PicturedRocks Documentation

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CHAPTER 1

Installing

Please ensure you have Python 3.6 or newer and have *numba* and *scikit-learn* installed. The best way to get Python and various dependencies is with Anaconda or Miniconda. Once you have a conda environment, run conda install numba scikit-learn. Then use pip to install PicturedRocks and all additional dependencies:

```
pip install picturedrocks
```

To install the latest code from github, clone our github repository. Once inside the project directory, instal by running pip install -e .. The -e option will point a symbolic link to the current directory instead of installing a copy on your system. This allows you to update picturedrocks by pulling the latest commit from github.

1.1 Feature Selection Tutorial

In this Jupyter notebook, we'll walk through the information-theoretic feature selection algorithms in PicturedRocks and demonstrate the interactive marker selection user interface.

If you are viewing this notebook inside the PicturedRocks documentation, the interactive marker selection tool will not work (as it needs a python backend to perform the computations. You can download this notebook from GitHub and run in on your own computer to try the interactive tool.)

```
[3]: adata
[3]: AnnData object with n_obs × n_vars = 2730 × 3451
    obs: 'paul15_clusters'
    uns: 'iroot'
```

The process_clusts method copies the cluster column and precomputes various indices, etc. If you have multiple columns that can be used as target labels (e.g., different treatments, clusters via different clustering algorithms or parameters, or demographics), this sets and processes the given columns as the one we're currently examining.

This is necessary for supervised analysis and visualization tools in PicturedRocks that use cluster labels.

```
[4]: pr.read.process_clusts(adata, "paul15_clusters")
[4]: AnnData object with n_obs × n_vars = 2730 × 3451
    obs: 'paul15_clusters', 'clust', 'y'
    uns: 'iroot', 'num_clusts', 'clusterindices'
```

The make_infoset method creates a SparseInformationSet object with a discretized version of the data matrix. It is useful to have only a small number of discrete states that each gene can take so that entropy is a reasonable measurement. By default, make_infoset performs an adaptive transform that we call a recursive quantile transform. This is implemented in pr.markers.mutualinformation.infoset. quantile_discretize. If you have a different discretization transformation, you can pass a transformed matrix directly to SparseInformationSet.

```
[5]: infoset = pr.markers.makeinfoset(adata, True)
```

Because this dataset only has 3451 features, it is computationally easy to do feature selection without restricting the number of features. If we wanted to, we could do either supervised or unsupervised univariate feature selection (i.e., without considering any interactions between features).

```
[6]: # supervised
mim = pr.markers.mutualinformation.iterative.MIM(infoset)
most_relevant_genes = mim.autoselect(1000)
```

```
[7]: # unsupervised
ue = pr.markers.mutualinformation.iterative.UniEntropy(infoset)
most_variable_genes = ue.autoselect(1000)
```

At this stage we can slice our adata object as adata[:,most_relevant_genes] or adata[:,most_variable_genes] and create a new InformationSet object for this sliced object. We don't need to do that here since there are not a lot of genes but will do so anyway for demonstration purposes.

1.1.1 Supervised Feature Selection

Let's jump straight into supervised feature selection. Here we will use the CIFE objective

```
[8]: adata_mr = adata[:,most_relevant_genes].copy()
  infoset_mr = pr.markers.makeinfoset(adata_mr, True)

[9]: cife = pr.markers.CIFE(infoset_mr)

[10]: cife.score[:20]
```

```
[10]: array([0.95022366, 0.93749845, 0.88470651, 0.86819372, 0.8634894, 0.80903075, 0.75775072, 0.75361203, 0.71991963, 0.7106652, 0.70321104, 0.6821289, 0.67109598, 0.65202536, 0.65192364, 0.6458561, 0.64569101, 0.63526239, 0.62452935, 0.62346646])
```

Let's select 'Mpo'

```
[12]: ind = adata_mr.var_names.get_loc('Mpo')
[13]: cife.add(ind)
```

Now, the top genes are

Observe that the order has changed based on redundancy (or lack thereof) with 'Mpo'. Let's add 'Car1'

```
[15]: ind = adata_mr.var_names.get_loc('Car1')
    cife.add(ind)
```

If we want to select the top gene repeatedly, we can use autoselect

```
[17]: cife.autoselect(5)
```

To look at the markers we've selected, we can examine cife.S

```
[18]: cife.S
[18]: [0, 5, 187, 23, 49, 931, 306]
[19]: adata_mr.var_names[cife.S]
[19]: Index(['Mpo', 'Carl', 'Actb', 'H2afy', 'Hsp90abl', 'Gpr56', 'Ly6e'], dtype='object')
```

User Interface

This process can also done manually with a user-interface allowing you to incorporate domain knowledge in this process. Use the View dropdown to look at heatplots for candidate genes and already selected genes.

Normalize per cell and log transform the data. We are doing this here only to generate familiar features. We do not recommend performing these transformations before make_infoset.

```
[20]: sc.pp.normalize_per_cell(adata_mr)
    sc.pp.log1p(adata_mr)

[21]: im = pr.markers.InteractiveMarkerSelection(adata_mr, cife, ['tsne', 'violin'])
    Running tsne on cells...

[22]: im.show()
    Output()
```

Note, that because we passed the same cife object, any genes added/removed in the interface will affect the cife object.

```
[23]: adata_mr.var_names[cife.S]
[23]: Index(['Mpo', 'Car1', 'Actb', 'H2afy', 'Hsp90ab1', 'Gpr56', 'Ly6e'], dtype='object')
```

1.1.2 Unsupervised Feature Selection

This works very similarly. In the example below, we'll autoselect 5 genes and then run the interface. Note that although the previous section would not work without cluster labels, the following code will.

```
[24]: cife_unsup = pr.markers.CIFEUnsup(infoset)
[25]: cife_unsup.autoselect(5)
```

If you ran the example above, this will load faster because the t_SNE coordinates for genes and cells have already been computed.

```
[27]: im_unsup.show()
Output()
```

1.1.3 Binary Feature Selection

We can also perform feature selection specifically for individual class labels (e.g., clusters). This is done by changing the SparseInformationSet's y array. In the example below, we will target the class label "2Ery". Notice that the features selected by MIM (MIM doesn't consider redundancy) are only those that are informative about "2Ery" in particular.

Binary (i.e., not multiclass) feature selection can be performed with any information-theoretic feature selection algorithm (e.g., CIFE, JMI, MIM).

1.2 Reading data

In addition to various functions for reading input data in scanpy, various methods in *picturedrocks* need cluster labels.

```
picturedrocks.read.process_clusts(adata, name='clust', copy=False)
```

Process cluster labels from an obs column

This copies adata.obs[name] into adata.obs["clust"] and precomputes cluster indices, number of clusters, etc for use by various functions in PicturedRocks.

Parameters

- adata (anndata. AnnData) -
- copy (bool) determines whether a copy of *AnnData* object is returned

Returns object with annotation

Return type anndata. Ann Data

Notes

The information computed here is lost when saving as a .loom file. If a .loom file has cluster information, you should run this function immediately after sc.read_loom.

Read cluster labels from a csv into an obs column

Parameters

- adata (anndata. AnnData) the AnnData object to read labels into
- **filename** (str) filename of the csv file with labels
- **sep** (str, optional) csv delimiter
- name (str, optional) destination for label is adata.obs[name]
- header (bool) deterimes whether csv has a header line. If false, it is assumed that data begins at the first line of csv
- copy (bool) determines whether a copy of *AnnData* object is returned

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Returns object with cluster labels

Return type anndata.AnnData

Notes

- Cluster ids will automatically be changed so they are 0-indexed
- csv can either be two columns (in which case the first column is treated as observation label and merging handled by pandas) or one column (only cluster labels, ordered as in adata)

1.3 Preprocessing

The preprocessing module provides basic preprocessing tools. To avoid reinventing the wheel, we won't repeat methods already in scanpy unless we need functionality not available there.

```
picturedrocks.preprocessing.pca (data, dim=3, center=True, copy=False)
Runs PCA
```

Parameters

- data (anndata. AnnData) input data
- dim(int, optional) number of PCs to compute
- center (bool, optional) determines whether to center data before running PCA
- copy determines whether a copy of AnnData object is returned

Returns object with obsm["X_pca"], and varm["PCs"] set

Return type anndata. Ann Data

1.4 Plotting

```
picturedrocks.plot.genericplot (celldata, coords, **scatterkwargs)

Generate a figure for some embedding of data
```

Senerate a figure for some embedding of data

This function supports both 2D and 3D plots. This may be used to plot data for any embedding (e.g., PCA or t-SNE). For example usage, see code for *pcafigure*.

Parameters

- celldata (anndata.AnnData) data to plot
- coords (numpy.ndarray) (N, 2) or (N, 3) shaped coordinates of the embedded data
- **scatterkwargs keyword arguments to pass to Scatter or Scatter3D in *plotly* (dictionaries are merged recursively)

```
picturedrocks.plot.genericwrongplot(celldata, coords, yhat, labels=None, **scatterkwargs)
Plot figure with incorrectly classified points highlighted
```

This can be used with any 2D or 3D embedding (e.g., PCA or t-SNE). For example code, see *pcawrongplot*.

Parameters

- celldata (anndata. AnnData) data to plot
- coords (numpy.ndarray) (N, 2) or (N, 3) shaped array with coordinates to plot

- yhat (numpy.ndarray) (N, 1) shaped array of predicted y values
- labels (list, optional) list of axis titles
- **scatterkwargs keyword arguments to pass to Scatter or Scatter3D in *plotly* (dictionaries are merged recursively)

picturedrocks.plot.pcafigure(celldata, **scatterkwargs)

Make a 3D PCA figure for an AnnData object

Parameters

- celldata (anndata. AnnData) data to plot
- **scatterkwargs keyword arguments to pass to Scatter or Scatter3D in plotly (dictionaries are merged recursively)

picturedrocks.plot.pcawrongplot (celldata, yhat, **scatterkwargs)

Generate a 3D PCA figure with incorrectly classified points highlighted

Parameters

- celldata (anndata. AnnData) data to plot
- **yhat** (numpy.ndarray) (N, 1) shaped array of predicted y values
- **scatterkwargs keyword arguments to pass to Scatter or Scatter3D in *plotly* (dictionaries are merged recursively)

picturedrocks.plot.plotgeneheat (celldata, coords, genes, hide_clusts=False, **scatterkwargs)

Generate gene heat plot for some embedding of AnnData

This generates a figure with multiple dropdown options. The first option is "Clust" for a plot similar to *genericplot*, and the remaining dropdown options correspond to genes specified in *genes*. When *celldata.genes* is defined, these drop downs are labeled with the gene names.

Parameters

- celldata (anndata.AnnData) data to plot
- **coords** (numpy.ndarray) (N, 2) or (N, 3) shaped coordinates of the embedded data (e.g., PCA or tSNE)
- **genes** (list) list of gene indices or gene names
- hide_clusts (bool) Determines if cluster labels are ignored even if they are available

picturedrocks.plot.umapfigure(adata, **scatterkwargs)

1.5 Selecting Markers

argmax

PicturedRocks can be used to implement numerous information-theoretic feature selection methods. In both the supervised and unsupervised cases, it is computationally intractable to optimize the true objective functions. We want to find a small set of genes S, with |S| < n such that, in the supervised case I(S;y) is maximized and in the unsupervised case H(S) is maximized. (For more information, see our paper)

Because these are computationally intractable, we have implemented the following approximations to $x_i I(S \cup \{x_i\}; y)$ (the ideal supervised objective function)

• The Mutual Information Maximization (MIM) is a univariate feature selection method—it does not consider the interaction

$$J_{\min}(x_i) = I(x_i; y)$$

• The Joint Mutual Information (JMI) algorithm proposed by Yang and Moody uses a diminishing penalty on redundancy.

$$J_{\text{jmi}}(x_i) = I(x_i; y) - \frac{1}{|S|} \sum_{x_j \in S} I(x_i; x_j; y)$$

· The (CIFE) algorithm proposed by Lin and Tang (and independently by others) does not diminish its redundancy penalty

$$J_{\text{cife}}(x_i) = I(x_i; y) - \sum_{x_j \in S} I(x_i; x_j; y)$$

1.5.1 Mutual information

Before running any mutual information based algorithms, we need a discretized version of the gene expression matrix, with a limited number of discrete values (because we do not make any assumptions about the distribution of gene expression). Such data is stored in <code>InformationSet</code>, but by default, we suggest using <code>makeinfoset()</code> to generate such an object. The <code>makeinfoset()</code> function uses the recursive quantile transform <code>quantile_discretize()</code>.

Iterative Feature Selection

All information-theoretic feature selection methods in PicturedRocks are greedy algorithms. In general, they implement the abstract class IterativeFeatureSelection class. See *Supervised Feature Selection* and *Unsupervised Feature Selection* below for specific algorithms.

```
class picturedrocks.markers.IterativeFeatureSelection (infoset)
```

Abstract Class for Iterative Feature Selection

add(ind)

Select specified feature

Parameters ind (int) – Index of feature to select

autoselect (n_feats)

Auto select features

This automatically selects n_{feats} features greedily by selecting the feature with the highest score at each iteration.

Parameters n_feats (int) - The number of features to select

remove (ind)

Remove specified feature

Parameters ind (int) – Index of feature to remove

Supervised Feature Selection

```
class picturedrocks.markers.MIM(infoset)
class picturedrocks.markers.CIFE(infoset)
class picturedrocks.markers.JMI(infoset)
```

Unsupervised Feature Selection

```
class picturedrocks.markers.UniEntropy(infoset)
class picturedrocks.markers.CIFEUnsup(infoset)
```

Auxiliary Classes and Methods

```
class picturedrocks.markers.InformationSet (X, has_y=False)
    Stores discrete gene expression matrix
```

Parameters

- X (numpy.ndarray) a (num_obs, num_vars) shape array with dtype int
- has_y (bool) whether the array X has a target label column (a y column) as its last column

class picturedrocks.markers.**SparseInformationSet** (*X*, *y=None*)

Stores sparse discrete gene expression matrix

Parameters

- X (scipy.sparse.csc_matrix) a (num_obs, num_vars) shape matrix with dtype int.
- has_y (bool) whether the array X has a target label column (a y column) as its last column

 $\verb|picturedrocks.markers.makeinfoset| (adata, include_y, k=5)$

Discretize data and make a Sparse InformationSet object

Parameters

- adata (anndata.AnnData) The data to discretize. By default data is discretized as round(log2(X + 1)).
- include_y (bool) Determines if the y (cluster label) column in included in the *InformationSet* object

Returns An object that can be used to perform information theoretic calculations.

Return type SparseInformationSet

```
picturedrocks.markers.mutualinformation.infoset.quantile_discretize (X, k=5) Discretize data matrix with a recursive quantile transform
```

Parameters

- **X** (Union[numpy.ndarray, scipy.sparse.spmatrix]) The input data matrix to transform.
- **k** (*int*) The number of bins to use in the discretization.

Returns The discretized data matrix

Return type np.ndarray

1.6 Interactive Marker Selection

PicturedRocks provides an interactive user interface for selecting markers that allows the practitioner to use both their domain knowledge and the information-theoretic methods to select a small set of genes.

The user can specify the tabs they want available through the *visuals* argument. It is also possible to write your own interface for a tab using any plotting library available to you. See below for more details.

Run an interactive marker selection GUI inside a jupyter notebook

Parameters

- adata (anndata.AnnData) The data to run marker selection on. If you want to restrict to a small number of genes, slice your anndata object.
- feature_selection (picturedrocks.markers.mutualinformation. iterative.IterativeFeatureSelection) — An instance of a interative feature selection algorithm class that corresponds to adata (i.e., the column indices in feature_selection should correspond to the column indices in adata)
- **visuals** (list) List of visualizations to display. These can either be shorthands for built-in visualizations (currently "tsne", "umap", and "violin"), or an instance of InteractiveVisualization (see GeneHeatmap and ViolinPlot for example implementations).
- **disp_genes** (*int*) Number of genes to display as options (by default, number of genes plotted on the tSNE plot is 3 * *disp_genes*, but can be changed by setting the *plot_genes* property after initializing.
- **connected** (bool) Parameter to pass to plotly.offline. init_notebook_mode. If your browser does not have internet access, you should set this to False.

Warning: This class requires modules not explicitly listed as dependencies of picturedrocks. Specifically, please ensure that you have *ipywidgets* installed and that you use this class only inside a jupyter notebook.

redraw()

Redraw jupyter widgets after a change

This is called internally and there should usually be no need for the user to explicitly call this method.

show()

Display the jupyter widgets

class picturedrocks.markers.interactive.**GeneHeatmap** ($dim_red='tsne', n_pcs=30$)
GeneHeatmap for Interactive Marker Selection

Parameters

• dim_red (str) – Dimensionality reduction algorithm to use. Currently available options are "umap" and "tsne"

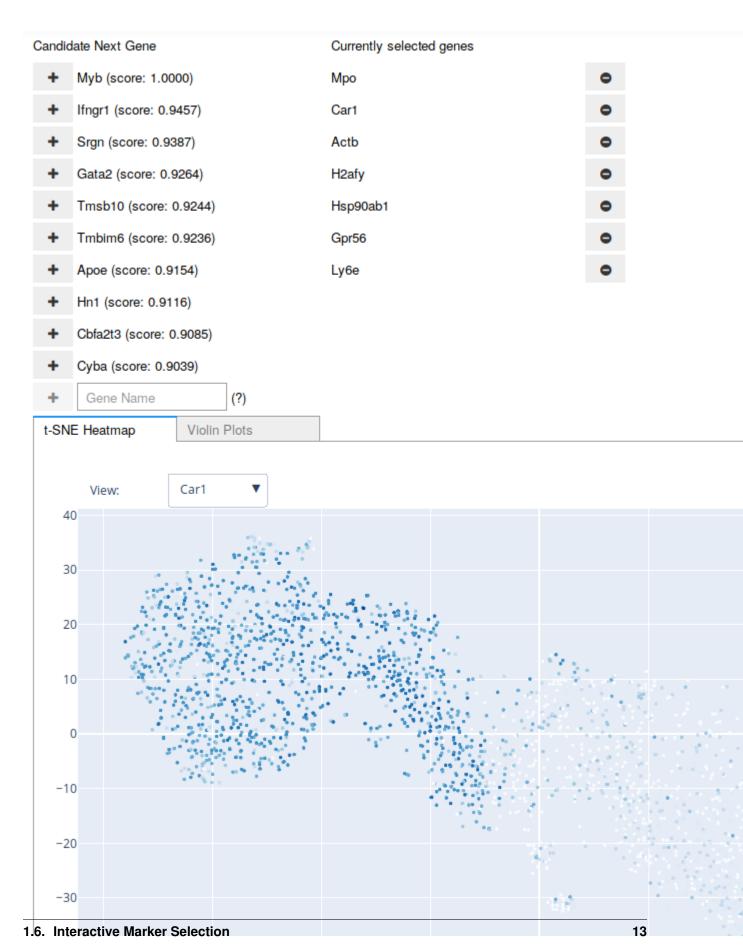


Fig. 1: A screenshot of the Interactive Marker Selection user interface being used on the Paul dataset. The "candidate" genes are updated after any addition to or removal from the "current" list of genes. The user may also type in the name

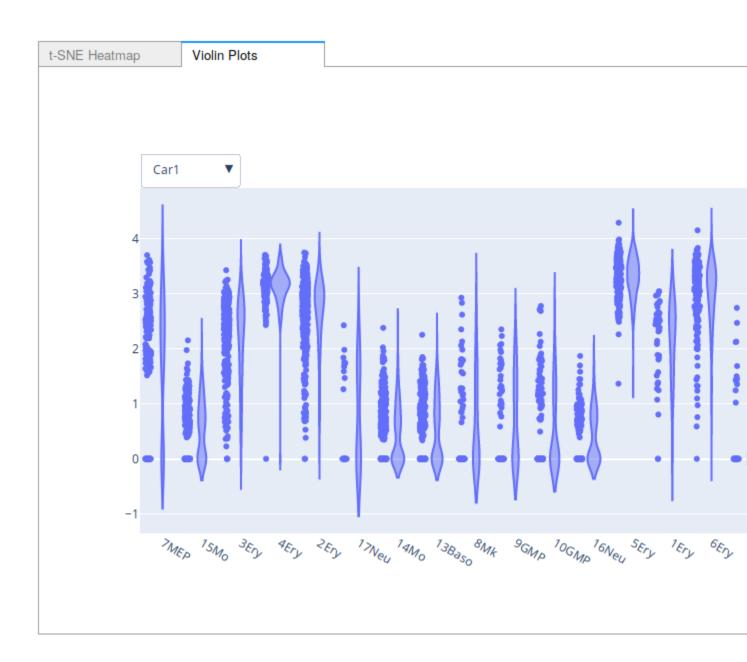


Fig. 2: A screenshot of the violin plot option for Interactive Marker Selection.

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n_pcs (int) – The number of principal components to map to before running dimensionality reduction

class picturedrocks.markers.interactive.ViolinPlot
 Violin Plots for each class label

1.6.1 Using Your Own Visualizations

To write your own visualization, simply extend the InteractiveVisualization. Use the source code for GeneHeatmap and ViolinPlot as a reference for implementation.

class picturedrocks.markers.interactive.InteractiveVisualization

Abstract base class for interactive visualizations

Extend this class and pass an instance of it to InteractiveMarkerSelection to use your own visualization. It is recommended that you begin your implementation of __init__ with:

```
super().__init__()
```

You are welcome to add parameters specific to your visualization in the __init__ method.

prepare (adata, out)

Prepare for visualization

This method is called when InteractiveMarkerSelection is initialized. It is recommended that you begin your implementation with

```
super().prepare(adata, out)
```

This stores adata and out in self.adata and self.out respectively.

redraw (next_gene_inds, cur_gene_inds)

Draw the visualization

You must implement this method. To display the plots in the appropriate widget, use:

```
with self.out:
   fig.show() # or your plotting library's equivalent
```

title

Title of the visualization

This should be a Python property, using the @property decorator.

1.7 Measuring Feature Selection Performance

This module can be used to evaluate feature selection methods via K-fold cross validation.

```
class picturedrocks.performance.FoldTester(adata)
```

Performs K-fold Cross Validation for Marker Selection

FoldTester can be used to evaluate various marker selection algorithms. It can split the data in K folds, run marker selection algorithms on these folds, and classify data based on testing and training data.

Parameters adata (anndata. AnnData) - data to slice into folds

classify (classifier)

Classify each cell using training data from other folds

For each fold, we project the data onto the markers selected for that fold, which we treat as test data. We also project the complement of the fold and treat that as training data.

Parameters classifier – a classifier that trains with a training data set and predicts labels of test data. See *NearestCentroidClassifier* for an example.

Note: The *classifier* should not attempt to modify data in-place. Any preprocessing should be done on a copy.

loadfolds(file)

Load folds from a file

The file can be one saved either by FoldTester.savefolds() or FoldTester. savefoldsandmarkers(). In the latter case, it will not load any markers.

See also:

FoldTester.loadfoldsandmarkers()

loadfoldsandmarkers(file)

Load folds and markers

Loads a folds and markers file saved by FoldTester.savefoldsandmarkers()

Parameters file (str) – filename to load from (typically with a .npz extension)

See also:

FoldTester.loadfolds()

makefolds(k=5, random=False)

Makes folds

Parameters

- **k** (int) the value of K
- random (bool) If true, *makefolds* will make folds randomly. Otherwise, the folds are made in order (i.e., the first ceil (N / k) cells in the first fold, etc.)

$\verb"savefolds"\,(file)$

Save folds to a file

Parameters file (str) – filename to save (typically with a .npz extension)

savefoldsandmarkers(file)

Save folds and markers for each fold

This saves folds, and for each fold, the markers previously found by FoldTester. selectmarkers().

Parameters file (str) – filename to save to (typically with a .npz extension)

selectmarkers (select_function)

Perform a marker selection algorithm on each fold

Parameters select_function (function) – a function that takes in an AnnData object and outputs a list of gene markers, given by their index

Note: The *select_function* should not attempt to modify data in-place. Any preprocessing should be done on a copy.

validatefolds()

Ensure that all observations are in exactly one fold

Returns

Return type bool

class picturedrocks.performance.NearestCentroidClassifier

Nearest Centroid Classifier for Cross Validation

Computes the centroid of each cluster label in the training data, then predicts the label of each test data point by finding the nearest centroid.

test (Xtest)

train(adata)

class picturedrocks.performance.PerformanceReport (y, yhat)

Report actual vs predicted statistics

Parameters

- y (numpy.ndarray) actual cluster labels, (N, 1)-shaped numpy array
- yhat (numpy.ndarray) predicted cluster labels, (N, 1)-shaped numpy array

confusionmatrixfigure()

Compute and make a confusion matrix figure

Returns confusion matrix

Return type plotly figure

getconfusionmatrix()

Get the confusion matrix for the latest run

Returns array of shape (K, K), with the [i, j] entry being the fraction of cells in cluster i that were predicted to be in cluster j

Return type numpy.ndarray

printscore()

Print a message with the score

show()

Print a full report

This uses *iplot*, so we assume this will only be run in a Jupyter notebook and that *init_notebook_mode* has already been run.

wrong()

Returns the number of cells misclassified.

picturedrocks.performance.kfoldindices (n, k, random=False)

Generate indices for k-fold cross validation

Parameters

- **n** (*int*) number of observations
- **k** (*int*) number of folds

• random (bool) – determines whether to randomize the order

Yields *numpy.ndarray* – array of indices in each fold

picturedrocks.performance.merge_markers (ft, n_markers)

picturedrocks.performance.truncatemarkers(ft, n_markers)

CHAPTER 2

Indices and tables

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