This tutorial demonstrates how to train a WAC-Net (Weighted average convolutional neural network) for refining the predicted properties from RU-Net. It covers information on: 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 → **j** z-distance vectors softmax(weights) NOTE: • If you're running this notebook on your local machine, please comment the code in the cell below In [8]: !git clone https://github.com/softmatterlab/Quantitative-Microplankton-Tracker.git %cd Quantitative-Microplankton-Tracker/training-tutorials/ %matplotlib inline In [9]: import sys sys.path.append("..") 1. Setup Imports the dependencies needed to run this tutorial. In [10]: import numpy as np import tensorflow as tf import matplotlib.pyplot as plt 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" coords 2d predator = np.load("../data/data figure3/predator 2d coords.npy" coords 2d prey = np.load("../data/data figure3/prey 2d coords.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 [11]: 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 [12]: 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) sphere main = dt.Sequential(sphere main, position=get position) 2.2.2 Defining noise planktons The noise planktons are planktons in the background. _sphere_noise = dt.Sphere(In [13]: 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 [14]: 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 [15]: 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 [16]: 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 [17]: 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 [18]: image_of_particles.update(skip_aug="False").plot(cmap="gray") Once Loop Reflect Out[18]: <matplotlib.animation.ArtistAnimation at 0x7fef76974fd0> 3. Creating target values 3.1. Vertical position values Function to generate normalized z values as outputs. In [19]: def get_z(image): z = image[0].get property("z") z = (z - variables["z range"][0] / variables["pix size"]) / (variables["z_range"][1] / variables["pix_size"] - variables["z_range"][0] / variables["pix_size"] return z 4. Training setup 4.1. Define WAC-Net using keras with tensorflow backend In [20]: **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)() # 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(1) (merged) model = Model(stack, out) model.summary() Model: "functional 1" Layer (type) Output Shape Param # Connected to _______ input 1 (InputLayer) [(None, None, 64, 64 0 time distributed (TimeDistribut (None, None, 128) 928768 input 1[0][0] conv1d (Conv1D) (None, None, 128) 16512 time distributed[0][0] conv1d 1 (Conv1D) (None, None, 128) 16512 conv1d[0][0] conv1d 2 (Conv1D) 129 conv1d 1[0][0] (None, None, 1) lambda (Lambda) (None, 128) time distributed[0][0] conv1d 2[0][0] (None, 32) dense 2 (Dense) 4128 lambda[0][0] dense_3 (Dense) 1056 (None, 32) dense 2[0][0] 33 dense 4 (Dense) (None, 1) dense 3[0][0] ______ Total params: 967,138 Trainable params: 967,138 Non-trainable params: 0 In [21]: model.compile(tf.keras.optimizers.Adam(lr=0.0001, amsgrad=True), loss="mae") 4.2. Defining batch function In [22]: def batch_function(image): for i in range(len(image)): image[i] = image[i] / np.median(image[i]) - 1 return 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 [23]: | generator = dt.generators.ContinuousGenerator(image_of_particles, get_z, batch size=64, batch function=batch function, min_data_size=2000, max data size=4000, 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 [24]: TRAIN MODEL = False 5.1. Generating validation data Skip this step if would not like to generate validation data. In [25]: 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_z(im)) b = np.array(b)l = np.array(1)5.2. Start the training Set the TRAIN MODEL to True to train the network from scratch. In [26]: if TRAIN MODEL: with generator: history = model.fit(generator, epochs=1000, steps_per_epoch=16, validation_data=(b, 1) model.load_weights("../pre-trained-models/WACNet vertical positions.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. In [27]: def Normalise(images, batch = 15): Normalised = [] for i in range(len(images)): Normalised.append(images[i]/np.median(images[i])) Normalised = np.array(Normalised)-1proc = [] #sliding window for i in range(len(Normalised)-batch+1): proc.append(Normalised[i:i+batch]) return np.expand dims(proc, axis = -1) In [28]: def real_z(p, a=700, b=2300): return (p*a+b) 6.2. Checking predictions In [29]: prediction predator = model.predict(Normalise(predator sequence, batch = 1prediction prey = model.predict(Normalise(prey sequence, batch = 1In [30]: z predator = real z(prediction predator[:,0]) z prey = real z(prediction prey[:,0]) In [31]: feeding at = 219 fig = plt.figure(figsize=(10,10)) ax = fig.gca(projection='3d') ax.plot(coords_2d_predator[:,0][:feeding_at], coords_2d_predator[:,1][:feeding_at], z_predator[:feeding _at], label='\$\it{Oxyrrhis\ marina}\$ pre-feeding', color="tab:orange", alpha=0.5, linestyle="solid", linewidth=2) ax.plot(coords_2d_prey[:,0][:feeding_at], coords_2d_prey[:,1][:feeding_at], z_prey[:feeding_at], label= '\$\it{Dunaliella\ tertiolecta}\$ pre-feeding', color="tab:blue", alpha=0.5, linestyle="solid", linewidth=2) ax.plot(coords 2d predator[:,0][feeding at:], coords 2d predator[:,1][feeding at:], z predator[feeding at:], label='\$\it{Oxyrrhis\ marina}\$ post-feeding', color="darkorange", alpha=0.5, linestyle="solid", linewidth=2) plt.legend() ax.set xlabel('\n\n\n Position X') ax.set ylabel('\n\n\n Position Y') ax.set zlabel(' $\n\n$ Position Z (\m mu m\$)') ax.azim = -50ax.elev = 40ax.dist = 10plt.show() Oxyrrhis marina pre-feeding Dunaliella tertiolecta pre-feeding Oxyrrhis marina post-feeding 2800 2700 2600 2500 2400 2300 700 650 700 600 750 800 Position x 850 900 400 950

Tutorial 3. Training WAC-Net to predict plankton vertical positions

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