SimpleViolins

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Contents

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 Hc

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`.
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## 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`.
```
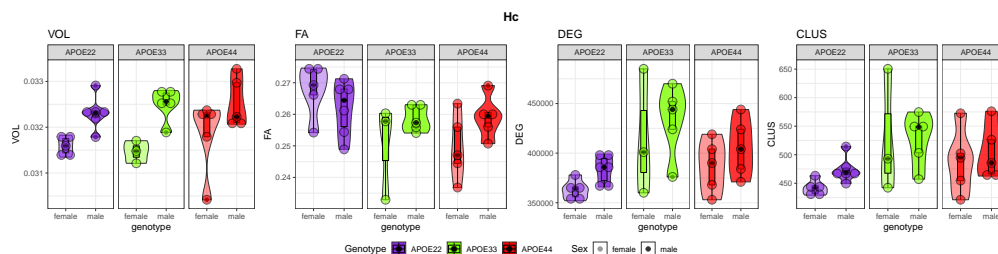


```
print('Hc')
```

```
## [1] "Hc"
```

```
lm_Hc_VOL <- lm(Hc ~ Genotype*Sex, geno_combined_VOL)
summary(lm_Hc_VOL)
```

```
## 
## Call:
## lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
## 
## Residuals:
##        Min         1Q     Median         3Q        Max
## -1.413e-03 -2.449e-04  1.222e-05  2.522e-04  7.467e-04
## 
## Coefficients:
##                        Estimate Std. Error t value Pr(>|t|)
## (Intercept)           3.159e-02  2.171e-04 145.497   <2e-16 ***
## GenotypeAPOE33       -1.273e-04  3.546e-04  -0.359   0.7228
## GenotypeAPOE44        2.371e-04  3.071e-04   0.772   0.4478
## Sexmale               7.183e-04  2.940e-04   2.443   0.0227 *
## GenotypeAPOE33:Sexmale 3.125e-04  4.606e-04   0.678   0.5043
## GenotypeAPOE44:Sexmale -1.995e-05 4.251e-04  -0.047   0.9630
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## 
## Residual standard error: 0.0004855 on 23 degrees of freedom
## Multiple R-squared:  0.4708, Adjusted R-squared:  0.3557
## F-statistic: 4.092 on 5 and 23 DF,  p-value: 0.00837
lm_Hc_VOL_f <- lm(Hc ~ Genotype, combo_f_FA)
#summary(lm_Hc_VOL_f)
lm_Hc_VOL_m <- lm(Hc ~ Genotype, combo_m_FA)
#summary(lm_Hc_VOL_f)


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

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

## Analysis of Variance Table
## 
## Response: Hc
##              Df     Sum Sq     Mean Sq F value     Pr(>F)
## Genotype      2 2.0430e-07 1.0210e-07  0.4333  0.6535491
## Sex           1 4.4749e-06 4.4749e-06 18.9821  0.0002313 ***
## Genotype:Sex  2 1.4360e-07 7.1800e-08  0.3047  0.7402957
## Residuals    23 5.4221e-06 2.3570e-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 = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                          Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0   0.0001273  0.0003546    0.359    0.931
## APOE22 - APOE44 == 0  -0.0002371  0.0003071   -0.772    0.723
## APOE33 - APOE44 == 0  -0.0003645  0.0003546   -1.028    0.566
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                          Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  -1.852e-04  2.940e-04   -0.630    0.805
## APOE22 - APOE44 == 0  -2.172e-04  2.940e-04   -0.739    0.743
## APOE33 - APOE44 == 0  -3.199e-05  3.071e-04   -0.104    0.994
## (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 = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                          Estimate Std. Error t value Pr(>|t|)
## female - male == 0  -0.0007183  0.0002940   -2.443   0.0227 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                          Estimate Std. Error t value Pr(>|t|)
## female - male == 0  -0.0010308  0.0003546   -2.907  0.00794 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
```

```
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.0006983  0.0003071  -2.274   0.0326 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
```

```
sink()
#3333

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

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

```
joint_tests(ref_grid(lm_Hc_VOL))
```

```
##  model term    df1 df2 F.ratio p.value
##  Genotype        2  23   0.645  0.5337
##  Sex             1  23  19.545  0.0002
##  Genotype:Sex    2  23   0.305  0.7403
```

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

```
## Sex = female:
##  model term df1 df2 F.ratio p.value
##  Genotype     2  23   0.594  0.5603
##
## Sex = male:
##  model term df1 df2 F.ratio p.value
##  Genotype     2  23   0.327  0.7241
```

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

```
## Genotype = APOE22:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   5.969  0.0227
##
## Genotype = APOE33:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   8.451  0.0079
##
## Genotype = APOE44:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   5.172  0.0326
```

```
'anova'
```

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

```
anova(lm_Hc_VOL)
```

```
## Analysis of Variance Table
##
## Response: Hc
```

```
##                 Df     Sum Sq    Mean Sq F value    Pr(>F)
## Genotype        2 2.0430e-07 1.0210e-07   0.4333 0.6535491
## Sex             1 4.4749e-06 4.4749e-06  18.9821 0.0002313 ***
## Genotype:Sex    2 1.4360e-07 7.1800e-08   0.3047 0.7402957
## Residuals      23 5.4221e-06 2.3570e-07
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
#anova(lm_Hc_VOL_f)
#anova(lm_Hc_VOL_m)

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

```
##  contrast            estimate       SE df t.ratio p.value
##  APOE22 female effect -0.000445 0.000200 23  -2.224  0.0544
##  APOE33 female effect -0.000572 0.000247 23  -2.317  0.0544
##  APOE44 female effect -0.000207 0.000200 23  -1.038  0.3103
##  APOE22 male effect    0.000274 0.000186 23   1.470  0.1863
##  APOE33 male effect    0.000459 0.000200 23   2.296  0.0544
##  APOE44 male effect    0.000491 0.000200 23   2.456  0.0544
##
## P value adjustment: fdr method for 6 tests
```

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

```
## $`simple contrasts for Genotype`
## Sex = female:
##  contrast       estimate       SE df t.ratio p.value
##  APOE22 effect -3.66e-05 0.000187 23  -0.196  0.8464
##  APOE33 effect -1.64e-04 0.000213 23  -0.769  0.6743
##  APOE44 effect  2.01e-04 0.000187 23   1.073  0.6743
##
## Sex = male:
##  contrast       estimate       SE df t.ratio p.value
##  APOE22 effect -1.34e-04 0.000167 23  -0.802  0.7728
##  APOE33 effect  5.11e-05 0.000175 23   0.292  0.7728
##  APOE44 effect  8.31e-05 0.000175 23   0.475  0.7728
##
## P value adjustment: fdr method for 3 tests
##
## $`simple contrasts for Sex`
## Genotype = APOE22:
##  contrast       estimate       SE df t.ratio p.value
##  female effect -0.000359 0.000147 23  -2.443  0.0227
##  male effect    0.000359 0.000147 23   2.443  0.0227
##
## Genotype = APOE33:
##  contrast       estimate       SE df t.ratio p.value
##  female effect -0.000515 0.000177 23  -2.907  0.0079
##  male effect    0.000515 0.000177 23   2.907  0.0079
##
## Genotype = APOE44:
##  contrast       estimate       SE df t.ratio p.value
```

```
##  female effect -0.000349 0.000154 23  -2.274  0.0326
##  male effect    0.000349 0.000154 23   2.274  0.0326
##
## P value adjustment: fdr method for 2 tests
```
```r
#post hoc tests
summary(glht(lm_Hc_VOL, emm(pairwise ~ Genotype*Sex, contr="sidak"))) #consider save to file
```
```
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                                Estimate Std. Error t value Pr(>|t|)
## APOE22 female - APOE33 female == 0  1.273e-04  3.546e-04   0.359   0.9991
## APOE22 female - APOE44 female == 0 -2.371e-04  3.071e-04  -0.772   0.9693
## APOE22 female - APOE22 male == 0   -7.183e-04  2.940e-04  -2.443   0.1819
## APOE22 female - APOE33 male == 0   -9.035e-04  3.071e-04  -2.942   0.0694 .
## APOE22 female - APOE44 male == 0   -9.355e-04  3.071e-04  -3.046   0.0560 .
## APOE33 female - APOE44 female == 0 -3.644e-04  3.546e-04  -1.028   0.9030
## APOE33 female - APOE22 male == 0   -8.456e-04  3.433e-04  -2.463   0.1755
## APOE33 female - APOE33 male == 0   -1.031e-03  3.546e-04  -2.907   0.0747 .
## APOE33 female - APOE44 male == 0   -1.063e-03  3.546e-04  -2.997   0.0617 .
## APOE44 female - APOE22 male == 0   -4.812e-04  2.940e-04  -1.637   0.5824
## APOE44 female - APOE33 male == 0   -6.663e-04  3.071e-04  -2.170   0.2874
## APOE44 female - APOE44 male == 0   -6.983e-04  3.071e-04  -2.274   0.2431
## APOE22 male - APOE33 male == 0     -1.852e-04  2.940e-04  -0.630   0.9874
## APOE22 male - APOE44 male == 0     -2.172e-04  2.940e-04  -0.739   0.9746
## APOE33 male - APOE44 male == 0     -3.199e-05  3.071e-04  -0.104   1.0000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
```
```r
res<-summary(glht(lm_Hc_VOL, emm(pairwise ~ Genotype*Sex))) #consider save to file
#summary(as.glht(pairs(lm_Hc_VOL.emm), by = NULL))

summary(glht(lm_Hc_VOL, emm(pairwise ~ Genotype | Sex)))
```
```
## $`Sex = female`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                  Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  0.0001273  0.0003546   0.359    0.931
## APOE22 - APOE44 == 0 -0.0002371  0.0003071  -0.772    0.723
## APOE33 - APOE44 == 0 -0.0003645  0.0003546  -1.028    0.566
## (Adjusted p values reported -- single-step method)
##
##
## $`Sex = male`
##
```

```
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                     Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0 -1.852e-04  2.940e-04  -0.630    0.805
## APOE22 - APOE44 == 0 -2.172e-04  2.940e-04  -0.739    0.743
## APOE33 - APOE44 == 0 -3.199e-05  3.071e-04  -0.104    0.994
## (Adjusted p values reported -- single-step method)
```

```
summary(glht(lm_Hc_VOL, emm(pairwise ~ Sex | Genotype)))
```

```
## $`Genotype = APOE22`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                     Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.0007183  0.0002940  -2.443   0.0227 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                     Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.0010308  0.0003546  -2.907  0.00794 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_VOL)
##
## Linear Hypotheses:
##                     Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.0006983  0.0003071  -2.274   0.0326 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
```

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

```
## Genotype = APOE22:
##  contrast       estimate       SE df t.ratio p.value
##  female - male -0.000718 0.000294 23  -2.443  0.0227
##
## Genotype = APOE33:
##  contrast       estimate       SE df t.ratio p.value
##  female - male -0.001031 0.000355 23  -2.907  0.0079
##
## Genotype = APOE44:
##  contrast       estimate       SE df t.ratio p.value
##  female - male -0.000698 0.000307 23  -2.274  0.0326
```

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

```
## Sex = female:
##  contrast         estimate       SE df t.ratio p.value
##  APOE22 - APOE33  0.000127 0.000355 23   0.359  0.9316
##  APOE22 - APOE44 -0.000237 0.000307 23  -0.772  0.7234
##  APOE33 - APOE44 -0.000364 0.000355 23  -1.028  0.5673
##
## Sex = male:
##  contrast         estimate       SE df t.ratio p.value
##  APOE22 - APOE33 -0.000185 0.000294 23  -0.630  0.8053
##  APOE22 - APOE44 -0.000217 0.000294 23  -0.739  0.7433
##  APOE33 - APOE44 -0.000032 0.000307 23  -0.104  0.9940
##
## P value adjustment: tukey method for comparing a family of 3 estimates
```

```
print('Hc VOL')
```

```
## [1] "Hc VOL"
```
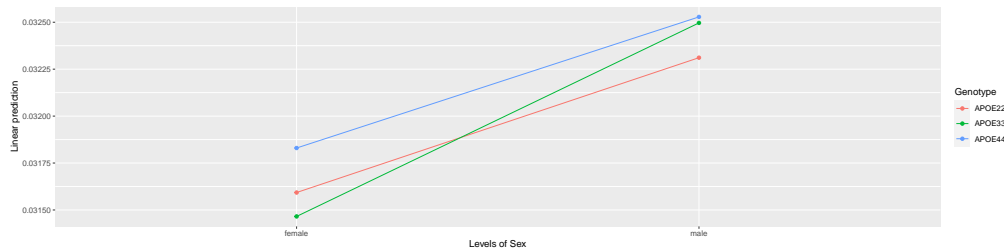
```
emmip(lm_Hc_VOL, ~ Genotype | Sex )
```
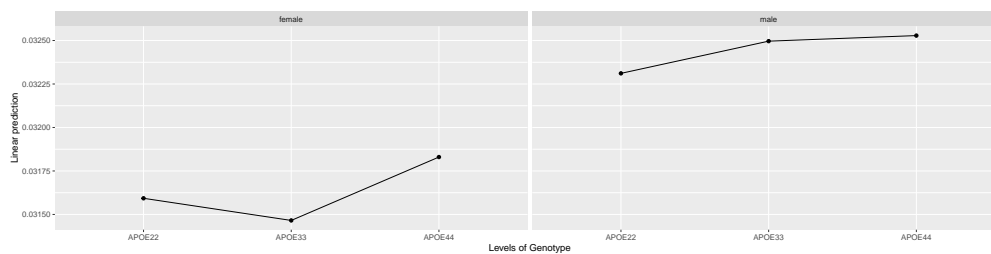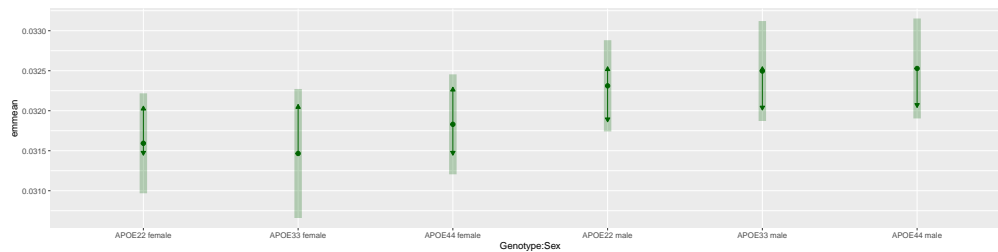


```
emmip(lm_Hc_VOL, ~ Sex | Genotype )
```
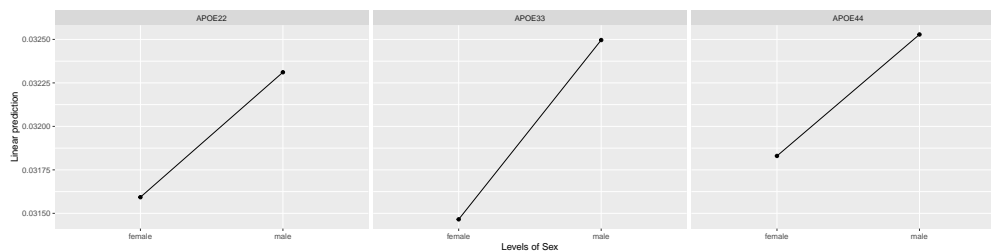
```
emmip(lm_Hc_VOL, Genotype~ Sex  )
```



```
sink()
```

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



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

```
'simple stats'
```

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

```
print('Hc')
```

```
## [1] "Hc"
```

```
lm_Hc_FA <- lm(Hc ~ Genotype*Sex, geno_combined_FA)
summary(lm_Hc_FA)
```

```
##
## Call:
## lm(formula = Hc ~ Genotype * Sex, data = geno_combined_FA)
##
## Residuals:
##        Min         1Q     Median         3Q        Max
## -0.0174129 -0.0045908  0.0002655  0.0064382  0.0138615
##
## Coefficients:
##                       Estimate Std. Error t value Pr(>|t|)
## (Intercept)           0.267691   0.003956  67.663  < 2e-16 ***
## GenotypeAPOE33       -0.017357   0.006460  -2.687  0.01317 *
## GenotypeAPOE44       -0.018166   0.005595  -3.247  0.00356 **
## Sexmale              -0.005776   0.005357  -1.078  0.29211
## GenotypeAPOE33:Sexmale  0.014094   0.008392   1.679  0.10661
## GenotypeAPOE44:Sexmale  0.015411   0.007746   1.990  0.05866 .
## ---
```

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```
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.008846 on 23 degrees of freedom
## Multiple R-squared:  0.3788, Adjusted R-squared:  0.2437
## F-statistic: 2.805 on 5 and 23 DF,  p-value: 0.04041
```

```r
lm_Hc_FA_f <- lm(Hc ~ Genotype, combo_f_FA)
#summary(lm_Hc_FA_f)
lm_Hc_FA_m <- lm(Hc ~ Genotype, combo_m_FA)
#summary(lm_Hc_FA_f)


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

```
## Analysis of Variance Table
##
## Response: Hc
##              Df     Sum Sq    Mean Sq F value  Pr(>F)
## Genotype      2 0.00064461 0.00032230  4.1185 0.02959 *
## Sex           1 0.00008065 0.00008065  1.0306 0.32059
## Genotype:Sex  2 0.00037217 0.00018609  2.3778 0.11517
## Residuals    23 0.00179995 0.00007826
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```r
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 = Hc ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0 0.0173571  0.0064605   2.687  0.03367 *
## APOE22 - APOE44 == 0 0.0181662  0.0055949   3.247  0.00968 **
## APOE33 - APOE44 == 0 0.0008091  0.0064605   0.125  0.99137
## ---
## 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 = Hc ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  0.0032628  0.0053568   0.609    0.817
```

```
## APOE22 - APOE44 == 0  0.0027551   0.0053568    0.514     0.865
## APOE33 - APOE44 == 0 -0.0005077   0.0055949   -0.091     0.995
## (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 = Hc ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## female - male == 0 0.005776   0.005357   1.078    0.292
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.008319   0.006460   -1.288    0.211
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.009635   0.005595   -1.722   0.0985 .
## ---
## 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, "Hc_FA_SimpleStats.txt",sep=''))
'omnibus testing'
```

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

```
joint_tests(ref_grid(lm_Hc_FA))
```

```
## model term    df1 df2 F.ratio p.value
## Genotype        2  23   4.641  0.0203
## Sex             1  23   1.458  0.2396
## Genotype:Sex    2  23   2.378  0.1152
```

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

```
## Sex = female:
##  model term df1 df2 F.ratio p.value
##  Genotype     2  23   6.281  0.0067
##
## Sex = male:
##  model term df1 df2 F.ratio p.value
##  Genotype     2  23   0.221  0.8034
```

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

```
## Genotype = APOE22:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   1.163  0.2921
##
## Genotype = APOE33:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   1.658  0.2107
##
## Genotype = APOE44:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   2.966  0.0985
```

```
'anova'
```

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

```
anova(lm_Hc_FA)
```

```
## Analysis of Variance Table
##
## Response: Hc
##              Df     Sum Sq    Mean Sq F value  Pr(>F)
## Genotype      2 0.00064461 0.00032230  4.1185 0.02959 *
## Sex           1 0.00008065 0.00008065  1.0306 0.32059
## Genotype:Sex  2 0.00037217 0.00018609  2.3778 0.11517
## Residuals    23 0.00179995 0.00007826
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
#anova(lm_Hc_FA_f)
#anova(lm_Hc_FA_m)

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

```
## contrast               estimate      SE df t.ratio p.value
## APOE22 female effect  0.009811 0.00364 23   2.694  0.0777
## APOE33 female effect -0.007546 0.00450 23  -1.678  0.2137
## APOE44 female effect -0.008355 0.00364 23  -2.294  0.0937
## APOE22 male effect    0.004036 0.00339 23   1.189  0.3699
```

```
##  APOE33 male effect     0.000773 0.00364 23    0.212  0.8338
##  APOE44 male effect     0.001281 0.00364 23    0.352  0.8338
##
## P value adjustment: fdr method for 6 tests
contrast(Hc_FA.emm, simple = list("Genotype", "Sex"))

## $`simple contrasts for Genotype`
## Sex = female:
##  contrast        estimate       SE df t.ratio p.value
##  APOE22 effect  0.011841 0.00340 23   3.478  0.0061
##  APOE33 effect -0.005516 0.00388 23  -1.421  0.1688
##  APOE44 effect -0.006325 0.00340 23  -1.858  0.1141
##
## Sex = male:
##  contrast        estimate       SE df t.ratio p.value
##  APOE22 effect  0.002006 0.00305 23   0.659  0.8161
##  APOE33 effect -0.001257 0.00319 23  -0.395  0.8161
##  APOE44 effect -0.000749 0.00319 23  -0.235  0.8161
##
## P value adjustment: fdr method for 3 tests
##
## $`simple contrasts for Sex`
## Genotype = APOE22:
##  contrast       estimate       SE df t.ratio p.value
##  female effect  0.00289 0.00268 23   1.078  0.2921
##  male effect   -0.00289 0.00268 23  -1.078  0.2921
##
## Genotype = APOE33:
##  contrast       estimate       SE df t.ratio p.value
##  female effect -0.00416 0.00323 23  -1.288  0.2107
##  male effect    0.00416 0.00323 23   1.288  0.2107
##
## Genotype = APOE44:
##  contrast       estimate       SE df t.ratio p.value
##  female effect -0.00482 0.00280 23  -1.722  0.0985
##  male effect    0.00482 0.00280 23   1.722  0.0985
##
## P value adjustment: fdr method for 2 tests
#post hoc tests
summary(glht(lm_Hc_FA, emm(pairwise ~ Genotype*Sex, contr="sidak"))) #consider save to file

##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##                                Estimate Std. Error t value Pr(>|t|)
## APOE22 female - APOE33 female == 0  0.0173571  0.0064605   2.687   0.1157
## APOE22 female - APOE44 female == 0  0.0181662  0.0055949   3.247   0.0364 *
## APOE22 female - APOE22 male == 0    0.0057758  0.0053568   1.078   0.8842
## APOE22 female - APOE33 male == 0    0.0090386  0.0055949   1.615   0.5954
## APOE22 female - APOE44 male == 0    0.0085309  0.0055949   1.525   0.6509
```

```
## APOE33 female - APOE44 female == 0   0.0008091   0.0064605    0.125   1.0000
## APOE33 female - APOE22 male == 0     -0.0115814  0.0062553   -1.851   0.4530
## APOE33 female - APOE33 male == 0     -0.0083185  0.0064605   -1.288   0.7872
## APOE33 female - APOE44 male == 0     -0.0088262  0.0064605   -1.366   0.7444
## APOE44 female - APOE22 male == 0     -0.0123904  0.0053568   -2.313   0.2277
## APOE44 female - APOE33 male == 0     -0.0091276  0.0055949   -1.631   0.5856
## APOE44 female - APOE44 male == 0     -0.0096353  0.0055949   -1.722   0.5300
## APOE22 male - APOE33 male == 0        0.0032628  0.0053568    0.609   0.9892
## APOE22 male - APOE44 male == 0        0.0027551  0.0053568    0.514   0.9950
## APOE33 male - APOE44 male == 0       -0.0005077  0.0055949   -0.091   1.0000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
```

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

summary(glht(lm_Hc_FA, emm(pairwise ~ Genotype | Sex)))
```

```
## $`Sex = female`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0 0.0173571  0.0064605    2.687  0.03385 *
## APOE22 - APOE44 == 0 0.0181662  0.0055949    3.247  0.00977 **
## APOE33 - APOE44 == 0 0.0008091  0.0064605    0.125  0.99136
## ---
## 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 = Hc ~ Genotype * Sex, data = geno_combined_FA)
##
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0  0.0032628  0.0053568    0.609    0.817
## APOE22 - APOE44 == 0  0.0027551  0.0053568    0.514    0.865
## APOE33 - APOE44 == 0 -0.0005077  0.0055949   -0.091    0.995
## (Adjusted p values reported -- single-step method)
```

```r
summary(glht(lm_Hc_FA, emm(pairwise ~ Sex | Genotype)))
```

```
## $`Genotype = APOE22`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_FA)
```

```
## 
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## female - male == 0 0.005776   0.005357   1.078    0.292
## (Adjusted p values reported -- single-step method)
## 
## 
## $`Genotype = APOE33`
## 
##   Simultaneous Tests for General Linear Hypotheses
## 
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_FA)
## 
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.008319   0.006460  -1.288    0.211
## (Adjusted p values reported -- single-step method)
## 
## 
## $`Genotype = APOE44`
## 
##   Simultaneous Tests for General Linear Hypotheses
## 
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_FA)
## 
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## female - male == 0 -0.009635   0.005595  -1.722   0.0985 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
```

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

```
## Genotype = APOE22:
##  contrast       estimate      SE df t.ratio p.value
##  female - male  0.00578 0.00536 23   1.078  0.2921
## 
## Genotype = APOE33:
##  contrast       estimate      SE df t.ratio p.value
##  female - male -0.00832 0.00646 23  -1.288  0.2107
## 
## Genotype = APOE44:
##  contrast       estimate      SE df t.ratio p.value
##  female - male -0.00964 0.00559 23  -1.722  0.0985
```

```r
pairs(Hc_FA.emm, by="Sex")
```

```
## Sex = female:
##  contrast        estimate      SE df t.ratio p.value
##  APOE22 - APOE33 0.017357 0.00646 23   2.687  0.0339
##  APOE22 - APOE44 0.018166 0.00559 23   3.247  0.0096
##  APOE33 - APOE44 0.000809 0.00646 23   0.125  0.9914
## 
```
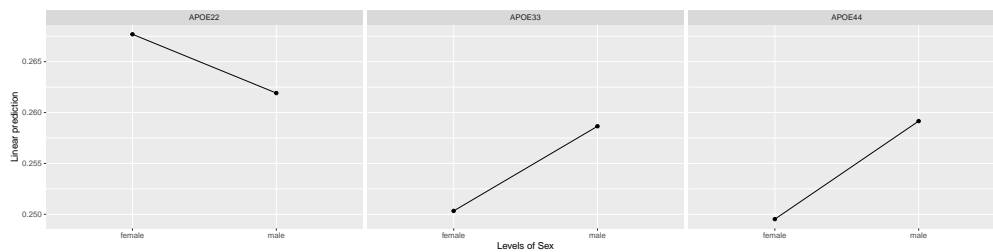
```
## Sex = male:
##  contrast          estimate      SE df t.ratio p.value
##  APOE22 - APOE33  0.003263 0.00536 23   0.609  0.8166
##  APOE22 - APOE44  0.002755 0.00536 23   0.514  0.8652
##  APOE33 - APOE44 -0.000508 0.00559 23  -0.091  0.9955
##
## P value adjustment: tukey method for comparing a family of 3 estimates
```

```
print('Hc FA')
```
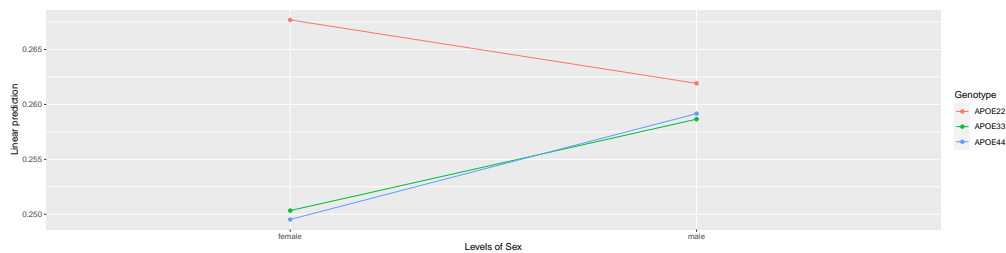
```
## [1] "Hc FA"
```

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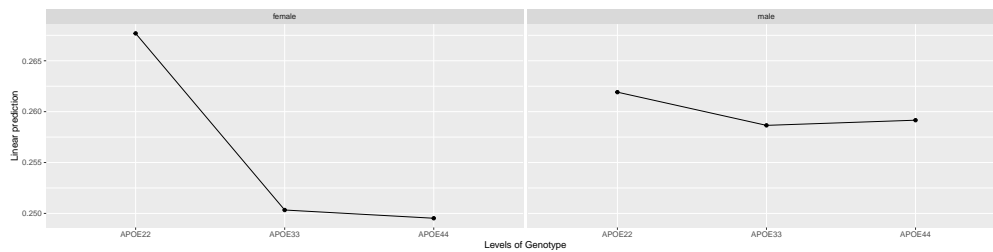


```
emmip(lm_Hc_FA, ~ Sex | Genotype )
```
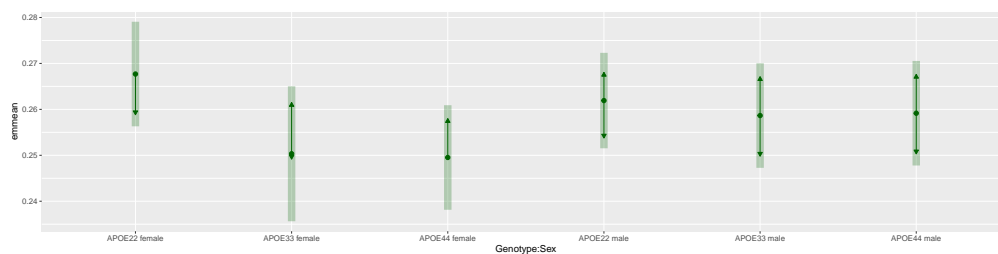


```
emmip(lm_Hc_FA, Genotype~ Sex  )
```



```
sink()
```

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


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

```r
'simple stats'
```

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

```r
print('Hc')
```

```
## [1] "Hc"
```

```r
lm_Hc_DEG <- lm(Hc ~ Genotype*Sex, geno_combined_DEG)
summary(lm_Hc_DEG)
```

```
##
## Call:
## lm(formula = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Residuals:
##    Min     1Q Median     3Q    Max
## -57200 -15583    917  15417  69667
##
## Coefficients:
##                       Estimate Std. Error t value Pr(>|t|)
## (Intercept)             363200      13515  26.873   <2e-16 ***
## GenotypeAPOE33           52133      22070   2.362    0.027 *
## GenotypeAPOE44           23600      19114   1.235    0.229
## Sexmale                  20383      18300   1.114    0.277
## GenotypeAPOE33:Sexmale   -2517      28670  -0.088    0.931
## GenotypeAPOE44:Sexmale   -1783      26462  -0.067    0.947
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 30220 on 23 degrees of freedom
## Multiple R-squared:  0.4199, Adjusted R-squared:  0.2938
## F-statistic:  3.33 on 5 and 23 DF,  p-value: 0.02083
```

```r
lm_Hc_DEG_f <- lm(Hc ~ Genotype, combo_f_FA)
#summary(lm_Hc_DEG_f)
lm_Hc_DEG_m <- lm(Hc ~ Genotype, combo_m_FA)
#summary(lm_Hc_DEG_f)

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

```
## Analysis of Variance Table
##
## Response: Hc
##             Df    Sum Sq    Mean Sq F value   Pr(>F)
## Genotype     2 1.2612e+10 6305878370  6.9043 0.004482 **
## Sex          1 2.5886e+09 2588593164  2.8343 0.105800
## Genotype:Sex 2 7.9682e+06    3984100  0.0044 0.995648
## Residuals   23 2.1006e+10  913325000
## ---
## 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 = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##                     Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0    -52133      22070  -2.362   0.0666 .
## APOE22 - APOE44 == 0    -23600      19114  -1.235   0.4443
## APOE33 - APOE44 == 0     28533      22070   1.293   0.4123
## ---
## 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 = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##                     Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0    -49617      18300  -2.711   0.0324 *
## APOE22 - APOE44 == 0    -21817      18300  -1.192   0.4694
## APOE33 - APOE44 == 0     27800      19114   1.454   0.3306
## ---
## 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 = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## female - male == 0    -20383      18300  -1.114    0.277
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
##    Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
```

```
##                   Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -17867      22070   -0.81    0.427
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
##    Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -18600      19114  -0.973    0.341
## (Adjusted p values reported -- single-step method)
```

```r
sink()
#3333

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

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

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

```r
joint_tests(ref_grid(lm_Hc_DEG))
```

```
##  model term   df1 df2 F.ratio p.value
##  Genotype       2  23   6.311  0.0065
##  Sex            1  23   2.722  0.1126
##  Genotype:Sex   2  23   0.004  0.9956
```

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

```
## Sex = female:
##  model term df1 df2 F.ratio p.value
##  Genotype     2  23   2.817  0.0805
##
## Sex = male:
##  model term df1 df2 F.ratio p.value
##  Genotype     2  23   3.677  0.0412
```

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

```
## Genotype = APOE22:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   1.241  0.2768
##
## Genotype = APOE33:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   0.655  0.4265
##
## Genotype = APOE44:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   0.947  0.3406
```

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```
'anova'
```

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

```
anova(lm_Hc_DEG)
```

```
## Analysis of Variance Table
##
## Response: Hc
##              Df     Sum Sq     Mean Sq F value    Pr(>F)
## Genotype      2 1.2612e+10 6305878370  6.9043  0.004482 **
## Sex           1 2.5886e+09 2588593164  2.8343  0.105800
## Genotype:Sex  2 7.9682e+06    3984100  0.0044  0.995648
## Residuals    23 2.1006e+10  913325000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
#anova(lm_Hc_DEG_f)
#anova(lm_Hc_DEG_m)

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

```
##  contrast             estimate    SE df t.ratio p.value
##  APOE22 female effect   -34719 12440 23  -2.791  0.0312
##  APOE33 female effect    17414 15360 23   1.134  0.4029
##  APOE44 female effect   -11119 12440 23  -0.894  0.4568
##  APOE22 male effect     -14336 11596 23  -1.236  0.4029
##  APOE33 male effect      35281 12440 23   2.836  0.0312
##  APOE44 male effect       7481 12440 23   0.601  0.5535
##
## P value adjustment: fdr method for 6 tests
```

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

```
## $`simple contrasts for Genotype`
## Sex = female:
##  contrast      estimate    SE df t.ratio p.value
##  APOE22 effect   -25244 11632 23  -2.170  0.0815
##  APOE33 effect    26889 13263 23   2.027  0.0815
##  APOE44 effect    -1644 11632 23  -0.141  0.8888
##
## Sex = male:
##  contrast      estimate    SE df t.ratio p.value
##  APOE22 effect   -23811 10404 23  -2.289  0.0474
##  APOE33 effect    25806 10881 23   2.372  0.0474
##  APOE44 effect    -1994 10881 23  -0.183  0.8562
##
## P value adjustment: fdr method for 3 tests
##
## $`simple contrasts for Sex`
## Genotype = APOE22:
##  contrast       estimate   SE df t.ratio p.value
##  female effect    -10192 9150 23  -1.114  0.2768
##  male effect       10192 9150 23   1.114  0.2768
```

```
##
## Genotype = APOE33:
##  contrast      estimate    SE df t.ratio p.value
##  female effect    -8933 11035 23  -0.810  0.4265
##  male effect       8933 11035 23   0.810  0.4265
##
## Genotype = APOE44:
##  contrast      estimate   SE df t.ratio p.value
##  female effect    -9300 9557 23  -0.973  0.3406
##  male effect       9300 9557 23   0.973  0.3406
##
## P value adjustment: fdr method for 2 tests
```

```
#post hoc tests
summary(glht(lm_Hc_DEG, emm(pairwise ~ Genotype*Sex, contr="sidak"))) #consider save to file
```

```
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##                              Estimate Std. Error t value Pr(>|t|)
## APOE22 female - APOE33 female == 0   -52133      22070  -2.362   0.2095
## APOE22 female - APOE44 female == 0   -23600      19114  -1.235   0.8143
## APOE22 female - APOE22 male == 0     -20383      18300  -1.114   0.8698
## APOE22 female - APOE33 male == 0     -70000      19114  -3.662   0.0144 *
## APOE22 female - APOE44 male == 0     -42200      19114  -2.208   0.2707
## APOE33 female - APOE44 female == 0    28533      22070   1.293   0.7845
## APOE33 female - APOE22 male == 0      31750      21370   1.486   0.6745
## APOE33 female - APOE33 male == 0     -17867      22070  -0.810   0.9625
## APOE33 female - APOE44 male == 0       9933      22070   0.450   0.9973
## APOE44 female - APOE22 male == 0       3217      18300   0.176   1.0000
## APOE44 female - APOE33 male == 0     -46400      19114  -2.428   0.1871
## APOE44 female - APOE44 male == 0     -18600      19114  -0.973   0.9212
## APOE22 male - APOE33 male == 0       -49617      18300  -2.711   0.1104
## APOE22 male - APOE44 male == 0       -21817      18300  -1.192   0.8349
## APOE33 male - APOE44 male == 0        27800      19114   1.454   0.6932
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
```

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

summary(glht(lm_Hc_DEG, emm(pairwise ~ Genotype | Sex)))
```

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

```
## APOE22 - APOE33 == 0    -52133        22070  -2.362    0.0667 .
## APOE22 - APOE44 == 0    -23600        19114  -1.235    0.4443
## APOE33 - APOE44 == 0     28533        22070   1.293    0.4123
## ---
## 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 = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##                      Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0    -49617      18300  -2.711   0.0321 *
## APOE22 - APOE44 == 0    -21817      18300  -1.192   0.4694
## APOE33 - APOE44 == 0     27800      19114   1.454   0.3306
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## (Adjusted p values reported -- single-step method)
```

```
summary(glht(lm_Hc_DEG, emm(pairwise ~ Sex | Genotype)))
```

```
## $`Genotype = APOE22`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -20383      18300  -1.114    0.277
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -17867      22070   -0.81    0.427
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_DEG)
##
```

```
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -18600      19114  -0.973    0.341
## (Adjusted p values reported -- single-step method)
```

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

```
## Genotype = APOE22:
##  contrast        estimate    SE df t.ratio p.value
##  female - male    -20383 18300 23  -1.114  0.2768
##
## Genotype = APOE33:
##  contrast        estimate    SE df t.ratio p.value
##  female - male    -17867 22070 23  -0.810  0.4265
##
## Genotype = APOE44:
##  contrast        estimate    SE df t.ratio p.value
##  female - male    -18600 19114 23  -0.973  0.3406
```

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

```
## Sex = female:
##  contrast          estimate    SE df t.ratio p.value
##  APOE22 - APOE33    -52133 22070 23  -2.362  0.0670
##  APOE22 - APOE44    -23600 19114 23  -1.235  0.4454
##  APOE33 - APOE44     28533 22070 23   1.293  0.4133
##
## Sex = male:
##  contrast          estimate    SE df t.ratio p.value
##  APOE22 - APOE33    -49617 18300 23  -2.711  0.0322
##  APOE22 - APOE44    -21817 18300 23  -1.192  0.4696
##  APOE33 - APOE44     27800 19114 23   1.454  0.3308
##
## P value adjustment: tukey method for comparing a family of 3 estimates
```

```
print('Hc DEG')
```
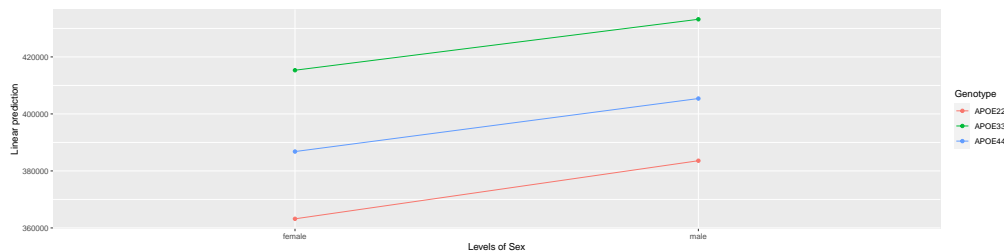
```
## [1] "Hc DEG"
```

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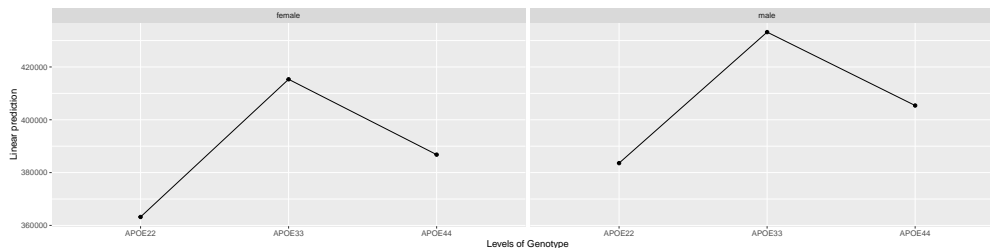


```
emmip(lm_Hc_DEG, ~ Sex | Genotype )
```
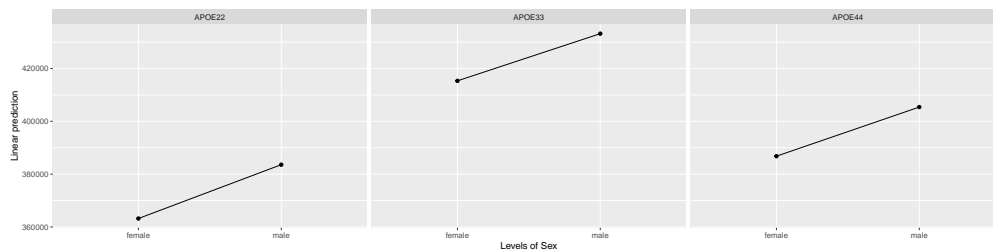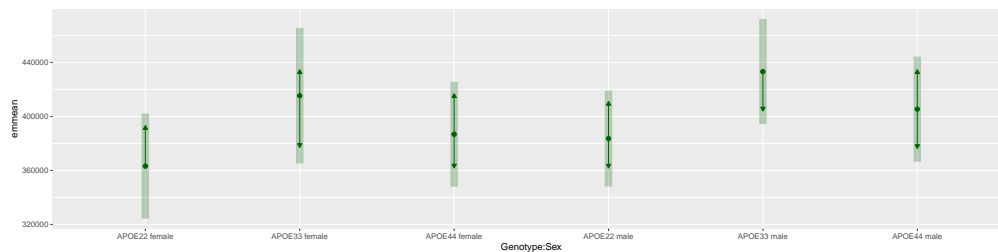
```
emmip(lm_Hc_DEG, Genotype~ Sex  )
```



```
sink()
```

```
plot(Hc_DEG.emm, by = NULL, comparisons = TRUE, adjust = "mvt",
horizontal = FALSE, colors = "darkgreen", main ="Hc DEG")
```



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

```
'simple stats'
```

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

```
print('Hc')
```

```
## [1] "Hc"
```

```
lm_Hc_CLUS <- lm(Hc ~ Genotype*Sex, geno_combined_CLUS)
summary(lm_Hc_CLUS)
```

```
##
## Call:
## lm(formula = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Residuals:
##     Min      1Q  Median      3Q     Max
## -86.377 -24.544  -3.834  21.724 121.923
##
## Coefficients:
##                     Estimate Std. Error t value Pr(>|t|)
## (Intercept)           442.45      22.38  19.769 6.23e-16 ***
## GenotypeAPOE33         86.11      36.55   2.356   0.0274 *
```

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```
## GenotypeAPOE44              46.66      31.65    1.474    0.1540
## Sexmale                     31.52      30.30    1.040    0.3091
## GenotypeAPOE33:Sexmale     -33.30      47.48   -0.701    0.4901
## GenotypeAPOE44:Sexmale     -17.24      43.82   -0.394    0.6976
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 50.05 on 23 degrees of freedom
## Multiple R-squared:  0.3022, Adjusted R-squared:  0.1505
## F-statistic: 1.992 on 5 and 23 DF,  p-value: 0.1179
```

```
lm_Hc_CLUS_f <- lm(Hc ~ Genotype, combo_f_FA)
#summary(lm_Hc_CLUS_f)
lm_Hc_CLUS_m <- lm(Hc ~ Genotype, combo_m_FA)
#summary(lm_Hc_CLUS_f)

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

```
## Analysis of Variance Table
##
## Response: Hc
##             Df Sum Sq Mean Sq F value  Pr(>F)
## Genotype     2  21720 10860.2  4.3361 0.02524 *
## Sex          1   1971  1971.3  0.7871 0.38416
## Genotype:Sex 2   1254   627.1  0.2504 0.78060
## Residuals   23  57605  2504.6
## ---
## 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 = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0   -86.11      36.55  -2.356   0.0676 .
## APOE22 - APOE44 == 0   -46.66      31.65  -1.474   0.3205
## APOE33 - APOE44 == 0    39.46      36.55   1.080   0.5349
## ---
## 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 = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
```

```
##
## Linear Hypotheses:
##                        Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0    -52.81      30.30  -1.743    0.211
## APOE22 - APOE44 == 0    -29.41      30.30  -0.971    0.602
## APOE33 - APOE44 == 0     23.40      31.65   0.739    0.743
## (Adjusted p values reported -- single-step method)
```

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

```
## $`Genotype = APOE22`
##
##    Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##                     Estimate Std. Error t value Pr(>|t|)
## female - male == 0    -31.52      30.30   -1.04    0.309
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE33`
##
##    Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##                     Estimate Std. Error t value Pr(>|t|)
## female - male == 0     1.781     36.548   0.049    0.962
## (Adjusted p values reported -- single-step method)
##
##
## $`Genotype = APOE44`
##
##    Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##                     Estimate Std. Error t value Pr(>|t|)
## female - male == 0    -14.28      31.65  -0.451    0.656
## (Adjusted p values reported -- single-step method)
```

```r
sink()
#3333

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

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

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

```
joint_tests(ref_grid(lm_Hc_CLUS))
```

```
##  model term    df1 df2 F.ratio p.value
##  Genotype       2   23   4.404  0.0240
##  Sex            1   23   0.595  0.4483
##  Genotype:Sex   2   23   0.250  0.7806
```

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

```
## Sex = female:
##  model term df1 df2 F.ratio p.value
##  Genotype     2  23   2.903  0.0752
##
## Sex = male:
##  model term df1 df2 F.ratio p.value
##  Genotype     2  23   1.539  0.2360
```

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

```
## Genotype = APOE22:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   1.082  0.3091
##
## Genotype = APOE33:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   0.002  0.9616
##
## Genotype = APOE44:
##  model term df1 df2 F.ratio p.value
##  Sex          1  23   0.203  0.6561
```

```
'anova'
```

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

```
anova(lm_Hc_CLUS)
```

```
## Analysis of Variance Table
##
## Response: Hc
##               Df Sum Sq Mean Sq F value  Pr(>F)
## Genotype       2  21720 10860.2  4.3361 0.02524 *
## Sex            1   1971  1971.3  0.7871 0.38416
## Genotype:Sex   2   1254   627.1  0.2504 0.78060
## Residuals     23  57605  2504.6
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
#anova(lm_Hc_CLUS_f)
#anova(lm_Hc_CLUS_m)

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

```
##  contrast               estimate   SE df t.ratio p.value
##  APOE22 female effect    -51.59 20.6 23  -2.504  0.1188
##  APOE33 female effect     34.52 25.4 23   1.357  0.3758
```

```
##  APOE44 female effect     -4.94 20.6 23  -0.240  0.8128
##  APOE22 male effect      -20.07 19.2 23  -1.045  0.4601
##  APOE33 male effect       32.74 20.6 23   1.589  0.3758
##  APOE44 male effect        9.34 20.6 23   0.453  0.7853
##
## P value adjustment: fdr method for 6 tests
contrast(Hc_CLUS.emm, simple = list("Genotype", "Sex"))

## $`simple contrasts for Genotype`
## Sex = female:
##  contrast       estimate   SE df t.ratio p.value
##  APOE22 effect   -44.26 19.3 23  -2.298  0.0931
##  APOE33 effect    41.86 22.0 23   1.906  0.1039
##  APOE44 effect     2.40 19.3 23   0.125  0.9019
##
## Sex = male:
##  contrast       estimate   SE df t.ratio p.value
##  APOE22 effect   -27.41 17.2 23  -1.591  0.2579
##  APOE33 effect    25.40 18.0 23   1.410  0.2579
##  APOE44 effect     2.01 18.0 23   0.111  0.9124
##
## P value adjustment: fdr method for 3 tests
##
## $`simple contrasts for Sex`
## Genotype = APOE22:
##  contrast       estimate   SE df t.ratio p.value
##  female effect   -15.76 15.2 23  -1.040  0.3091
##  male effect      15.76 15.2 23   1.040  0.3091
##
## Genotype = APOE33:
##  contrast       estimate   SE df t.ratio p.value
##  female effect     0.89 18.3 23   0.049  0.9616
##  male effect      -0.89 18.3 23  -0.049  0.9616
##
## Genotype = APOE44:
##  contrast       estimate   SE df t.ratio p.value
##  female effect    -7.14 15.8 23  -0.451  0.6561
##  male effect       7.14 15.8 23   0.451  0.6561
##
## P value adjustment: fdr method for 2 tests
#post hoc tests
summary(glht(lm_Hc_CLUS, emm(pairwise ~ Genotype*Sex, contr="sidak"))) #consider save to file

##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##                               Estimate Std. Error t value Pr(>|t|)
## APOE22 female - APOE33 female == 0   -86.115     36.548  -2.356    0.212
## APOE22 female - APOE44 female == 0   -46.658     31.652  -1.474    0.681
## APOE22 female - APOE22 male == 0     -31.522     30.304  -1.040    0.899
```

```
## APOE22 female - APOE33 male == 0    -84.334    31.652  -2.664   0.121
## APOE22 female - APOE44 male == 0    -60.936    31.652  -1.925   0.411
## APOE33 female - APOE44 female == 0   39.457    36.548   1.080   0.884
## APOE33 female - APOE22 male == 0     54.593    35.388   1.543   0.640
## APOE33 female - APOE33 male == 0      1.781    36.548   0.049   1.000
## APOE33 female - APOE44 male == 0     25.179    36.548   0.689   0.981
## APOE44 female - APOE22 male == 0     15.136    30.304   0.499   0.996
## APOE44 female - APOE33 male == 0    -37.676    31.652  -1.190   0.836
## APOE44 female - APOE44 male == 0    -14.278    31.652  -0.451   0.997
## APOE22 male - APOE33 male == 0      -52.812    30.304  -1.743   0.517
## APOE22 male - APOE44 male == 0      -29.414    30.304  -0.971   0.922
## APOE33 male - APOE44 male == 0       23.398    31.652   0.739   0.975
## (Adjusted p values reported -- single-step method)
```

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

summary(glht(lm_Hc_CLUS, emm(pairwise ~ Genotype | Sex)))
```

```
## $`Sex = female`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0   -86.11      36.55  -2.356   0.0673 .
## APOE22 - APOE44 == 0   -46.66      31.65  -1.474   0.3204
## APOE33 - APOE44 == 0    39.46      36.55   1.080   0.5349
## ---
## 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 = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
##
## Linear Hypotheses:
##                   Estimate Std. Error t value Pr(>|t|)
## APOE22 - APOE33 == 0   -52.81      30.30  -1.743   0.211
## APOE22 - APOE44 == 0   -29.41      30.30  -0.971   0.602
## APOE33 - APOE44 == 0    23.40      31.65   0.739   0.743
## (Adjusted p values reported -- single-step method)
```

```r
summary(glht(lm_Hc_CLUS, emm(pairwise ~ Sex | Genotype)))
```

```
## $`Genotype = APOE22`
##
##   Simultaneous Tests for General Linear Hypotheses
##
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
```

```
## 
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -31.52      30.30   -1.04    0.309
## (Adjusted p values reported -- single-step method)
## 
## 
## $`Genotype = APOE33`
## 
##   Simultaneous Tests for General Linear Hypotheses
## 
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
## 
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## female - male == 0    1.781     36.548   0.049    0.962
## (Adjusted p values reported -- single-step method)
## 
## 
## $`Genotype = APOE44`
## 
##   Simultaneous Tests for General Linear Hypotheses
## 
## Fit: lm(formula = Hc ~ Genotype * Sex, data = geno_combined_CLUS)
## 
## Linear Hypotheses:
##                    Estimate Std. Error t value Pr(>|t|)
## female - male == 0   -14.28      31.65  -0.451    0.656
## (Adjusted p values reported -- single-step method)
```

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

```
## Genotype = APOE22:
##  contrast      estimate   SE df t.ratio p.value
##  female - male   -31.52 30.3 23  -1.040  0.3091
## 
## Genotype = APOE33:
##  contrast      estimate   SE df t.ratio p.value
##  female - male     1.78 36.5 23   0.049  0.9616
## 
## Genotype = APOE44:
##  contrast      estimate   SE df t.ratio p.value
##  female - male   -14.28 31.7 23  -0.451  0.6561
```

```r
pairs(Hc_CLUS.emm, by="Sex")
```

```
## Sex = female:
##  contrast        estimate   SE df t.ratio p.value
##  APOE22 - APOE33    -86.1 36.5 23  -2.356  0.0678
##  APOE22 - APOE44    -46.7 31.7 23  -1.474  0.3215
##  APOE33 - APOE44     39.5 36.5 23   1.080  0.5359
## 
## Sex = male:
##  contrast          estimate   SE df t.ratio p.value
```
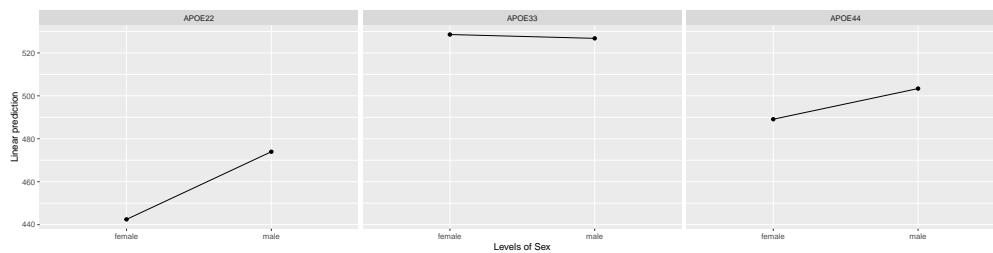
```
##  APOE22 - APOE33     -52.8 30.3 23  -1.743  0.2113
##  APOE22 - APOE44     -29.4 30.3 23  -0.971  0.6023
##  APOE33 - APOE44      23.4 31.7 23   0.739  0.7430
##
## P value adjustment: tukey method for comparing a family of 3 estimates
```

```
print('Hc CLUS')
```
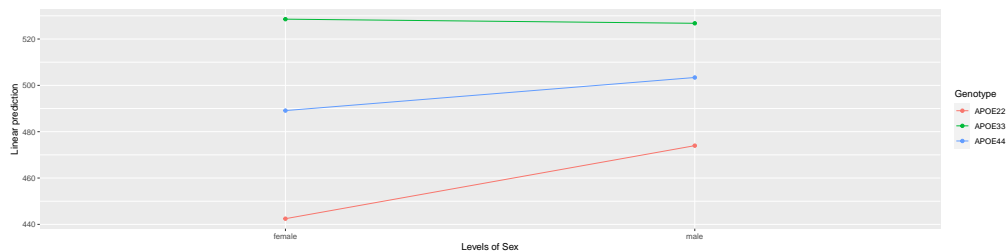
```
## [1] "Hc CLUS"
```

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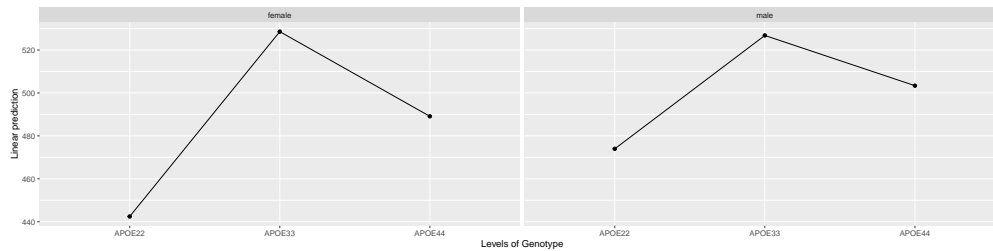


```
emmip(lm_Hc_CLUS, ~ Sex | Genotype )
```
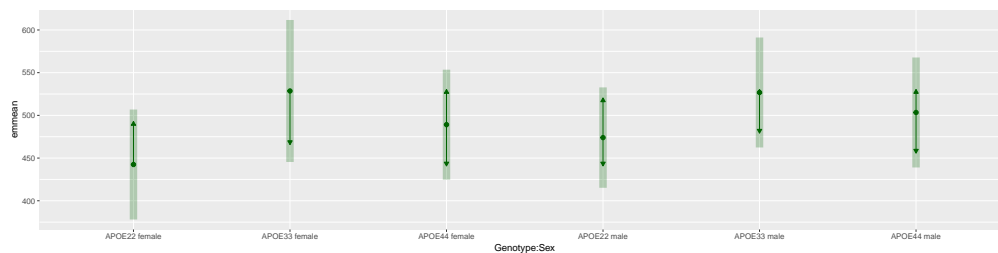


```
emmip(lm_Hc_CLUS, Genotype~ Sex  )
```



```
sink()
```

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



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