# **Documentation: trainsegmentation v 0.1.5**

## Importing training data

Training data should be 2d-numpy arrays. Labeled images should be the same size as the corresponding training image and can be imported from binary masks with the filenames from a separate directory using import training data.

**import\_training\_data**(imgdir,maskdir,ext = '.tif'): imports images and labels from different directories to use for generating feature sets and training data.

#### Parameters:

imgdir: str

path to the image directory or image file as string

maskdir: list of str

list of strings indicating paths to directories or files containing labeled images (<u>if paths to directories the files must have matching filenames to the images in the image directory to ensure corresponding images and labels.</u>

ext: str

extension of image filenames. Default is '.tif'

#### Returns:

IMG: list of ndarray

list containing images (ndarrays) imported from imgdir

LABELS: list of ndarray

list containing labeled images corresponding to IMG. Label numbers match the order of paths listed in maskdir (e.g. 1, 2, 3, etc.)

## Generating feature sets for training data

Once training data is imported, feature sets are generated to create the input needed for pixel classification. Functions to create feature sets are passed in through a list: featureselect. The input to all of these functions will be the image (ndarray). To define other variables to pass through the function for all images, create a new function or use functools.partial().

**get\_training\_data**(IMG,LABELS,featureselect,loaddatafile = None, savedatafile = None, returnmeta = False): takes lists of images, corresponding labels and selected features (see Defining Features) to generate feature sets then output training data to use with a classifier..

## Parameters:

IMG: list of ndarray or ndarray

list containing images (ndarrays) imported from imgdir

LABELS: list of ndarray or ndarray

list containing labeled ndarrays with shapes corresponding to IMG. Label numbers match the order of paths listed in maskdir (e.g. 1, 2, 3, etc.)

featureselect: list of functions

list of functions used to determine features, each expecting a single image (ndarray) as the input. See Defining Features.

loaddatafile: str

string with path to a previously generated data file which will be appended to the data

generated from the input images and labels

savedatafile: str

string with path to save data file as a pickle file

returnmeta: Boolean

if true adds meta (list of strings corresponding to metadata from each feature)

## Returns:

traininglabels: ndarray

a flattened (1d) ndarray contining the training labels

trainingfeatures: ndarray

a flattened (2d) ndarray containing the feature sets, wherein len(axis = 1) is equal to the

number of features

meta: list of str

list of strings output as metadata from each feature set, only output if returnmeta = true

**load\_training\_data**(loaddatafile, returnmeta = False): loads previously generated training data from the datafile

#### Parameters:

loaddatafile: str

string with path to a previously generated data file which will be appended to the data

generated from the input images and labels

returnmeta: Boolean

if true adds meta (list of strings corresponding to metadata from each feature)

#### Returns:

traininglabels: ndarray

a flattened (1d) ndarray contining the training labels

trainingfeatures: *ndarray* 

a flattened (2d) ndarray containing the feature sets, wherein len(axis = 1) is equal to the

number of features

featureselect: function or list of functions

functions used to determine features, the function(s) will be called with each image

(ndarray) as the input. See Defining Features.

meta: list of str

list of strings output as metadata from each feature set, only output if returnmeta = true

# Training a classifier

The training data is then used to train an sklearn classifier.

**train\_classifier**(traininglabels, trainingfeatures, featureselect, saveclftofile = None, clf = None). Trains a classifier using labels and features generated by get\_training\_data.

## **Parameters:**

traininglabels: ndarray

a flattened (1d) ndarray contining the training labels

trainingfeatures: ndarray

a flattened (2d) ndarray containing the feature sets, wherein len(axis = 1) is equal to the

number of features

featureselect: function or list of functions

functions used to determine features, the function(s) will be called with each image

(ndarray) as the input. See Defining Features.

saveclftofile: str

string with path to classifier as a pickle file

clf: sklearn classifier

A sklearn classifier, default (None) is clf =

 $sklearn.ensemble.RandomForestClassifier (n\_estimators = 50, n\_jobs = -1, max\_depth = 10, n\_jobs = -1, max\_depth = -1, ma$ 

max\_samples=0.05)

#### **Returns:**

clf: sklearn classifier trained sklearn classifier

## Applying a classifier

Once trained, classifiers can be applied to similar images to generate probability or binary masks.

load\_classifier(clffile): loads a classifier (pickle file) saved by train\_classifier

#### Parameters:

clffile: str

string with path to pickle file containing classifier

## Returns:

clf: sklearn classifier

trained sklearn classifier

**classify\_image**(img,clf,featureselect): generates feature set for an input image and outputs predicted classification.

#### Parameters:

img: *ndarray* input image clf: *sklearn classifier* 

classifier trained by train classifier

featureselect: list of functions

list of functions used to determine features, each expecting a single image (ndarray) as the input. Must match those used when training the classifier.

### **Returns:**

result: ndarray

labeled ndarray with shape img.shape

**classify\_image\_probability**(img,clf,featureselect): generates feature set for an input image and outputs probability of classification.

#### Parameters:

img: *ndarray* input image clf: *sklearn classifier* 

classifier trained by train\_classifier

featureselect: list of functions

list of functions used to determine features, each expecting a single image (ndarray) as the input. <u>Must match those used when training the classifier</u>

#### Returns:

result: ndarray

labeled ndarray with shape (img.shape[0], img.shape[1], # of labels)

**classify\_image\_label**(img, clf, featureselect, selectlabel = 1): generates feature set for an input image and outputs predicted classification for the indicated label.

## Parameters:

img: ndarray input image clf: sklearn classifier

classifier trained by train\_classifier

featureselect: list of functions

list of functions used to determine features, each expecting a single image (ndarray) as the input. Must match those used when training the classifier

selectlabel: int

integer indicating the label to output classification

#### Returns:

result: ndarray

labeled ndarray with shape img.shape

**classify\_image\_label\_probability**(img,clf,featureselect,selectlabel = 1): generates feature set for an input image and outputs probability of classification for the indicated label.

#### Parameters:

img: ndarray input image clf: sklearn classifier

classifier trained by train\_classifier

featureselect: list of functions

list of functions used to determine features, each expecting a single image (ndarray) as

the input. Must match those used when training the classifier

selectlabel: int

integer indicating the label to output classification

#### Returns:

label: ndarray

labeled ndarray with shape img.shape

**threshold\_mask**(img, threshmethod = sklearn.filters.threshold\_minimum): generates a mask by thresholding the input image with the given threshold method.

#### Parameters:

img: ndarray
input image
threshmethod: function
a thresholding function

#### **Returns:**

mask: *ndarray* 

labeled ndarray with shape img.shape; outputs a binary mask 0 or 255.

# **Defining Features**

The following functions are used to generate image features. To utilize parameters other than *img* with variables other than the default, use function.partial() or define a new function to initiate those parameters as desired then call the newly defined partial function.

**Neighbors**(img, minSigma = 1, maxSigma = 16)\*\*: shifts the image in 8 directions by a certain number of pixels,  $\sigma$ . Creates 8n feature images where  $(2^{n-1})$ \*minSigma  $\leq$  maxSigma.

## **Parameters:**

img: *ndarray*input image
minSigma: *int*minimum value of σ
maxSigma: *int*maximum value of σ

#### Returns:

meta: list of str

Meta data for features, wherein strings indicate feature of each ndarray in features. e.g.

'Neighbors\_1\_0' wherein the integers indicate  $\sigma$  and direction respectively.

features: list of ndarray

List containing feature images each of shape(img)

**Membrane\_projections**(img,nAngles = 30, patchSize = 19,membraneSize = 1)\*\*: enhances membrane-like structures of the image through directional filtering. The initial kernel for this operation is patchsize<sup>2</sup> zero matrix with the membraneSize number of middle column entries set to 1. Multiple kernels are created by rotating the original kernel 180 degrees/nAngles for a number of kernels = nAngles. Each kernel is convolved with the image and then the set of images are Z-projected into a single image via 6 methods:

- sum of the pixels in each image
- mean of the pixels in each image
- standard deviation of the pixels in each image
- median of the pixels in each image
- maximum of the pixels in each image
- minimum of the pixels in each image

Each of the 6 resulting images is a feature. Hence pixels in lines of similarly valued pixels in the image that are different from the average image intensity will stand out in the Z-projections.

### Parameters:

img: *ndarray* input image nAngles: *int* 

number of kernels

patchSize: int

size of matrix (patchSize x patchSize) to convolve image

membraneSize: int

number of columns in middle of patch to set to one before rotating and convolving

#### **Returns:**

meta: list of str

Meta data for *features*, wherein strings indicate feature of each ndarray in *features*. e.g.

'Membrane\_projections\_0' wherein the integer indicates the method.

features: list of ndarray

List containing feature images each of shape(img)

**Guassian\_blur**(img,minSigma = 1,maxSigma = 16)\*\*: performs n individual convolutions with Gaussian kernels with the normal n variations of  $\sigma$ . The larger the radius the more blurred the image becomes until the pixels are homogeneous

#### Parameters:

img: *ndarray* input image minSigma: *int* 

minimum value of  $\sigma$ 

maxSigma: int

maximum value of  $\sigma$ 

#### Returns:

meta: list of str

Meta data for *features*, wherein strings indicate feature of each ndarray in *features*. e.g. 'Gaussian blur 1' wherein the integer indicates  $\sigma$ .

features: *list of ndarray* 

List containing feature images each of shape(img)

**Difference\_of\_Gaussians**(img,minSigma = 1,maxSigma = 16)\*\*: performs convolutions with Gaussian kernels with the normal n variations of  $\sigma$  and subtracts the previous iteration to obtain the feature image, with the first image to be subtracted being the original image.

#### Parameters:

img: *ndarray*input image
minSigma: *int*minimum value of σ

minimum value of o

maxSigma: int

maximum value of σ

#### **Returns:**

meta: list of str

Meta data for *features*, wherein strings indicate feature of each ndarray in *features*. e.g. 'Difference\_of\_Gaussian\_1\_0' wherein the integer indicates  $\sigma$  for initial image and  $\sigma$  for the subtracted image respectively (the original image is 0).

features: list of ndarray

List containing feature images each of shape(img)

**Sobel\_filter**(img, minSigma = 1, maxSigma = 16)\*\*: calculates an approximation of the gradient of the image intensity at each pixel using ndimage.sobel. Gaussian blurs with  $\sigma$  varying as usual are performed prior to the filter.

## Parameters:

img: *ndarray* input image minSigma: *int* 

minimum value of σ maxSigma: *int* maximum value of σ

### **Returns:**

meta: list of str

Meta data for features, wherein strings indicate feature of each ndarray in features. e.g.

'Sobel\_filter\_1' wherein the integer indicates  $\sigma$ .

features: list of ndarray

List containing feature images each of shape(img)

**Hessian**(img, minSigma = 1, maxSigma = 16)\*\*: runs a Hessian filter (sklearn.filters.hessian). Gaussian blurs with  $\sigma$  varying as usual are performed prior to the filter.

#### Parameters:

img: *ndarray* input image minSigma: *int* 

minimum value of  $\sigma$ 

maxSigma: int

maximum value of  $\boldsymbol{\sigma}$ 

#### Returns:

meta: list of str

Meta data for features, wherein strings indicate feature of each ndarray in features. e.g.

'Hessian\_1.0' wherein the integer indicates  $\sigma$ .

features: list of ndarray

List containing feature images each of shape(img)

**Watershed\_distance**(img, threshmethod = filters.threshold\_yen): thresholds the image with the indicated threshold method and transforms the image based on distances between objects (see Distances output in example: <a href="https://scikit-image.org/docs/stable/auto\_examples/segmentation/plot\_watershed.html">https://scikit-image.org/docs/stable/auto\_examples/segmentation/plot\_watershed.html</a>). Default threshmethod is skimage.filters.threshold\_yen. Other threshold methods can be applied from skimage.filters

## Parameters:

img: *ndarray* input image

threshmethod: function

select threshold method from skimage.filters. Default is skimage.filters.threshold\_yen.

#### Returns:

meta: list of str

Meta data for features, ['Watershed distance']

features: *list of ndarray* 

List containing feature image of shape(img)

**Meijering\_filter**(img, minSigma = 1, maxSigma = 16): applies Meijering neuriteness filter on n images, with Gaussian blurs with  $\sigma$  varying as usual are performed prior to the filter.

#### Parameters:

img: *ndarray* input image minSigma: int

minimum value of  $\sigma$ 

maxSigma: int

maximum value of  $\sigma$ 

#### Returns:

meta: list of str

Meta data for features, wherein strings indicate feature of each ndarray in features. e.g.

'Meijering filter 1' wherein the integer indicates  $\sigma$ .

features: list of ndarray

List containing feature images each of shape(img)

**Sklearn\_basic**(img): runs sklearn. feature.multiscale\_basic\_features(img) to image and returns results.

#### Parameters:

img: ndarray input image

#### Returns:

meta: list of str

Meta data for features. Repeating list of strings 'Sklearn basic' with length of features

features: list of ndarray

List containing feature images each of shape(img)

**Mean**(img, minsigma = 1, maxsigma = 16): gets the mean of each pixel and surrounding pixels distance  $\sigma$  with the normal n variations of  $\sigma$ .

## **Parameters:**

img: *ndarray* input image minSigma: int minimum value of  $\sigma$ 

maxSigma: int

#### maximum value of σ

#### Returns:

meta: list of str

Meta data for features, wherein strings indicate feature of each ndarray in features. e.g.

'Mean 1' wherein the integer indicates  $\sigma$ .

features: list of ndarray

List containing feature images each of shape(img)

**Variance**(img, minsigma = 1, maxsigma = 16): gets the variance of each pixel and surrounding pixels distance  $\sigma$  with the normal n variations of  $\sigma$ .

#### **Parameters:**

img: *ndarray* input image minSigma: *int* 

minimum value of  $\sigma$ 

maxSigma: int

maximum value of  $\sigma$ 

#### Returns:

meta: list of str

Meta data for features, wherein strings indicate feature of each ndarray in features. e.g.

'Variance 1' wherein the integer indicates  $\sigma$ .

features: list of ndarray

List containing feature images each of shape(img)

**Median**(img, minsigma = 1, maxsigma = 16): gets the median of each pixel and surrounding pixels distance  $\sigma$  with the normal n variations of  $\sigma$ .

## Parameters:

img: *ndarray* input image minSigma: *int* 

minimum value of  $\sigma$ 

maxSigma: int

maximum value of  $\sigma$ 

#### Returns:

meta: list of str

Meta data for *features*, wherein strings indicate feature of each ndarray in *features*. e.g.

'Median\_1' wherein the integer indicates  $\sigma$ .

features: list of ndarray

List containing feature images each of shape(img)

**Maximum**(img, minsigma = 1, maxsigma = 16): gets the maximum of each pixel and surrounding pixels distance  $\sigma$  with the normal n variations of  $\sigma$ .

#### Parameters:

img: ndarray
input image
minSigma: int

minimum value of  $\boldsymbol{\sigma}$ 

maxSigma: int

maximum value of  $\boldsymbol{\sigma}$ 

## Returns:

meta: list of str

Meta data for *features*, wherein strings indicate feature of each ndarray in *features*. e.g. 'Maximum 1' wherein the integer indicates  $\sigma$ .

features: list of ndarray

List containing feature images each of shape(img)

**Minimum**(img, minsigma = 1, maxsigma = 16): gets the minimum of each pixel and surrounding pixels distance  $\sigma$  with the normal n variations of  $\sigma$ .

#### Parameters:

img: *ndarray* input image minSigma: *int* 

minimum value of  $\sigma$ 

maxSigma: int

maximum value of σ

## Returns:

meta: list of str

Meta data for *features*, wherein strings indicate feature of each ndarray in *features*. e.g. 'Minimum 1' wherein the integer indicates  $\sigma$ .

features: *list of ndarray* 

List containing feature images each of shape(img)