

# SimpleViolins

Alex Badea

10/29/2021

## Contents

Data sources	1
Plots for CPu	1

---

## Data sources

We evaluate genotype and sex differences in mouse models of Alzheimer's disease risk, based on APOE alleles: APOE2 is protective, APOE3 is control, and APOE4 represents genetic risk for AD. One allele increases risk by 3 times, and two APOE4 alleles increase risk by 10 times (ref) in relation to APOE3 . We use multiple data sources:

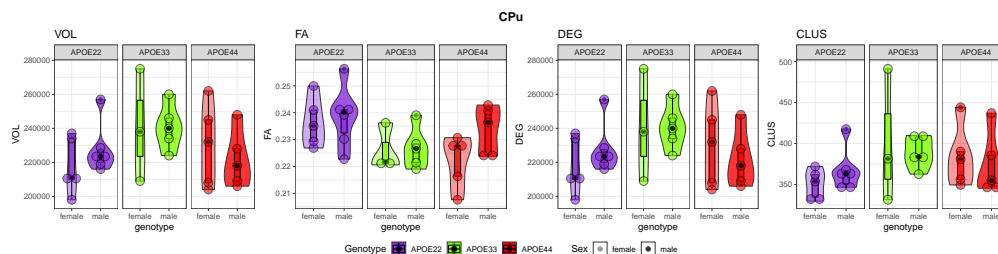
Morris Water Maze - testing spatial navigation memory Segmentation: regional volumes Microstructural properties: FA Tractography Based Connectivity: Degree, and Clustering Coefficient

We read the data and normalize volumes to the whole brain.

## Plots for CPu

We embed plots for day d8 probe trials:

```
## Bin width defaults to 1/30 of the range of the data. Pick better value with `binwidth`.
## Bin width defaults to 1/30 of the range of the data. Pick better value with `binwidth`.
## Bin width defaults to 1/30 of the range of the data. Pick better value with `binwidth`.
## Bin width defaults to 1/30 of the range of the data. Pick better value with `binwidth`.
## Bin width defaults to 1/30 of the range of the data. Pick better value with `binwidth`.
```



```
print('CPu')
```

```
## [1] "CPu"
```

```
lm_CPu_VOL <- lm(CPu ~ Genotype*Sex, geno_combined_VOL)
summary(lm_CPu_VOL)
```

```
##
## Call:
## lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -7.026e-04 -3.106e-04 -6.856e-05  1.250e-04  1.486e-03
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)    2.476e-02  2.420e-04 102.283 < 2e-16 ***
## GenotypeAPOE33  -7.692e-04  3.952e-04  -1.946  0.06395 .
## GenotypeAPOE44   9.720e-04  3.423e-04   2.840  0.00928 **
## Sexmale         -4.568e-05  3.277e-04  -0.139  0.89036
## GenotypeAPOE33:Sexmale 2.317e-04  5.134e-04   0.451  0.65601
## GenotypeAPOE44:Sexmale -5.205e-04  4.739e-04  -1.098  0.28336
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.0005412 on 23 degrees of freedom
## Multiple R-squared:  0.5711, Adjusted R-squared:  0.4778
## F-statistic: 6.124 on 5 and 23 DF,  p-value: 0.0009559

lm_CPu_VOL_f <- lm(CPu ~ Genotype, combo_f_FA)
#summary(lm_CPu_VOL_f)
lm_CPu_VOL_m <- lm(CPu ~ Genotype, combo_m_FA)
#summary(lm_CPu_VOL_f)

#omnibus testing
#https://cran.r-project.org/web/packages/emmeans/vignettes/confidence-intervals.html#adjust

#33333
sink(paste(outpath, "CPu_VOL_SimpleModels.txt"))
mydata.lm <- lm(CPu ~ Genotype*Sex, data = geno_combined_VOL)
anova(mydata.lm)

## Analysis of Variance Table
##
## Response: CPu
##           Df      Sum Sq    Mean Sq F value    Pr(>F)
## Genotype    2 8.0970e-06 4.0485e-06 13.8218 0.0001142 ***
## Sex          1 1.9980e-07 1.9980e-07  0.6822 0.4173028
## Genotype:Sex  2 6.7230e-07 3.3610e-07  1.1476 0.3349205
## Residuals   23 6.7368e-06 2.9290e-07
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

summary(glht(mydata.lm, emm(pairwise ~ Genotype|Sex, adjust="sidak")))
```

*#consider save to file*

```
## $`Sex = female`
##
## Simultaneous Tests for General Linear Hypotheses
##
```

```

## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  0.0007692  0.0003952   1.946   0.1479
## APOE22 - APOE44 == 0 -0.0009720  0.0003423  -2.840   0.0241 *
## APOE33 - APOE44 == 0 -0.0017413  0.0003952  -4.406   <0.001 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  0.0005375  0.0003277   1.640   0.2494
## APOE22 - APOE44 == 0 -0.0004515  0.0003277  -1.378   0.3685
## APOE33 - APOE44 == 0 -0.0009890  0.0003423  -2.889   0.0217 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
summary(glht(mydata.lm, emm(pairwise ~ Sex| Genotype)))

## $`Genotype = APOE22`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0 4.568e-05 3.277e-04  0.139   0.89
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.0001860  0.0003952  -0.471   0.642
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##

```

```
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## female - male == 0 0.0005662 0.0003423  1.654    0.112
## (Adjusted p values reported -- single-step method)
```

```
sink()
#3333

sink(paste(outpath, "CPu_VOL_SimpleStats.txt", sep=''))
'omnibus testing'
```

```
## [1] "omnibus testing"
```

```
joint_tests(ref_grid(lm_CPu_VOL))
```

```
## model term    df1 df2 F.ratio p.value
## Genotype      2  23  13.818 0.0001
## Sex           1  23   0.476 0.4970
## Genotype:Sex   2  23   1.148 0.3349
```

```
joint_tests(ref_grid(lm_CPu_VOL), by = "Sex", adjust = "sidak")
```

```
## Sex = female:
## model term df1 df2 F.ratio p.value
## Genotype   2  23  10.239 0.0007
##
## Sex = male:
## model term df1 df2 F.ratio p.value
## Genotype   2  23   4.186 0.0282
```

```
joint_tests(ref_grid(lm_CPu_VOL), by = "Genotype", adjust = "sidak" )
```

```
## Genotype = APOE22:
## model term df1 df2 F.ratio p.value
## Sex        1  23   0.019 0.8904
##
## Genotype = APOE33:
## model term df1 df2 F.ratio p.value
## Sex        1  23   0.222 0.6423
##
## Genotype = APOE44:
## model term df1 df2 F.ratio p.value
## Sex        1  23   2.736 0.1117
```

```
'anova'
```

```
## [1] "anova"
```

```
anova(lm_CPu_VOL)
```

```
## Analysis of Variance Table
##
```

```
## Response: CPu
```

```
##           Df      Sum Sq    Mean Sq F value    Pr(>F)
## Genotype    2 8.0970e-06 4.0485e-06 13.8218 0.0001142 ***
```

```
## Sex          1 1.9980e-07 1.9980e-07 0.6822 0.4173028
## Genotype:Sex 2 6.7230e-07 3.3610e-07 1.1476 0.3349205
## Residuals    23 6.7368e-06 2.9290e-07
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

#anova(lm_CPu_VOL_f)
#anova(lm_CPu_VOL_m)

#post hoc comparison
CPu_VOL.emm <- emmeans(lm_CPu_VOL , ~ Genotype | Sex, adjust = "sidak")
contrast(CPu_VOL.emm, simple = c("Genotype", "Sex")) #FDR

## contrast          estimate          SE df t.ratio p.value
## APOE22 female effect  3.37e-06 0.000223 23   0.015 0.9881
## APOE33 female effect -7.66e-04 0.000275 23  -2.784 0.0316
## APOE44 female effect  9.75e-04 0.000223 23   4.378 0.0013
## APOE22 male effect   -4.23e-05 0.000208 23  -0.204 0.9881
## APOE33 male effect   -5.80e-04 0.000223 23  -2.603 0.0318
## APOE44 male effect    4.09e-04 0.000223 23   1.837 0.1188
##
## P value adjustment: fdr method for 6 tests
contrast(CPu_VOL.emm, simple = list("Genotype", "Sex"))

## $`simple contrasts for Genotype`
## Sex = female:
## contrast          estimate          SE df t.ratio p.value
## APOE22 effect   -6.76e-05 0.000208 23  -0.325 0.7484
## APOE33 effect   -8.37e-04 0.000238 23  -3.523 0.0027
## APOE44 effect    9.04e-04 0.000208 23   4.342 0.0007
##
## Sex = male:
## contrast          estimate          SE df t.ratio p.value
## APOE22 effect    2.87e-05 0.000186 23   0.154 0.8791
## APOE33 effect   -5.09e-04 0.000195 23  -2.611 0.0324
## APOE44 effect    4.80e-04 0.000195 23   2.464 0.0324
##
## P value adjustment: fdr method for 3 tests
##
## $`simple contrasts for Sex`
## Genotype = APOE22:
## contrast          estimate          SE df t.ratio p.value
## female effect    2.28e-05 0.000164 23   0.139 0.8904
## male effect     -2.28e-05 0.000164 23  -0.139 0.8904
##
## Genotype = APOE33:
## contrast          estimate          SE df t.ratio p.value
## female effect   -9.30e-05 0.000198 23  -0.471 0.6423
## male effect     9.30e-05 0.000198 23   0.471 0.6423
##
## Genotype = APOE44:
## contrast          estimate          SE df t.ratio p.value
## female effect    2.83e-04 0.000171 23   1.654 0.1117
## male effect     -2.83e-04 0.000171 23  -1.654 0.1117
```

```
##
## P value adjustment: fdr method for 2 tests
#post hoc tests
summary(glht(lm_CPu_VOL, emm(pairwise ~ Genotype*Sex, contr="sidak"))) #consider save to file

##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##
## Estimate Std. Error t value Pr(>|t|)
## APOE22 female - APOE33 female == 0 7.692e-04 3.952e-04 1.946 0.39953
## APOE22 female - APOE44 female == 0 -9.720e-04 3.423e-04 -2.840 0.08562 .
## APOE22 female - APOE22 male == 0 4.568e-05 3.277e-04 0.139 0.99999
## APOE22 female - APOE33 male == 0 5.832e-04 3.423e-04 1.704 0.54111
## APOE22 female - APOE44 male == 0 -4.058e-04 3.423e-04 -1.186 0.83799
## APOE33 female - APOE44 female == 0 -1.741e-03 3.952e-04 -4.406 0.00248 **
## APOE33 female - APOE22 male == 0 -7.235e-04 3.827e-04 -1.891 0.43045
## APOE33 female - APOE33 male == 0 -1.860e-04 3.952e-04 -0.471 0.99671
## APOE33 female - APOE44 male == 0 -1.175e-03 3.952e-04 -2.973 0.06521 .
## APOE44 female - APOE22 male == 0 1.018e-03 3.277e-04 3.105 0.04932 *
## APOE44 female - APOE33 male == 0 1.555e-03 3.423e-04 4.544 0.00170 **
## APOE44 female - APOE44 male == 0 5.662e-04 3.423e-04 1.654 0.57147
## APOE22 male - APOE33 male == 0 5.375e-04 3.277e-04 1.640 0.58017
## APOE22 male - APOE44 male == 0 -4.515e-04 3.277e-04 -1.378 0.73782
## APOE33 male - APOE44 male == 0 -9.890e-04 3.423e-04 -2.889 0.07733 .
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
res<-summary(glht(lm_CPu_VOL, emm(pairwise ~ Genotype*Sex))) #consider save to file
#summary(as.glht(pairs(lm_CPu_VOL.emm), by = NULL))

summary(glht(lm_CPu_VOL, emm(pairwise ~ Genotype | Sex)))

## $`Sex = female`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##
## Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0 0.0007692 0.0003952 1.946 0.1479
## APOE22 - APOE44 == 0 -0.0009720 0.0003423 -2.840 0.0241 *
## APOE33 - APOE44 == 0 -0.0017413 0.0003952 -4.406 <0.001 ***
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
```

```
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  0.0005375  0.0003277   1.640   0.2495
## APOE22 - APOE44 == 0 -0.0004515  0.0003277  -1.378   0.3686
## APOE33 - APOE44 == 0 -0.0009890  0.0003423  -2.889   0.0217 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
summary(glht(lm_CPu_VOL, emm(pairwise ~ Sex | Genotype)))
```

```
## $`Genotype = APOE22`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0 4.568e-05 3.277e-04  0.139   0.89
## (Adjusted p values reported -- single-step method)
```

```
##
## $`Genotype = APOE33`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.0001860 0.0003952  -0.471   0.642
## (Adjusted p values reported -- single-step method)
```

```
##
## $`Genotype = APOE44`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0 0.0005662 0.0003423   1.654   0.112
## (Adjusted p values reported -- single-step method)
```

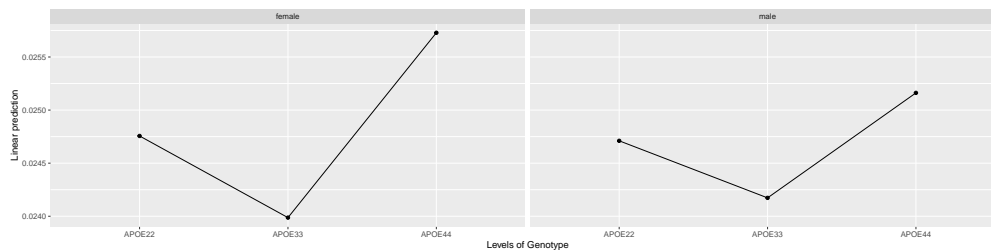
```
mypairs<-pairs(CPu_VOL.emm) #_consider save to file
pairs(CPu_VOL.emm, by="Genotype")
```

```
## Genotype = APOE22:
## contrast      estimate      SE df t.ratio p.value
```

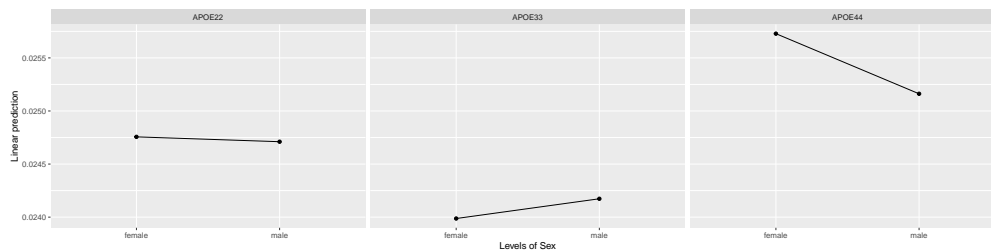
```
## female - male 4.57e-05 0.000328 23 0.139 0.8904
##
## Genotype = APOE33:
## contrast estimate SE df t.ratio p.value
## female - male -1.86e-04 0.000395 23 -0.471 0.6423
##
## Genotype = APOE44:
## contrast estimate SE df t.ratio p.value
## female - male 5.66e-04 0.000342 23 1.654 0.1117
pairs(CPu_VOL.emm, by="Sex")
```

```
## Sex = female:
## contrast estimate SE df t.ratio p.value
## APOE22 - APOE33 0.000769 0.000395 23 1.946 0.1486
## APOE22 - APOE44 -0.000972 0.000342 23 -2.840 0.0242
## APOE33 - APOE44 -0.001741 0.000395 23 -4.406 0.0006
##
## Sex = male:
## contrast estimate SE df t.ratio p.value
## APOE22 - APOE33 0.000538 0.000328 23 1.640 0.2496
## APOE22 - APOE44 -0.000451 0.000328 23 -1.378 0.3687
## APOE33 - APOE44 -0.000989 0.000342 23 -2.889 0.0217
##
## P value adjustment: tukey method for comparing a family of 3 estimates
print('CPu VOL')
```

```
## [1] "CPu VOL"
emmip(lm_CPu_VOL, ~ Genotype | Sex )
```

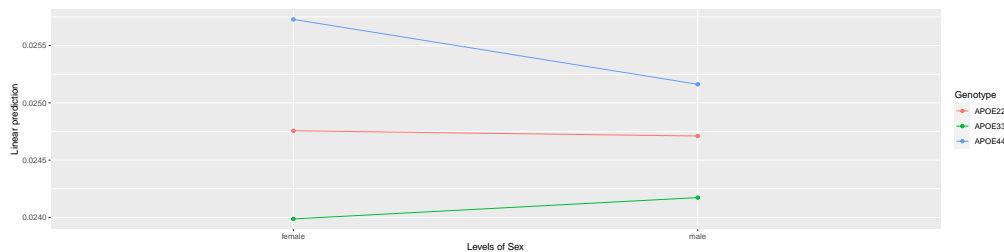


```
emmip(lm_CPu_VOL, ~ Sex | Genotype )
```



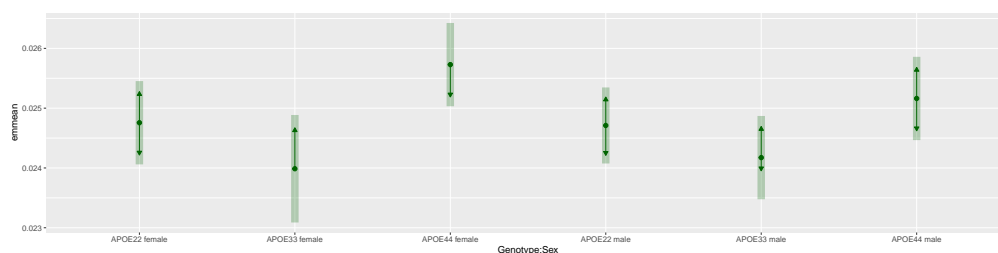
```
emmip(lm_CPu_VOL, Genotype~ Sex )
```





```
sink()
```

```
plot(CPu_VOL.emm, by = NULL, comparisons = TRUE, adjust = "mvt",
horizontal = FALSE, colors = "darkgreen", main = "CPu VOL")
```



```
ggsave(paste(outpath, 'CPu_VOL_emmeans.pdf', sep=''), plot = last_plot(), device='pdf', scale=1, width=16
```

```
'simple stats'
```

```
## [1] "simple stats"
```

```
print('CPu')
```

```
## [1] "CPu"
```

```
lm_CPu_FA <- lm(CPu ~ Genotype*Sex, geno_combined_FA)
summary(lm_CPu_FA)
```

```
##
```

```
## Call:
```

```
## lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
```

```
##
```

```
## Residuals:
```

```
##      Min       1Q   Median       3Q      Max
## -0.0157390 -0.0071250  0.0007236  0.0057157  0.0178542
```

```
##
```

```
## Coefficients:
```

```
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)      0.236449   0.004266  55.423  <2e-16 ***
## GenotypeAPOE33    -0.010244   0.006967  -1.470   0.1550
## GenotypeAPOE44    -0.014434   0.006033  -2.392   0.0253 *
## Sexmale           0.002017   0.005777   0.349   0.7302
## GenotypeAPOE33:Sexmale -0.001301  0.009050  -0.144   0.8869
## GenotypeAPOE44:Sexmale  0.009600  0.008353   1.149   0.2623
```

```
## ---
```

```
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
##
```

```
## Residual standard error: 0.00954 on 23 degrees of freedom
```

```
## Multiple R-squared:  0.3388, Adjusted R-squared:  0.195
## F-statistic: 2.357 on 5 and 23 DF,  p-value: 0.07251

lm_CPu_FA_f <- lm(CPu ~ Genotype, combo_f_FA)
#summary(lm_CPu_FA_f)
lm_CPu_FA_m <- lm(CPu ~ Genotype, combo_m_FA)
#summary(lm_CPu_FA_m)

#33333
sink(paste(outpath, "CPu_FA_SimpleModels.txt"))
mydata.lm <- lm(CPu ~ Genotype*Sex, data = geno_combined_FA)
anova(mydata.lm)

## Analysis of Variance Table
##
## Response: CPu
##           Df      Sum Sq   Mean Sq F value    Pr(>F)
## Genotype    2 0.00072293 0.00036146   3.9719 0.03298 *
## Sex          1 0.00018127 0.00018127   1.9919 0.17153
## Genotype:Sex  2 0.00016811 0.00008405   0.9236 0.41131
## Residuals   23 0.00209312 0.00009101
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

summary(glht(mydata.lm, emm(pairwise ~ Genotype|Sex, adjust="sidak")))
```

*#consider save to file*

```
## $`Sex = female`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0 0.010244  0.006967   1.470  0.3222
## APOE22 - APOE44 == 0 0.014434  0.006033   2.392  0.0628 .
## APOE33 - APOE44 == 0 0.004190  0.006967   0.601  0.8202
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0 0.011545  0.005777   1.999  0.135
## APOE22 - APOE44 == 0 0.004835  0.005777   0.837  0.684
## APOE33 - APOE44 == 0 -0.006711  0.006033  -1.112  0.516
## (Adjusted p values reported -- single-step method)
```

```

summary(glht(mydata.lm, emm(pairwise ~ Sex| Genotype)))

## $`Genotype = APOE22`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.002017  0.005777  -0.349    0.73
## (Adjusted p values reported -- single-step method)
##
## $`Genotype = APOE33`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.0007154  0.0069668  -0.103    0.919
## (Adjusted p values reported -- single-step method)
##
## $`Genotype = APOE44`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.011616  0.006033  -1.925    0.0666 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)

sink()
#3333

#omnibus testing
#https://cran.r-project.org/web/packages/emmeans/vignettes/confidence-intervals.html#adjust

sink(paste(outpath, "CPu_FA_SimpleStats.txt", sep=''))
'omnibus testing'

## [1] "omnibus testing"

joint_tests(ref_grid(lm_CPu_FA))

##   model term    df1 df2 F.ratio p.value
##   Genotype      2   23   3.860  0.0359
##   Sex           1   23   1.740  0.2001

```

```
## Genotype:Sex  2  23  0.924  0.4113
joint_tests(ref_grid(lm_CPu_FA), by = "Sex", adjust = "sidak")

## Sex = female:
## model term df1 df2 F.ratio p.value
## Genotype    2  23   2.978  0.0708
##
## Sex = male:
## model term df1 df2 F.ratio p.value
## Genotype    2  23   2.001  0.1581
joint_tests(ref_grid(lm_CPu_FA), by = "Genotype", adjust = "sidak" )

## Genotype = APOE22:
## model term df1 df2 F.ratio p.value
## Sex        1  23   0.122  0.7302
##
## Genotype = APOE33:
## model term df1 df2 F.ratio p.value
## Sex        1  23   0.011  0.9191
##
## Genotype = APOE44:
## model term df1 df2 F.ratio p.value
## Sex        1  23   3.707  0.0666
'anova'

## [1] "anova"
anova(lm_CPu_FA)

## Analysis of Variance Table
##
## Response: CPu
##           Df      Sum Sq    Mean Sq F value    Pr(>F)
## Genotype    2 0.00072293  0.00036146   3.9719  0.03298 *
## Sex         1 0.00018127  0.00018127   1.9919  0.17153
## Genotype:Sex  2 0.00016811  0.00008405   0.9236  0.41131
## Residuals   23 0.00209312  0.00009101
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

#anova(lm_CPu_FA_f)
#anova(lm_CPu_FA_m)

#post hoc comparison
CPu_FA.emm <- emmeans(lm_CPu_FA , ~ Genotype | Sex, adjust = "sidak")
contrast(CPu_FA.emm, simple = c("Genotype", "Sex")) #FDR

## contrast            estimate      SE df t.ratio p.value
## APOE22 female effect  0.00583 0.00393 23   1.486  0.3018
## APOE33 female effect -0.00441 0.00485 23  -0.909  0.4471
## APOE44 female effect -0.00860 0.00393 23  -2.190  0.1282
## APOE22 male effect    0.00785 0.00366 23   2.145  0.1282
## APOE33 male effect   -0.00369 0.00393 23  -0.941  0.4471
## APOE44 male effect    0.00302 0.00393 23   0.768  0.4502
##
```

```
## P value adjustment: fdr method for 6 tests
contrast(CPu_FA.emm, simple = list("Genotype", "Sex"))

## $`simple contrasts for Genotype`
## Sex = female:
## contrast      estimate      SE df t.ratio p.value
## APOE22 effect  0.008226 0.00367 23   2.240  0.1051
## APOE33 effect -0.002018 0.00419 23  -0.482  0.6343
## APOE44 effect -0.006208 0.00367 23  -1.691  0.1566
##
## Sex = male:
## contrast      estimate      SE df t.ratio p.value
## APOE22 effect  0.005460 0.00328 23   1.663  0.1650
## APOE33 effect -0.006085 0.00343 23  -1.772  0.1650
## APOE44 effect  0.000625 0.00343 23   0.182  0.8571
##
## P value adjustment: fdr method for 3 tests
##
## $`simple contrasts for Sex`
## Genotype = APOE22:
## contrast      estimate      SE df t.ratio p.value
## female effect -0.001008 0.00289 23  -0.349  0.7302
## male effect   0.001008 0.00289 23   0.349  0.7302
##
## Genotype = APOE33:
## contrast      estimate      SE df t.ratio p.value
## female effect -0.000358 0.00348 23  -0.103  0.9191
## male effect   0.000358 0.00348 23   0.103  0.9191
##
## Genotype = APOE44:
## contrast      estimate      SE df t.ratio p.value
## female effect -0.005808 0.00302 23  -1.925  0.0666
## male effect   0.005808 0.00302 23   1.925  0.0666
##
## P value adjustment: fdr method for 2 tests
#post hoc tests
summary(glht(lm_CPu_FA, emm(pairwise ~ Genotype*Sex, contr="sidak"))) #consider save to file

##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##
##               Estimate Std. Error t value Pr(>|t|)
## APOE22 female - APOE33 female == 0  0.0102443  0.0069668   1.470  0.6837
## APOE22 female - APOE44 female == 0  0.0144341  0.0060334   2.392  0.1988
## APOE22 female - APOE22 male == 0   -0.0020165  0.0057766  -0.349  0.9992
## APOE22 female - APOE33 male == 0    0.0095289  0.0060334   1.579  0.6176
## APOE22 female - APOE44 male == 0    0.0028180  0.0060334   0.467  0.9968
## APOE33 female - APOE44 female == 0  0.0041898  0.0069668   0.601  0.9898
## APOE33 female - APOE22 male == 0   -0.0122608  0.0067456  -1.818  0.4727
## APOE33 female - APOE33 male == 0   -0.0007154  0.0069668  -0.103  1.0000
```

```

## APOE33 female - APOE44 male == 0    -0.0074263    0.0069668    -1.066    0.8889
## APOE44 female - APOE22 male == 0    -0.0164506    0.0057766    -2.848    0.0841 .
## APOE44 female - APOE33 male == 0    -0.0049051    0.0060334    -0.813    0.9618
## APOE44 female - APOE44 male == 0    -0.0116161    0.0060334    -1.925    0.4110
## APOE22 male - APOE33 male == 0        0.0115455    0.0057766     1.999    0.3712
## APOE22 male - APOE44 male == 0        0.0048346    0.0057766     0.837    0.9569
## APOE33 male - APOE44 male == 0     -0.0067109    0.0060334    -1.112    0.8704
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)

res<-summary(glht(lm_CPu_FA, emm(pairwise ~ Genotype*Sex))) #consider save to file
#summary(as.glht(pairs(lm_CPu_FA.emm), by = NULL))

summary(glht(lm_CPu_FA, emm(pairwise ~ Genotype | Sex)))

## $`Sex = female`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  0.010244   0.006967   1.470   0.3223
## APOE22 - APOE44 == 0  0.014434   0.006033   2.392   0.0626 .
## APOE33 - APOE44 == 0  0.004190   0.006967   0.601   0.8202
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  0.011545   0.005777   1.999   0.135
## APOE22 - APOE44 == 0  0.004835   0.005777   0.837   0.684
## APOE33 - APOE44 == 0 -0.006711   0.006033  -1.112   0.516
## (Adjusted p values reported -- single-step method)

summary(glht(lm_CPu_FA, emm(pairwise ~ Sex | Genotype)))

## $`Genotype = APOE22`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)

```

```

## female - male == 0 -0.002017 0.005777 -0.349 0.73
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.0007154 0.0069668 -0.103 0.919
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.011616 0.006033 -1.925 0.0666 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)

mypairs<-pairs(CPu_FA.emm) #_consider save to file
pairs(CPu_FA.emm, by="Genotype")

## Genotype = APOE22:
## contrast      estimate      SE df t.ratio p.value
## female - male -0.002017 0.00578 23 -0.349 0.7302
##
## Genotype = APOE33:
## contrast      estimate      SE df t.ratio p.value
## female - male -0.000715 0.00697 23 -0.103 0.9191
##
## Genotype = APOE44:
## contrast      estimate      SE df t.ratio p.value
## female - male -0.011616 0.00603 23 -1.925 0.0666

pairs(CPu_FA.emm, by="Sex")

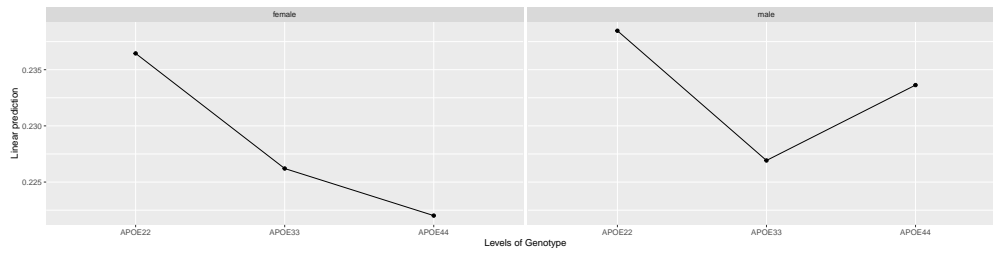
## Sex = female:
## contrast      estimate      SE df t.ratio p.value
## APOE22 - APOE33 0.01024 0.00697 23 1.470 0.3232
## APOE22 - APOE44 0.01443 0.00603 23 2.392 0.0630
## APOE33 - APOE44 0.00419 0.00697 23 0.601 0.8208
##
## Sex = male:
## contrast      estimate      SE df t.ratio p.value
## APOE22 - APOE33 0.01155 0.00578 23 1.999 0.1351

```

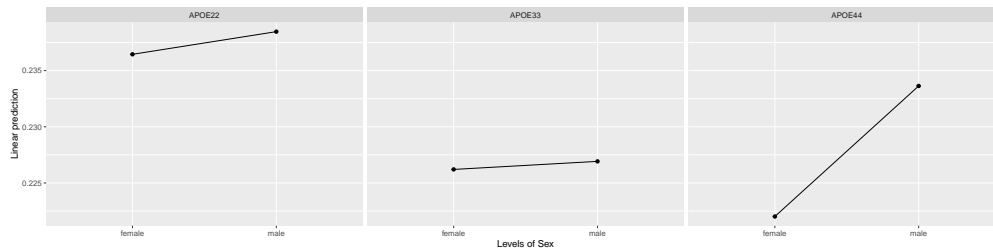
```
## APOE22 - APOE44 0.00483 0.00578 23 0.837 0.6844
## APOE33 - APOE44 -0.00671 0.00603 23 -1.112 0.5164
##
## P value adjustment: tukey method for comparing a family of 3 estimates
print('CPu FA')
```

```
## [1] "CPu FA"
```

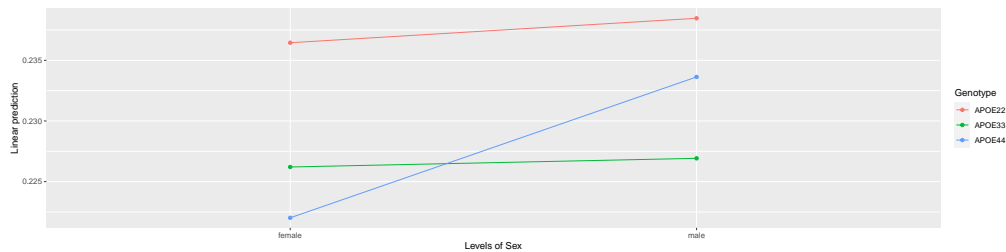
```
emmip(lm_CPu_FA, ~ Genotype | Sex )
```



```
emmip(lm_CPu_FA, ~ Sex | Genotype )
```

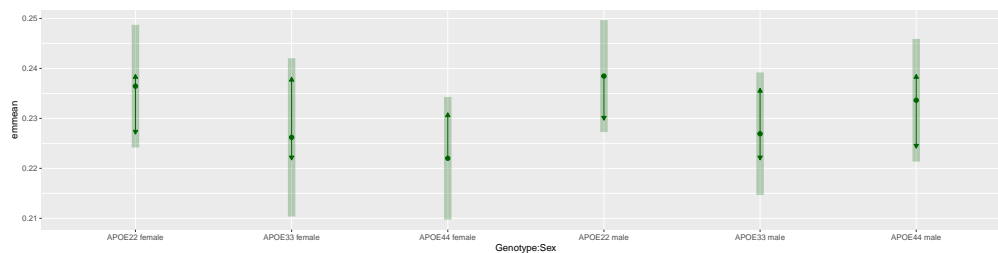


```
emmip(lm_CPu_FA, Genotype~ Sex )
```



```
sink()
```

```
plot(CPu_FA.emm, by = NULL, comparisons = TRUE, adjust = "mvt",
horizontal = FALSE, colors = "darkgreen", main = "CPu FA")
```



```
ggsave(paste(outpath, 'CPu_FA_emmeans.pdf', sep = ''), plot = last_plot(), device = 'pdf', scale = 1, width = 16,
```



```

'simple stats'

## [1] "simple stats"
print('CPu')

## [1] "CPu"

lm_CPu_DEG <- lm(CPu ~ Genotype*Sex, geno_combined_DEG)
summary(lm_CPu_DEG)

##
## Call:
## lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -31667 -11917  -3800   14800   34333
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)      218000      8706   25.040  <2e-16 ***
## GenotypeAPOE33      22667     14217    1.594    0.125
## GenotypeAPOE44      12200     12312    0.991    0.332
## Sexmale           9917      11788    0.841    0.409
## GenotypeAPOE33:Sexmale -9783     18468   -0.530    0.601
## GenotypeAPOE44:Sexmale -18317     17045   -1.075    0.294
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 19470 on 23 degrees of freedom
## Multiple R-squared:  0.1853, Adjusted R-squared:  0.008217
## F-statistic: 1.046 on 5 and 23 DF,  p-value: 0.4148

lm_CPu_DEG_f <- lm(CPu ~ Genotype, combo_f_FA)
#summary(lm_CPu_DEG_f)
lm_CPu_DEG_m <- lm(CPu ~ Genotype, combo_m_FA)
#summary(lm_CPu_DEG_f)

#33333
sink(paste(outpath, "CPu_DEG_SimpleModels.txt"))
mydata.lm <- lm(CPu ~ Genotype*Sex, data = geno_combined_DEG)
anova(mydata.lm)

## Analysis of Variance Table
##
## Response: CPu
##          Df      Sum Sq   Mean Sq F value Pr(>F)
## Genotype    2 1538125392 769062696  2.0294 0.1543
## Sex          1   5580291    5580291  0.0147 0.9045
## Genotype:Sex  2  439053800 219526900  0.5793 0.5683
## Residuals   23  8716275000 378968478

summary(glht(mydata.lm, emm(pairwise ~ Genotype|Sex, adjust="sidak")))
```

*#consider save to file*

```

## $`Sex = female`
##

```

```

## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0   -22667     14217  -1.594    0.267
## APOE22 - APOE44 == 0   -12200     12312  -0.991    0.589
## APOE33 - APOE44 == 0    10467     14217   0.736    0.744
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0   -12883     11788  -1.093    0.528
## APOE22 - APOE44 == 0     6117     11788   0.519    0.863
## APOE33 - APOE44 == 0    19000     12312   1.543    0.290
## (Adjusted p values reported -- single-step method)
summary(glht(mydata.lm, emm(pairwise ~ Sex| Genotype)))

## $`Genotype = APOE22`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -9917     11788  -0.841    0.409
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -133.3    14216.8  -0.009    0.993
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
## Simultaneous Tests for General Linear Hypotheses
##

```

```
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0      8400      12312   0.682   0.502
## (Adjusted p values reported -- single-step method)

sink()
#3333

#omnibus testing
#https://cran.r-project.org/web/packages/emmeans/vignettes/confidence-intervals.html#adjust

sink(paste(outpath, "CPu_DEG_SimpleStats.txt", sep=''))
'omnibus testing'

## [1] "omnibus testing"

joint_tests(ref_grid(lm_CPu_DEG))

##   model term    df1 df2 F.ratio p.value
##   Genotype      2  23   2.004  0.1577
##   Sex           1  23   0.006  0.9414
##   Genotype:Sex   2  23   0.579  0.5683

joint_tests(ref_grid(lm_CPu_DEG), by = "Sex", adjust = "sidak")

## Sex = female:
##   model term df1 df2 F.ratio p.value
##   Genotype   2  23   1.327  0.2849
##
## Sex = male:
##   model term df1 df2 F.ratio p.value
##   Genotype   2  23   1.247  0.3060

joint_tests(ref_grid(lm_CPu_DEG), by = "Genotype", adjust = "sidak" )

## Genotype = APOE22:
##   model term df1 df2 F.ratio p.value
##   Sex        1  23   0.708  0.4089
##
## Genotype = APOE33:
##   model term df1 df2 F.ratio p.value
##   Sex        1  23   0.000  0.9926
##
## Genotype = APOE44:
##   model term df1 df2 F.ratio p.value
##   Sex        1  23   0.465  0.5019

'anova'

## [1] "anova"

anova(lm_CPu_DEG)

## Analysis of Variance Table
##
## Response: CPu
##           Df      Sum Sq   Mean Sq F value Pr(>F)
```

```
## Genotype      2 1538125392 769062696 2.0294 0.1543
## Sex           1   5580291   5580291 0.0147 0.9045
## Genotype:Sex  2  439053800 219526900 0.5793 0.5683
## Residuals    23 8716275000 378968478

#anova(lm_CPu_DEG_f)
#anova(lm_CPu_DEG_m)

#post hoc comparison
CPu_DEG.emm <- emmeans(lm_CPu_DEG , ~ Genotype | Sex, adjust = "sidak")
contrast(CPu_DEG.emm, simple = c("Genotype", "Sex")) #FDR

## contrast            estimate    SE df t.ratio p.value
## APOE22 female effect  -11897 8013 23  -1.485  0.4842
## APOE33 female effect   10769 9894 23   1.088  0.4842
## APOE44 female effect    303 8013 23   0.038  0.9702
## APOE22 male effect    -1981 7469 23  -0.265  0.9519
## APOE33 male effect    10903 8013 23   1.361  0.4842
## APOE44 male effect    -8097 8013 23  -1.010  0.4842
##
## P value adjustment: fdr method for 6 tests
contrast(CPu_DEG.emm, simple = list("Genotype", "Sex"))

## $`simple contrasts for Genotype`
## Sex = female:
## contrast            estimate    SE df t.ratio p.value
## APOE22 effect      -11622 7493 23  -1.551  0.3134
## APOE33 effect       11044 8543 23   1.293  0.3134
## APOE44 effect        578 7493 23   0.077  0.9392
##
## Sex = male:
## contrast            estimate    SE df t.ratio p.value
## APOE22 effect      -2256 6702 23  -0.337  0.7395
## APOE33 effect       10628 7009 23   1.516  0.3667
## APOE44 effect      -8372 7009 23  -1.195  0.3667
##
## P value adjustment: fdr method for 3 tests
##
## $`simple contrasts for Sex`
## Genotype = APOE22:
## contrast            estimate    SE df t.ratio p.value
## female effect     -4958.3 5894 23  -0.841  0.4089
## male effect       4958.3 5894 23   0.841  0.4089
##
## Genotype = APOE33:
## contrast            estimate    SE df t.ratio p.value
## female effect      -66.7 7108 23  -0.009  0.9926
## male effect        66.7 7108 23   0.009  0.9926
##
## Genotype = APOE44:
## contrast            estimate    SE df t.ratio p.value
## female effect     4200.0 6156 23   0.682  0.5019
## male effect     -4200.0 6156 23  -0.682  0.5019
##
```

```
## P value adjustment: fdr method for 2 tests
#post hoc tests
summary(glht(lm_CPu_DEG, emm(pairwise ~ Genotype*Sex, contr="sidak"))) #consider save to file

##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##
## Estimate Std. Error t value Pr(>|t|)
## APOE22 female - APOE33 female == 0 -22666.7 14216.8 -1.594 0.608
## APOE22 female - APOE44 female == 0 -12200.0 12312.1 -0.991 0.916
## APOE22 female - APOE22 male == 0 -9916.7 11787.9 -0.841 0.956
## APOE22 female - APOE33 male == 0 -22800.0 12312.1 -1.852 0.453
## APOE22 female - APOE44 male == 0 -3800.0 12312.1 -0.309 1.000
## APOE33 female - APOE44 female == 0 10466.7 14216.8 0.736 0.975
## APOE33 female - APOE22 male == 0 12750.0 13765.3 0.926 0.935
## APOE33 female - APOE33 male == 0 -133.3 14216.8 -0.009 1.000
## APOE33 female - APOE44 male == 0 18866.7 14216.8 1.327 0.766
## APOE44 female - APOE22 male == 0 2283.3 11787.9 0.194 1.000
## APOE44 female - APOE33 male == 0 -10600.0 12312.1 -0.861 0.952
## APOE44 female - APOE44 male == 0 8400.0 12312.1 0.682 0.982
## APOE22 male - APOE33 male == 0 -12883.3 11787.9 -1.093 0.878
## APOE22 male - APOE44 male == 0 6116.7 11787.9 0.519 0.995
## APOE33 male - APOE44 male == 0 19000.0 12312.1 1.543 0.640
## (Adjusted p values reported -- single-step method)

res<-summary(glht(lm_CPu_DEG, emm(pairwise ~ Genotype*Sex))) #consider save to file
#summary(as.glht(pairs(lm_CPu_DEG.emm), by = NULL))

summary(glht(lm_CPu_DEG, emm(pairwise ~ Genotype | Sex)))

## $`Sex = female`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##
## Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0 -22667 14217 -1.594 0.267
## APOE22 - APOE44 == 0 -12200 12312 -0.991 0.589
## APOE33 - APOE44 == 0 10467 14217 0.736 0.744
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
```

```

##               Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0   -12883      11788  -1.093    0.528
## APOE22 - APOE44 == 0    6117      11788   0.519    0.863
## APOE33 - APOE44 == 0   19000      12312   1.543    0.290
## (Adjusted p values reported -- single-step method)
summary(glht(lm_CPu_DEG, emm(pairwise ~ Sex | Genotype)))

## $`Genotype = APOE22`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -9917      11788  -0.841    0.409
## (Adjusted p values reported -- single-step method)
##
## $`Genotype = APOE33`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -133.3    14216.8  -0.009    0.993
## (Adjusted p values reported -- single-step method)
##
## $`Genotype = APOE44`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0    8400      12312   0.682    0.502
## (Adjusted p values reported -- single-step method)
mypairs<-pairs(CPu_DEG.emm) #_consider save to file
pairs(CPu_DEG.emm, by="Genotype")

## Genotype = APOE22:
##   contrast      estimate      SE df t.ratio p.value
## female - male   -9917 11788 23  -0.841  0.4089
##
## Genotype = APOE33:
##   contrast      estimate      SE df t.ratio p.value
## female - male   -133 14217 23  -0.009  0.9926
##
## Genotype = APOE44:

```

```
## contrast      estimate      SE df t.ratio p.value
## female - male      8400 12312 23   0.682  0.5019
```

```
pairs(CPu_DEG.emm, by="Sex")
```

```
## Sex = female:
## contrast      estimate      SE df t.ratio p.value
## APOE22 - APOE33 -22667 14217 23   -1.594  0.2682
## APOE22 - APOE44 -12200 12312 23   -0.991  0.5899
## APOE33 - APOE44  10467 14217 23    0.736  0.7448
##
```

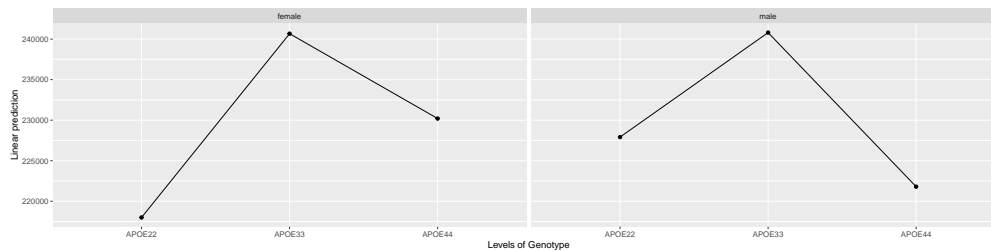
```
## Sex = male:
## contrast      estimate      SE df t.ratio p.value
## APOE22 - APOE33 -12883 11788 23   -1.093  0.5279
## APOE22 - APOE44   6117 11788 23    0.519  0.8629
## APOE33 - APOE44  19000 12312 23    1.543  0.2901
##
```

```
## P value adjustment: tukey method for comparing a family of 3 estimates
```

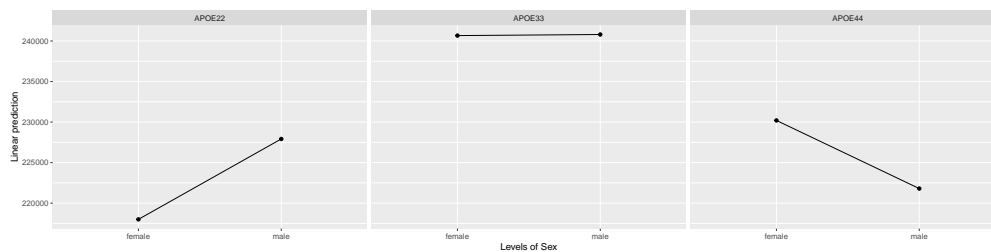
```
print('CPu DEG')
```

```
## [1] "CPu DEG"
```

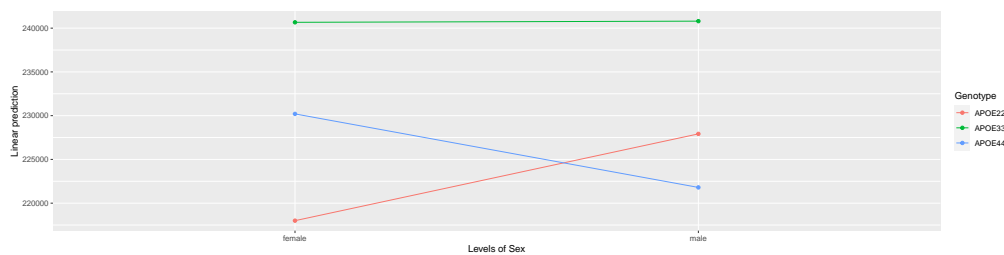
```
emmip(lm_CPu_DEG, ~ Genotype | Sex )
```



```
emmip(lm_CPu_DEG, ~ Sex | Genotype )
```



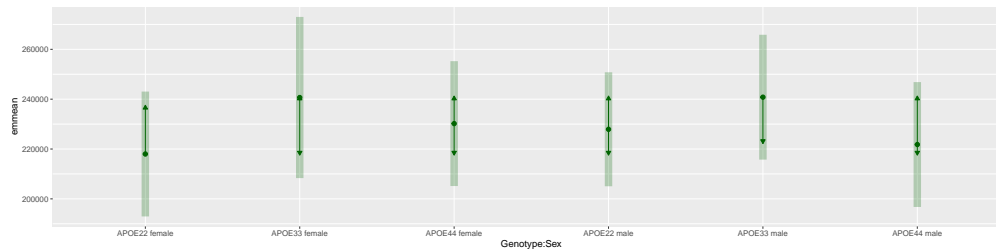
```
emmip(lm_CPu_DEG, Genotype~ Sex )
```



```
sink()
```

```
plot(CPu_DEG.emm, by = NULL, comparisons = TRUE, adjust = "mvt",
```

```
horizontal = FALSE, colors = "darkgreen", main = "CPu DEG")
```



```
ggsave(paste(outpath, 'CPu_DEG_emmeans.pdf', sep=''), plot = last_plot(), device='pdf', scale=1, width=16
```

```
'simple stats'
```

```
## [1] "simple stats"
```

```
print('CPu')
```

```
## [1] "CPu"
```

```
lm_CPu_CLUS <- lm(CPu ~ Genotype*Sex, geno_combined_CLUS)
summary(lm_CPu_CLUS)
```

```
##
## Call:
## lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -70.017 -20.873  -5.233  11.696  89.933
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)       350.33      16.50   21.229  <2e-16 ***
## GenotypeAPOE33        50.99      26.95    1.892   0.0711 .
## GenotypeAPOE44        33.83      23.34    1.449   0.1607
## Sexmale             17.17      22.34    0.768   0.4501
## GenotypeAPOE33:Sexmale -29.15      35.01   -0.833   0.4135
## GenotypeAPOE44:Sexmale -27.20      32.31   -0.842   0.4086
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 36.9 on 23 degrees of freedom
## Multiple R-squared:  0.1803, Adjusted R-squared:  0.002162
## F-statistic: 1.012 on 5 and 23 DF,  p-value: 0.4332

lm_CPu_CLUS_f <- lm(CPu ~ Genotype, combo_f_FA)
#summary(lm_CPu_CLUS_f)
lm_CPu_CLUS_m <- lm(CPu ~ Genotype, combo_m_FA)
#summary(lm_CPu_CLUS_f)

#33333
sink(paste(outpath, "CPu_CLUS_SimpleModels.txt"))
mydata.lm <- lm(CPu ~ Genotype*Sex, data = geno_combined_CLUS)
anova(mydata.lm)
```



```

## Analysis of Variance Table
##
## Response: CPu
##           Df Sum Sq Mean Sq F value Pr(>F)
## Genotype    2  5566.0  2783.01   2.0439 0.1524
## Sex          1    0.1    0.07   0.0001 0.9943
## Genotype:Sex  2  1324.6   662.30   0.4864 0.6210
## Residuals   23 31317.3  1361.62

summary(glht(mydata.lm, emm(pairwise ~ Genotype|Sex, adjust="sidak")))
```

*#consider save to file*

```

## $`Sex = female`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0   -50.99     26.95  -1.892   0.163
## APOE22 - APOE44 == 0   -33.83     23.34  -1.449   0.332
## APOE33 - APOE44 == 0    17.17     26.95   0.637   0.801
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  -21.839     22.344  -0.977   0.598
## APOE22 - APOE44 == 0   -6.629     22.344  -0.297   0.953
## APOE33 - APOE44 == 0   15.210     23.338   0.652   0.793
## (Adjusted p values reported -- single-step method)

summary(glht(mydata.lm, emm(pairwise ~ Sex| Genotype)))

## $`Genotype = APOE22`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##           Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -17.17     22.34  -0.768   0.45
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
##   Simultaneous Tests for General Linear Hypotheses

```

```

##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##             Estimate Std. Error t value Pr(>|t|)
## female - male == 0    11.98     26.95   0.445   0.661
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##             Estimate Std. Error t value Pr(>|t|)
## female - male == 0    10.03     23.34   0.43   0.671
## (Adjusted p values reported -- single-step method)

sink()
#3333

#omnibus testing
#https://cran.r-project.org/web/packages/emmeans/vignettes/confidence-intervals.html#adjust

sink(paste(outpath, "CPu_CLUS_SimpleStats.txt", sep=''))
'omnibus testing'

## [1] "omnibus testing"

joint_tests(ref_grid(lm_CPu_CLUS))

##   model term   df1 df2 F.ratio p.value
##   Genotype     2  23   2.233  0.1299
##   Sex           1  23   0.013  0.9094
##   Genotype:Sex   2  23   0.486  0.6210

joint_tests(ref_grid(lm_CPu_CLUS), by = "Sex", adjust = "sidak")

## Sex = female:
##   model term df1 df2 F.ratio p.value
##   Genotype   2  23   2.035  0.1536
##
## Sex = male:
##   model term df1 df2 F.ratio p.value
##   Genotype   2  23   0.491  0.6181

joint_tests(ref_grid(lm_CPu_CLUS), by = "Genotype", adjust = "sidak" )

## Genotype = APOE22:
##   model term df1 df2 F.ratio p.value
##   Sex        1  23   0.590  0.4501
##
## Genotype = APOE33:
##   model term df1 df2 F.ratio p.value
##   Sex        1  23   0.198  0.6607

```

```
##
## Genotype = APOE44:
## model term df1 df2 F.ratio p.value
## Sex          1  23   0.185  0.6714

'anova'

## [1] "anova"

anova(lm_CPu_CLUS)

## Analysis of Variance Table
##
## Response: CPu
##           Df Sum Sq Mean Sq F value Pr(>F)
## Genotype    2  5566.0  2783.01   2.0439 0.1524
## Sex          1    0.1    0.07   0.0001 0.9943
## Genotype:Sex  2  1324.6   662.30   0.4864 0.6210
## Residuals   23 31317.3  1361.62

#anova(lm_CPu_CLUS_f)
#anova(lm_CPu_CLUS_m)

#post hoc comparison
CPu_CLUS.emm <- emmeans(lm_CPu_CLUS , ~ Genotype | Sex, adjust = "sidak")
contrast(CPu_CLUS.emm, simple = c("Genotype", "Sex")) #FDR

## contrast          estimate    SE df t.ratio p.value
## APOE22 female effect  -27.47 15.2 23  -1.808  0.5020
## APOE33 female effect   23.53 18.8 23   1.254  0.6668
## APOE44 female effect    6.36 15.2 23   0.419  0.8113
## APOE22 male effect   -10.30 14.2 23  -0.727  0.7116
## APOE33 male effect    11.54 15.2 23   0.760  0.7116
## APOE44 male effect    -3.67 15.2 23  -0.241  0.8113
##
## P value adjustment: fdr method for 6 tests

contrast(CPu_CLUS.emm, simple = list("Genotype", "Sex"))

## $`simple contrasts for Genotype`
## Sex = female:
## contrast          estimate    SE df t.ratio p.value
## APOE22 effect   -28.27 14.2 23  -1.991  0.1756
## APOE33 effect    22.72 16.2 23   1.403  0.2610
## APOE44 effect     5.55 14.2 23   0.391  0.6994
##
## Sex = male:
## contrast          estimate    SE df t.ratio p.value
## APOE22 effect    -9.49 12.7 23  -0.747  0.6940
## APOE33 effect    12.35 13.3 23   0.930  0.6940
## APOE44 effect    -2.86 13.3 23  -0.215  0.8314
##
## P value adjustment: fdr method for 3 tests
##
## $`simple contrasts for Sex`
## Genotype = APOE22:
## contrast          estimate    SE df t.ratio p.value
```

```

## female effect      -8.58 11.2 23  -0.768  0.4501
## male effect        8.58 11.2 23   0.768  0.4501
##
## Genotype = APOE33:
## contrast      estimate    SE df t.ratio p.value
## female effect    5.99 13.5 23   0.445  0.6607
## male effect    -5.99 13.5 23  -0.445  0.6607
##
## Genotype = APOE44:
## contrast      estimate    SE df t.ratio p.value
## female effect    5.01 11.7 23   0.430  0.6714
## male effect    -5.01 11.7 23  -0.430  0.6714
##
## P value adjustment: fdr method for 2 tests
#post hoc tests
summary(glht(lm_CPu_CLUS, emm(pairwise ~ Genotype*Sex, contr="sidak"))) #consider save to file

##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##
##               Estimate Std. Error t value Pr(>|t|)
## APOE22 female - APOE33 female == 0 -50.993    26.948  -1.892    0.430
## APOE22 female - APOE44 female == 0 -33.826    23.338  -1.449    0.696
## APOE22 female - APOE22 male == 0  -17.169    22.344  -0.768    0.970
## APOE22 female - APOE33 male == 0  -39.008    23.338  -1.671    0.561
## APOE22 female - APOE44 male == 0  -23.798    23.338  -1.020    0.906
## APOE33 female - APOE44 female == 0   17.167    26.948   0.637    0.987
## APOE33 female - APOE22 male == 0   33.823    26.092   1.296    0.783
## APOE33 female - APOE33 male == 0   11.985    26.948   0.445    0.997
## APOE33 female - APOE44 male == 0   27.195    26.948   1.009    0.909
## APOE44 female - APOE22 male == 0   16.657    22.344   0.745    0.974
## APOE44 female - APOE33 male == 0   -5.182    23.338  -0.222    1.000
## APOE44 female - APOE44 male == 0   10.028    23.338   0.430    0.998
## APOE22 male - APOE33 male == 0   -21.839    22.344  -0.977    0.920
## APOE22 male - APOE44 male == 0    -6.629    22.344  -0.297    1.000
## APOE33 male - APOE44 male == 0    15.210    23.338   0.652    0.985
## (Adjusted p values reported -- single-step method)

res<-summary(glht(lm_CPu_CLUS, emm(pairwise ~ Genotype*Sex))) #consider save to file
#summary(as.glht(pairs(lm_CPu_CLUS.emm), by = NULL))

summary(glht(lm_CPu_CLUS, emm(pairwise ~ Genotype | Sex)))

## $`Sex = female`
##
## Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##
##               Estimate Std. Error t value Pr(>|t|)

```

```

## APOE22 - APOE33 == 0   -50.99      26.95   -1.892    0.163
## APOE22 - APOE44 == 0   -33.83      23.34   -1.449    0.332
## APOE33 - APOE44 == 0    17.17      26.95    0.637    0.801
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  -21.839     22.344  -0.977    0.598
## APOE22 - APOE44 == 0   -6.629     22.344  -0.297    0.953
## APOE33 - APOE44 == 0   15.210     23.338   0.652    0.793
## (Adjusted p values reported -- single-step method)
summary(glht(lm_CPu_CLUS, emm(pairwise ~ Sex | Genotype)))

## $`Genotype = APOE22`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -17.17      22.34  -0.768    0.45
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0    11.98      26.95   0.445    0.661
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = CPu ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##               Estimate Std. Error t value Pr(>|t|)
## female - male == 0    10.03      23.34   0.43     0.671
## (Adjusted p values reported -- single-step method)

```

```
mypairs<-pairs(CPu_CLUS.emm) #_consider save to file
pairs(CPu_CLUS.emm, by="Genotype")
```

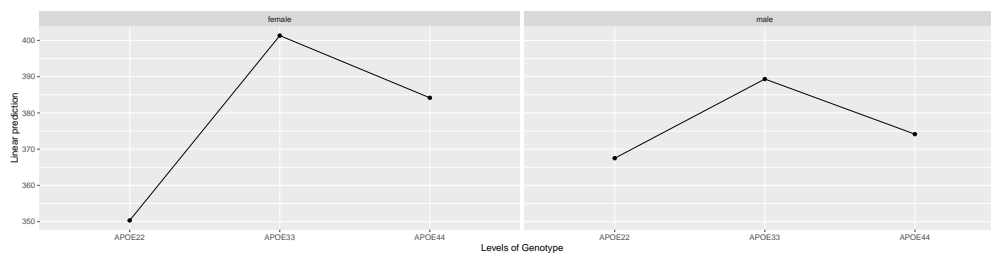
```
## Genotype = APOE22:
## contrast      estimate    SE df t.ratio p.value
## female - male   -17.2 22.3 23  -0.768  0.4501
##
## Genotype = APOE33:
## contrast      estimate    SE df t.ratio p.value
## female - male    12.0 26.9 23   0.445  0.6607
##
## Genotype = APOE44:
## contrast      estimate    SE df t.ratio p.value
## female - male    10.0 23.3 23   0.430  0.6714
```

```
pairs(CPu_CLUS.emm, by="Sex")
```

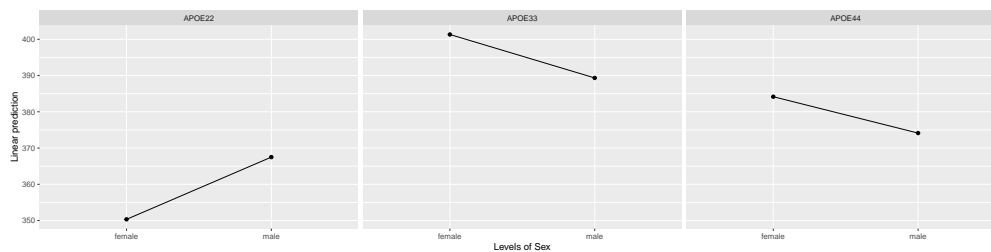
```
## Sex = female:
## contrast      estimate    SE df t.ratio p.value
## APOE22 - APOE33  -50.99 26.9 23  -1.892  0.1636
## APOE22 - APOE44  -33.83 23.3 23  -1.449  0.3332
## APOE33 - APOE44   17.17 26.9 23   0.637  0.8014
##
## Sex = male:
## contrast      estimate    SE df t.ratio p.value
## APOE22 - APOE33  -21.84 22.3 23  -0.977  0.5981
## APOE22 - APOE44   -6.63 22.3 23  -0.297  0.9527
## APOE33 - APOE44   15.21 23.3 23   0.652  0.7933
##
## P value adjustment: tukey method for comparing a family of 3 estimates
print('CPu CLUS')
```

```
## [1] "CPu CLUS"
```

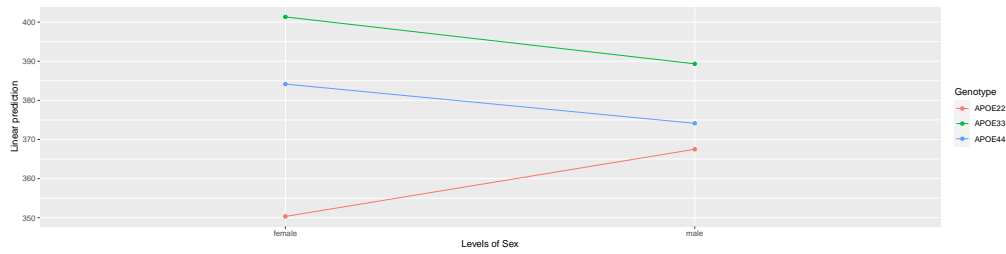
```
emmip(lm_CPu_CLUS, ~ Genotype | Sex )
```



```
emmip(lm_CPu_CLUS, ~ Sex | Genotype )
```

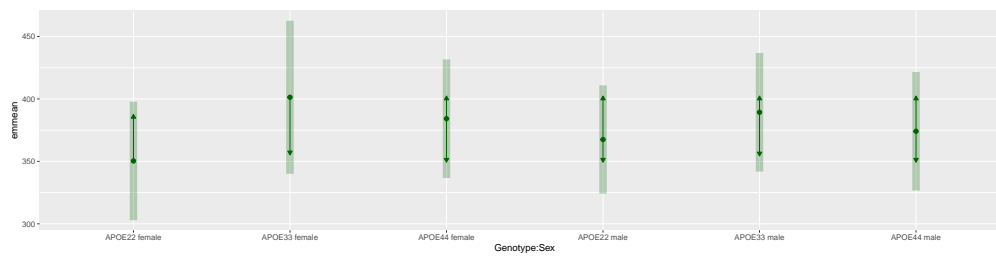


```
emmip(lm_CPu_CLUS, Genotype~ Sex )
```



```
sink()
```

```
plot(CPu_CLUS.emm, by = NULL, comparisons = TRUE, adjust = "mvt",  
horizontal = FALSE, colors = "darkgreen", main = "CPu CLUS")
```



```
ggsave(paste(outpath, 'CPu_CLUS_emmeans.pdf', sep=''), plot = last_plot(), device='pdf', scale=1, width=1000, height=1000)
```