# main

July 10, 2021

# 1 Calculate RXE for public dataset, females only based on Apua's script

This has been edited from Apua's script.

```
[1]: import re
     import functools
     import numpy as np
     import pandas as pd
     from os import environ
     from plotnine import *
     from gtfparse import read_gtf
     from warnings import filterwarnings
     from scipy.stats import mannwhitneyu
     from matplotlib.cbook import mplDeprecation
[2]: filterwarnings("ignore", category=mplDeprecation)
     filterwarnings('ignore', category=UserWarning, module='plotnine.*')
     filterwarnings('ignore', category=DeprecationWarning, module='plotnine.*')
[3]: environ['NUMEXPR_MAX_THREADS'] = '16'
[4]: config = {
         'pheno_file': '/ceph/users/jbenja13/projects/sex_sz_ria/input/phenotypes/_m/
      ⇔phenotypes.csv',
         'insert_size': '/ceph/users/jbenja13/projects/sex_sz_ria/input/gene_tpm/_m/
      →picard_insert_size_metrics.csv',
         'gtf': '/ceph/genome/human/gencode26/gtf.ALL/_m/gencode.v26.
     →chr_patch_hapl_scaff.annotation.gtf',
[5]: @functools.lru_cache()
     def get_pheno():
         return pd.read_csv(config['pheno_file'], index_col=0)
     @functools.lru_cache()
```

```
def get_insert_size():
         return pd.read_csv(config['insert_size'], index_col=0)
     @functools.lru_cache()
     def get_gtf(gtf_file):
         return read_gtf(gtf_file)
[6]: def gene_annotation():
         gtf0 = get_gtf(config['gtf'])
         gtf = gtf0[gtf0["feature"] == "gene"]
         return gtf[["gene_id", "gene_name", "seqname", "start", "end", "strand"]]
     def chrom_type(chrom):
         if chrom=='chrX':
             return 'X'
         elif re.search('^chr(\d+)$', chrom):
             return 'autosome'
         else:
             return 'other'
     def save_ggplot(p, fn):
         for ext in ['.pdf', '.png', '.svg']:
             p.save(fn+ext, width=10, height=7)
     def rge(df, chrom):
         return df[df['seqname'] == chrom] [samples].mean() \
         - df[(df['chrom_type'] == 'autosome') & (df['seqname']!=chrom)][samples].
      \rightarrowmean()
[7]: gene_annot = gene_annotation()
     gene_annot.head()
    INFO:root:Extracted GTF attributes: ['gene_id', 'gene_type', 'gene_name',
    'level', 'havana_gene', 'transcript_id', 'transcript_type', 'transcript_name',
    'transcript_support_level', 'tag', 'havana_transcript', 'exon_number',
    'exon_id', 'ont', 'protein_id', 'ccdsid']
[7]:
                              gene_name seqname start
                                                          end strand
                   gene_id
     0
         ENSG00000223972.5
                                DDX11L1
                                           chr1
                                                 11869 14409
     12 ENSG00000227232.5
                                           chr1 14404 29570
                                 WASH7P
     25 ENSG00000278267.1
                              MIR6859-1
                                           chr1 17369 17436
     28 ENSG00000243485.5 MIR1302-2HG
                                           chr1 29554 31109
     36 ENSG00000284332.1
                             MIR1302-2
                                           chr1 30366 30503
```

```
[8]: log2tpm = pd.read_csv('/ceph/users/jbenja13/projects/sex_sz_ria/input/gene_tpm/
      log2tpm.iloc[0:2, 0:5]
 [8]:
                          R12864
                                   R12865
                                             R12866
                                                       R12867
                                                                 R12868
     Geneid
     ENSG00000223972.5 0.000000 0.000000 0.000000 0.000000
                                                               0.000000
     ENSG00000227232.5 2.294585 2.349649 2.983534 2.694415
                                                               2.364178
 [9]: df0 = log2tpm.merge(gene_annot.loc[:, ["gene_id", "gene_name", "seqname"]],
                         left_index=True, right_on="gene_id")
     df0['chrom_type'] = df0['seqname'].apply(chrom_type)
     df0.shape
 [9]: (51740, 1443)
[10]: df0.groupby('chrom_type').size()
[10]: chrom_type
                  1818
     autosome
                 44719
     other
                  5203
     dtype: int64
[11]: samples = [x for x in log2tpm.columns if re.match('R\d+', x)]
     df = df0[(df0[samples].sum(axis=1) >= 0.2 * len(samples)) & (df0['chrom_type'] !
      →= 'other')]
     df1 = df.groupby('chrom_type').mean().transpose()
     df1['sample'] = df1.index
     df1['RXE'] = df1['X'] - df1['autosome']
     df1.head(2)
[11]: chrom_type
                        X autosome sample
                                                 RXF.
     R12864
                 3.076746 2.791519 R12864 0.285227
     R12865
                 3.201302 2.986970 R12865 0.214332
[12]: df2 = df1.merge(get_pheno().loc[:, ['Sex', 'Dx', 'Region']], left_index=True,
      →right_index=True)
     df2.loc[:, ['Region', 'Sex', 'RXE']].groupby(['Region', 'Sex']).describe()
[12]:
                    RXE
                                                           25%
                                                                     50%
                  count
                             mean
                                       std
                                                 min
     Region Sex
     Caudate F
                  121.0 0.201658 0.080121 -0.010120 0.160207 0.218018
             М
                  273.0 0.201201 0.062586 -0.053587
                                                      0.177907 0.211707
     DLPFC
                  114.0 0.221461 0.061893 0.028849
                                                      0.202148 0.242446
             F
             М
                  246.0 0.217834 0.038949 0.038942 0.199114 0.221585
```

```
HIPPO
             F
                  121.0 0.225148 0.052878 0.069767 0.194949 0.229280
                   255.0 0.217129 0.049547 0.072521 0.192701 0.222089
             М
                        75%
                                  max
      Region Sex
                  0.261447 0.316335
      Caudate F
             M
                  0.246972 0.317737
     DLPFC
                  0.261521 0.314081
             F
                  0.244747 0.291408
                  0.261691 0.332028
     HIPPO
             F
                  0.245014 0.358289
[13]: from scipy.stats import variation
[14]: for tissue in ['Caudate', 'DLPFC', 'HIPPO']:
          for sex in ['F', 'M']:
             var = variation(df2[(df2['Region']==tissue) & (df2['Sex']==sex)].RXE)
              print("There is %.3f variation for %s in %s." % (var, sex, tissue))
     There is 0.396 variation for F in Caudate.
     There is 0.310 variation for M in Caudate.
     There is 0.278 variation for F in DLPFC.
     There is 0.178 variation for M in DLPFC.
     There is 0.234 variation for F in HIPPO.
     There is 0.228 variation for M in HTPPO.
     1.0.1 Separate out tissues
[15]: caudate = df2[(df2['Region']=='Caudate')].copy()
      dlpfc = df2[(df2['Region']=='DLPFC')].copy()
      hippo = df2[(df2['Region']=='HIPPO')].copy()
[16]: for tissue in ['caudate', 'DLPFC', "hippocampus"]:
          df_config = {'caudate': caudate,
                       'DLPFC': dlpfc,
                       'hippocampus': hippo}
          stat, pval =
       →mannwhitneyu(df_config[tissue][(df_config[tissue]['Sex']=='F')].RXE,

→df_config[tissue][(df_config[tissue]['Sex']=='M')].RXE)
          print("Mann-WhitneyU for female vs male (RXE) for %s: %.4f" % (tissue, __
       →pval))
     Mann-WhitneyU for female vs male (RXE) for caudate: 0.1118
     Mann-WhitneyU for female vs male (RXE) for DLPFC: 0.0017
     Mann-WhitneyU for female vs male (RXE) for hippocampus: 0.0523
```

```
[17]: for tissue in ['caudate', 'DLPFC', "hippocampus"]:
          df_config = {'caudate': caudate,
                       'DLPFC': dlpfc,
                       'hippocampus': hippo}
          stat, pval =
       →mannwhitneyu(df_config[tissue][(df_config[tissue]['Dx']=='Control')].RXE,

→df_config[tissue][(df_config[tissue]['Dx']=='Schizo')].RXE)
          print("Mann-WhitneyU for ctl vs sz (RXE) for %s: %.2e" % (tissue, pval))
     Mann-WhitneyU for ctl vs sz (RXE) for caudate: 4.18e-01
     Mann-WhitneyU for ctl vs sz (RXE) for DLPFC: 2.02e-01
     Mann-WhitneyU for ctl vs sz (RXE) for hippocampus: 3.83e-05
[18]: for tissue in ['caudate', 'DLPFC', "hippocampus"]:
          df_config = {'caudate': caudate,
                       'DLPFC': dlpfc,
                       'hippocampus': hippo}
          female = df_config[tissue] [(df_config[tissue] ['Sex'] == 'F')].copy()
          male = df_config[tissue][(df_config[tissue]['Sex']=='M')].copy()
          stat_f, pval_f = mannwhitneyu(female[(female['Dx']=='Control')].RXE,
                                        female[(female['Dx']=='Schizo')].RXE)
          stat_m, pval_m = mannwhitneyu(male[(male['Dx']=='Control')].RXE,
                                        male[(male['Dx']=='Schizo')].RXE)
          print("Mann-WhitneyU of female, ctl vs sz (RXE) for %s: %.4f" % (tissue, ⊔
       →pval_f))
          print("Mann-WhitneyU of male, ctl vs sz (RXE) for %s: %.4f" % (tissue, __
       →pval_m))
```

Mann-WhitneyU of female, ctl vs sz (RXE) for caudate: 0.1962
Mann-WhitneyU of male, ctl vs sz (RXE) for caudate: 0.1802
Mann-WhitneyU of female, ctl vs sz (RXE) for DLPFC: 0.2686
Mann-WhitneyU of male, ctl vs sz (RXE) for DLPFC: 0.2438
Mann-WhitneyU of female, ctl vs sz (RXE) for hippocampus: 0.0164
Mann-WhitneyU of male, ctl vs sz (RXE) for hippocampus: 0.0003

# 1.0.2 X chromosome expression differences

```
print("Mann-WhitneyU of female, ctl vs sz (RXE) for %s: %.4f" % (tissue, ⊔ ⇔pval_f))
print("Mann-WhitneyU of male, ctl vs sz (RXE) for %s: %.4f" % (tissue, ⊔ ⇒pval_m))
```

```
Mann-WhitneyU of female, ctl vs sz (RXE) for caudate: 0.0321
Mann-WhitneyU of male, ctl vs sz (RXE) for caudate: 0.1929
Mann-WhitneyU of female, ctl vs sz (RXE) for DLPFC: 0.0383
Mann-WhitneyU of male, ctl vs sz (RXE) for DLPFC: 0.0203
Mann-WhitneyU of female, ctl vs sz (RXE) for hippocampus: 0.0510
Mann-WhitneyU of male, ctl vs sz (RXE) for hippocampus: 0.0005
```

#### 1.0.3 Autosome expression

```
Mann-WhitneyU of female, ctl vs sz (RXE) for caudate: 0.1135
Mann-WhitneyU of male, ctl vs sz (RXE) for caudate: 0.1579
Mann-WhitneyU of female, ctl vs sz (RXE) for DLPFC: 0.0072
Mann-WhitneyU of male, ctl vs sz (RXE) for DLPFC: 0.0069
Mann-WhitneyU of female, ctl vs sz (RXE) for hippocampus: 0.0574
Mann-WhitneyU of male, ctl vs sz (RXE) for hippocampus: 0.0041
```

#### 1.0.4 Interaction model

```
[21]: df2.Sex = df2.Sex.astype("category").cat.rename_categories({"F": "Female", "M":

→"Male"})

df2.Dx = df2.Dx.astype("category").cat.rename_categories({"Schizo": "SZ",

→"Control": "CTL"})

df2.head(2)
```

```
[21]: X autosome sample RXE Sex Dx Region R12864 3.076746 2.791519 R12864 0.285227 Female SZ Caudate R12865 3.201302 2.986970 R12865 0.214332 Male SZ Caudate
```

```
[22]: df3 = df2[['RXE', 'Sex', 'Dx', 'Region']]
  df3.Sex = df3.Sex.cat.codes
  df3.Dx = df3.Dx.cat.codes
  df3.Region = df3.Region.astype("category").cat.codes
  df3.head(2)
```

/home/jbenja13/.local/lib/python3.9/site-packages/pandas/core/generic.py:5494: SettingWithCopyWarning:

A value is trying to be set on a copy of a slice from a DataFrame. Try using .loc[row\_indexer,col\_indexer] = value instead

See the caveats in the documentation: https://pandas.pydata.org/pandas-docs/stable/user\_guide/indexing.html#returning-a-view-versus-a-copy

[22]: RXE Sex Dx Region R12864 0.285227 0 1 0 R12865 0.214332 1 1 0

```
import statsmodels.api as sm
from statsmodels.formula.api import ols
anova_df = df2.loc[:,['RXE','Sex','Dx','Region']]

# ANOVA results with combinations of 2 groups:
formula = 'RXE ~ C(Sex) + C(Dx) + C(Region) + C(Sex):C(Dx) + C(Sex):C(Region) + C(Dx):C(Region)'
lm = ols(formula, anova_df).fit()
table = sm.stats.anova_lm(lm, typ=2)
print(table)
```

```
df
                                            F
                                                 PR(>F)
                   sum_sq
C(Sex)
                 0.005023
                               1.0
                                    1.594365 0.206966
C(Dx)
                 0.036769
                              1.0 11.670248 0.000658
C(Region)
                 0.081754
                              2.0 12.974264 0.000003
                              1.0
C(Sex):C(Dx)
                 0.005657
                                   1.795461 0.180534
C(Sex):C(Region) 0.003553
                              2.0
                                    0.563890 0.569153
                                    4.561611 0.010640
C(Dx):C(Region)
                 0.028744
                              2.0
Residual
                 3.528694 1120.0
                                         {\tt NaN}
                                                   NaN
```

#### By tissue interaction

```
# ANOVA results with combinations of 2 groups:
    formula = 'RXE ~ C(Sex) + C(Dx) + C(Sex):C(Dx)'
    lm = ols(formula, anova_df).fit()
    table = sm.stats.anova_lm(lm, typ=2)
    print(tissue)
    print(table)
    print("\n")
    anova_df['combination'] = anova_df.Sex.astype("str") + " / " + anova_df.Dx.
 →astype("str")
    anova_df['combination'] = anova_df.Sex.astype("str") + " / " + anova_df.Dx.
 →astype("str")
    # perform multiple pairwise comparison (Tukey HSD)
    m_comp = pairwise_tukeyhsd(endog=anova_df['RXE'],__

¬groups=anova_df['combination'], alpha=0.05)
    # coerce the tukeyhsd table to a DataFrame
    tukey_data = pd.DataFrame(data=m_comp._results_table.data[1:], columns =__
 →m_comp._results_table.data[0])
    if tukey_data[(tukey_data['p-adj'] < 0.05)].shape[0] == 0:</pre>
        print("There is no significant interactions!")
    else:
        print(tukey_data[(tukey_data['p-adj'] < 0.05)])</pre>
    print("\n")
caudate
                                           PR(>F)
                         df
                                      F
                sum_sq
C(Sex)
             0.000029 1.0 0.006205 0.937254
C(Dx)
             0.001563
                          1.0 0.333410 0.563991
C(Sex):C(Dx) 0.005784
                          1.0 1.233748 0.267364
Residual
             1.828405 390.0
                                    \mathtt{NaN}
                                              NaN
```

There is no significant interactions!

DLPFC

	sum_sq	df	F	PR(>F)
C(Sex)	0.001069	1.0	0.474628	0.491314
C(Dx)	0.002436	1.0	1.081793	0.299003
C(Sex):C(Dx)	0.000603	1.0	0.267795	0.605136
Residual	0.801500	356.0	NaN	NaN

There is no significant interactions!

hippocampus

sum\_sq df F PR(>F)

```
      C(Sex)
      0.007208
      1.0
      2.988436
      8.469084e-02

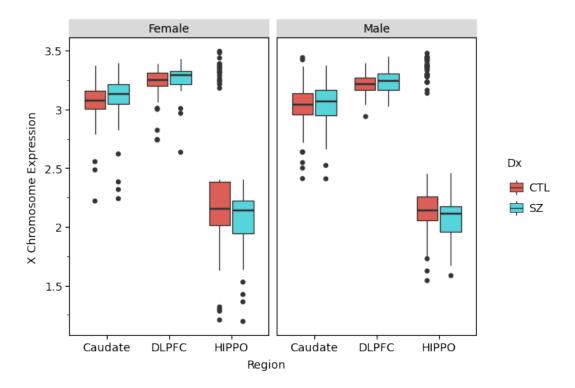
      C(Dx)
      0.061027
      1.0
      25.302216
      7.641565e-07

      C(Sex):C(Dx)
      0.000824
      1.0
      0.341619
      5.592505e-01

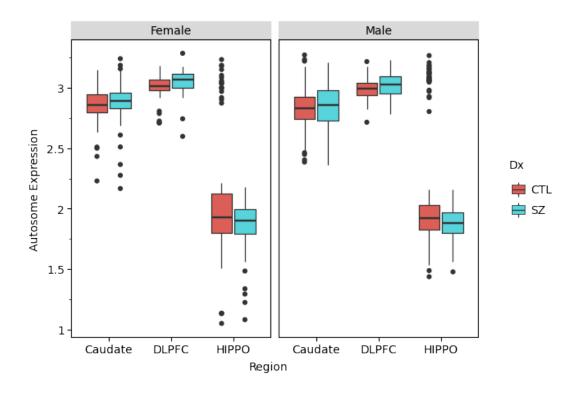
      Residual
      0.897235
      372.0
      NaN
      NaN
```

```
group1 group2 meandiff p-adj lower upper reject
2 F / Control M / Schizo -0.0358 0.001 -0.0559 -0.0157 True
5 M / Control M / Schizo -0.0289 0.001 -0.0457 -0.0121 True
```

#### 1.0.5 Plot RXE



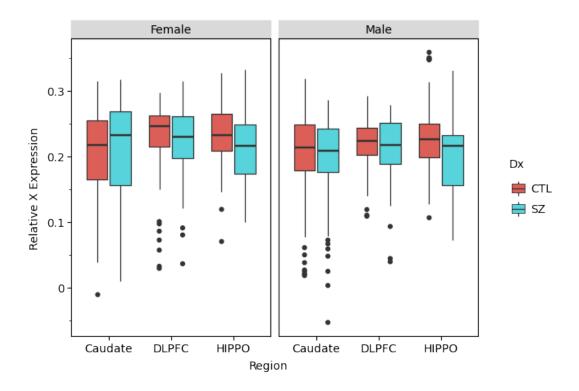
## [25]: <ggplot: (8738604889567)>



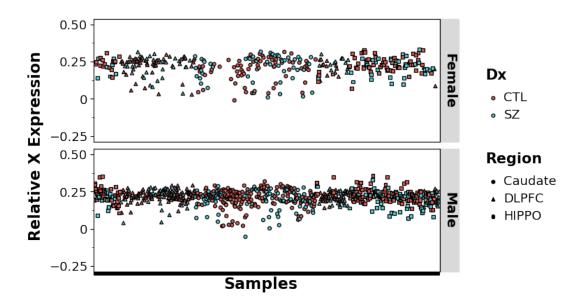
## [26]: <ggplot: (8738604804476)>

/home/jbenja13/.local/lib/python3.9/sitepackages/plotnine/facets/facet\_grid.py:136: FutureWarning: Index.\_\_and\_\_
operating as a set operation is deprecated, in the future this will be a logical
operation matching Series.\_\_and\_\_. Use index.intersection(other) instead
/home/jbenja13/.local/lib/python3.9/sitepackages/plotnine/facets/facet\_grid.py:137: FutureWarning: Index.\_\_and\_\_
operating as a set operation is deprecated, in the future this will be a logical

operation matching Series.\_\_and\_\_. Use index.intersection(other) instead



# [27]: <ggplot: (8738604890793)>



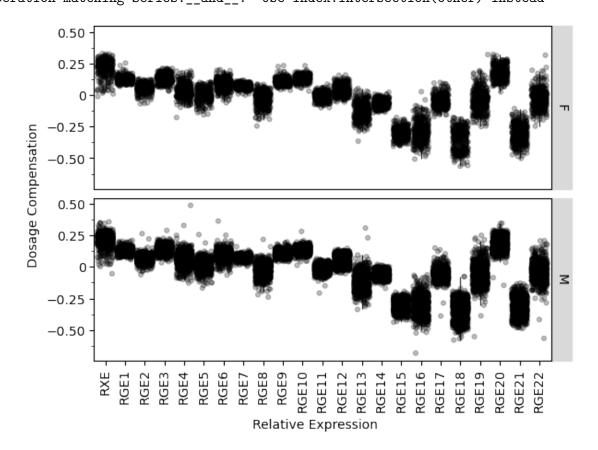
[28]: <ggplot: (8738603931140)>

```
[29]: df2.to_csv('RXE_public.csv')
      save_ggplot(p, "RXE_public_bySex")
     /home/jbenja13/.local/lib/python3.9/site-
     packages/plotnine/facets/facet_grid.py:136: FutureWarning: Index.__and__
     operating as a set operation is deprecated, in the future this will be a logical
     operation matching Series.__and__. Use index.intersection(other) instead
     /home/jbenja13/.local/lib/python3.9/site-
     packages/plotnine/facets/facet_grid.py:137: FutureWarning: Index.__and__
     operating as a set operation is deprecated, in the future this will be a logical
     operation matching Series.__and__. Use index.intersection(other) instead
     /home/jbenja13/.local/lib/python3.9/site-
     packages/plotnine/facets/facet_grid.py:136: FutureWarning: Index.__and__
     operating as a set operation is deprecated, in the future this will be a logical
     operation matching Series.__and__. Use index.intersection(other) instead
     /home/jbenja13/.local/lib/python3.9/site-
     packages/plotnine/facets/facet grid.py:137: FutureWarning: Index. and
     operating as a set operation is deprecated, in the future this will be a logical
     operation matching Series.__and__. Use index.intersection(other) instead
     /home/jbenja13/.local/lib/python3.9/site-
     packages/plotnine/facets/facet_grid.py:136: FutureWarning: Index.__and__
     operating as a set operation is deprecated, in the future this will be a logical
     operation matching Series.__and__. Use index.intersection(other) instead
     /home/jbenja13/.local/lib/python3.9/site-
     packages/plotnine/facets/facet grid.py:137: FutureWarning: Index. and
     operating as a set operation is deprecated, in the future this will be a logical
     operation matching Series.__and__. Use index.intersection(other) instead
```

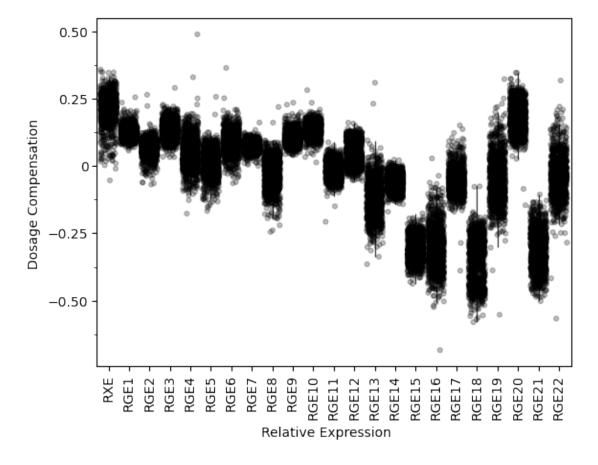
#### 1.1 Other chromosomes

```
[30]: for chrom in range (1,23):
        df1['RGE%d' % chrom] = rge(df, 'chr%d' % chrom)
    rge = df1[[x for x in df1.columns if 'RGE' in x or 'RXE' in x]]
    rge.head()
[30]: chrom type
                  RXE
                         RGE1
                                 RGE2
                                          RGE3
                                                  RGE4
                                                          RGE5 \
    R12864
              R12865
              R12866
    R12867
              0.144386 0.102819 0.063515 0.147857 0.012888 -0.003432
    R12868
              0.071929 0.096117 -0.028140 0.085753 -0.073615 -0.100151
                 RGE6
                         RGE7
                                 RGE8
                                          RGE9 ...
                                                   RGE13
    chrom_type
                                                           RGE14
              R12864
    R12865
              0.098431 0.073774 -0.114831 0.140709 ... -0.226386 -0.045472
    R12866
              0.039514 0.094366 -0.098518 0.143308 ... -0.173592 -0.024457
    R12867
              0.069605 0.044507 -0.058054 0.137344
                                              ... -0.117058 -0.047924
    R12868
             RGE17
                                                 RGE19
    chrom_type
                RGE15
                         RGF16
                                         RGE18
                                                         RGE20 \
    R12864
             -0.355004 -0.280154 -0.038059 -0.450791 0.023306 0.273079
    R12865
             -0.388450 -0.279795 0.002795 -0.489906 0.066002 0.247618
             -0.294699 -0.214212 -0.020374 -0.488748 0.060728 0.256096
    R12866
    R12867
             -0.277289 -0.212308 -0.045132 -0.396895 -0.044963 0.208471
             R12868
                RGE21
    chrom_type
                         RGE22
    R12864
             -0.339813 -0.009248
    R12865
             -0.319956 -0.032715
    R12866
             -0.285757 0.029614
    R12867
             -0.227589 -0.047968
             -0.163413 0.126474
    R12868
    [5 rows x 23 columns]
[31]: rge2 = rge.stack().reset_index()
    rge2.columns = ['sample', 'var', 'value']
    var2 = pd.Categorical(rge2['var'], categories=['RXE'] + ['RGE%d' % chrom for_
     \rightarrowchrom in range(1,23)])
    rge2 = rge2.assign(var2 = var2)
    rge2 = rge2.merge(get_pheno().loc[:, ['Sex', 'Dx']], left_on='sample',_
     →right_index=True)
    rge2.head()
```

```
[31]:
        sample
                         value var2 Sex
                                              Dx
                 var
     0 R12864
                 RXE 0.285227
                                 RXE
                                       F
                                          Schizo
     1 R12864 RGE1 0.137221
                                RGE1
                                          Schizo
     2 R12864
                RGE2 0.048619
                                RGE2
                                          Schizo
                                       F
                      0.124203
     3 R12864 RGE3
                                RGE3
                                          Schizo
     4 R12864
                RGE4 0.027022 RGE4
                                          Schizo
[32]: pp = ggplot(rge2, aes(x='var2', y='value')) \
     + geom_boxplot(outlier_alpha=0) \
     + geom_jitter(alpha=0.25) + facet_grid('Sex~')\
     + ylab("Dosage Compensation") + xlab("Relative Expression")
     + theme matplotlib() \
     + theme(axis_text_x=element_text(rotation=90))
     pp
```



```
[32]: <ggplot: (8738595709175)>
```



```
[33]: <ggplot: (8738595273198)>
```

```
[34]: rge.to_csv('RGE_public.csv')
save_ggplot(pp, "RGE_public_subset")
save_ggplot(ww, "RGE_public_all")
```

/home/jbenja13/.local/lib/python3.9/site-packages/plotnine/facets/facet\_grid.py:136: FutureWarning: Index.\_\_and\_\_

```
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