Real data: CAPE Negative

In: A guide to test association between Polygenic Risk Scores and psychological and psychiatric traits: practical examples

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Working flow and code

In this real data set there are 106 PRS, and a **continuous Trait** ($CAPE_{Negative}$), with gender, age, and two Principal Components as covariates. For more details see Section 7 in the paper.

- Data reading

```
dat <- read.table("Real_data_Negative.csv", header=TRUE, sep="\t", dec=".")
names(dat) #
```

```
##   [1] "ID"            "Sex"           "Age"           "CAPE_Negative"
##   [5] "PRS.1"         "PRS.2"         "PRS.3"         "PRS.4"
##   [9] "PRS.5"         "PRS.6"         "PRS.7"         "PRS.8"
##  [13] "PRS.9"         "PRS.10"        "PRS.11"        "PRS.12"
##  [17] "PRS.13"        "PRS.14"        "PRS.15"        "PRS.16"
##  [21] "PRS.17"        "PRS.18"        "PRS.19"        "PRS.20"
##  [25] "PRS.21"        "PRS.22"        "PRS.23"        "PRS.24"
##  [29] "PRS.25"        "PRS.26"        "PRS.27"        "PRS.28"
##  [33] "PRS.29"        "PRS.30"        "PRS.31"        "PRS.32"
##  [37] "PRS.33"        "PRS.34"        "PRS.35"        "PRS.36"
##  [41] "PRS.37"        "PRS.38"        "PRS.39"        "PRS.40"
##  [45] "PRS.41"        "PRS.42"        "PRS.43"        "PRS.44"
##  [49] "PRS.45"        "PRS.46"        "PRS.47"        "PRS.48"
##  [53] "PRS.49"        "PRS.50"        "PRS.51"        "PRS.52"
##  [57] "PRS.53"        "PRS.54"        "PRS.55"        "PRS.56"
##  [61] "PRS.57"        "PRS.58"        "PRS.59"        "PRS.60"
##  [65] "PRS.61"        "PRS.62"        "PRS.63"        "PRS.64"
##  [69] "PRS.65"        "PRS.66"        "PRS.67"        "PRS.68"
##  [73] "PRS.69"        "PRS.70"        "PRS.71"        "PRS.72"
##  [77] "PRS.73"        "PRS.74"        "PRS.75"        "PRS.76"
##  [81] "PRS.77"        "PRS.78"        "PRS.79"        "PRS.80"
##  [85] "PRS.81"        "PRS.82"        "PRS.83"        "PRS.84"
##  [89] "PRS.85"        "PRS.86"        "PRS.87"        "PRS.88"
##  [93] "PRS.89"        "PRS.90"        "PRS.91"        "PRS.92"
##  [97] "PRS.93"        "PRS.94"        "PRS.95"        "PRS.96"
## [101] "PRS.97"        "PRS.98"        "PRS.99"        "PRS.100"
## [105] "PRS.101"       "PRS.102"       "PRS.103"       "PRS.104"
## [109] "PRS.105"       "PRS.106"       "PC1"           "PC2"
```

```
dat <- dat[, -1]
```

Important! Check that all variables you are interested in are properly read and that there are not other variables you do not need.

- Do not forget to declare the categorical variables as factors.

```
dat$Sex <- as.factor(dat$Sex)
```

1. What full model should be considered?

First, given a particular PRS (named PRS.i), consider all the possible full models:

- FM_{WI} : Trait versus PRS.i + Sex + Age + PC1 + PC2
- FM_{Sex}: Trait versus PRS.i + Sex + PRS.i $\cdot$ Sex + Age + PC1 + PC2

2. How to make a PRS ranking to find the important ones?

As is described in the paper, for each model, calculate the coefficient of determination R^2 and calculate the sum: $S = R^2_{WI} + R^2_{Sex}$.

According to S, list the PRSs in decreasing order:

```
out <- orderR2(dat, yname="CAPE_Negative", prsname = "PRS.")
head(out)
```

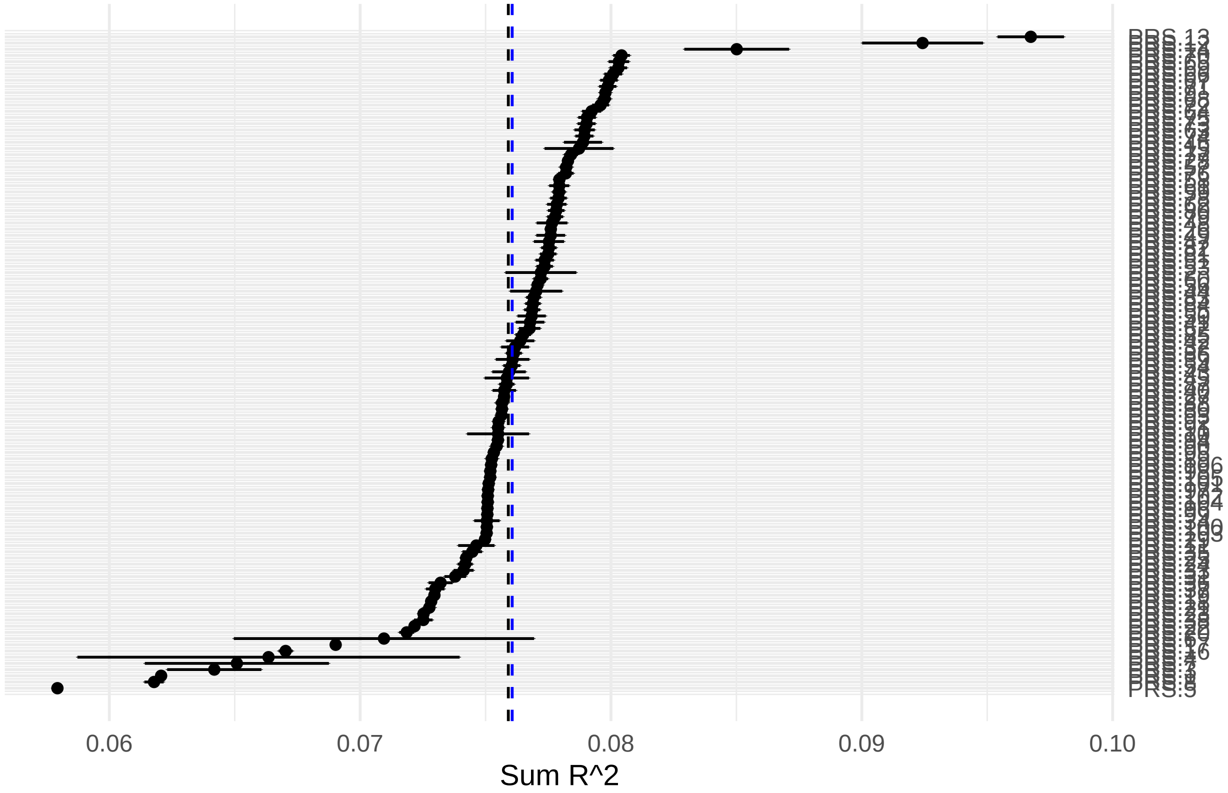
```
##             Model1     Model2        Sum
## PRS.13 0.04743339 0.04930427 0.09673766
## PRS.12 0.04450825 0.04791485 0.09242311
## PRS.14 0.04102906 0.04398352 0.08501259
## PRS.70 0.03997513 0.04044623 0.08042136
## PRS.65 0.03986674 0.04044801 0.08031474
## PRS.69 0.03990297 0.04039181 0.08029478
```

```
mainfilename <- "Real_example_CAPE_Negative"
filename <- paste0(mainfilename, "_Ordered_PRS.csv")
write.csv2(out,file=filename)
```

Plot the sum of coefficients of determination S_{R^2}. Lines: in blue the median; in black the mean.

```
out <- data.frame(out)
nPRS <- dim(out)[1]
select <- grep("Model", names(out), value=FALSE)
out$effect <- out$Sum
sds <- apply(out[, select], 1, sd)
out$lower <- out$effect - sds
out$upper <- out$effect + sds
out$rank <- nPRS:1

n <- dim(out)[1]
ggplot(data=out, aes(y=rank, x=effect, xmin=lower, xmax=upper)) +
  geom_point() +
  geom_errorbarh(height=.1) +
  scale_y_continuous(name=NULL, breaks= n:1, labels=row.names(out), position="right") +
  labs(title='', x='Sum R^2', y = 'PRS') +
  geom_vline(xintercept=mean(out$effect), color='black', linetype='dashed') +
  geom_vline(xintercept=median(out$effect), color='blue', linetype='dashed') +
  theme_minimal()
```

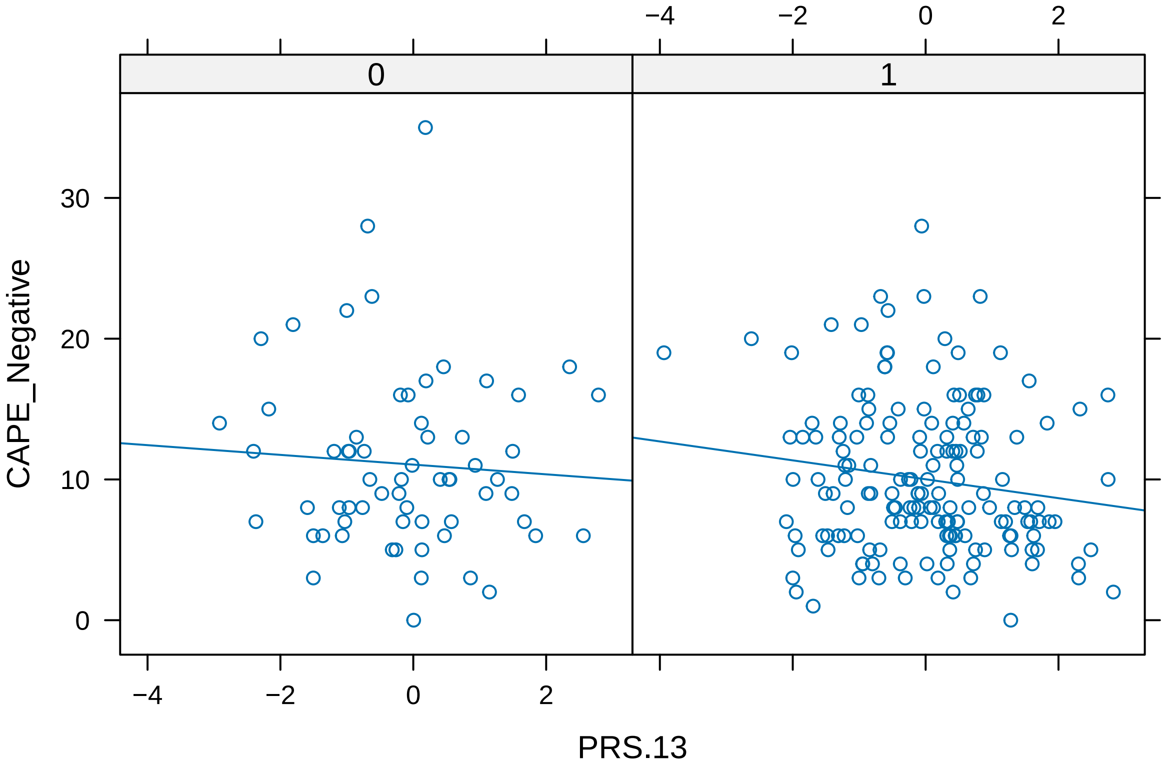


According to the obtained results, first, PRS.13 is selected to analyse its possible association with the $CAPE_{Negative}$.

3. Which model, of all the possible ones, should be used?

The following Figure represents the scatter plot of $CAPE_{Negative}$ versus PRS.13 separated by Sex group.

```
# First candidate PRS.13
# Plot it
library(lattice)
xyplot(CAPE_Negative~PRS.13|Sex, data=dat,  type=c("p", "r"))
```



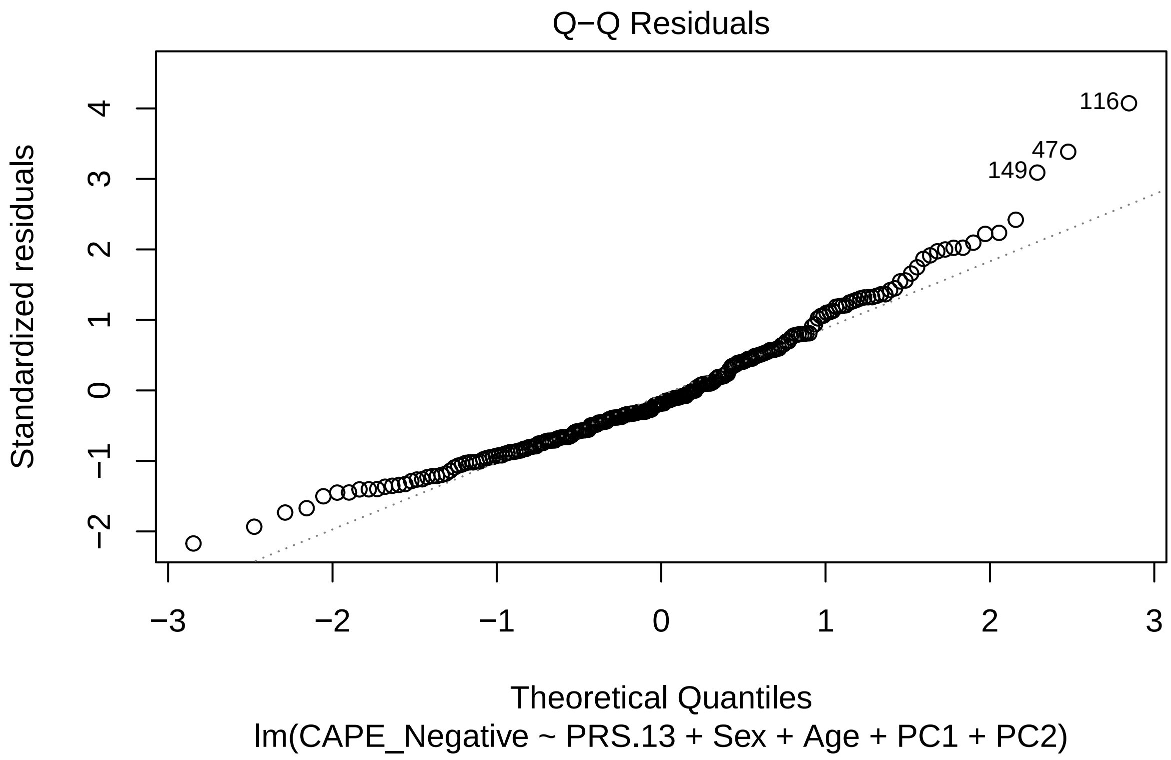
The plots suggest that the interaction between the PRS.13 and the sex is not relevant. Thus, we set the full model candidate (FM): $CAPE_{Negative} \sim PRS + Sex + Age + PC1 + PC2$.

4. For a continuous trait, what steps should be followed for a correct analysis?

- **4.1. How is the candidate model validated?**

First, we validate the normality of the errors and the constant variance conditions (see the figures and the results of Shapiro test and Levene test).

```
#model
FM <- lm(CAPE_Negative ~ PRS.13 + Sex + Age + PC1 + PC2, data=dat)
#qq-plot for normality
plot(FM,2)
```



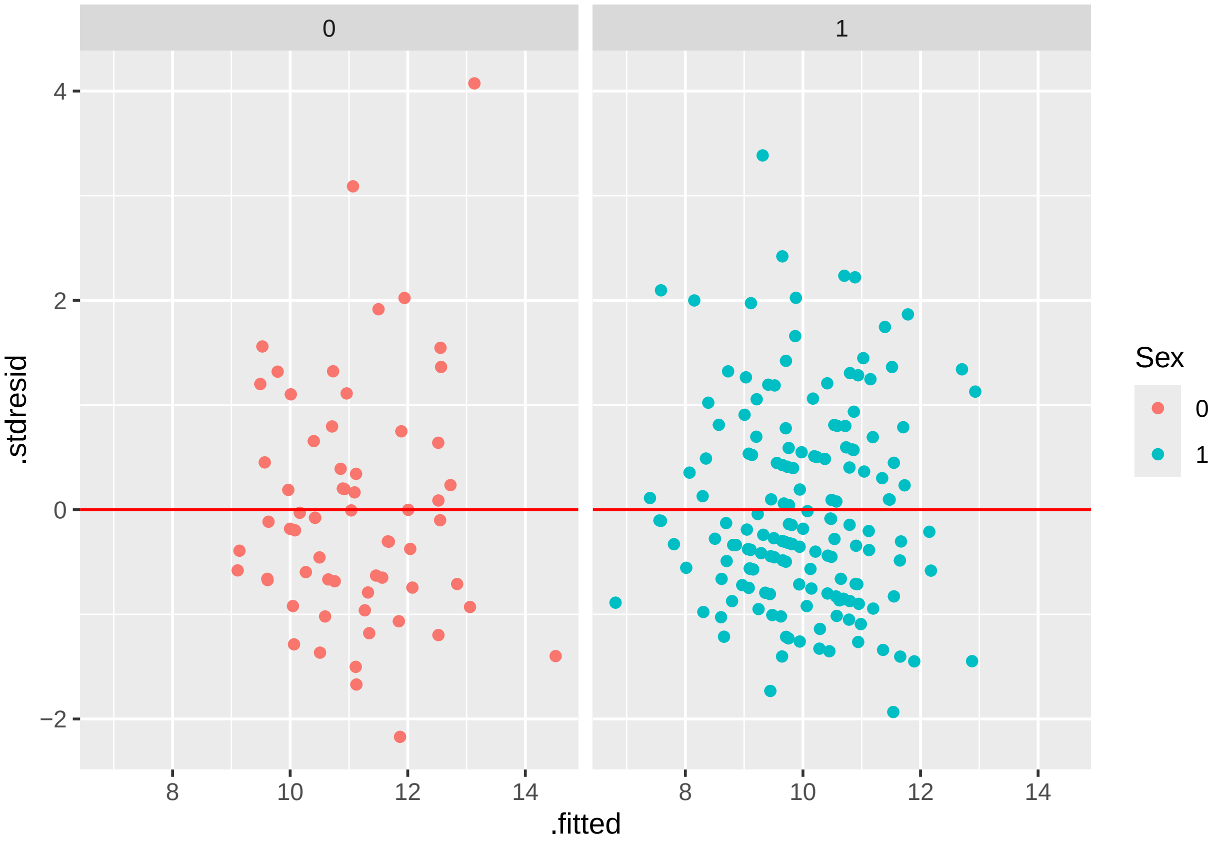
The lack of normality of residuals is suggested by this last plot.

This supported by Shapiro's test:

```
#Shapiro-Wilk test
shapiro.test(FM$residuals)
```

```
##
##  Shapiro-Wilk normality test
##
## data:  FM$residuals
## W = 0.95885, p-value = 4.335e-06
```

```
#plot for variances
d <- fortify(FM)
ggplot(d,aes(x=.fitted, y=.stdresid, colour=Sex)) + geom_point() + geom_hline(yintercept=0, col="red")+
  facet_wrap(.~Sex)
```



```
#Levene's test
leveneTest(.stdresid ~ Sex, data=d)
```

```
## Levene's Test for Homogeneity of Variance (center = median)
##        Df F value Pr(>F)
## group   1  0.7613 0.3839
##       224
```

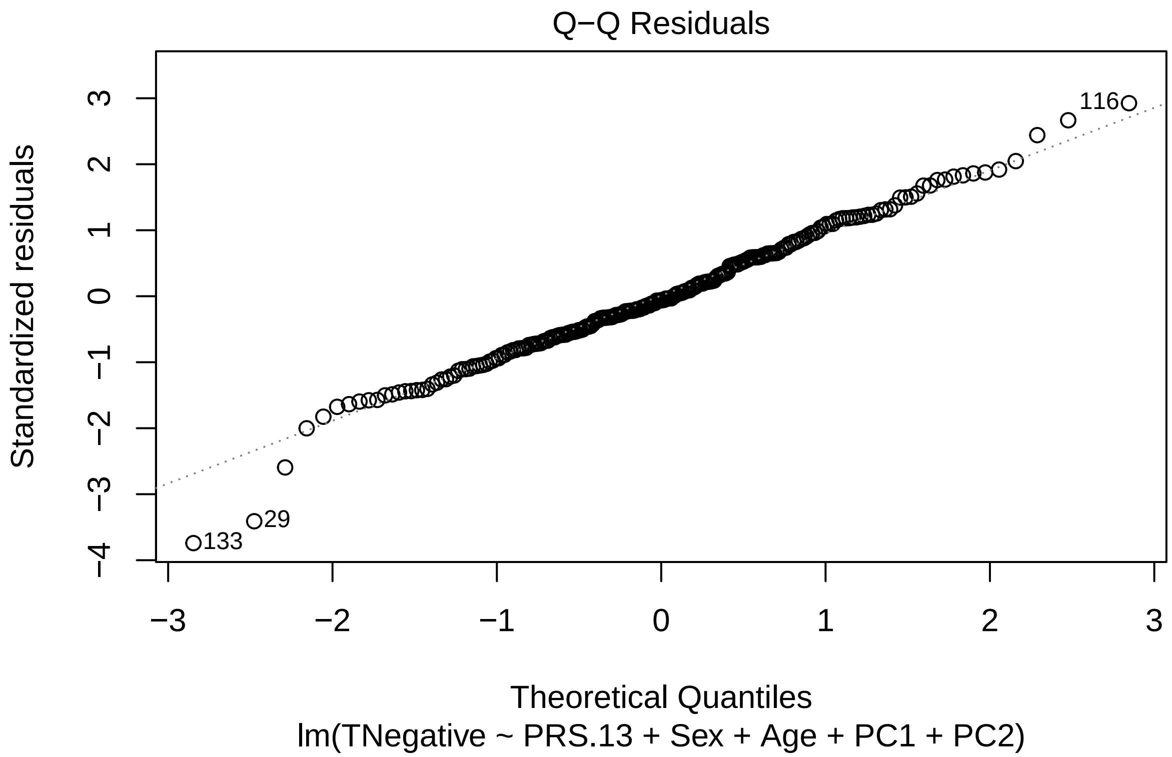
All in all, it seems that there is lack of normality of residuals but linearity and homocedasticity assumptions hold.

- **4.2. What can be done if any validation condition fails?**

We have two approaches to assess the possible association with PRS.13 and $CAPE_{Negative}$: try a transformation or perform a permutation test.

First we try the squared root transformation for the dependent variable:

```
dat$TNegative <- sqrt(dat$CAPE_Negative)
FM <- lm(TNegative ~ PRS.13 + Sex + Age + PC1 + PC2, data=dat)
#qq-plot for normality
plot(FM,2)
```

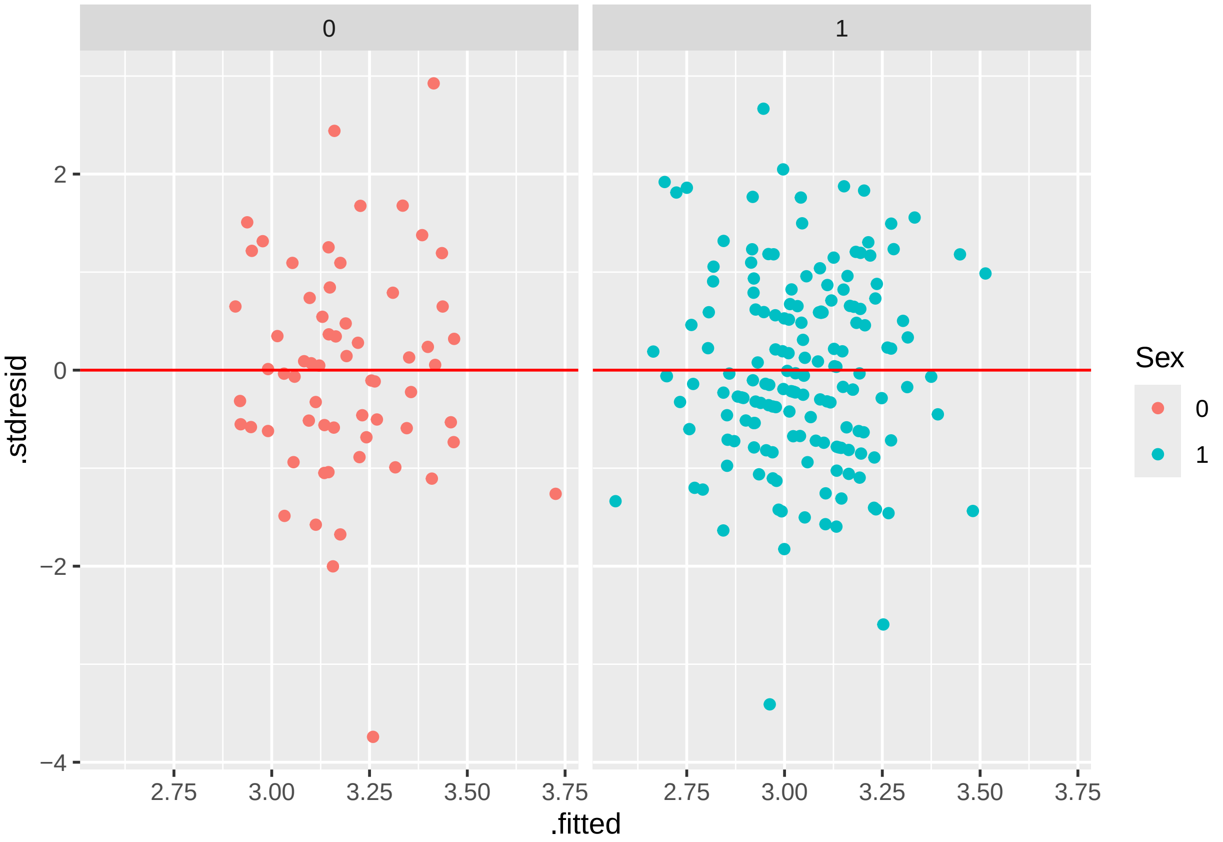


```
#Shapiro-Wilk test
shapiro.test(FM$residuals)
```

```
##
##  Shapiro-Wilk normality test
##
## data:  FM$residuals
## W = 0.98898, p-value = 0.08108
```

It is suggested the transformation offered a solution for the lack of normality. Furthermore,. . .

```
#plot for variances
d <- fortify(FM)
ggplot(d,aes(x=.fitted, y=.stdresid, colour=Sex)) +
  geom_point() +
  geom_hline(yintercept=0, col="red")+
  facet_wrap(.~Sex)
```



```
#Levene's test
leveneTest(.stdresid ~ Sex, data=d)
```

```
## Levene's Test for Homogeneity of Variance (center = median)
##        Df F value Pr(>F)
## group   1  0.2998 0.5846
##       224
```

. . . results do not indicate evidence against homoscedasticity, neither a pattern is observed that could indicate a lack of linearity.

Therefore, the model we build is given by:

```
summary(FM)
```

```
##
## Call:
## lm(formula = TNegative ~ PRS.13 + Sex + Age + PC1 + PC2, data = dat)
##
## Residuals:
##     Min      1Q  Median      3Q     Max
## -3.2590 -0.5534 -0.0529  0.5640  2.5020
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)  3.013198   0.467855   6.440 7.39e-10 ***
## PRS.13      -0.101131   0.050141  -2.017   0.0449 *
## Sex1        -0.142137   0.133729  -1.063   0.2890
## Age          0.008634   0.021935   0.394   0.6943
## PC1          3.796970   4.232105   0.897   0.3706
## PC2          8.408721   4.206204   1.999   0.0468 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.8837 on 220 degrees of freedom
##   (1 observation deleted due to missingness)
## Multiple R-squared:  0.04105,    Adjusted R-squared:  0.01926
## F-statistic: 1.884 on 5 and 220 DF,  p-value: 0.09824
```

The results show that PRS.13 is related with the $\sqrt{Trait}$ in the following way:

$$\widehat{\sqrt{Trait}} = 3.013 - 0.101 \times PRS.13 - 0.142 \times Sex + 0.009 \times Age + 3.797 \times PC1 + 8.409 \times PC2,$$

where Sex takes values 0 or 1, depending on whether the individual under study is male or female. See section 7.1 in the paper for more details.

On the other hand, based on the permutation approach:

```
NM <- lm(CAPE_Negative ~  Sex + Age + PC1 + PC2, data=dat)
FM <- lm(CAPE_Negative ~ PRS.13 + Sex + Age + PC1 + PC2, data=dat)
outperm <- dR2(NullModel=NM, FullModel=FM, B=5000, seed=165)
outperm
```

```
## $dR2
## [1] 0.01964777
##
## $pvalue
## [1] 0.033
```

We observe an increase of 0.0196 in the coefficient of determination when the PRS.13 is included in the model and the permutation test indicates it is significant.

- **Last step: We move to the next PRS.**