

# ELF\_Cytokine\_Analysis PCA, Apr 23, 2024. R (v.4.3.2)

#

# ELF\_Cytokine Principal Component Analysis  
# Apr 23, 2024 R (v.4.3.2) PCA by Teiji Sawa, MD, PhD  
# Kyoto Prefectural University of Medicine, Japan  
# For the analyses of the following publication:  
# Authors: Sazuki Sudo, others & Teiji Sawa.  
# Title: Case study observational research: inflammatory  
# cytokines in the bronchial epithelial lining fluid of COVID-19  
# patients with acute hypoxemic respiratory failure.  
# Journal: Critical Care volume 28: 134, 2024.  
# DOI: doi:10.1186/s13054-024-04921-3

#

#-[R Code #1]

```
install.packages("FactoMineR")
install.packages("factoextra")
library(ggplot2)
library(GGally)
library(FactoMineR)
library(factoextra)
library(tidyverse)
```

#-[END]

#-[R Code #2]

```
data1 <- read_csv("pMOL_ELF4_r25_p27_20240301_PCA2.csv", show_col_types = FALSE)
data2 <- read_csv("pMOL_Plasma4_r25_p27_20240301_PCA2.csv", show_col_types = FALSE)
```

#-[END]

#-[R Code #3]

```
pca_d1 <- PCA(data1[,9:29], graph = FALSE)
pca_d2 <- PCA(data2[,9:22], graph = FALSE)
```

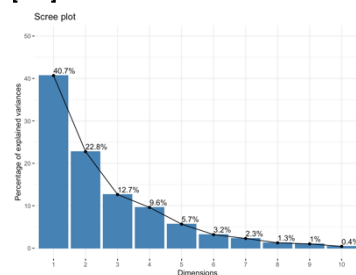
#-[END]

#-[R Code #4]

```
fviz_screplot(pca_d1, addlabels = TRUE, ylim = c(0, 50))
```

#-[END]

[out]

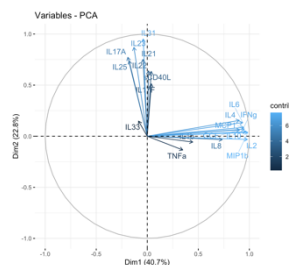


#-[R Code #5]

```
fviz_pca_var(pca_d1, axes = c(1, 2), col.var = "contrib", repel = TRUE)
```

#-[END]

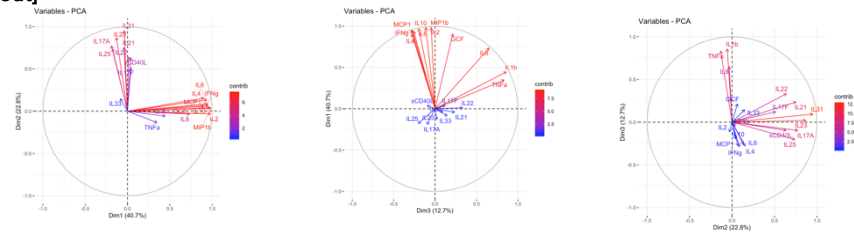
[out]



#-[R Code #6]

```
fviz_pca_var(pca_d1, axes = c(1, 2), col.var = "contrib", repel = TRUE) + scale_color_gradient2(low = "white", mid = "blue", high = "red", midpoint = 0.6)
fviz_pca_var(pca_d1, axes = c(2, 3), col.var = "contrib", repel = TRUE) + scale_color_gradient2(low = "white", mid = "blue", high = "red", midpoint = 0.6)
fviz_pca_var(pca_d1, axes = c(3, 1), col.var = "contrib", repel = TRUE) + scale_color_gradient2(low = "white", mid = "blue", high = "red", midpoint = 0.6)
```

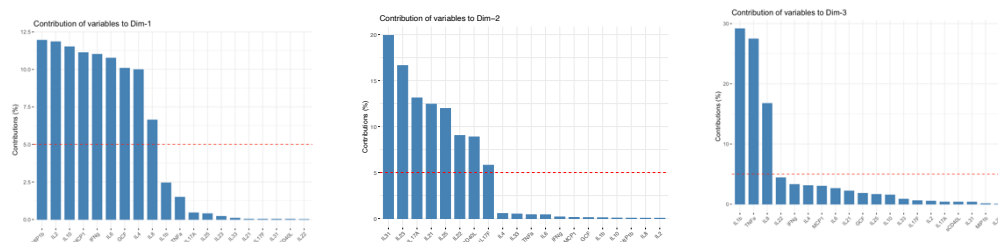
```
#[END]-----
[out]
```



```
#[R Code #7]-----
```

```
fviz_contrib(pca_d1, choice = "var", axes = 1)
fviz_contrib(pca_d1, choice = "var", axes = 2)
fviz_contrib(pca_d1, choice = "var", axes = 3)
```

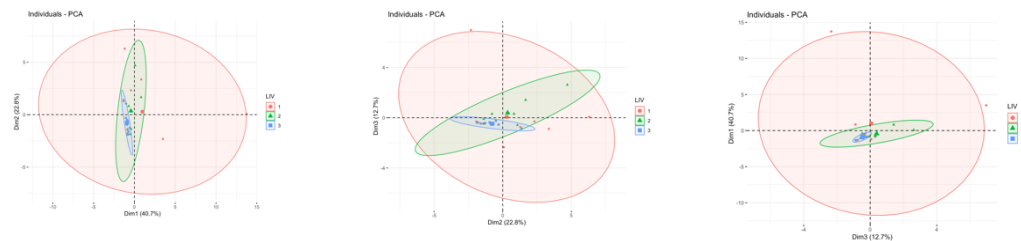
```
#[END]-----
[out]
```



```
#[R Code #8]-----
```

```
fviz_pca_ind(pca_d1, geom.ind="point", axes= c(1,2), habillage =as.factor(data1$liv_group), legend.title = "LIV",
repel=TRUE, addEllipses =TRUE)
fviz_pca_ind(pca_d1, geom.ind="point", axes= c(2,3), habillage =as.factor(data1$liv_group), legend.title = "LIV",
repel=TRUE, addEllipses =TRUE)
fviz_pca_ind(pca_d1, geom.ind="point", axes= c(3,1), habillage =as.factor(data1$liv_group), legend.title = "LIV",
repel=TRUE, addEllipses =TRUE)
```

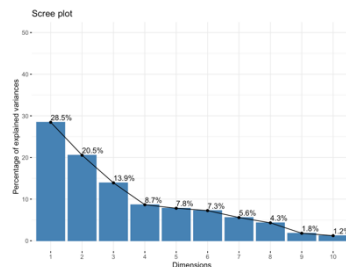
```
#[END]-----
[out]
```



```
#[R Code #9]-----
```

```
fviz_screplot(pca_d2, addlabels =TRUE, ylim =c(0, 50))
```

```
#[END]-----
```



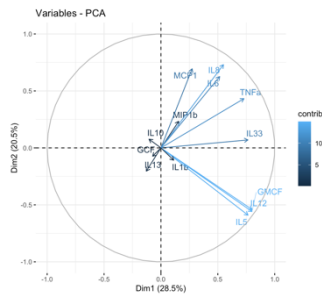
```
[out]
```

```
#[R Code #10]-----
```

```
fviz_pca_var(pca_d2, axes= c(1,2), col.var = "contrib", repel =TRUE)
```

```
#[END]-----
```

[out]

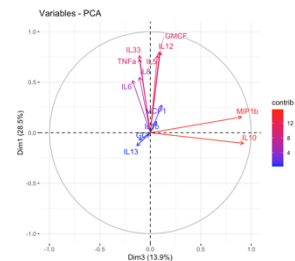
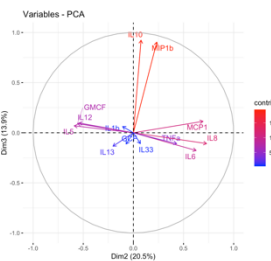
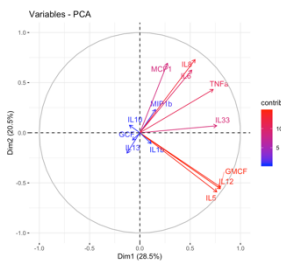


#[R Code #11]

```
fviz_pca_var(pca_d2, axes= c(1,2), col.var = "contrib", repel = TRUE) + scale_color_gradient2(low="white", mid="blue",
high="red", midpoint=0.6)
fviz_pca_var(pca_d2, axes= c(2,3), col.var = "contrib", repel = TRUE) + scale_color_gradient2(low="white", mid="blue",
high="red", midpoint=0.6)
fviz_pca_var(pca_d2, axes= c(3,1), col.var = "contrib", repel = TRUE) + scale_color_gradient2(low="white", mid="blue",
high="red", midpoint=0.6)
```

#[END]

[out]

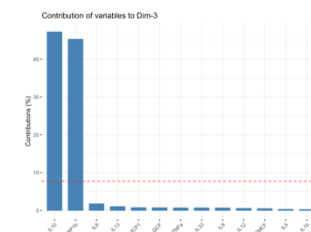
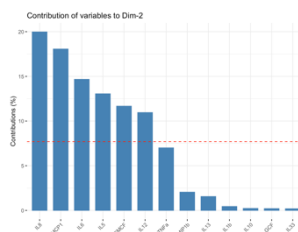
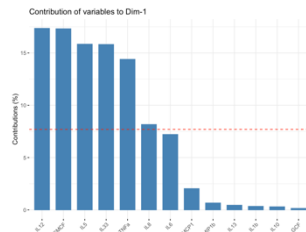


#[R Code #12]

```
fviz_contrib(pca_d2, choice = "var", axes = 1)
fviz_contrib(pca_d2, choice = "var", axes = 2)
fviz_contrib(pca_d2, choice = "var", axes = 3)
```

#[END]

[out]

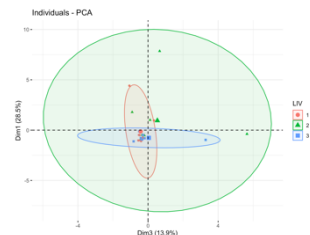
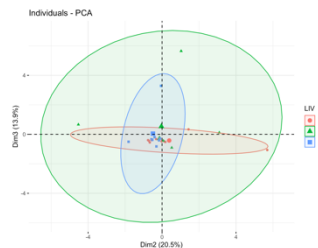
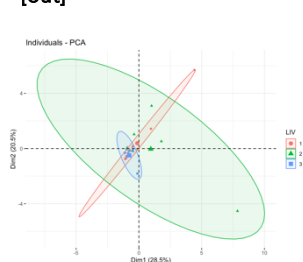


#[R Code #13]

```
fviz_pca_ind(pca_d2, geom.ind="point", axes= c(1,2), habillage =as.factor(data1$liv_group), legend.title = "LIV",
repel=TRUE, addEllipses=TRUE)
fviz_pca_ind(pca_d2, geom.ind="point", axes= c(2,3), habillage =as.factor(data1$liv_group), legend.title = "LIV",
repel=TRUE, addEllipses=TRUE)
fviz_pca_ind(pca_d2, geom.ind="point", axes= c(3,1), habillage =as.factor(data1$liv_group), legend.title = "LIV",
repel=TRUE, addEllipses=TRUE)
```

#[END]

[out]



```

#-[R Code #14]-----
library(corrplot)
corrplot 0.92 loaded
#-[END]-----

```

```

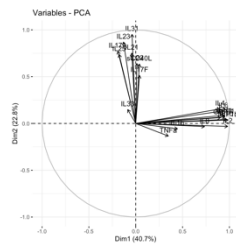
#-[R Code #15]-----
var1 <- get_pca_var(pca_d1)
#-[END]-----

```

```

#-[R Code #16]-----
pca_d1 %>% fviz_pca_var(col.var1 = "black")
#-[END]-----
[out]

```



```

#-[R Code #17]-----
var1$cos2 %>% round(4)
#-[END]-----
[out]

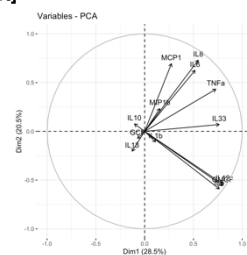
```

	Dim.1	Dim.2	Dim.3	Dim.4	Dim.5
IL6	0.8750	0.0179	0.0661	0.0051	0.0056
IFNg	0.8956	0.0075	0.0829	0.0030	0.0001
MCP1	0.9046	0.0049	0.0759	0.0017	0.0022
MIP1b	0.9718	0.0014	0.0022	0.0014	0.0160
IL10	0.9362	0.0019	0.0388	0.0025	0.0150
IL2	0.9634	0.0010	0.0129	0.0003	0.0138
IL4	0.8124	0.0240	0.0783	0.0001	0.0448
GCF	0.8201	0.0039	0.0458	0.0048	0.0001
IL8	0.5395	0.0011	0.4235	0.0185	0.0035
IL1b	0.1986	0.0033	0.7375	0.0483	0.0035
TNFa	0.1210	0.0181	0.6948	0.1233	0.0061
IL31	0.0016	0.9055	0.0089	0.0072	0.0380
IL17A	0.0362	0.5964	0.0092	0.1284	0.0575
IL23	0.0171	0.7562	0.0004	0.0307	0.1268
IL25	0.0315	0.5440	0.0412	0.2675	0.0037
sCD40L	0.0015	0.4033	0.0091	0.1935	0.1780
IL21	0.0018	0.5657	0.0556	0.1136	0.1781
IL22	0.0001	0.4101	0.1111	0.3238	0.0056
IL17F	0.0016	0.2636	0.0148	0.1539	0.4360
IL33	0.0073	0.0220	0.0213	0.4929	0.0001

```

#-[R Code #18]-----
var1$cos2 %>% corrplot(is.corr = FALSE)
#-[END]-----
[out]

```



```

#-[R Code #19]-----
var2 <- get_pca_var(pca_d2)
#-[END]-----

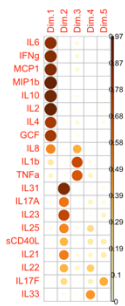
```

```

#-[R Code #20]-----
pca_d2 %>% fviz_pca_var(col.var2 = "black")
#-[END]-----

```

[out]



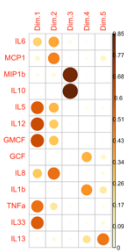
```
#[R Code #21]-----
var2$cos2 %>% round(4)
#[END]-----
```

[out]

	Dim.1	Dim.2	Dim.3	Dim.4	Dim.5
IL6	0.2667	0.3918	0.0313	0.0090	0.0021
MCP1	0.0753	0.4825	0.0130	0.0241	0.1022
MIP1b	0.0244	0.0545	0.8181	0.0060	0.0012
IL10	0.0110	0.0058	0.8529	0.0001	0.0012
IL5	0.5859	0.3485	0.0048	0.0231	0.0173
IL12	0.6420	0.2926	0.0096	0.0080	0.0055
GMCF	0.6405	0.3116	0.0084	0.0013	0.0081
GCF	0.0053	0.0053	0.0125	0.3630	0.0791
IL8	0.3020	0.5335	0.0116	0.0112	0.0341
IL1b	0.0126	0.0116	0.0039	0.4438	0.1748
TNFα	0.5324	0.1869	0.0119	0.0027	0.0258
IL33	0.5850	0.0049	0.0118	0.0211	0.0501
IL13	0.0165	0.0415	0.0180	0.2116	0.5142

```
#[R Code #22]-----
var2$cos2 %>% corplot(is.corr = FALSE)
#[END]-----
```

[out]



## ELF\_Cytokine\_統計解析 PCA, Apr 23, 2024 Python 3.18

```
#[Python Code #1]-----
from sklearn.preprocessing import StandardScaler
import pandas as pd
import numpy as np
import matplotlib.pyplot as plt
from matplotlib.colors import ListedColormap
#pip install pca
#[END]-----

#[Python Code #2]-----
data_elf_all = pd.read_csv('Data5_ELF_PCA_20240426_selected.csv')
data_plasma_all = pd.read_csv('Data6_Plasma_PCA_20240426_selected.csv')
#[END]-----

#[Python Code #3]-----
data_all_elf = data_all[data_all.source=='elf']
data_all_plasma = data_all[data_all.source=='plasma']

data_elf = data_all_elf.iloc[:,3:23]
data_plasma = data_all_plasma.iloc[:,3:16]
data_LIV = data_all.iloc[0:27,2]
#[END]-----

#[Python Code #4]-----
elf = data_elf.to_numpy()
plasma = data_plasma.to_numpy()
LIV_group = data_LIV.to_numpy()
#[END]-----

#[Python Code #5]-----
scaler1 = StandardScaler(with_mean=True, with_std=True)
scaler2 = StandardScaler(with_mean=True, with_std=True)
elf_std = scaler1.fit_transform(elf)
plasma_std = scaler2.fit_transform(plasma)
#[END]-----

#[Python Code #6]-----
Cytokines_elf= ['IL-6','IFN-g','MCP-1','MIP-1b','IL-10','IL-2','IL-5','IL-12p70','IL-7',\
               'GM-CSF','IL-4','G-CSF','IL-8','IL-1b','TNF-a','IL-31','IL-17A',\
               'IL-23','IL-25','sCD40L','IL-21','IL-22','IL-17F','IL-33','IL-13']

cytokines_plasma= ['IL-6','MCP-1','MIP-1b','IL-10',\
                  'IL-5','IL-12p70','GM-CSF','G-CSF','IL-8','IL-1b','TNF-a',\
                  'IL-33','IL-13']
#[END]-----

#[Python Code #7]-----
from pca import pca
# Initialize
model = pca(normalize = True)
#[END]-----

#[Python Code #8]-----
from pca import pca
# Initialize
model2 = pca(normalize = True)
#[END]-----

#[Python Code #9]-----
# Fit transform and include the column labels and row labels
results_elf = model.fit_transform(elf_std, col_labels=cytokines_elf, \
                                row_labels=LIV_group)
#[END]-----

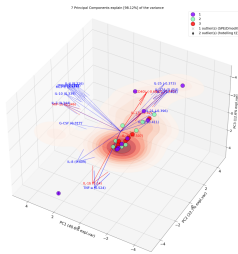
#[Python Code #10]-----
# Fit transform and include the column labels and row labels
results_plasma = model2.fit_transform(plasma_std, col_labels=cytokines_plasma, \
                                    row_labels=LIV_group)
#[END]-----

#[Python Code #11]-----
fig, ax = model.plot()
fig, ax = model.biplot3d(SPE=True,
                        HT2=True, arrowdict={'scale_factor': 3, \
                        'color_strong': 'red', 'color_weak': 'blue'}, s=500,
                        cmap="rainbow", density= True)

ax.set_xlim(-5, 5)
ax.set_ylim(-5, 5)
ax.set_zlim(-5, 5)
```

```
ax.legend(loc=1)
plt.savefig('Fig1_3D_ELF.svg')
plt.savefig('Fig1_3D_ELF.png')
plt.show()
#-[END]-----
```

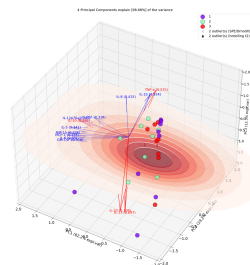
[out]



```
#-[Python Code #12]-----
fig, ax = model2.biplot3d(SPE=True,
                          HT2=True, arrowdict={'scale_factor': 3, \
                          'color_strong': 'red', 'color_weak': 'blue'}, s=500,
                          cmap="rainbow", density=True)

ax.set_xlim(-5, 5)
ax.set_ylim(-5, 5)
ax.set_zlim(-5, 5)
ax.legend(loc=1)
plt.savefig('Fig2_3D_plasma.svg')
plt.savefig('Fig2_3D_plasma.png')
plt.show()
#-[END]-----
```

[Out]



```
#-[Python Code #13]-----
# All available markers
markers = np.array(['o:blue', 'o:orange', 'o:red'])
# Generate random integers
random_integers = np.random.randint(0, len(markers), size=ELF_std.shape[0])
# Draw markers
marker = markers
# Set alpha
alpha = np.random.rand(1, ELF_std.shape[0])[0][random_integers]

fig, ax = model.biplot(PC=[0, 1],
                       SPE=True,
                       HT2=True,
                       s=400,
                       cmap="rainbow",
                       marker=marker,
                       n_feat=27,
                       fontsize=16,
                       fontweight='normal',
                       arrowdict={'fontsize': 6, 'scale_factor': 3, \
                       'color_strong': 'r', 'color_weak': 'b'},
                       density=True,
                       density_on_top=False,
```

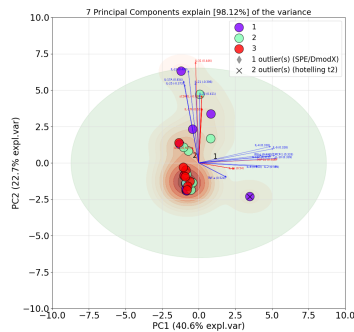
```

figsize= (12,12)
)
ax.set_xlim(-8, 8)
ax.set_ylim(-8, 8)

ax.legend(loc=1)
plt.savefig('Fig3_ELF_DIM1&2.svg')
plt.savefig('Fig3_ELF_DIM1&2.png')
plt.show()
#-[END]-----

```

[Out]



```

#-[Python Code #14]-----
# All available markers
# Generate random integers
random_integers = np.random.randint(0, len(markers), size=ELF_std.shape[0])
# Draw markers
marker = markers
# Set alpha
alpha = np.random.rand(1, ELF_std.shape[0])[0][random_integers]

fig, ax = model.biplot(PC= [1, 2],
                       SPE= True,
                       HT2= True,
                       s=400,
                       cmap="rainbow",
                       marker=marker,
                       n_feat=27,
                       fontsize=16,
                       fontweight='normal',
                       arrowdict={'fontsize': 6, 'scale_factor': 3, \
                                  'color_strong': 'r', 'color_weak': 'b'},
                       density= True,
                       density_on_top=False,
                       figsize= (12,12)
                       )

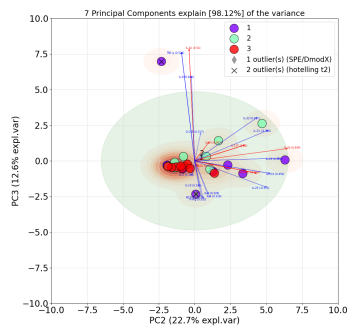
ax.set_xlim(-8, 8)
ax.set_ylim(-8, 8)

ax.legend(loc=1)
plt.savefig('Fig4_ELF_DIM2&3.svg')
plt.savefig('Fig4_ELF_DIM2&3.png')
plt.show()
#-[END]-----

```



[Out]



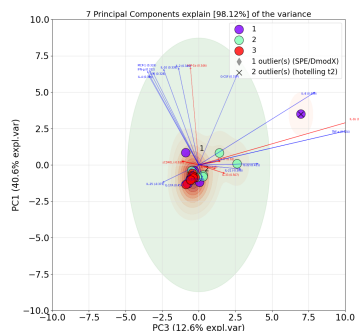
```
#-[Python Code #15]-----
# All available markers
# Generate random integers
random_integers = np.random.randint(0, len(markers), size=ELF_std.shape[0])
# Draw markers
marker = markers
# Set alpha
alpha = np.random.rand(1, ELF_std.shape[0])[0][random_integers]

fig, ax = model.biplot(PC=[2, 0],
                       SPE=True,
                       HT2=True,
                       s=400,
                       cmap="rainbow",
                       marker=marker,
                       n_feat=27,
                       fontsize=16,
                       fontweight='normal',
                       arrowdict={'fontsize': 6, 'scale_factor': 3, \
                                  'color_strong': 'r', 'color_weak': 'b'},
                       density=True,
                       density_on_top=False,
                       figsize=(12,12)
                      )

ax.set_xlim(-8, 8)
ax.set_ylim(-8, 8)

ax.legend(loc=1)
plt.savefig('Fig5_ELF_DIM3&1.svg')
plt.savefig('Fig5_ELF_DIM3&1.png')
plt.show()
#-[END]-----
```

[Out]



```
#-[Python Code #16]-----
# All available markers
markers = np.array(['o:blue', 'o:orange', 'o:red'])
# Generate random integers
random_integers = np.random.randint(0, len(markers), size=plasma_std.shape[0])
# Draw markers
marker = markers
# Set alpha
```

```

alpha = np.random.rand(1, plasma_std.shape[0])[0][random_integers]

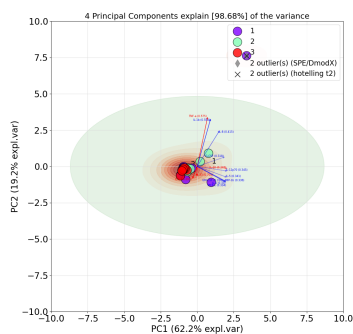
fig, ax = model2.biplot(PC=[0, 1],
                        SPE=True,
                        HT2=True,
                        s=400,
                        cmap="rainbow",
                        marker=marker,
                        n_feat=27,
                        fontsize=16,
                        fontweight='normal',
                        arrowdict={'fontsize': 6, 'scale_factor': 1, \
                                'color_strong': 'r', 'color_weak': 'b'},
                        density=True,
                        density_on_top=False,
                        figsize=(12,12)
                        )

ax.set_xlim(-8, 8)
ax.set_ylim(-8, 8)

ax.legend(loc=1)
plt.savefig('Fig6_plasma_DIM1&2.svg')
plt.savefig('Fig6_plasma_DIM1&2.png')
plt.show()
#-[END]-----

```

[Out]



```

#-[Python Code #17]-----
# All available markers
markers = np.array(['o:blue', 'o:orange', 'o:red'])
# Generate random integers
random_integers = np.random.randint(0, len(markers), size=plasma_std.shape[0])
# Draw markers
marker = markers
# Set alpha
alpha = np.random.rand(1, plasma_std.shape[0])[0][random_integers]

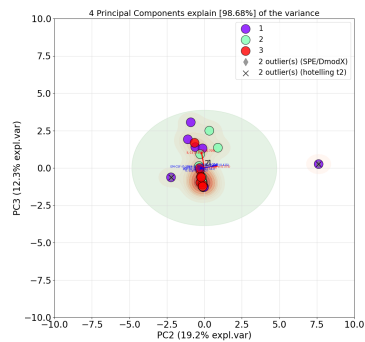
fig, ax = model2.biplot(PC=[1, 2],
                        SPE=True,
                        HT2=True,
                        s=400,
                        cmap="rainbow",
                        marker=marker,
                        n_feat=27,
                        fontsize=16,
                        fontweight='normal',
                        arrowdict={'fontsize': 6, 'scale_factor': 1, \
                                'color_strong': 'r', 'color_weak': 'b'},
                        density=True,
                        density_on_top=False,
                        figsize=(12,12)
                        )

ax.set_xlim(-8, 8)
ax.set_ylim(-8, 8)

ax.legend(loc=1)
plt.savefig('Fig7_plasma_DIM2&3.svg')
plt.savefig('Fig7_plasma_DIM2&3.png')
plt.show()
#-[END]-----

```

[Out]



```
#-[Python Code #18]-----
# All available markers
markers = np.array(['o:blue', 'o:orange', 'o:red'])
# Generate random integers
random_integers = np.random.randint(0, len(markers), size=plasma_std.shape[0])
# Draw markers
marker = markers
# Set alpha
alpha = np.random.rand(1, plasma_std.shape[0])[0][random_integers]

fig, ax = model2.biplot(PC= [2, 0],
                        SPE= True,
                        HT2= True,
                        s=400,
                        cmap="rainbow",
                        marker=marker,
                        n_feat=27,
                        fontsize=16,
                        fontweight='normal',
                        arrowdict={'fontsize': 6, 'scale_factor': 1, \
                                  'color_strong': 'r', 'color_weak': 'b'},
                        density= True,
                        density_on_top=False,
                        figsize= (12,12)
                        )

ax.set_xlim(-8, 8)
ax.set_ylim(-8, 8)

ax.legend(loc=1)
plt.savefig('Fig8_plasma_DIM3&1.svg')
plt.savefig('Fig8_plasma_DIM3&1.png')
plt.show()
#-[END]-----
```

[Out]

