Defining Simulation parameters Defining WAC-Net model using tensorflow keras Training WAC-Net model Testing the model on an experimental images Time Distributed (n samples) weighted average Dry mass 32, 32, 2 softmax(weights) vectors NOTE: • If you're running this notebook on your local machine, please comment the code in the cell below In [60]: !git clone https://github.com/softmatterlab/Quantitative-Microplankton-Tracker.git %cd Quantitative-Microplankton-Tracker/training-tutorials/ In [36]: %matplotlib inline import sys sys.path.append("..") 1. Setup Imports the dependencies needed to run this tutorial. In [37]: import numpy as np import tensorflow as tf import matplotlib.pyplot as plt from tqdm.notebook import tqdm import deeptrack as dt # Load exp data predator sequence = np.load( "../data/data figure3/predator sequence.npy" prey sequence = np.load( "../data/data figure3/prey\_sequence.npy" 2. Defining training dataset 2.1. Defining simulation parameters We generate 15-frame sequences of simulated holgoraphic images to train the WAC-Net, where each image is of size 64 x 64 pixels. Each image contains plankton holograms of different properties as defined below. In [38]: variables = { "pix size": 3.6, # in micro meters "radius\_range": [1.5e-6, 10e-6], "ri\_range": [1.35, 1.38], "z range": [2300, 3000], #in microns "apod val": 0.07, #Gaussian apodization value 2.2. Defining scatterrer properties Each plankton is a scatterrer. We use the Sphere feature of DeepTrack to generate spherical scatterrers. Since we are using lenslessholographic setup, planktons can be approximated as spherical objects. All the other properties are automatically filled from the variables defined above in the cell above. 2.2.1. Defining main plankton The main plankton is the plankton that is always centered in a sequnce, and it is the plankton for which for which the dry mass and radius are predicted. In [39]: \_sphere\_main = dt.Sphere( position=(32, 32),position unit="pixel", cam pix size=variables["pix size"], radius=lambda: variables["radius\_range"][0] + np.random.rand() \* (variables["radius range"][1] - variables["radius range"][0]), refractive index=lambda: variables["ri range"][0] + np.random.rand() \* (variables["ri\_range"][1] - variables["ri\_range"][0]), z=lambda: variables["z\_range"][0] / variables["pix\_size"] + np.random.rand() \* (variables["z range"][1] - variables["z range"][0]) / variables["pix\_size"], find me=True, def get position(previous value): return previous value + np.random.uniform(-1 / 8, 1 / 8, size=2) def get z(previous value): return previous value + np.random.uniform( -100 / variables["pix size"], 100 / variables["pix size"] sphere main = dt.Sequential( sphere main, z=get z, position=get position) 2.2.2 Defining noise planktons The noise planktons are planktons in the background. In [40]: \_sphere\_noise = dt.Sphere( position=lambda: np.random.rand(2) \* 64, position unit="pixel", cam\_pix\_size=\_sphere\_main.cam\_pix\_size, z=\_sphere\_main.z, radius=\_sphere\_main.radius, refractive index= sphere main.refractive index, find me=False, def get position2(previous value): return previous\_value + np.random.uniform(0, 7, size=2) sphere noise = dt.Sequential( sphere noise, position=get position2) 2.3. Defining the optical device The scatterers are imaged with an optical device. We use the Brightfield feature of DeepTrack to define a microscope operating at wavelength of 633 nm. In [41]: optics = dt.Brightfield( wavelength=633e-9, NA=1, resolution=variables["pix size"] \* 1e-6, magnification=1, refractive\_index\_medium=1.33, apod val=variables["apod val"], upscale=4, aberration=lambda apod val: dt.GaussianApodization(apod val + np.random.uniform(-1,1)\*0.01), output\_region=(0, 0, 64, 64), 2.4. Defining noises We add gaussian noise to the generated images with a blur factor. In [42]: from deeptrack.noises import Noise from skimage.filters import gaussian class Blurred\_Gaussian(Noise): """Adds IID Gaussian noise to an image Parameters mu : float The mean of the distribution. sigma : float The root of the variance of the distribution. def \_\_init\_\_(self, mu=0, sigma=1,blur=2, \*\*kwargs): super(). init (mu=mu, sigma=sigma,blur=blur, \*\*kwargs) def get(self, image, mu, sigma,blur, \*\*kwargs): noisy\_image = mu + np.random.randn(\*image.shape) \* sigma noisy image=gaussian(noisy image, sigma=blur) noisy image=image+noisy image return noisy\_image noise = Blurred Gaussian(mu=0, sigma= lambda: .025, blur=0.9+np.random.uniform(0,1)\*0.1) 2.5. Defining number of planktons (main and noise) The main plankton is always set to 1, and number of noise planktons is set to vary between 0 to 3. In [43]: | sphere\_main\_no = lambda: np.random.randint(1, 2) sphere noise no = lambda: np.random.randint(0, 3) sample normal = sphere main \*\* sphere main no sample\_with\_noise = sample\_normal + sphere\_noise \*\* sphere\_noise\_no 2.6. Combining all the properties defined above Passing all the features to the optical device to generate sample sequences. In [61]: image formed = optics(sample with noise) seq length = 15image of particles = dt.ConditionalSetFeature( on true=dt.Sequence(image formed, sequence length=seq length) + noise, on false=dt.FlipUD( dt.FlipLR( dt.FlipDiagonal( dt.Sequence(image formed, sequence length=seq length) + noise, condition="skip aug", skip\_aug=False, 2.7. Visualising an example sequence In [45]: image of particles.update(skip aug="False").plot(cmap="gray") OnceLoopReflect Out[45]: <matplotlib.animation.ArtistAnimation at 0x7f9ad5b34bb0> 3. Creating target values 3.1. Dry mass values Function to check the range of possible dry mass values for parameters defined above. In [62]: def dm range(rad range, ri range): m = lambda rad, ri: ((4 \* np.pi) / 3) \* ((rad \* 1e6) \*\* 3) \* (ri - 1.33)return m(rad\_range[0], ri\_range[0]), m(rad\_range[1], ri\_range[1]) dm vals = dm range(variables["radius range"], variables["ri range"]) dm\_vals Out[62]: (0.2827433388230816, 209.4395102393188) 3.2. Normalising target images Function to generate the normalized dry mass and radius value as outputs. In [47]: **def** get drymass radius(image): rad = image[0].get property("radius") ri = image[0].get\_property("refractive\_index") dm = ((4 \* np.pi) / 3) \* ((rad \* 1e6) \*\* 3) \* (ri - 1.33)return (dm - dm vals[0]) / (dm vals[1] - dm vals[0]), ( rad - variables["radius range"][0] ) / (variables["radius range"][1] - variables["radius range"][0]) 4. Training setup 4.1. Define WAC-Net using keras with tensorflow backend. In [48]: **from tensorflow import** keras Sequential = keras.models.Sequential Model = keras.models.Model Dense = keras.layers.Dense # Dropout = keras.layers.Dropout Conv = keras.layers.Conv2D Conv1D = keras.layers.Conv1D ConvL = keras.layers.LocallyConnected2D Pool = keras.layers.MaxPooling2D Input = keras.layers.Input Concat = keras.layers.Concatenate TimeDistributed = keras.layers.TimeDistributed Flatten = keras.layers.Flatten Lambda = keras.layers.Lambda K = keras.backendmodel = Sequential() model.add( ( Conv ( 32, kernel size=3, strides=1, activation="relu", input shape=(64, 64, 1), model.add(Pool(2)) model.add((Conv(64, kernel size=3, strides=1, activation="relu"))) model.add(Pool(2)) model.add((Conv(128, kernel size=3, strides=1, activation="relu"))) model.add(Pool(2)) model.add((Conv(256, kernel\_size=3, strides=1, activation="relu"))) model.add((Flatten())) model.add((Dense(128, activation="relu"))) model.add((Dense(128, activation="relu"))) stack = Input(model.input shape) vectors = TimeDistributed(model)( stack ) # time distributed applies a layer to every temporal slice of the input weights = Conv1D(128, 1, padding="same") (vectors) weights = Conv1D(128, 1, padding="same") (weights) weights = Conv1D(1, 1, padding="same") (weights) def merge function(tensors): x = tensors[0]weights = tensors[1] weights = K.softmax(weights, axis=1) merged = K.sum(x \* weights, axis=1)return merged merge\_layer = Lambda(merge\_function) merged = merge\_layer([vectors, weights]) merged = Dense(32, activation="relu") (merged) merged = Dense(32, activation="relu") (merged) out = Dense(2) (merged) model = Model(stack, out) model.summary() Model: "functional\_3" Layer (type) Output Shape Param # Connected to \_\_\_\_\_\_ input\_2 (InputLayer) [(None, None, 64, 64 0 time distributed 1 (TimeDistrib (None, None, 128) input 2[0][0] 928768 conv1d 3 (Conv1D) 16512 time distributed 1[0][0] (None, None, 128) conv1d 4 (Conv1D) 16512 conv1d 3[0][0] (None, None, 128) 129 conv1d\_4[0][0] conv1d 5 (Conv1D) (None, None, 1) (None,  $1\overline{28}$ ) lambda 1 (Lambda) time distributed 1[0][0] conv1d\_5[0][0] 4128 dense 7 (Dense) (None, 32) lambda 1[0][0] dense 8 (Dense) (None, 32) 1056 dense 7[0][0] dense 9 (Dense) (None, 2) dense 8[0][0] \_\_\_\_\_\_\_ Total params: 967,171 Trainable params: 967,171 Non-trainable params: 0 In [49]: model.compile(tf.keras.optimizers.Adam(lr=0.0001, amsgrad=True), loss="mae") 4.2. Defining batch function In [50]: def batch function(image): for i in range(len(image)): image[i] = image[i] / np.median(image[i]) - 1return image 4.3. Defining generator The generator generates 2000 image sequences before the training begins, and continous to generate another 2000 sample sequences during the training process In [51]: | generator = dt.generators.ContinuousGenerator( image\_of\_particles, get\_drymass\_radius, batch size=64, batch function=batch\_function, min\_data\_size=2000, max\_data\_size=4000, ndim=5, 5. Training the model Set the TRAIN MODEL = True to train the model from scratch. Set the TRAIN MODEL = False to load a pretrained model. In [52]: TRAIN MODEL = False 5.1. Generating validation data Skip this step if would not like to generate validation data. In [53]: if TRAIN MODEL: print("Generating validation data...") b = []1 = [] for i in tqdm(range(1000)): im = image\_of\_particles.update(skip\_aug=False).resolve() b.append(batch function(im)) l.append(get\_drymass radius(im)) b = np.array(b) l = np.array(l)5.2. Start the training Set the TRAIN MODEL to True to train the network from scratch. In [63]: if TRAIN MODEL: with generator: history = model.fit( generator, epochs=500, steps\_per\_epoch=16#, validation\_data=(b, 1) else: model.load weights("../pre-trained-models/WACNet dry mass.h5") 6. Testing the trained model on experimental sequences 6.1. Normalising the experimental images Functions normalise the images, and to convert predicted dry mass values to real values. def Normalise(images, batch = 15): Normalised = [] for i in range(len(images)): Normalised.append(images[i]/np.median(images[i])) Normalised = np.array(Normalised)-1 proc = [] #sliding window for i in range(len(Normalised)-batch+1): proc.append(Normalised[i:i+batch]) return np.expand dims(proc, axis = -1) In [56]: **def** real dm(p, a=209.16, b=0.28, sp ri inc = 0.21): return (p\*a+b)/sp\_ri\_inc 6.2. Checking predictions In [57]: prediction\_predator = model.predict( Normalise( predator sequence, batch = 1prediction prey = model.predict( Normalise( prey\_sequence, batch = 1In [58]: drymass predator = real dm(prediction predator[:,0]) drymass prey = real dm(prediction prey[:,0]) In [59]: feeding at = 219 fig, ax = plt.subplots(figsize=(15, 5)) plt.title("Feeding event") plt.plot(np.arange(0, feeding at,1), drymass predator[0:feeding at], linestyle='dashed',color='orange', marker='.', label='\$\it{Oxyrrhis\ Marina}\$ pre-feeding', alpha = 1, markersize = '5') plt.plot(np.arange(feeding at,len(drymass predator), 1),drymass predator[feeding at:], linestyle='dashe d',color='darkorange', marker='.', label='\$\it{Oxyrrhis\ Marina}\$ post-feeding', alpha = 1, markersize = '5') plt.plot(drymass prey[:feeding at], linestyle='dashed',color='blue', marker='.', label='\$\it{Dunaliella \ Teriolecta}\$', alpha=1, markersize = '5') plt.xlabel('Time (seconds)') plt.ylabel('Drymass (in picograms)') xticks = np.arange(0,550,50)ax.set xticks(xticks) ax.set xticklabels([int(x\*(1/10)) for x in xticks]) plt.legend(prop={'size': 16}) plt.show() Feeding event 700 Oxyrrhis Marina pre-feeding 600 Oxyrrhis Marina post-feeding Dunaliella Teriolecta ymass (in picograms) 500 400 100

25

Time (seconds)

30

40

**Tutorial 2. Training WAC-Net to predict plankton dry masses** 

from RU-Net. It covers information on:

Open in Colab

This tutorial demonstrates how to train a WAC-Net (Weighted average convolutional neural network) for refining the predicted properties