Project 2

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Scope and Goals

The goal of the project is to practice data analysis in Python. To that end, each student should choose a paper and reproduce at least two of its figures. It is important to make sure that you have access to the data. The data could have been deposited in a public repository or available from the authors upon request. Unless you have interest in processing raw data, it is often better to start with processed data. This is not mandatory, only recommended. Processing data could be the main aspect of the project especially if it is done using your own code.

Selected Paper

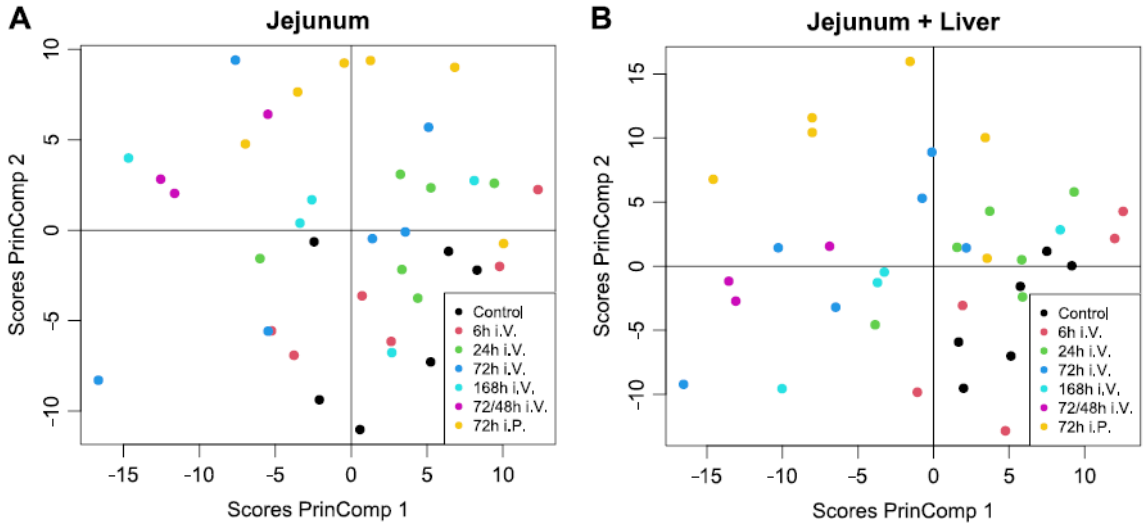
Orthogonality in Principal Component Analysis Allows the Discovery of Lipids in the Jejunum That Are Independent of Ad Libitum Feeding

https://www.mdpi.com/2218-1989/12/9/866 (https://www.mdpi.com/2218-1989/12/9/866)

Figure 1

Based on Figure S1 in the article. Scatterplot of scores of PCAs in Figure 2.

Original



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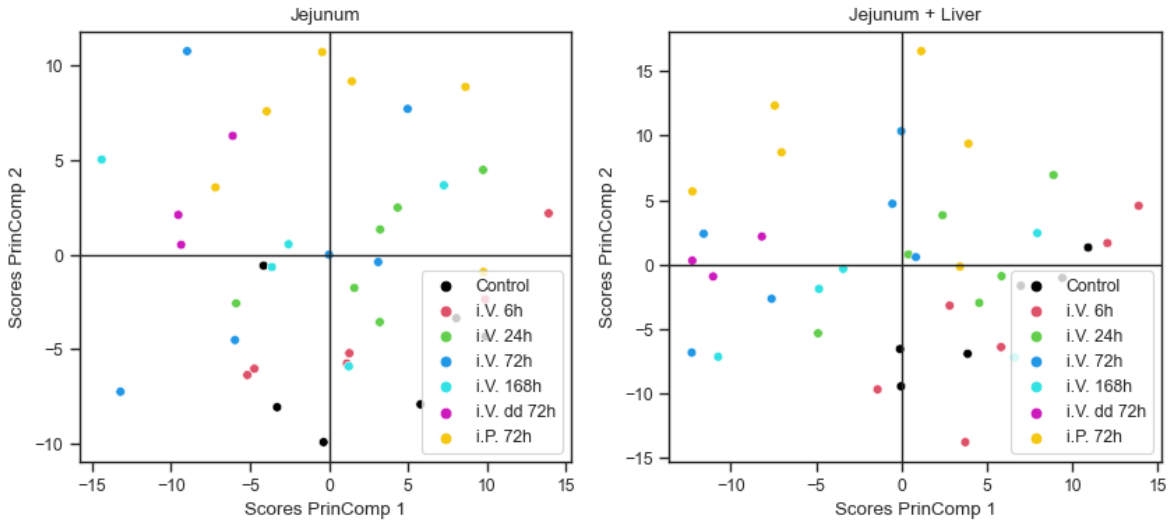
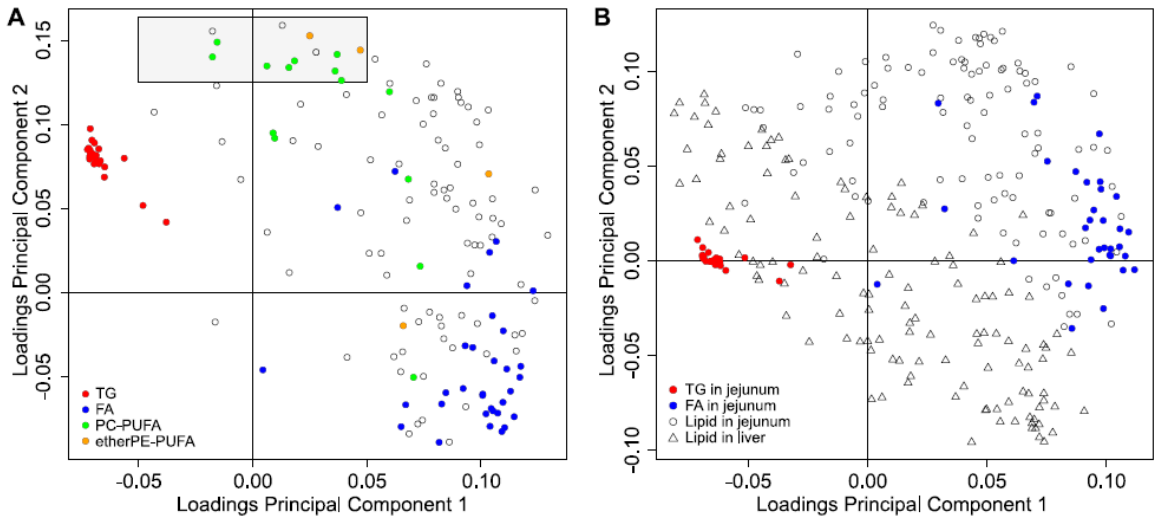


Scatterplot of the scores of the principal component analyses in Figure 2. Left: Scores of the first two principal components of the lipids identified in the jejunum (the loadings are in Figure 2, Left) Principal Component 1 represents 30% of the variance and Principal Component 2 represents 20% of the variation. Right: Scores of the first two principal component analysis of the lipids indentified in the jejunum and the liver (the loadings are in Figure 2, Right). Principal Component 1 represents 19% of the variance and Principal Component 2 represents 15% of the variation. i.V., intra venous treatment; i.P., intra peritoneal treatment.

Figure 2

Figure 2 in the article. Scatterplot of PCA of variables in the lipidome.

Original



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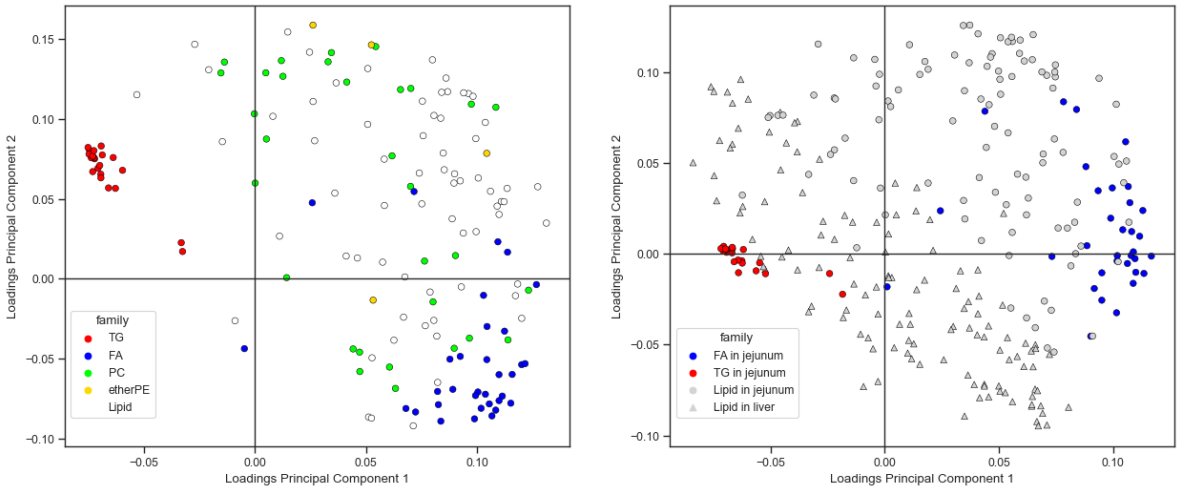
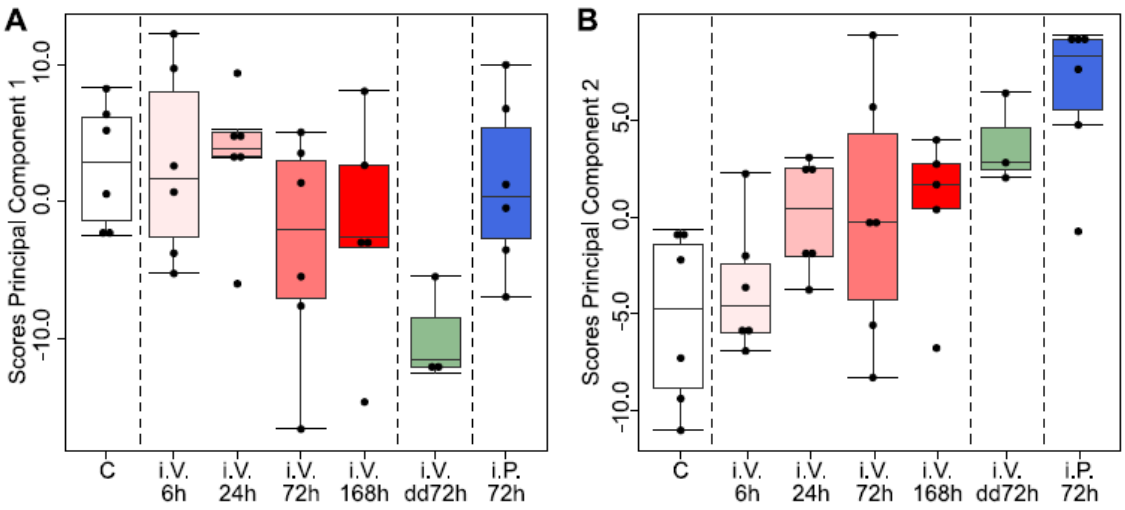


Figure 2. Loadings of the PCA of the jejunal and hepatic lipidomes. Left: Loadings plot of 1st and 2nd PCs of lipidome in jejunum. Triacylglycerols TGs in red; free fatty acids FAs in blue; phosphatidylcholines PCs in lime green; and ether phospholipids of ethanolamine in gold. Right: Loadings of the 1st and 2nd PCs of joint lipidome in jejunum (circles) and liver (triangles). Jenunal TGs in red, jej FAs in blue, everything else in light gray. Scores are in Figure 1.

Figure 3

Figure 3 in the article. Categorical plot of PCA Scores of the lipidome in the jejunum.

Original



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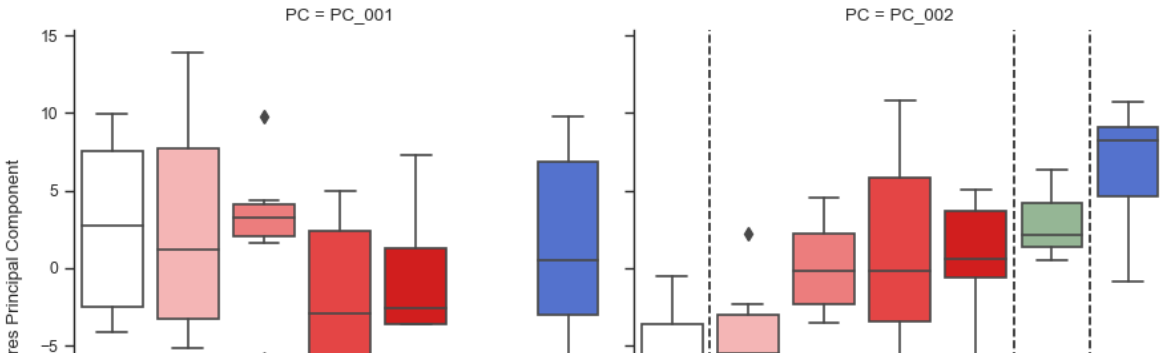


Figure 3. Scores of the PCA of lipidome in the jejunum. Left facet: Boxplots of scores of 1st PC in jejunal lipidome. Manipulation is according to treatment of animals. Right facet: Boxplots of scores of 2nd PC in jejunal lipidome; manipulation is according to treatment of animals. dd, double dose; i.V. intravenous treatment; i.P. intra-peritoneal treatment.

Results shows that PC2 better explains variation from treatment.

Data Processing and Plots

In [103]:

```
import matplotlib.pyplot as plt
import matplotlib.ticker as ticks
import numpy as np
import pandas as pd
import seaborn as sns
sns.set(context = "paper", style = "ticks")

from sklearn.decomposition import PCA
from sklearn.preprocessing import StandardScaler

## code goes here
def load_data(filepath):
    "Loads in data from filepath as Dataframe"
    return pd.read_csv(filepath, index_col = 0)

# load signals as df
signals = load_data("Signal.csv")
print(signals.head())
# load in variables df
variables = load_data("Supplementary Material_1.csv")
variables = variables.loc[variables.index.dropna()] # now removing NA-
# validate
variables
```

```
              Var_001      Var_002      Var_003      Var_004      Var_00
5  \
Sample.ID
Control    3743702.719  124938.2934  34342.77148  3237221.918  2276153.52
4
Control    4545331.531  162651.0564  40449.35386  3890073.086  2257931.67
2
Control    3142453.278  103221.2317  29182.41142  2627075.939  1540910.81
7
Control    3773137.835  104932.8221  34026.15821  2841866.205  1662993.43
8
Control    3685081.420  174503.3075  29162.54888  2856276.477  2873556.31
6

              Var_006      Var_007       Var_008      Var_009      Var_0
10  \
Sample.ID
Control    1839203.560  34907.60723   89996.94420  16844.08597  226153.19
61
Control    3007269.456  33246.50355  127398.01300  13204.65459  271822.45
23
Control    2041865.953  25659.92653   98065.68270  11812.44116  235509.96
10
Control    2293310.164  31054.23498  101527.72310  11785.19430  183577.12
88
Control    2286710.131  26944.36637   97810.56002  15314.62265  194008.13
24

           ...      Var_273      Var_274      Var_275      Var_276  \
Sample.ID  ...
Control    ...  39349.09464  6411240.422  8243266.131  3076412.342
Control    ...  29369.19504  6363726.217  5798585.701  2158111.661
Control    ...  33187.68543  5945555.542  5600472.579  2195959.362
Control    ...  69340.06999  6789465.677  7698740.799  3039580.526
Control    ...  48330.13841  6126494.691  7161421.093  2801062.139

              Var_277      Var_278      Var_279      Var_280      Var_28
1  \
Sample.ID
Control    2455682.233  468789.3121  949536.2033  3570645.673  4383111.64
5
Control    1785544.841  352497.8170  690960.0751  2488716.981  3200070.18
7
Control    1721910.928  306371.7150  717631.6318  2254738.609  2695616.74
4
Control    2313397.924  455964.3334  997880.8836  3242282.592  4038371.54
8
Control    2673249.837  466810.0367  991277.1944  3351846.491  4238467.08
1

              Var_282
Sample.ID
Control    2158538.825
Control    1619211.355
Control    1367342.733
Control    2141077.111
Control    1897258.803
```

```
[5 rows x 282 columns]
```

Out[103]:

Sample.ID	Tissue	family	lipid	Carbons	Unsaturations	RT/s	Adduct	mz	ad
Var_001	Liver	FA	FA(16:0)	16.0	0.0	248.0	[M-H]-	255.2311	
Var_002	Liver	FA	FA(16:1)	16.0	1.0	189.2	[M-H]-	253.2150	
Var_003	Liver	FA	FA(17:0)	17.0	0.0	291.6	[M-H]-	269.2463	
Var_004	Liver	FA	FA(18:0)	18.0	0.0	337.6	[M-H]-	283.2626	
Var_005	Liver	FA	FA(18:1)	18.0	1.0	264.0	[M-H]-	281.2468	
...	...	...	...	...	...	...	...	...	
Var_278	Jejunum	SM	SM(d40:2)	40.0	2.0	670.0	[M+H]+	785.6508	
Var_279	Jejunum	SM	SM(d41:1)	41.0	1.0	731.8	[M+H]+	801.6826	
Var_280	Jejunum	SM	SM(d42:1)	42.0	1.0	751.9	[M+H]+	815.6988	
Var_281	Jejunum	SM	SM(d42:2)	42.0	2.0	704.3	[M+H]+	813.6840	
Var_282	Jejunum	SM	SM(d42:3)	42.0	3.0	665.8	[M+H]+	811.6664	

282 rows × 12 columns

In [104]:

```
# Make jejunum-only values

print(variables.dtypes)

tissue_mask = variables.loc[:, "Tissue"] == "Jejunum" # keep values w
print(tissue_mask.tail())

"Making a copy of signals where the only non-zero variables are Jejunu
jejunum_signals_df = signals.T.where(tissue_mask, other = 0.0, axis =

jejunum_signals_df.T.head()
```

```
Tissue                                          object
family                                          object
lipid                                           object
Carbons                                        float64
Unsaturations                                  float64
RT/s                                           float64
Adduct                                          object
mz                                             float64
ppm m/z adduct                                 float64
MS/MS: m/z and fragment identification          object
Unnamed: 11                                    float64
Unnamed: 12                                    float64
dtype: object
Sample.ID
Var_278    True
Var_279    True
Var_280    True
Var_281    True
Var_282    True
Name: Tissue, dtype: bool
```

Out[104]:

Sample.ID	Var_001	Var_002	Var_003	Var_004	Var_005	Var_006	Var_007	Var_008	Var_0(
Control	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0
Control	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0
Control	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0
Control	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0
Control	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0

5 rows × 282 columns

In [105]:

```
# Principal Component Analysis

# init std scaler
# standardize features by recentering at zero and scaling to unit vari
signals_standardized = StandardScaler().fit_transform(signals)

# Now apply PCA to selected features
num_components = 2
signals_PCA = PCA(n_components = num_components)
#
signals_PCA_fit = signals_PCA.fit_transform(signals_standardized)
print(f"{signals_PCA.components_.shape = }")

sample_id = signals.index

signals_PCA_scores = pd.DataFrame(signals_PCA_fit[:, :2], columns = ['
print(f"signals_PCA_scores.head \n{signals_PCA_scores.head()}")


# Do over all steps but for the jejunum signals only

# standardize features
j_signals_standardized = StandardScaler().fit_transform(jejunum_signal
#
jejunum_PCA = PCA(n_components = num_components)
jejunum_PCA_fit = jejunum_PCA.fit_transform(j_signals_standardized)
jejunum_PCA_fit[..., 0] = - jejunum_PCA_fit[..., 0] # reverse eigenvec
jejunum_PCA_scores = pd.DataFrame(jejunum_PCA_fit, columns = ['PC_001'
print(f"jejunum_PCA_scores.head \n{jejunum_PCA_scores.head()}")
```

```
signals_PCA.components_.shape = (2, 282)
signals_PCA_scores.head
             PC_001    PC_002
Sample.ID
Control    9.413856 -1.022951
Control   -0.028738 -9.427465
Control    3.870471 -6.907008
Control    6.982983 -1.638149
Control   -0.087840 -6.539702
jejunum_PCA_scores.head
             PC_001    PC_002
Sample.ID
Control    8.068391 -3.348617
Control   -3.282909 -8.059449
Control   -0.336738 -9.910995
Control    5.803622 -7.912033
Control   -4.138326 -0.571479
```

In [125]:

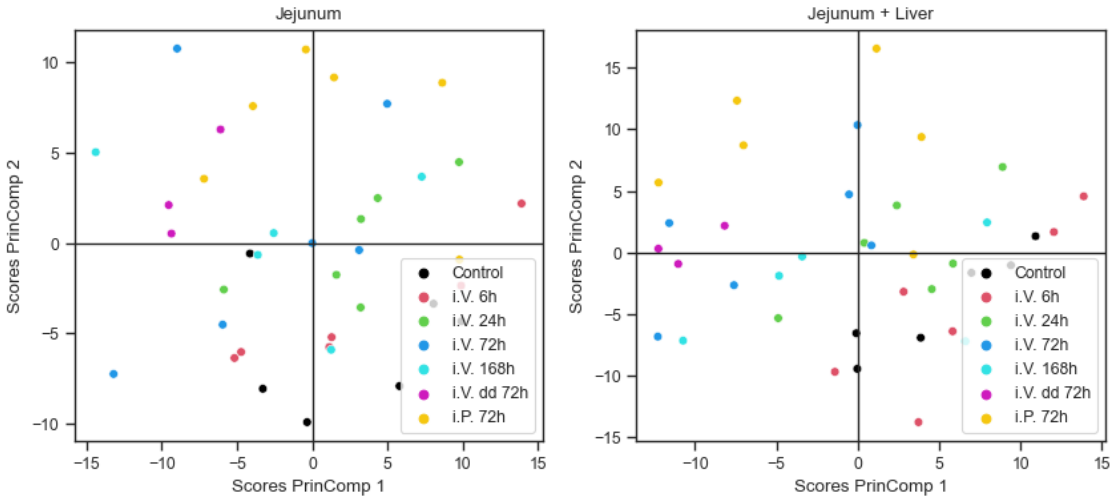
```
# Plotting PCA Scores
pc_one = signals_PCA_scores.loc[:, "PC_001"]
pc_two = signals_PCA_scores.loc[:, "PC_002"]
sample_id = signals.index.array
# print(sample_id)

# init figure
fig, axs = plt.subplots(1, 2, figsize = (10, 4))

with sns.axes_style("ticks", rc = {'axis.grid': False}): # temporarily
    with sns.color_palette(["#000000", "#DE5168", "#61CF4C", "#2197E7"
        sns.scatterplot(data = jejunum_PCA_scores, x = "PC_001", y = "
        sns.scatterplot(data = signals_PCA_scores, x = "PC_001", y = "
sns.move_legend(axs[0], "lower right")
axs[0].set_title("Jejunum")
axs[0].set_xlabel("Scores PrinComp 1")
axs[0].set_ylabel("Scores PrinComp 2")
axs[0].axhline(0, c = "#262626", linewidth = 1)
axs[0].axvline(0, c = "#262626", linewidth = 1)
sns.move_legend(axs[1], "lower right")
axs[1].set_title("Jejunum + Liver")
axs[1].set_xlabel("Scores PrinComp 1")
axs[1].set_ylabel("Scores PrinComp 2")
axs[1].axhline(0, c = "#262626", linewidth = 1)
axs[1].axvline(0, c = "#262626", linewidth = 1)

j_v_explained = jejunum_PCA.explained_variance_ratio_
s_v_explained = signals_PCA.explained_variance_ratio_
var_explnd = f"Explained Variance: Jejunum PC1 {j_v_explained[0]:.2f};
Joint Liver + Jejunum PC1 {s_v_explained[0]:.2f}; Joint Liver + Jejunu
print(var_explnd)
```

```
Explained Variance: Jejunum PC1 0.30; Jejunum PC2 0.20; Joint Liver + Jej
unum PC1 0.19; Joint Liver + Jejunum PC2 0.15
```



In [107]:

```
# More pca loadings stuff

loadings = signals_PCA.components_.T # Transpose to have loadings be i
# create df from loadings
loadings_df = pd.DataFrame(data=loadings, columns=['PC1_loadings', 'PC
loadings_df.head()

# combining pca loadings with variables df
loadings_variables_merged = pd.concat((loadings_df, variables), axis =
print(loadings_variables_merged.dtypes)

# mask out Lipid families not belonging to FAs or TGs
lipid_mask = loadings_variables_merged.family.isin(("TG", "FA"))
tissue_mask = loadings_variables_merged.Tissue.eq("Jejunum") # keep va

loadings_variables_merged["family"] =  np.where(lipid_mask, loadings_v
loadings_variables_merged["family"] =  np.where(tissue_mask,
                                                loadings_variables_mer
                                                "Lipid" + " in liver")

print((loadings_variables_merged["family"]).unique())
loadings_variables_merged
```

```
PC1_loadings                                float64
PC2_loadings                                float64
Tissue                                       object
family                                       object
lipid                                        object
Carbons                                     float64
Unsaturations                               float64
RT/s                                        float64
Adduct                                       object
mz                                          float64
ppm m/z adduct                              float64
MS/MS: m/z and fragment identification       object
Unnamed: 11                                 float64
Unnamed: 12                                 float64
dtype: object
['Lipid in liver' 'FA in jejunum' 'Lipid in jejunum' 'TG in jejunum']
```

Out[107]:

	PC1_loadings	PC2_loadings	Tissue	family	lipid	Carbons	Unsaturations
Var_001	-0.005098	-0.052275	Liver	Lipid in liver	FA(16:0)	16.0	0.0
Var_002	0.043666	-0.071817	Liver	Lipid in liver	FA(16:1)	16.0	1.0
Var_003	-0.009278	-0.073087	Liver	Lipid in liver	FA(17:0)	17.0	0.0
Var_004	-0.045011	-0.001792	Liver	Lipid in liver	FA(18:0)	18.0	0.0
Var_005	0.039243	-0.071956	Liver	Lipid in liver	FA(18:1)	18.0	1.0
...	...	...	...	...	...	...	...
Var_278	0.046389	0.108161	Jejunum	Lipid in jejunum	SM(d40:2)	40.0	2.0
Var_279	0.044894	0.103075	Jejunum	Lipid in jejunum	SM(d41:1)	41.0	1.0
Var_280	0.040907	0.120961	Jejunum	Lipid in jejunum	SM(d42:1)	42.0	1.0
Var_281	0.050726	0.118803	Jejunum	Lipid in jejunum	SM(d42:2)	42.0	2.0
Var_282	0.059375	0.117524	Jejunum	Lipid in jejunum	SM(d42:3)	42.0	3.0

282 rows × 14 columns

In [116]:

```
# Repeat loadings df steps for jejunum

# Transpose
jejunum_loadings = jejunum_PCA.components_.T

# create df from loadings
jejunum_loadings_df = pd.DataFrame(data=jejunum_loadings, columns=['PC
jejunum_loadings_df["PC1_loadings"] = - jejunum_loadings_df.PC1_loadir

#  keep values w Tissue=="Jejunum"
tissue_mask = variables.Tissue == "Jejunum"
jejunum_variables = variables.where(tissue_mask, axis = 0)

# Generate merged jejunum loadings & variables
jejunum_loadings_variables_merged = pd.concat((jejunum_loadings_df, je
jejunum_loadings_variables_merged = jejunum_loadings_variables_merged[

# ~mask~ rename values in 'family'
lipid_mask = jejunum_loadings_variables_merged.family.isin(["TG", "FA"
jejunum_loadings_variables_merged.family = np.where(lipid_mask,
                                                    jejunum_loadings_variables
print(jejunum_loadings_variables_merged.family.unique())

jejunum_loadings_variables_merged
```

```
['FA' 'Lipid' 'PC' 'etherPE' 'TG']
```

Out[116]:

	PC1_loadings	PC2_loadings	Tissue	family	lipid	Carbons	Unsaturations
Var_125	0.082502	-0.078743	Jejunum	FA	FA(16:1)	16.0	1.0
Var_126	0.067909	-0.081143	Jejunum	FA	FA(16:1)	16.0	1.0
Var_127	0.089006	-0.069148	Jejunum	FA	FA(17:0)	17.0	0.0
Var_128	0.119961	-0.053704	Jejunum	FA	FA(18:0)	18.0	0.0
Var_129	0.087701	-0.050282	Jejunum	FA	FA(18:1)	18.0	1.0
...	...	...	...	...	...	...	...
Var_278	0.083989	0.116764	Jejunum	Lipid	SM(d40:2)	40.0	2.0
Var_279	0.083558	0.100558	Jejunum	Lipid	SM(d41:1)	41.0	1.0
Var_280	0.085964	0.125693	Jejunum	Lipid	SM(d42:1)	42.0	1.0
Var_281	0.093873	0.116447	Jejunum	Lipid	SM(d42:2)	42.0	2.0
Var_282	0.103641	0.097914	Jejunum	Lipid	SM(d42:3)	42.0	3.0

158 rows × 14 columns

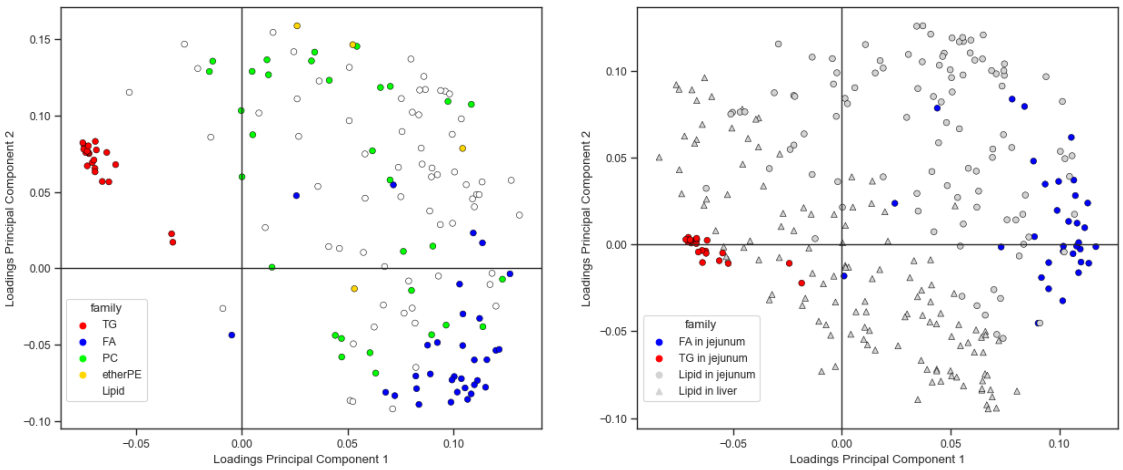
In [117]:

```
from matplotlib.text import Text

# Plot Loadings of Principal Components
labels = ["FA in jejunum", "TG in jejunum", "Lipid in jejunum", "Lipic
# init figure
fig2, loadings_axs = plt.subplots(1, 2, figsize = (15, 6))

with sns.axes_style("ticks"):
    with sns.color_palette(["blue", "red", "lightgray", "lightgray"]):
        # plot jejunum lipidome
        sns.scatterplot(jejunum_loadings_variables_merged, x = "PC1_lo
                hue = "family", hue_order = ("TG", "FA", "PC", "etherP
                legend = 'auto', edgecolor = "black", palette = ["red"
                ax = loadings_axs[0])
        # plot lipidome in jejunum + liver
        sns.scatterplot(loadings_variables_merged, x = "PC1_loadings",
                hue = "family", hue_order = labels, edgecolor = ["blac
                style = "family", markers = ["^", "o", "o", "o"], lege
                ax = loadings_axs[1])

sns.move_legend(loadings_axs[0], "lower left", bbox_to_anchor = (0.0,
loadings_axs[0].axhline(0, color = "#262626", linewidth = 1)
loadings_axs[0].axvline(0, color = "#262626", linewidth = 1)
loadings_axs[0].set_xlabel("Loadings Principal Component 1")
loadings_axs[0].set_ylabel("Loadings Principal Component 2")
sns.move_legend(loadings_axs[1], "lower left", bbox_to_anchor = (0.0,
loadings_axs[1].xaxis.set_major_locator(ticks.MultipleLocator(0.050))
loadings_axs[1].axhline(0, color = "#262626", linestyle = '-', linewic
loadings_axs[1].axvline(0, color = "#262626", linestyle = '-', linewic
loadings_axs[1].set_xlabel("Loadings Principal Component 1")
loadings_axs[1].set_ylabel("Loadings Principal Component 2")
# Text(x = -0.075, y = 0.15, text = 'B')
plt.show()
```



In [110]:

```
# Prepare long form of signals_PCA_scores for the categorical plot

jejunum_long_PCA = jejunum_PCA_scores.melt(var_name = "PC",
                                           value_name = "Scores Princ
                                           ignore_index = False)
sample_id = jejunum_long_PCA.index.to_frame() # creates new dataframe
jejunum_long_PCA = jejunum_long_PCA.assign(Sample_Id = sample_id)

jejunum_long_PCA.head()
```

Out[110]:

Sample.ID	PC	Scores Principal Component	Sample_Id
Control	PC_001	8.068391	Control
Control	PC_001	-3.282909	Control
Control	PC_001	-0.336738	Control
Control	PC_001	5.803622	Control
Control	PC_001	-4.138326	Control

In [127]:

```
# Plot box of Jejunum Scores

with sns.color_palette(palette = ["#FFF", "#FFAAAA", "#FF6A6A", "#FF2A
    box = sns.catplot(data = jejunum_long_PCA, kind = "box", col = "PC
                      aspect = 1, height = 4.5)
    plt.ylabel("Scores Principal Component")
    plt.axvline(0.5, c = "#222", linestyle = "--")
    plt.axvline(4.5, c = "#222", linestyle = "--")
    plt.axvline(5.5, c = "#222", linestyle = "--")
```

```
c:\Users\truma\anaconda3\Lib\site-packages\seaborn\axisgrid.py:118: UserW
arning: The figure layout has changed to tight
  self._figure.tight_layout(*args, **kwargs)
```