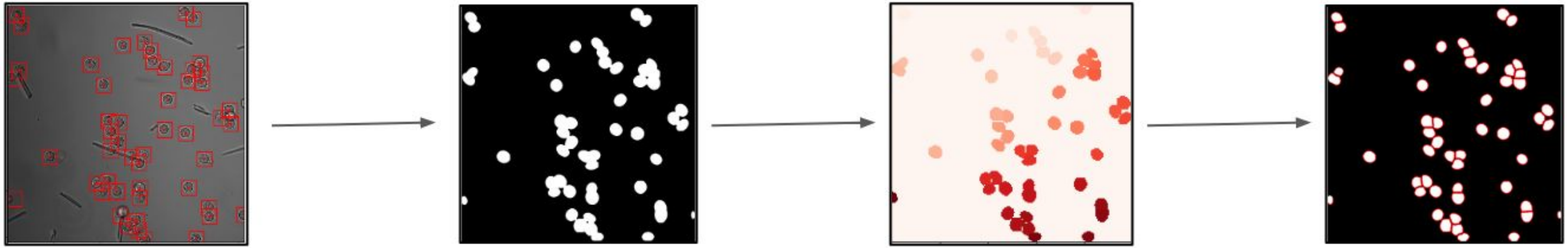


DATA PREPARATION
SEGMENTATION

SEGMENTATION MAPS

- Not available!!

- Ground truth maps created:



1. Manually marked centers
2. Extracted (64 x 64) patch

3. Fit an ellipse to each patch: Binary maps
4. Cluttered cells: big blobs

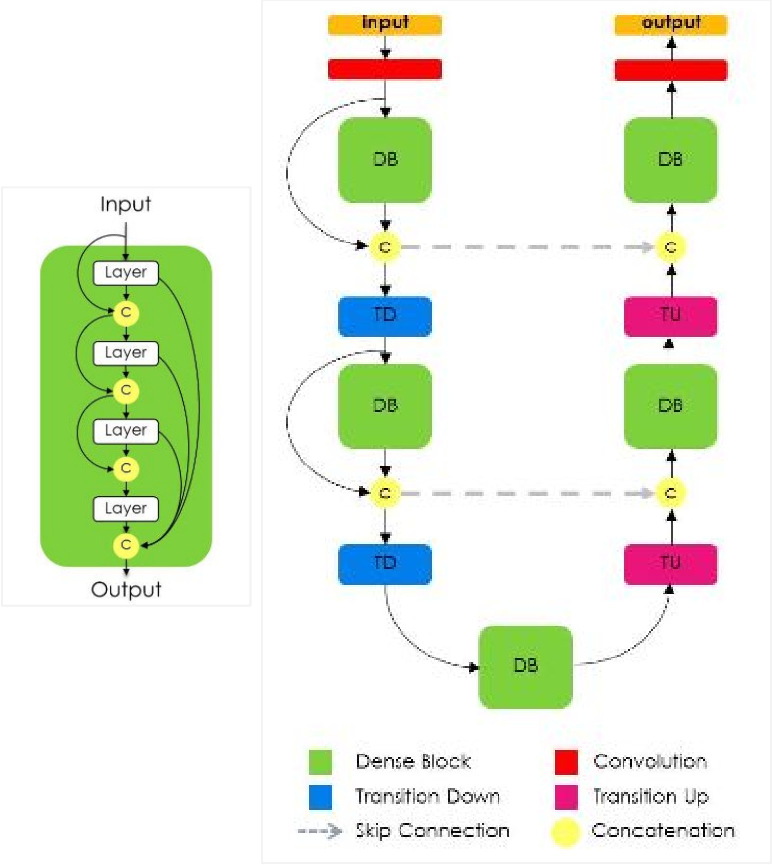
5. Marker based Watershed - separated each instance

6. Final segmentation map

AUTOMATIC SEGMENTATION

- Manual center marking - not possible
- Fully Convolutional NN + DenseNets
    - Downsampling: Extract features
    - Upsampling: Project onto the pixel space
    - Dense Blocks: Concatenate feature maps in the same block
    - Skip Connections

Credits: FCN DenseNet Architecture [22]



TRAINING

- Total #images: 412, Holdout test set: 46 images
- 3-fold cross validation from the remaining: 90% - train, 10% - validation splits
- Preprocessing:
  - Resized: 224 x 224
  - Mean subtracted, standardized
- Data Augmentation:
  - Random vertical/horizontal flips
- Challenge - Separation of touching objects of the same class
  - Pixelwise cross-entropy + Weighted loss

Layer
Batch Normalization
ReLU
3×3 Convolution
Dropout $p = 0.2$

Transition Down (TD)
Batch Normalization
ReLU
1×1 Convolution
Dropout $p = 0.2$
2×2 Max Pooling

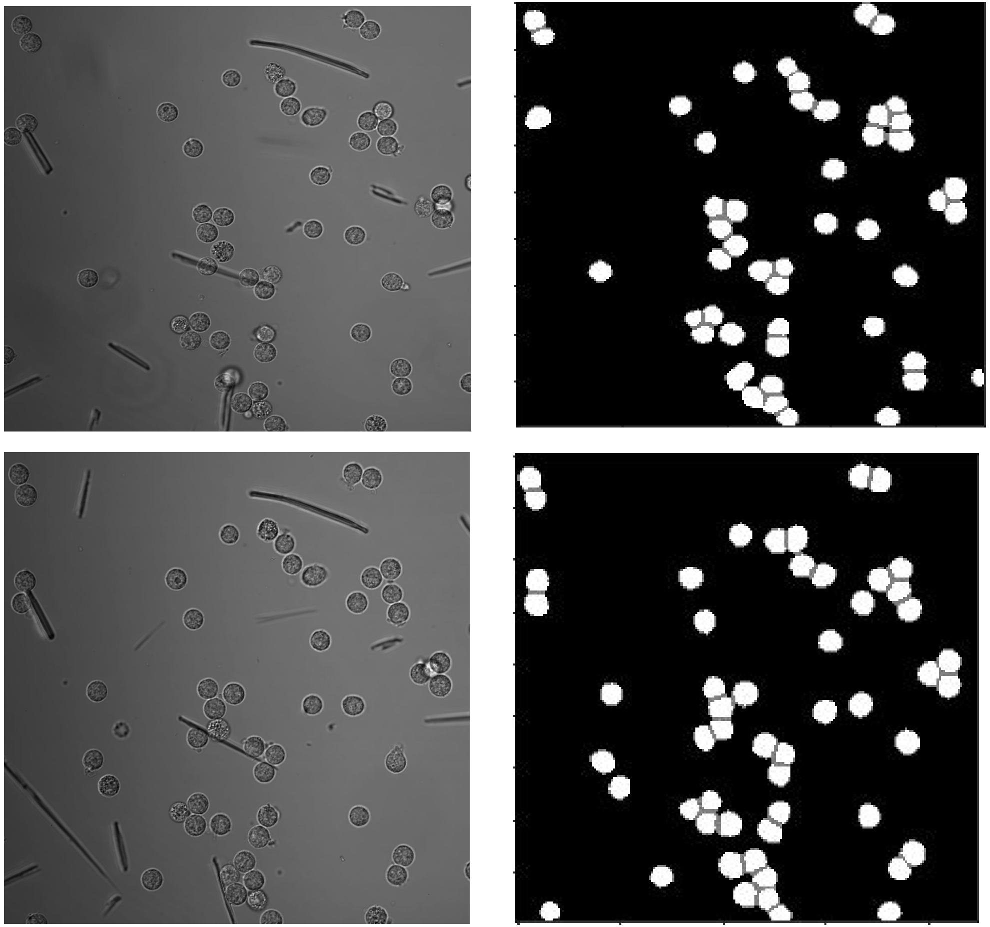
Transition Up (TU)
3×3 Transposed Convolution $stride = 2$

Architecture
Input, $m = 3$
3×3 Convolution, $m = 48$
DB (4 layers) + TD, $m = 112$
DB (5 layers) + TD, $m = 192$
DB (7 layers) + TD, $m = 304$
DB (10 layers) + TD, $m = 464$
DB (12 layers) + TD, $m = 656$
DB (15 layers), $m = 896$
TU + DB (12 layers), $m = 1088$
TU + DB (10 layers), $m = 816$
TU + DB (7 layers), $m = 578$
TU + DB (5 layers), $m = 384$
TU + DB (4 layers), $m = 256$
1×1 Convolution, $m = c$
Softmax

Credits: FC-DenseNet103 [22]

- Weights initialized: HeUniform
- Optimizer: RMSProp, Learning Rate = 1e - 3
- Regularized: Weight decay = 1e - 4, Dropout = 0.2
- Trained for: 100 epochs
- Test accuracy (pixel classification): 96%
- Dice Coefficient:

Neutrophil border	0.478704134139
Neutrophil	0.877087162892
Background	0.988928566552



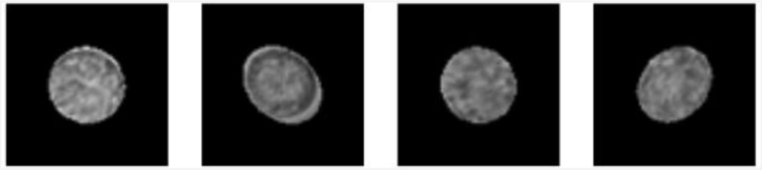
Images

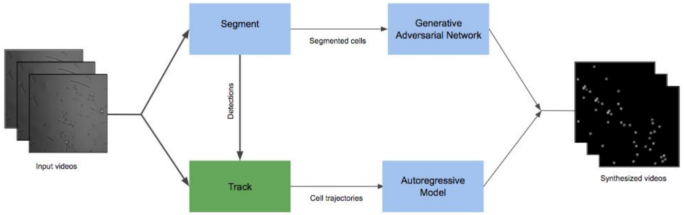
Predicted maps

EXTRACTING SEGMENTED CELLS

	mean	std
area	1430.736241	376.554888
eccentricity	0.507366	0.159627
major axis length	46.581709	6.338599
minor axis length	38.994317	6.169081

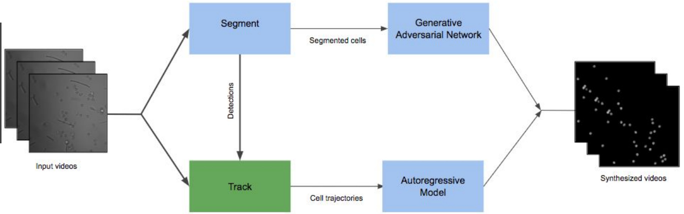
Area, Major and minor axis: ± 2 std deviations
Eccentricity: [0,+1] std deviations





DATA PREPARATION
TRACKING

TRACKING



- Associate segmented cells across frames

- Online tracking
    - Detections from: only previous & current frame

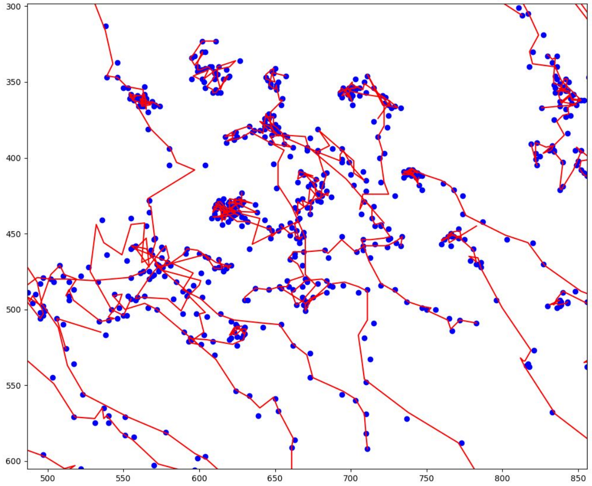
- Kalman Filters
    - Recursive filter. Estimates states from a series of noisy measurements
    - Constant Velocity

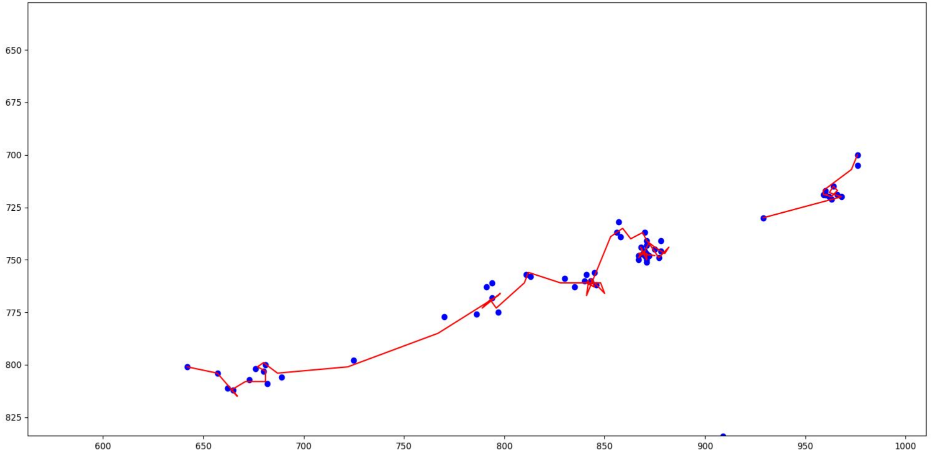
- Hungarian Matching
    - Optimally matches the Kalman filter predicted location with the detection

- Predict ⟵⟶ Correct

SAMPLE TRAJECTORIES

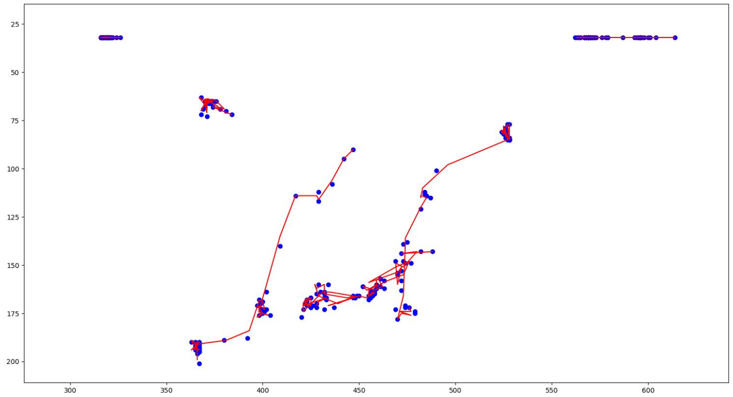
Sample #1



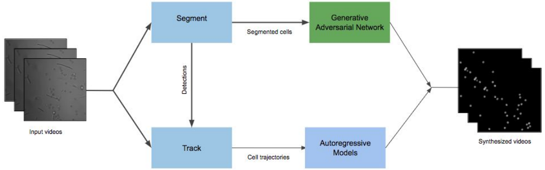


Sample #3

Sample #2



Blue: Ground truth center coordinates
Red: Trajectories



GENERATIVE MODELING

CONTENT SYNTHESIS

GENERATIVE ADVERSARIAL NETWORKS (GANs)

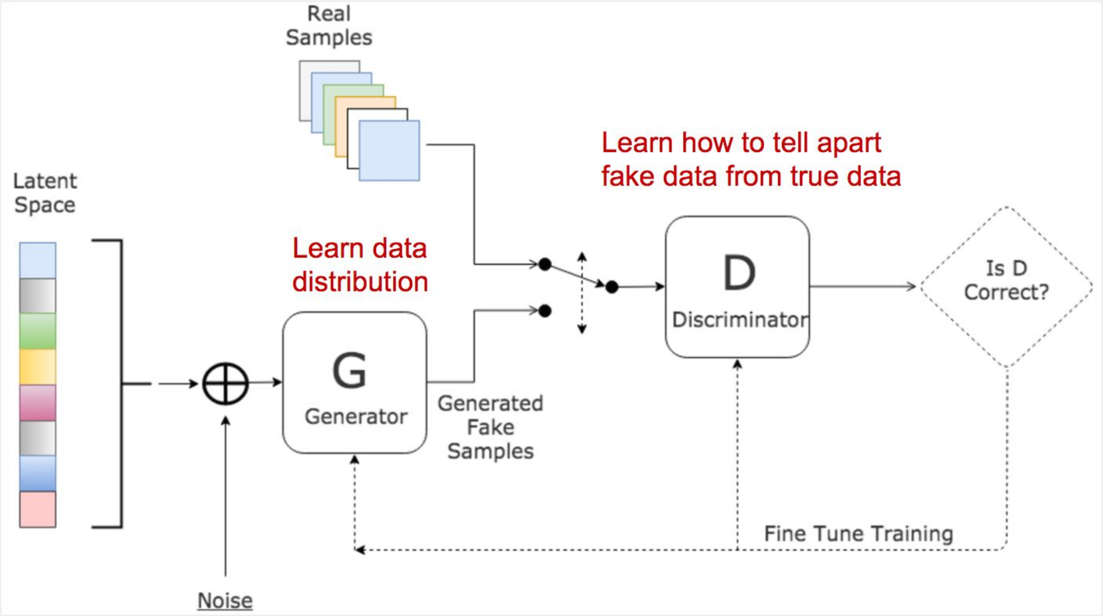
- Framework: Minimax game

- Vanilla GAN

$$\min_{G} \max_{D} \mathop{\mathbb{E}}_{\boldsymbol{x}\sim\mathbb{P}_r} [\log(D(\boldsymbol{x}))] + \mathop{\mathbb{E}}_{\tilde{\boldsymbol{x}}\sim\mathbb{P}_g} [\log(1 - D(\tilde{\boldsymbol{x}}))].$$

- Issues:
  - Scalability
  - Vanishing gradient
  - Mode Collapse
  - Lack of Evaluation Metric

- Active Research:
  - Novel applications
  - Optimize network architecture
  - Improve cost function



Credits: GAN Architecture [24]

GAN Types

Vanilla GAN:

$$\min_G \max_D \mathop{\mathbb{E}}_{\boldsymbol{x}\sim\mathbb{P}_r}[\log(D(\boldsymbol{x}))] + \mathop{\mathbb{E}}_{\tilde{\boldsymbol{x}}\sim\mathbb{P}_g}[\log(1 - D(\tilde{\boldsymbol{x}}))].$$

Jensen-Shannon distance

$$JS(P_r, P_g) = \frac{1}{2}KL(P_r \| P_m) + \frac{1}{2}KL(P_g \| P_m)$$

Wasserstein GAN (WGAN):

$$\min_G \max_{D\in\mathcal{D}} \mathop{\mathbb{E}}_{\boldsymbol{x}\sim\mathbb{P}_r}\big[D(\boldsymbol{x})\big] - \mathop{\mathbb{E}}_{\tilde{\boldsymbol{x}}\sim\mathbb{P}_g}\big[D(\tilde{\boldsymbol{x}}))\big]$$

Earth-Mover distance aka Wasserstein

$$W(P_r, P_g) = \inf_{\gamma\in\Pi(P_r,P_g)} \mathbb{E}_{(x,y)\sim\gamma}\big[\, \|x - y\| \,\big]$$

- How much mass to move to convert: P_g to P_r, at minimum cost

- Enforce constraints on weights of D [$-c$, c]

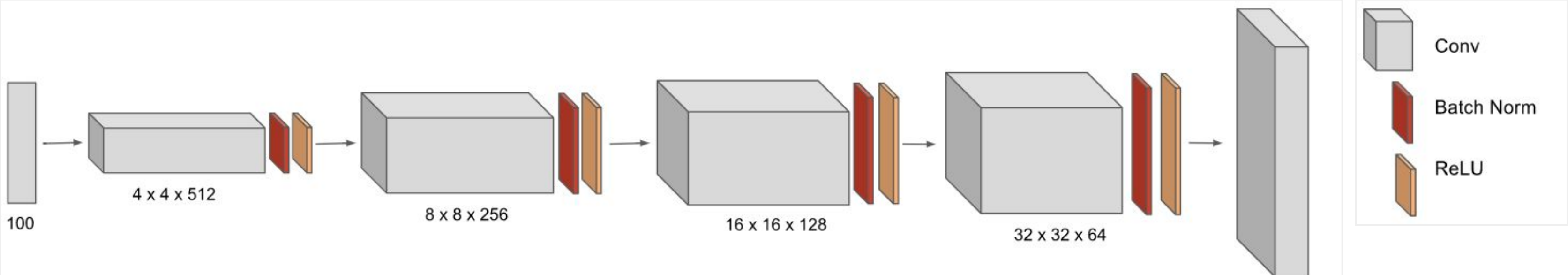
Improved WGAN:

$$L = \underbrace{\mathop{\mathbb{E}}_{\tilde{\boldsymbol{x}}\sim\mathbb{P}_g}[D(\tilde{\boldsymbol{x}})] - \mathop{\mathbb{E}}_{\boldsymbol{x}\sim\mathbb{P}_r}[D(\boldsymbol{x})]}_{\text{Original critic loss}} + \underbrace{\lambda \mathop{\mathbb{E}}_{\hat{\boldsymbol{x}}\sim\mathbb{P}_{\hat{\boldsymbol{x}}}}\left[(\|\nabla_{\hat{\boldsymbol{x}}} D(\hat{\boldsymbol{x}})\|_2 - 1)^2\right]}_{\text{Our gradient penalty}}$$

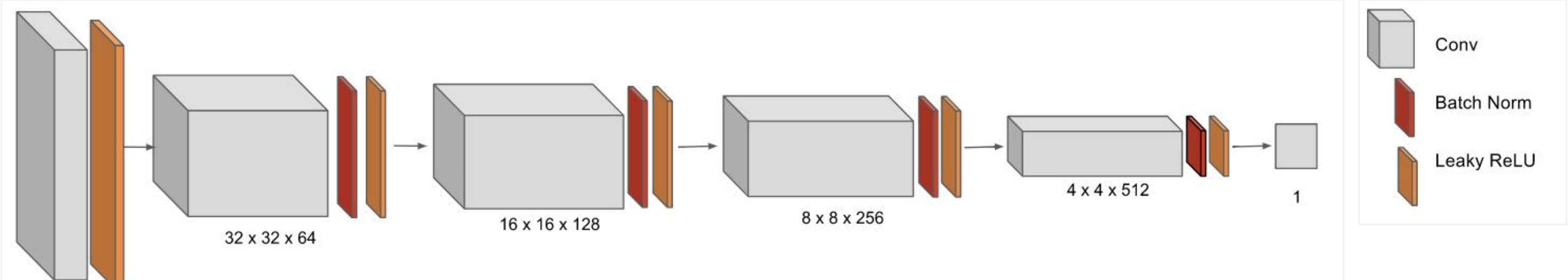
- Replace weight clipping with gradient penalty

TRAINING

- Trained models:
    - Deep Convolutional GAN(DCGAN) Architecture
    - With Vanilla GAN, Wasserstein GAN (WGAN) and Improved WGAN loss



Generator (G)



Discriminator (D)

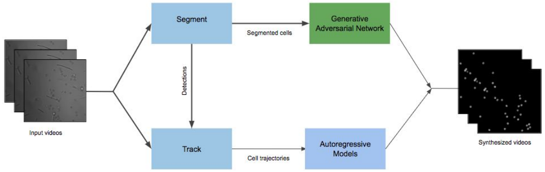
	Vanilla GAN	WGAN	Improved WGAN
Optimizer	Adam	RMSProp	Adam
Learning Rate	*D, G* = 0.0002	*D, G* = 0.00005	*D, G* = 0.0001
Decay	beta1 = 0.5 beta2 = 0.999	-	beta1 = 0 beta2 = 0.9
Iterations of D	-	5	5
# Epochs	10000	10000	10000
Loss functions	(1)	(2)	(3)

$$\min_G \max_D V(D,G) = \mathbb{E}_{x\sim p_{data(x)}}[\log D(x)] + \mathbb{E}_{z\sim p_{z(z)}}[\log 1 - D(G(z))]$$

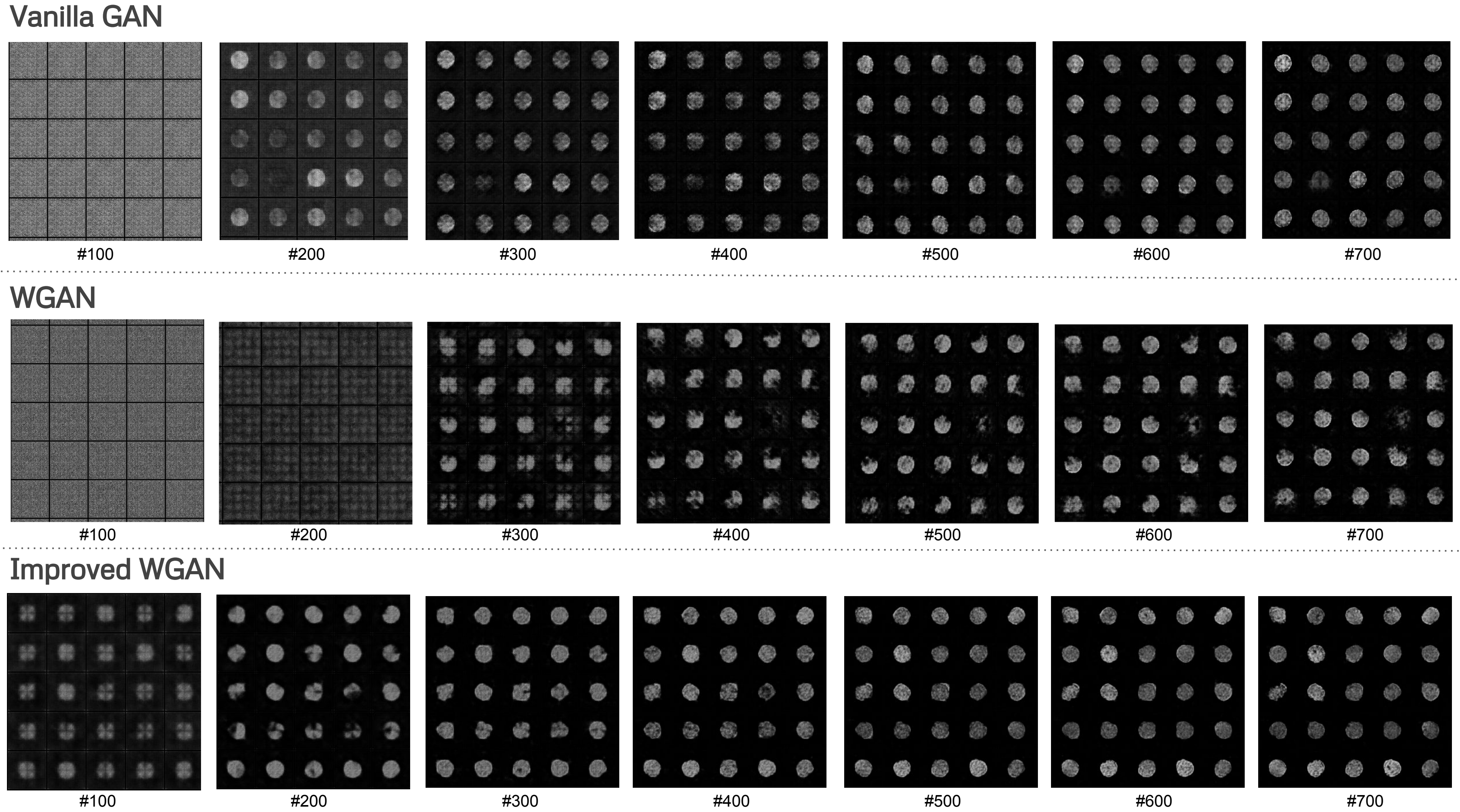
$$\min_G \max_{D \in D} \mathbb{E}_{x\sim P_r}[D(x)] - \mathbb{E}_{\tilde{x}\sim P_g}[D(\tilde{x}))]$$

$$L = \underbrace{\mathbb{E}_{\tilde{x}\sim\mathbb{P}_g}[D(\tilde{x})] - \mathbb{E}_{x\sim\mathbb{P}_r}[D(x)]}_{\text{Original Critic Loss}} + \underbrace{\lambda \mathbb{E}_{\hat{x}\sim\mathbb{P}_{\hat{x}}}[(\|\nabla D(\hat{x})\|_2 - 1)^2]}_{\text{Gradient Penalty}}$$

- Vanilla GAN loss (1)
- WGAN loss (2)
- Improved WGAN loss (3)



GANs RESULTS AND EVALUATION


Vanilla GAN
#100
#200
#300
#400
#500
#600
#700
WGAN
#100
#200
#300
#400
#500
#600
#700
Improved WGAN
#100
#200
#300
#400
#500
#600
#700

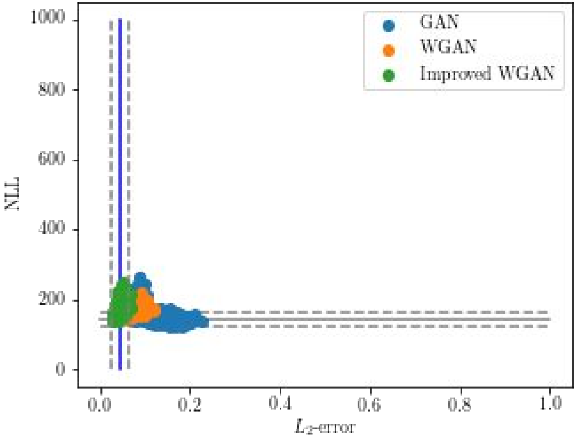
EVALUATION

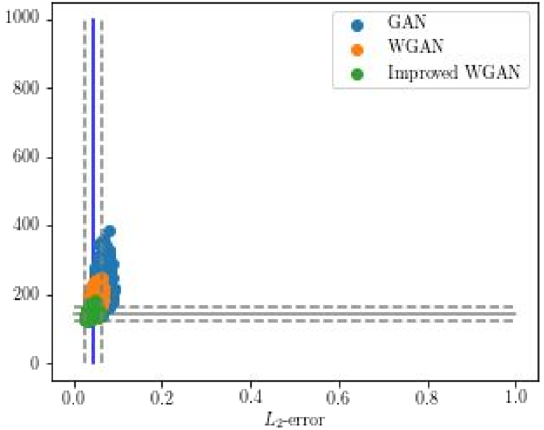
- Mode Collapse
  - Reconstruction Error: Can the test samples be reconstructed well?

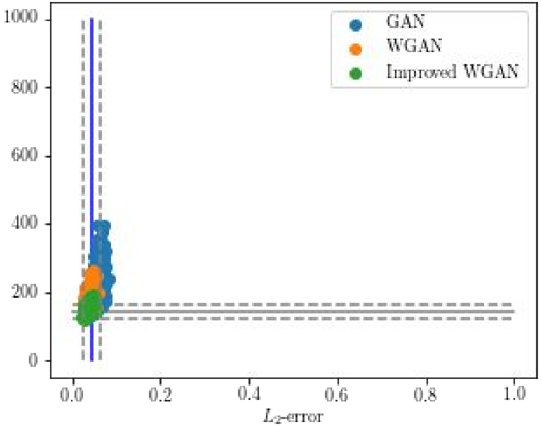
- Latent Space Walk
  - Has the network memorized or learned?

MODE COLLAPSE

- Minimize the *L2*-distance (lower the better)
- 50 iterations of L-BFGS, selected best reconstruction out of 3
- Negative log likelihood (NLL) w.r.t. the prior P_z of the noise vectors *z* (lower the better)



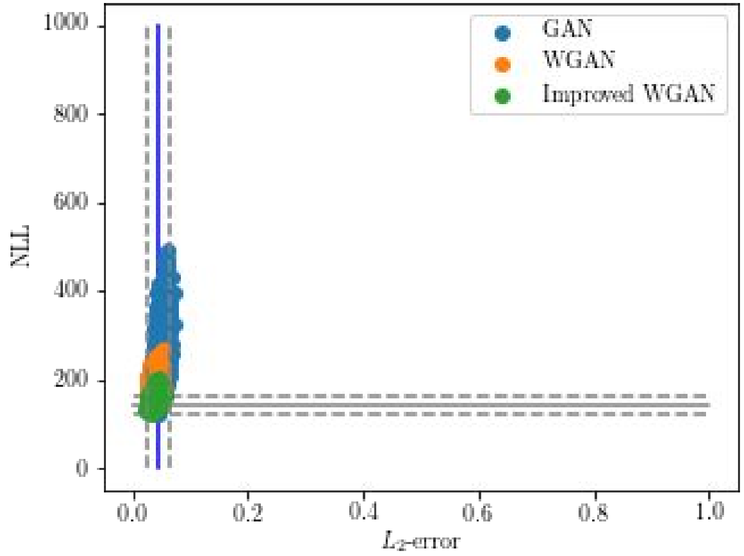




#500	Vanilla GAN	WGAN	Improved WGAN
L2	0.142 ± 0.25	0.073 ± 0.013	0.041 ± 0.006
NLL	146 ± 12	172 ± 9	170 ± 14

#1000	Vanilla GAN	WGAN	Improved WGAN
L2	0.064 ± 0.008	0.044 ± 0.007	0.037 ± 0.007
NLL	242 ± 29	191 ± 13	141 ± 7

#5000	Vanill GAN	WGAN	Improved WGAN
L2 - error	0.054 ± 0.006	0.034 ± 0.005	0.031 ± 0.005
NLL	204 ± 36	165 ± 17	143 ± 10



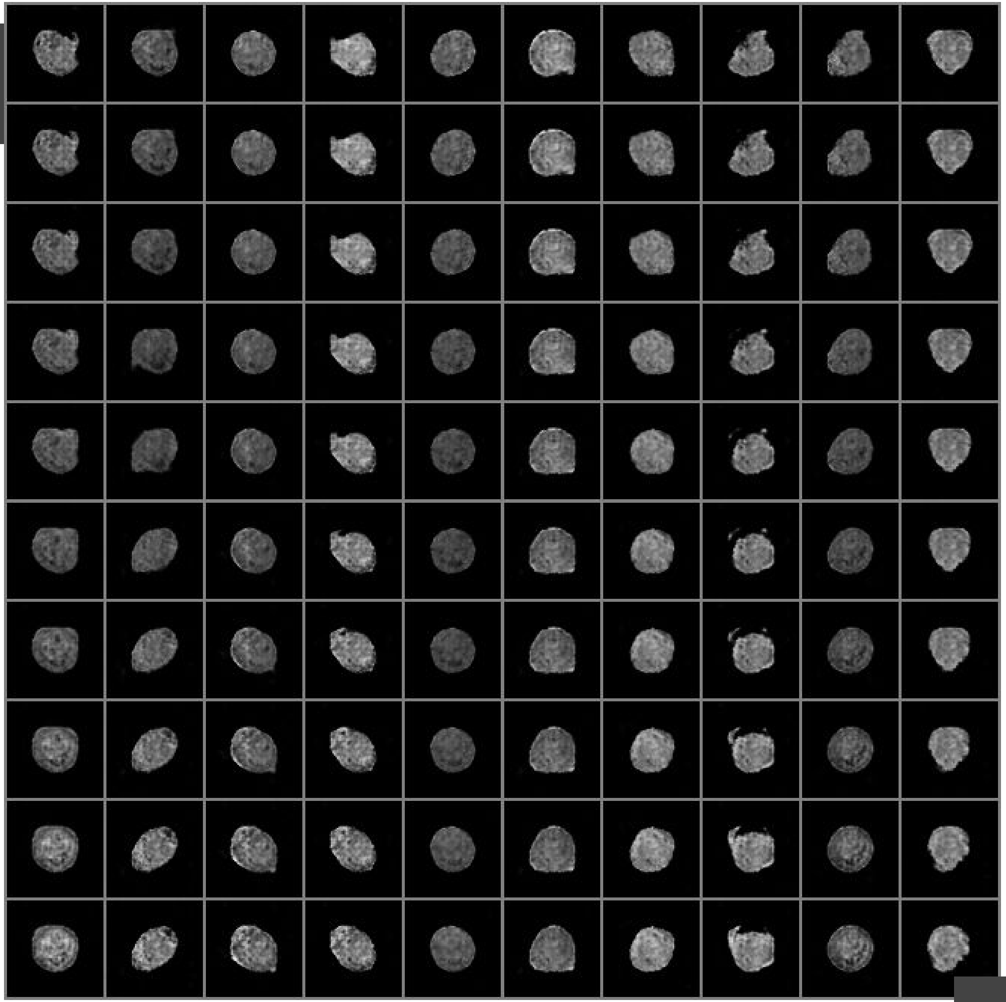
#10000	Vanilla GAN	WGAN	Improved WGAN
L2 - error	0.044 ± 0.006	0.033 ± 0.005	0.033 ± 0.005
NLL	230 ± 49	178 ± 18	149 ± 10

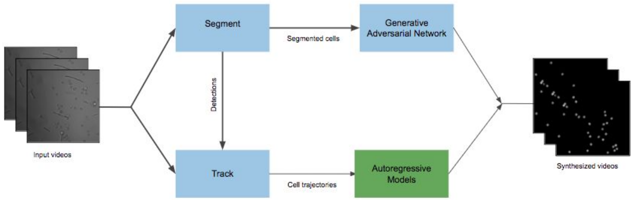
LATENT SPACE WALK

- Interpolate between points in the latent space

- Identify for existence of sharp transitions

- Smooth transitions = validates model has learned relevant representations

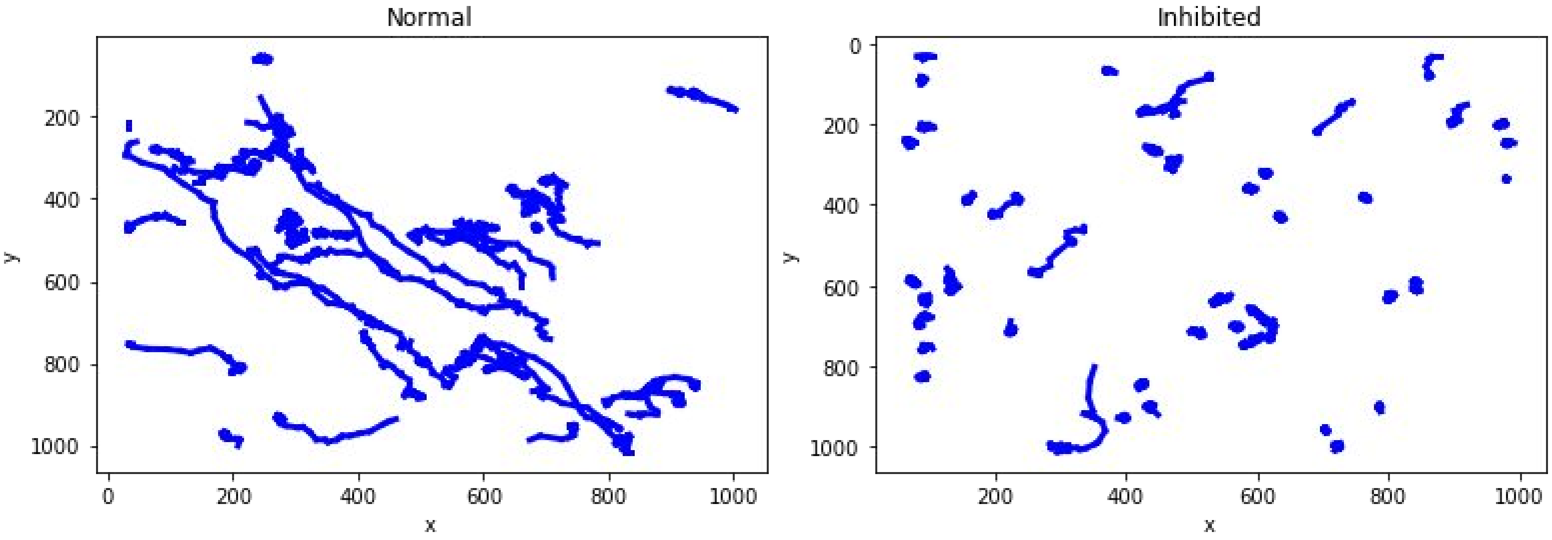




GENERATIVE MODELING
MOTION SYNTHESIS

AUTOREGRESSIVE (AR) MODELS

- Normal and Inhibited - have different motion patterns
- Build a global motion model for cells under the same conditions
- Synthesize new sequences - based on the existing motion characteristics



Original trajectories

AUTOREGRESSIVE MODELS

- Parametric models
  - Every point in sequence = linear combination of previous

- Components:
  - Appearance - Determines state of the system
    Decomposes: original space -> low-dimensional state space

$$\overrightarrow{y_t} = C\overrightarrow{x_t} + \overrightarrow{u_t}$$

  - Dynamic - Captures how the states are change
    Denotes: new state = (sum of *d* of its previous states)

$$\overrightarrow{x_t} = B_1\overrightarrow{x}_{t-1} + B_2\overrightarrow{x}_{t-2} + .. + B_d\overrightarrow{x}_{t-d} + \overrightarrow{v_t}$$

TRAINING

- Each neutrophil - Single compartment

- Extracted trajectories - consists of center Cartesian coordinates

- All trajectories - truncated to 61 frames
  Example: (34 x 61 x 2) -> x, y centers of 34 neutrophils across 61 frames

- Pooled separately: normal and inhibitor-treated

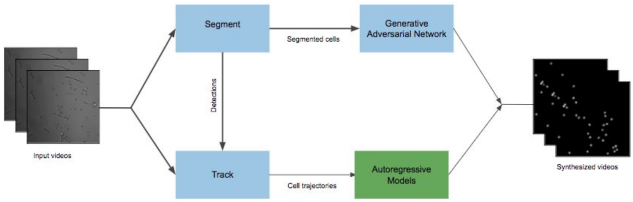
$$\overrightarrow{y_t} = C\overrightarrow{x_t} + \overrightarrow{u_t}$$

- Projected trajectories into low-dimensional space *C,* using Principal Component Analysis (PCA)

$$\overrightarrow{x_t} = B_1\overrightarrow{x}_{t-1} + B_2\overrightarrow{x}_{t-2} + .. + B_d\overrightarrow{x}_{t-d} + \overrightarrow{v_t}$$

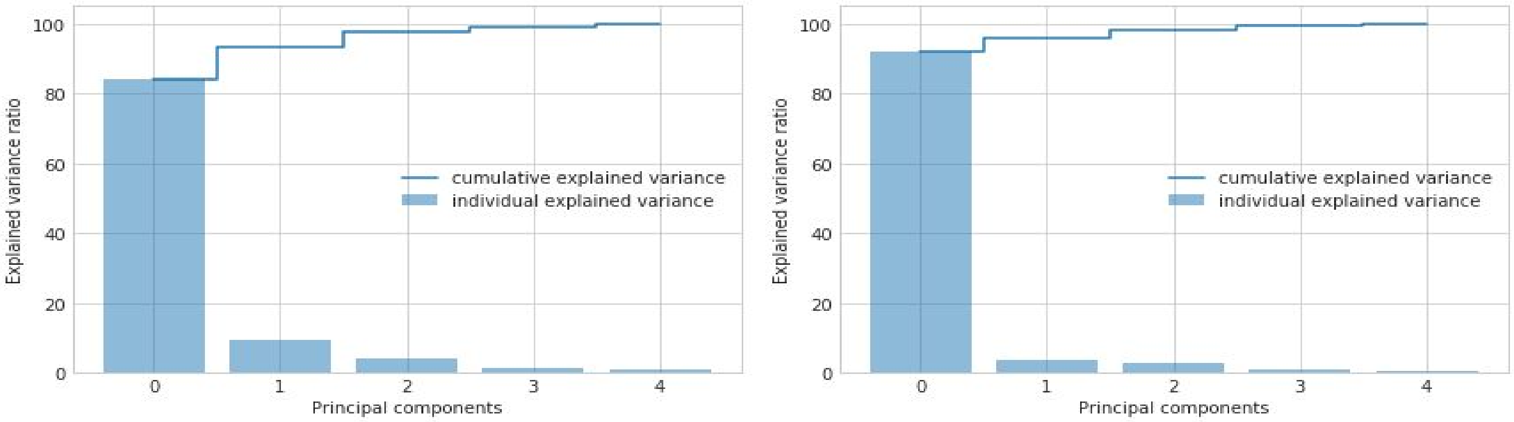
- Transition matrices determined: Least square

- Final model: (3-principal components, 2-previous states)

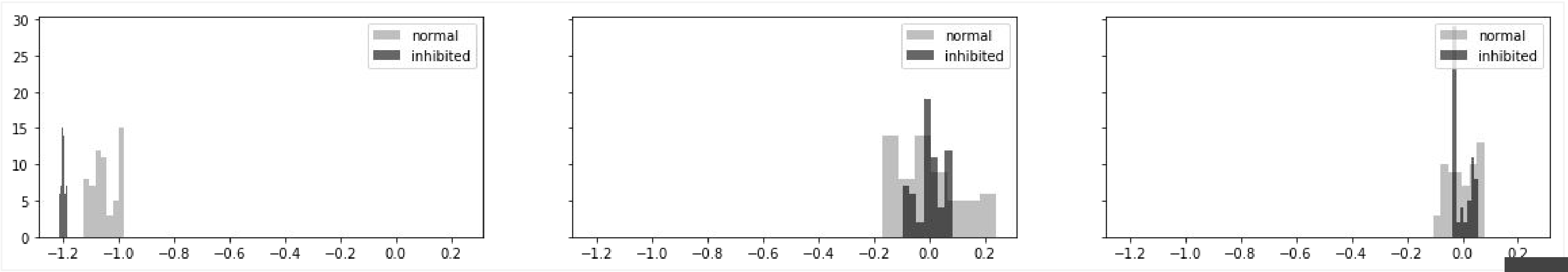


AR RESULTS AND EVALUATION

APPEARANCE COMPONENT (PCA)

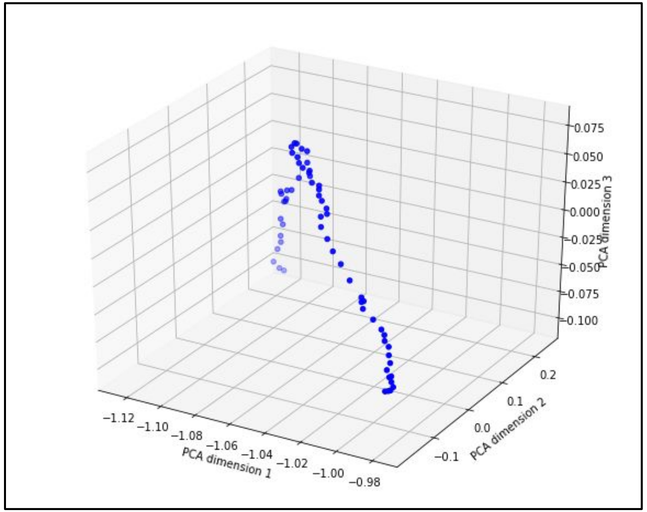


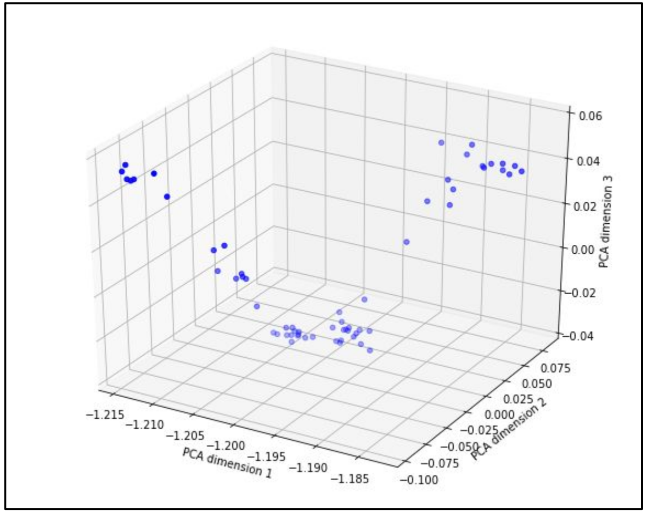
% of variance explained by top 5 principal components. (left) normal and (right) inhibited



Distribution of values of top three principal components

APPEARANCE COMPONENT (PCA)

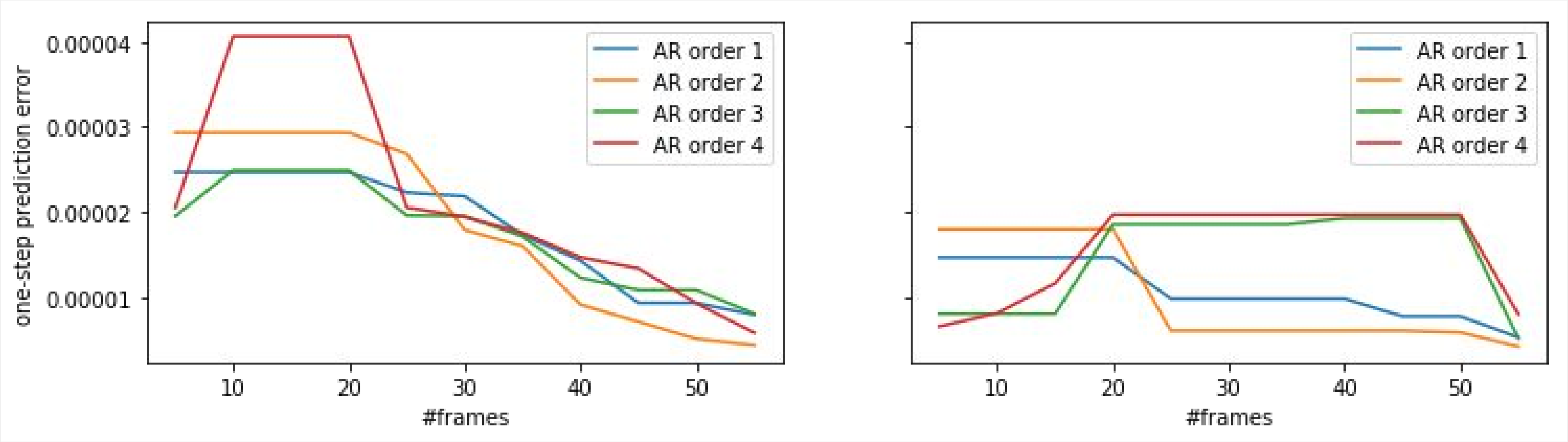




Original trajectories can be viewed as **digital signatures** through the low-dimensional space.
(left) normal and (right) inhibited

DYNAMIC COMPONENT

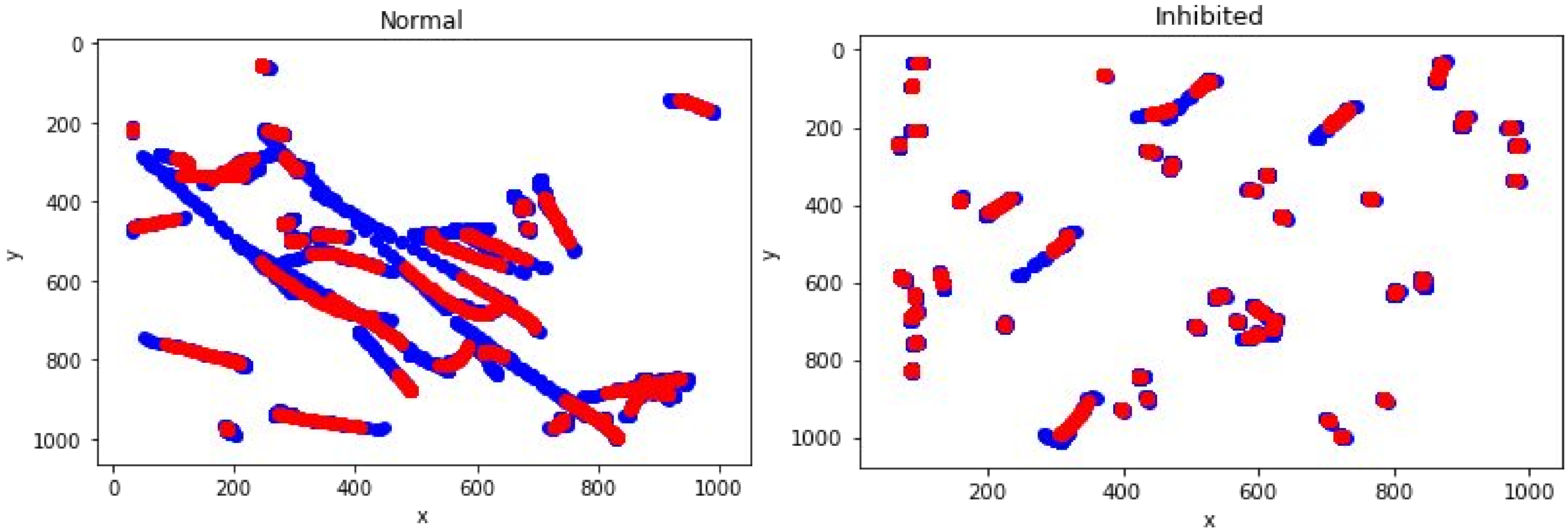
- One-step prediction error
  - train with *d* frames
  - generate *d + 1* frame
  - Calculate the difference: MSE



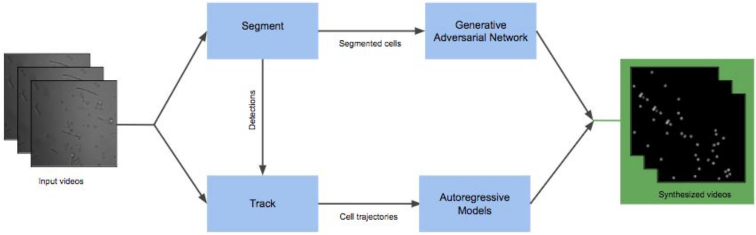
(left) normal and (right) inhibited

SYNTHESIZED SEQUENCES

Using the learned transition matrices *B,* new trajectories can be synthesized.



Synthesized(red) trajectories superimposed over the original(blue) trajectories



SYNTHESIS
CONTENT + MOTION

SYNTHESIS

Synthesized behavior consists:

1. content and appearance sampled from: Improved WGAN generator G
2. motion sampled from: a point in subspace C + AR transition matrices
3. new sequences $\rightarrow$ project back into original space

SYNTHESIZED VIDEO

CONCLUSION and FUTURE WORK

- Our two-stream approach
    - each component: identifies respective key features
    - GAN: spatial
    - AR: temporal
    - controlled video generation

- Recently proposed two-stream approaches -> entirely based on GANs
    - Learn to model the appearance and motion -> unsupervised manner
    - Disentangle these two factors -> through adversarial training
    - Data hungry!

BIBLIOGRAPHY

https://drive.google.com/file/d/1WBdv0AtcFUYrza0wVckCLrK5aP-RGBWb/view?usp=sharing