R-code for 'Effects of hunting on black grouse inbreeding and dispersal'

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Contents

This document contains all R-code used in the workflow for the manuscript *"Effects of hunting on genetic diversity, inbreeding and dispersal in Finnish black grouse (Lyrurus tetrix)"* by Rebecca Shuhua Chen, Carl Soulsbury, Christophe Lebigre, Kees van Oers and Joseph Hoffman (in review). The raw data can be found in the public GitHub repository together with full R scripts and processed datafiles. Please contact me at rebecca.chen@uni-bielefeld.de for any questions.

Within this markdown file, we follow the same order of analyses as described in the Materials and Methods. However, not all analyses are executed in R, and other softwares were used in combination with our workflow in R to collect all results presented in the manuscript.

Libraries

The following packages were used in the analyses:

```
library(tidyverse);library(adegenet); library(pegas)
library(data.table);library(hierfstat); library(plot.matrix); library(lme4)
library(forcats); library(ape); library(ParallelStructure)
library(pophelper); library(inbreedR); library(lmerTest); library(DHARMa)
library(performance);library(MuMIn); library(readxl); library(glmmTMB)
library(RColorBrewer); library(extrafont); library(devtools);library(gridExtra)
```

Data

Raw datasheets are provided in three different formats: 1) unsplit genotypes with one row per individual, 2) STRUCTURE (.stru) datafiles with two consecutive rows per individual, one row per allele. In both these files, populations are coded as integers. Thirdly, a full, easy to read .RData can be found including all details on the sites and hunted status, as well as sex.

```
base::load("data/rawdata/Fulldata_adults.RData")
hunted.ad$site <- as.factor(hunted.ad$site)
summary(hunted.ad[,c(3,7,8,9)])
```

```
##    year     sex           hunt             site
##  2001: 83   F: 813   hunted  : 544   Kummunsuo:308
##  2002:240   M:1065   unhunted:1334   Teerisuo :307
##  2003:214                            Nyrölä   :290
##  2004:234                            Koskenpää:248
##  2005:252                            Lehtusuo :194
##  2006:498                            Saarisuo :161
##  2007:357                            (Other)  :370
```

```
#structure file to summarise genotypes
all.raw <- read.structure("data/rawdata/Microsat.adults.plus.unrelated.chicks.forstructure.stru",
                         n.ind = 2078, n.loc = 14, onerowperind = F,
                         col.lab = 1, col.pop = 2, col.others = NULL,
                         row.marknames = 0, NA.char = "-9", pop = NULL,
                         sep = NULL,ask = F, quiet = TRUE)

summary(all.raw)
```

```
##
## // Number of individuals: 2078
## // Group sizes: 274 363 46 211 320 59 62 40 91 184 350 78
## // Number of alleles per locus: 8 11 14 13 9 23 11 9 23 9 10 11 21 5
## // Number of alleles per group: 132 131 99 136 130 96 101 103 102 125 142 119
## // Percentage of missing data: 9.4 %
## // Observed heterozygosity: 0.44 0.81 0.83 0.73 0.78 0.8 0.81 0.17 0.86 0.71 0.81 0.78 0.71 0.19
## // Expected heterozygosity: 0.8 0.8 0.83 0.75 0.8 0.8 0.82 0.18 0.87 0.72 0.8 0.79 0.82 0.19
```

Test for Hardy-Weinberg equilibrium

We tested for Hardy-Weinberg equilibrium in the combined dataset consisting of adults and unrelated chicks (one chick per brood).

```
#### Calculate Hardy-Weinberg equilibrium ####

# First all together
allHWE.all <- pegas::hw.test(all.raw, B = 1000)
#B = 1000 for 1000 Monte Carlo permutations
summary(allHWE.all )
```

```
##      chi^2              df          Pr(chi^2 >)        Pr.exact
##  Min.   :   7.96   Min.   : 10.00   Min.   :0.00000   Min.   :0.0000
##  1st Qu.:  65.45   1st Qu.: 36.00   1st Qu.:0.00000   1st Qu.:0.0160
##  Median : 121.30   Median : 55.00   Median :0.08816   Median :0.0615
##  Mean   : 631.64   Mean   : 88.64   Mean   :0.33660   Mean   :0.1786
##  3rd Qu.: 490.24   3rd Qu.: 87.75   3rd Qu.:0.69941   3rd Qu.:0.1265
##  Max.   :2771.65   Max.   :253.00   Max.   :0.99999   Max.   :0.8520
```

```
# Then per population using a for loop
allpop <- seppop(all.raw)
# Run loop
allHWE = NULL
for(i in 1:length(allpop)) {
  hwt <- pegas::hw.test(allpop[[i]], B=1000)
  smry <- summary(allpop[[i]])
  Hobs <- smry[[6]]
  Hexp <- smry[[7]]
  pexact <- hwt[,4] #hw.test does chi2 test and exact test.
  #We use p-values of exact test which are given in 4th col
  qval.FDR <- p.adjust(pexact, method = "fdr")
  qval.bon <- p.adjust(pexact, method = "bonferroni")
  allHWE <- as.data.frame(cbind(allHWE, Hobs, Hexp, pexact, qval.FDR, qval.bon))

}

#then we transform the resulting allpop dataframe into something readable (hidden)
```

```
#### Results Hardy-Weinberg ####

head(allHWE.t) #HWE per locus per site
```

```
##          Site  L01  L02  L03  L04  L05  L06  L07  L08  L09  L10  L11  L12  L13
## Hobs        1 0.34 0.83 0.82 0.73 0.79 0.84 0.84 0.21 0.83 0.67 0.81 0.75 0.78
## Hexp        1 0.80 0.82 0.81 0.76 0.79 0.81 0.81 0.21 0.89 0.70 0.81 0.77 0.83
## pexact      1 0.00 0.07 0.61 0.58 0.98 0.15 0.53 0.47 0.45 0.10 0.40 0.42 0.01
## qval.FDR    1 0.00 0.31 0.66 0.66 0.98 0.42 0.66 0.66 0.66 0.35 0.66 0.66 0.06
## qval.bon    1 0.00 0.92 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 1.00 0.11
## Hobs.1      2 0.36 0.82 0.84 0.70 0.74 0.79 0.80 0.27 0.88 0.69 0.81 0.74 0.71
##           L14
## Hobs     0.19
## Hexp     0.19
## pexact   0.57
## qval.FDR 0.66
## qval.bon 1.00
## Hobs.1   0.28
```

This table includes observed heterozygosity (Hobs), expected heterozygosity (Hexp), pexact, qval.FDR and qval.bon for each locus in (columns) for each site (1-12). The full output of the table can be found in Supplementary Table 3. We included a threshold in which a locus was excluded if the FDR-corrected value was lower than 0.05 in over 70% of the sites. Subsequently, we exclude locus 1 and 13 (locus 1 is not included in the manuscript as it resulted in PCA plots for females with very unexpected patterns (erogenous clustering)), most likely due to some genotyping and/or scoring error.

Analysing population structure

To investigate patterns of genetic differentiation, we calculated pairwise F_{ST} values and conducted a STRUCTURE analysis. Here, we go through calculating summary statistics, constructing a PCA to get a grasp of the distribution of our data and identify potential outliers, and calculating the pairwise F_{ST} values for adult males, adult females and chicks separately.

Moreover, we compare observed and expected heterozygosity as well as allelic richness between hunted and unhunted sites, through modelling the effect of hunting on these three genetic diversity measures.

```
#using filtered structure files excluding loci out of HWE
all <- read.structure("data/cleandata/Microsat.adults.plus.unrelated.chicks.noLOCUS1+13.forstructure.st
                      n.ind = 2078, n.loc = 12, onerowperind = F,
                      col.lab = 1, col.pop = 2, col.others = NULL,
                      row.marknames = 0, NA.char = "-9",
                      pop = NULL, sep = NULL,
                      ask = F, quiet = FALSE)
```

```
##
##  Converting data from a STRUCTURE .stru file to a genind object...
```

```
#### Summary statistics ####

# This is based on adults and unrelated chicks across all loci
basicstat.all <- basic.stats(all, diploid = TRUE, digits = 2)
allelic.richness.all <- allelic.richness(all, diploid = TRUE)

#Ar
allelic.richness.all.df <- as.data.frame(allelic.richness.all$Ar)
head(allelic.richness.all.df)
```

```
##            1        2        3        4        5        6        7        8
## L01 1.824264 1.814686 1.795509 1.769247 1.793099 1.808096 1.789011 1.775000
## L02 1.812080 1.821476 1.803870 1.832592 1.821816 1.840070 1.834645 1.841139
## L03 1.759738 1.721630 1.723841 1.728597 1.762764 1.767348 1.778652 1.700000
## L04 1.791671 1.793210 1.787625 1.803537 1.799412 1.774880 1.800813 1.807278
## L05 1.810506 1.800365 1.778548 1.806768 1.790072 1.814429 1.850643 1.809177
## L06 1.811080 1.808226 1.833731 1.794407 1.820189 1.839345 1.840808 1.795538
##            9       10       11       12
## L01 1.806083 1.773232 1.801402 1.788668
## L02 1.832251 1.844913 1.826493 1.816873
## L03 1.748892 1.737102 1.739391 1.773284
## L04 1.811973 1.799742 1.786466 1.781969
## L05 1.763402 1.802837 1.773211 1.784864
## L06 1.846761 1.830337 1.807819 1.837800
```

```
# full table of allelic richness can be found in Supplementary Table 1

#get mean Ho and Hx per pop
x.pop = seppop(all)
summary.by.pop = lapply(x.pop, summary)
Hobs.ls = rep(NA, length(summary.by.pop))
for (i in 1:length(summary.by.pop)){
  Hobs.ls[i] = mean(summary.by.pop[[i]]$Hobs, na.rm=TRUE)
}
Hobs.ls
```

```
##  [1] 0.6923074 0.6972209 0.6702899 0.6904110 0.6842245 0.7007807 0.7653959
##  [8] 0.6875874 0.6822344 0.6964826 0.6758077 0.6983662
```

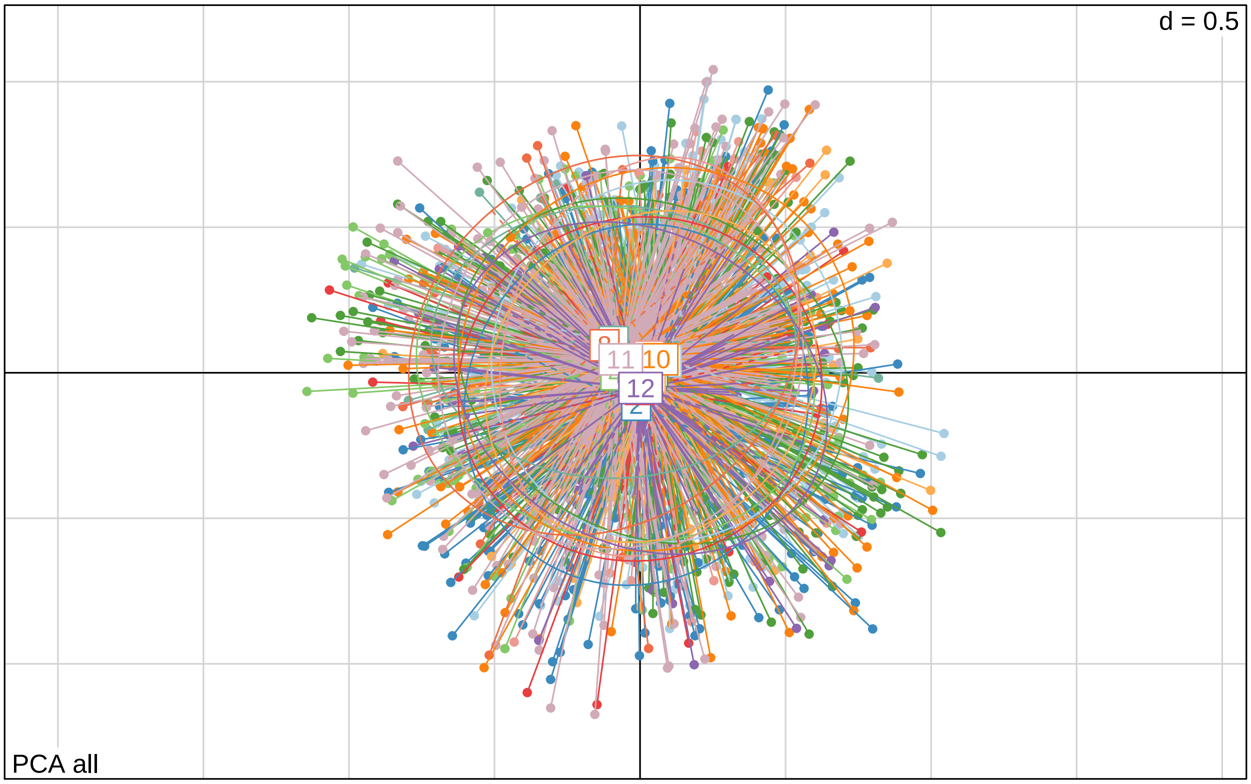
```
Hexp.ls = rep(NA, length(summary.by.pop))
for (i in 1:length(summary.by.pop)){
  Hexp.ls[i] = mean(summary.by.pop[[i]]$Hexp, na.rm=TRUE)
}
Hexp.ls
```

```
##  [1] 0.6979300 0.7025220 0.6666470 0.6883917 0.6843798 0.6878527 0.7656413
##  [8] 0.6808575 0.6722769 0.6940767 0.6779083 0.6976739
```

```
#### PCA ####
basic.stats(all)
```

```
## $perloc
##         Ho     Hs     Ht     Dst    Htp    Dstp     Fst    Fstp     Fis    Dest
## L01 0.8094 0.7948 0.8000  0.0052 0.8005  0.0057  0.0065  0.0071 -0.0184  0.0278
## L02 0.8432 0.8273 0.8321  0.0048 0.8325  0.0053  0.0058  0.0063 -0.0193  0.0305
## L03 0.7240 0.7452 0.7500  0.0047 0.7504  0.0052  0.0063  0.0069  0.0285  0.0203
## L04 0.7796 0.7950 0.8037  0.0087 0.8045  0.0095  0.0108  0.0118  0.0193  0.0463
## L05 0.8069 0.7987 0.8039  0.0052 0.8044  0.0057  0.0065  0.0071 -0.0103  0.0282
## L06 0.8060 0.8222 0.8259  0.0037 0.8263  0.0040  0.0045  0.0049  0.0197  0.0228
## L07 0.1461 0.1575 0.1508 -0.0067 0.1502 -0.0073 -0.0441 -0.0488  0.0721 -0.0087
## L08 0.8773 0.8669 0.8711  0.0042 0.8715  0.0046  0.0048  0.0052 -0.0119  0.0343
## L09 0.7338 0.7206 0.7256  0.0050 0.7261  0.0055  0.0069  0.0076 -0.0183  0.0197
## L10 0.7940 0.7926 0.7966  0.0040 0.7970  0.0044  0.0051  0.0055 -0.0018  0.0213
## L11 0.7853 0.7931 0.7951  0.0020 0.7953  0.0022  0.0025  0.0027  0.0098  0.0105
## L12 0.1839 0.1856 0.1859  0.0003 0.1860  0.0004  0.0019  0.0020  0.0090  0.0005
##
## $overall
##     Ho     Hs     Ht    Dst    Htp   Dstp    Fst   Fstp    Fis   Dest
## 0.6908 0.6916 0.6951 0.0034 0.6954 0.0038 0.0050 0.0054 0.0012 0.0122
```

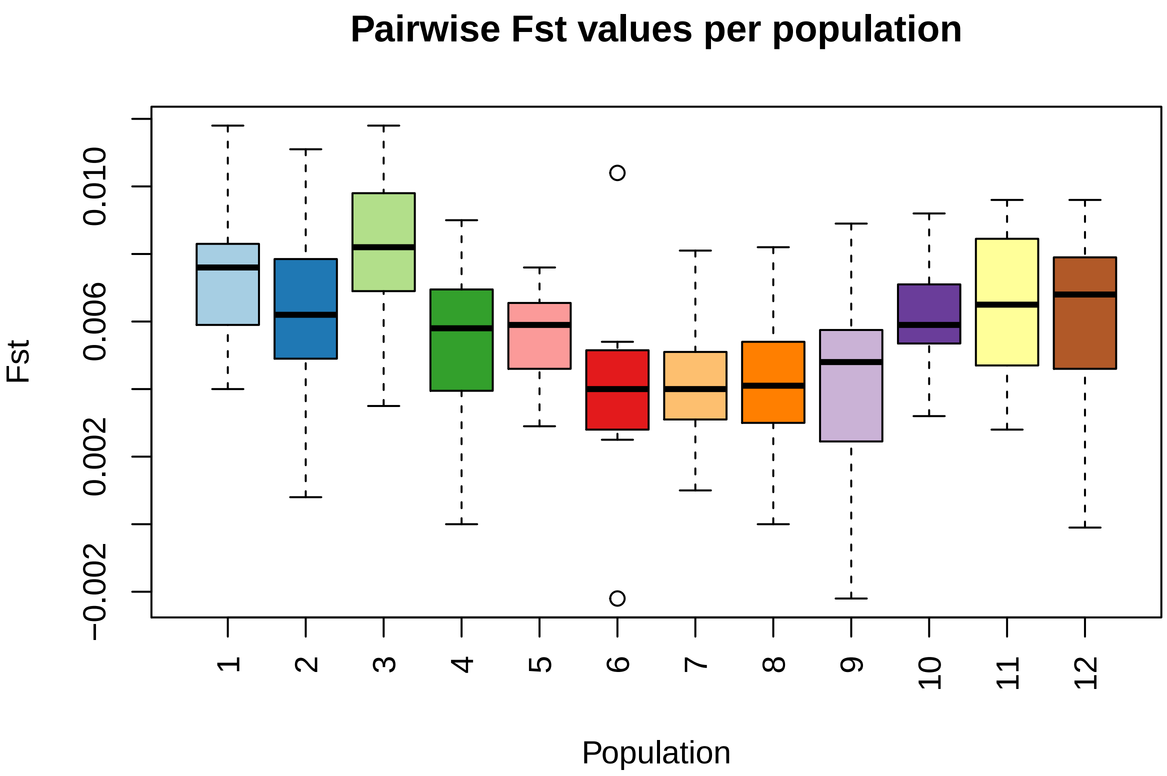
```
x <- tab(all, freq=TRUE, NA.method="mean")
pca <- dudi.pca(df = x, center = TRUE, scale = FALSE, scannf = FALSE, nf = 3)
s.class(pca$li, fac=pop(all), col=funky(15), sub = "PCA all")
```



```
#### Calculate Fst ####

#convert to hfstat object
all.hfstat <- genind2hierfstat(all)
#calculate statsf
basicstat.all <- basic.stats(all, diploid = TRUE, digits = 2)

# per locus
fst.all.perlocus <- basicstat.all$perloc$Fst
fst.all.perlocus <- data.frame(Locus = seq(from = 1, to = 12), Fst = fst.all.perlocus)

# Pairwise Fst
fst.all <- pairwise.neifst(all.hfstat)
head(fst.all)
```

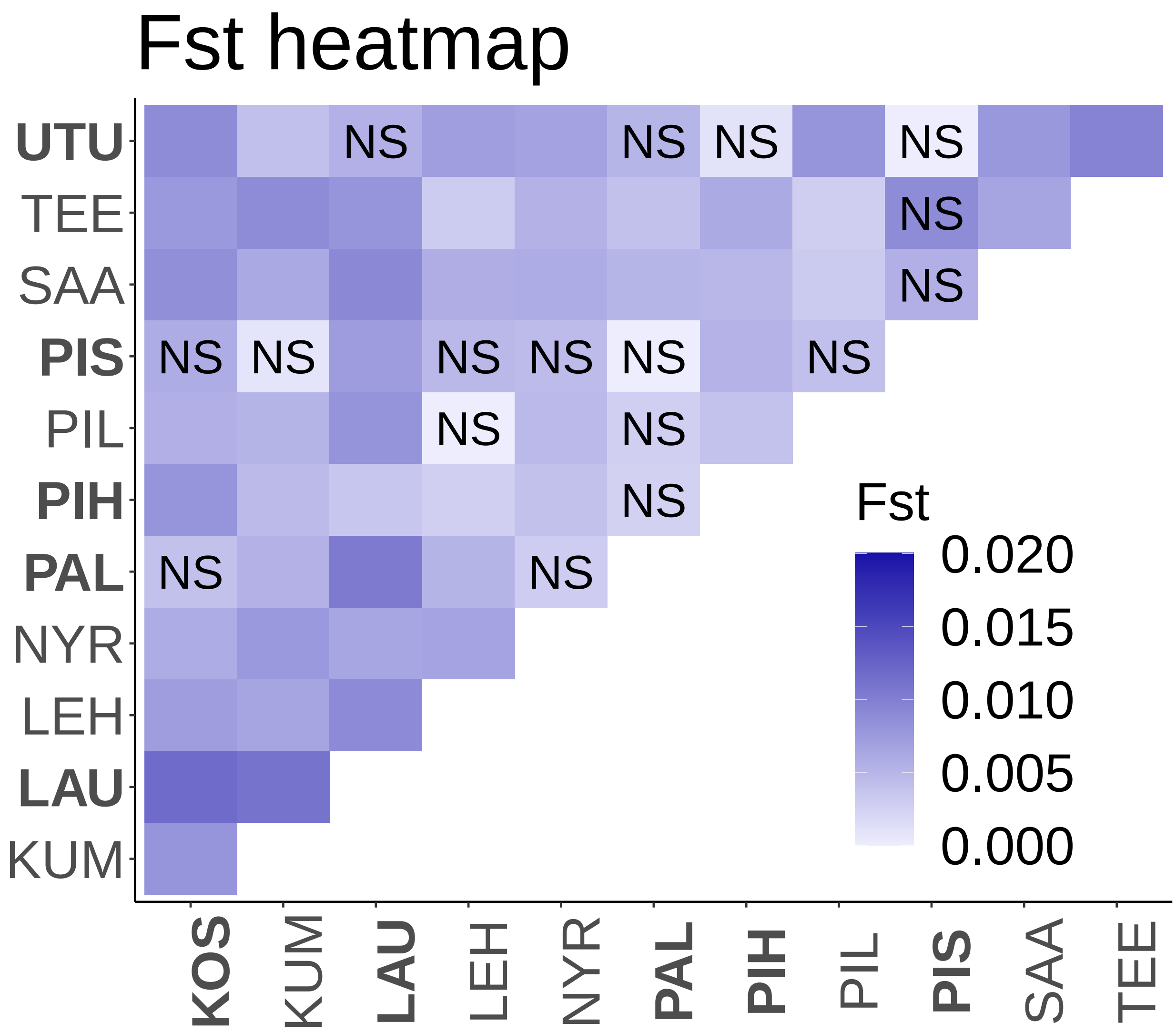
```
##        1      2      3      4      5      6      7      8       9     10     11
## 1     NA 0.0081 0.0118 0.0073 0.0059 0.0040 0.0081 0.0056  0.0059 0.0085 0.0076
## 2 0.0081     NA 0.0111 0.0066 0.0076 0.0054 0.0046 0.0052  0.0008 0.0062 0.0088
## 3 0.0118 0.0111     NA 0.0090 0.0064 0.0104 0.0035 0.0082  0.0074 0.0092 0.0081
## 4 0.0073 0.0066 0.0090     NA 0.0067 0.0052 0.0027 0.0000  0.0048 0.0058 0.0031
## 5 0.0059 0.0076 0.0064 0.0067     NA 0.0029 0.0040 0.0047  0.0045 0.0059 0.0054
## 6 0.0040 0.0054 0.0104 0.0052 0.0029     NA 0.0025 0.0027 -0.0022 0.0051 0.0040
##       12
## 1 0.0089
## 2 0.0041
## 3 0.0055
## 4 0.0072
## 5 0.0068
## 6 0.0051
```

```
# Fst per population
boxplot(fst.all, col=funky(nPop(all)), las=3,
        xlab="Population", ylab="Fst", main = "Pairwise Fst values per population")
```



```
#Bootstrap
boot.fst.all <- boot.ppfst(all.hfstat, nboot = 1000)
```

```
 #purples from structure
ggplot(pairwise.fst, aes(abb.x, abb.y, fill = Fst)) + geom_tile() + theme_classic() +
  scale_fill_gradientn(colors = mypalette3, limits = c(0,0.02)) +
  geom_text(aes(label = Sig), size = 8)+
  theme(text = element_text(size = 22),
        axis.text.x = element_text(angle = 90, size = 26,
                                   face = c("bold", "plain", "bold", "plain", "plain",
                                            "bold", "bold", "plain", "bold",
                                            "plain", "plain")),
        axis.text.y = element_text(size = 26,
                                   face = c("plain", "bold", "plain", "plain",
                                            "bold", "bold", "plain", "bold",
                                            "plain", "plain", "bold")),
        axis.title.x = element_blank(),
        axis.title.y = element_blank(),
        legend.text = element_text(size = 26),
        legend.title = element_text(size = 26),
        legend.key.size = unit(1, 'cm'),
        plot.title = element_text(size = 38),
        legend.position = c(0.8, 0.3)) +
  ggtitle('Fst heatmap')
```



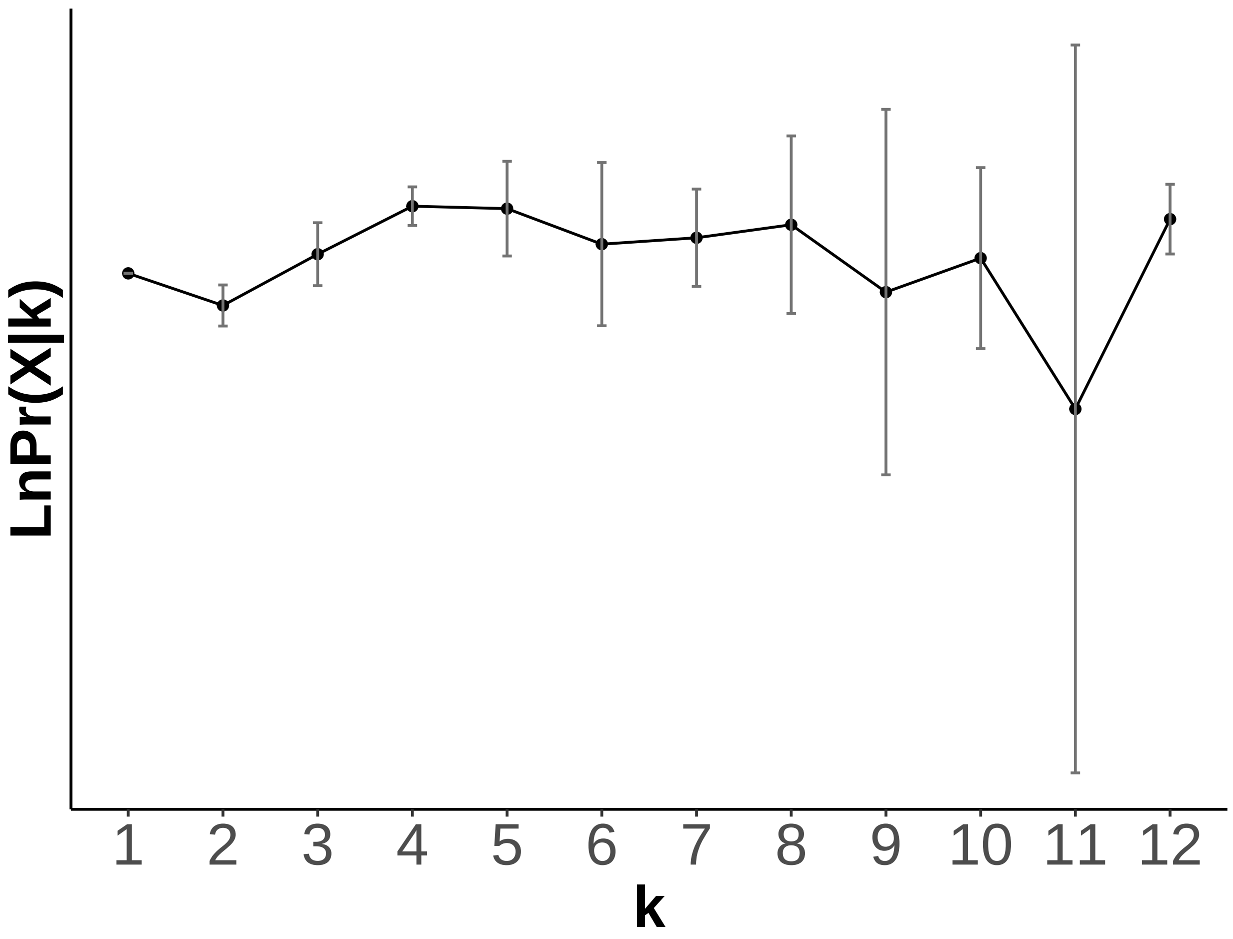
Structure analysis

We used ParallelStructure to run STRUCTURE on multiple cores. Details on inferring the highest likelihood K and plotting the barcharts can be found in the full script within the GitHub directory called 3.STRUCTUREanalysis.R.

Ensure you have enough computational power to conduct this analysis.

```r
##### Running structure #####

all <- fread("data/cleandata/Microsat.adults.plus.unrelated.
             chicks.noLOCUS1+13.forstructure.stru")

infile <- "/data/home/rchen/Hunting/blackgrouse-hunting/data/
cleandata/Microsat.adults.plus.unrelated.chicks.noLOCUS1+13.forstructure.stru"

outpath <- "/data/home/rchen/Hunting/blackgrouse-hunting/analyses/
structure/results/"

# job matrix and write to job file
nrep <- 10
burnin <- 10000
niter <- 10000
up_to_k <- 12

# job matrix
k_var <- rep(1:up_to_k, each = nrep)
ID_var <- as.character(sapply(c(1:up_to_k), function(k)
  sapply(c(1:nrep), function(x) paste0("T",k, "_", x))))

# make the job matrix
pop <- "1,2,3,4,5,6,7,8,9,10,11,12" #number of pops in the file

hunt_jobs <- matrix(c(ID_var, rep(pop, nrep * up_to_k), k_var,
                      rep(burnin, nrep * up_to_k),
                      rep(niter, nrep * up_to_k)), nrow = nrep * up_to_k)

write(t(hunt_jobs), ncol = length(hunt_jobs[1,]),
      file = "analyses/structure/structure_jobs.txt")

# file path to structure

STR_path='/usr/local/bin/'

# Run Parallel Structure

# Run structure (from terminal, do not run this last part in Rstudio)

parallel_structure(structure_path=STR_path,
                                      joblist='analyses/structure/structure_jobs.txt',
                   n_cpu=45, infile=infile,
                   outpath=outpath,
                   numinds = nrow(all)/2,
                   numloci=ncol(all)-2,
                   noadmix = 0, alpha = 1.0, freqscorr=1,
                   lambda = 1,
                   printqhat=1, plot_output=0,
                   onerowperind=0, locprior = 0)
```

These STRUCTURE analyses gave us the following results:



Sex biased dispersal

Next, we executed Mantel tests and spatial auto-correlation in GenAlEx (Excel add-in) for the sexes separately. To infer sex-biased dispersal directly, we also executed a sex-biased dispersal test in GenAlEx. These analyses were not conducted in R, but the raw data sheets can be found in the analyses/genalex directory, as well as the GenAlEx excel sheets.

Effect of hunting on genetic diversity

To understand the effect of hunting on genetic diversity, we first conduct a t-test comparing observed heterozygosity, expected heterozygosity and allelic richness between hunted and unhunted populations. Next, we construct a lm to include random effects of locus and population on the genetic diversity measures.

```
## First: t-test to compare differences in genetic diversity between
## hunted vs unhunted sites (where we have the mean value of all loci per site)

head(measures)
```

```
##   pop      Hobs      Hexp       Ar     hunt
## 1   1 0.6923074 0.6979300 1.699265   hunted
## 2   2 0.6972209 0.7025220 1.703523 unhunted
## 3   3 0.6702899 0.6666470 1.673973   hunted
## 4   4 0.6904110 0.6883917 1.690081 unhunted
## 5   5 0.6842245 0.6843798 1.685471 unhunted
## 6   6 0.7007807 0.6878527 1.694508   hunted
```

```
t.test(measures$Hobs[which(measures$hunt == "hunted")],
       measures$Hobs[which(measures$hunt == "unhunted")])
```

```
##
##  Welch Two Sample t-test
##
## data:  measures$Hobs[which(measures$hunt == "hunted")] and measures$Hobs[which(measures$hunt == "unhu
## t = 0.92695, df = 5.5838, p-value = 0.3923
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -0.02184505  0.04772519
## sample estimates:
## mean of x mean of y
## 0.7015624 0.6886223
```

```
t.test(measures$Hexp[which(measures$hunt == "hunted")],
       measures$Hexp[which(measures$hunt == "unhunted")])
```

```
##
##  Welch Two Sample t-test
##
## data:  measures$Hexp[which(measures$hunt == "hunted")] and measures$Hexp[which(measures$hunt == "unhu
## t = 0.66604, df = 5.6518, p-value = 0.5316
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -0.02724204  0.04720395
## sample estimates:
## mean of x mean of y
## 0.6980036 0.6880227
```

```
t.test(measures$Ar[which(measures$hunt == "hunted")],
       measures$Ar[which(measures$hunt == "unhunted")])
```

```
##
##  Welch Two Sample t-test
##
## data:  measures$Ar[which(measures$hunt == "hunted")] and measures$Ar[which(measures$hunt == "unhunte
## t = 0.74388, df = 5.9353, p-value = 0.4853
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -0.02019666  0.03777352
## sample estimates:
## mean of x mean of y
##  1.699395  1.690606
## Then, we model the effect of hunting on the three genetic diversity measures,
## where we take each measure per locus per site and perform a likelihood
## ratio test to assess the difference between the null and alternative model

head(measures.all)
```

```
##   locus pop      Hobs      Hexp       Ar   hunt
## 1   L01   1 0.8284672 0.8227596 1.824264 hunted
## 2   L02   1 0.8175182 0.8105986 1.812080 hunted
## 3   L03   1 0.7335766 0.7583515 1.759738 hunted
## 4   L04   1 0.7919708 0.7902259 1.791671 hunted
## 5   L05   1 0.8357664 0.8090269 1.810506 hunted
## 6   L06   1 0.8357664 0.8095996 1.811080 hunted
```

```
# excluding BG20 (out of HWE)
Ho_model <- lmerTest::lmer(Hobs ~ hunt + (1|locus) + (1|pop),
                           data = subset(measures.all, locus != "L13"))

Ho_model_null <- lmerTest::lmer(Hobs ~ (1|locus) + (1|pop),
                                data = subset(measures.all, locus != "L13"))

He_model <- lmerTest::lmer(Hexp ~ hunt + (1|locus) + (1|pop),
                           data = subset(measures.all, locus != "L13"))
He_model_null <- lmerTest::lmer(Hexp ~ (1|locus) + (1|pop),
                                data = subset(measures.all, locus != "L13"))

Ar_model <- lmerTest::lmer(Ar ~ hunt + (1|locus) + (1|pop),
                           data = subset(measures.all, locus != "L13"))
Ar_model_null <- lmerTest::lmer(Ar ~ (1|locus) + (1|pop),
                                data = subset(measures.all, locus != "L13"))

#LRT
anova(Ho_model, Ho_model_null) # no significant difference
```

```
## Data: subset(measures.all, locus != "L13")
## Models:
## Ho_model_null: Hobs ~ (1 | locus) + (1 | pop)
## Ho_model: Hobs ~ hunt + (1 | locus) + (1 | pop)
##               npar     AIC     BIC logLik deviance Chisq Df Pr(>Chisq)
## Ho_model_null    4 -392.20 -380.35 200.10  -400.20
## Ho_model         5 -390.51 -375.70 200.26  -400.51 0.314  1     0.5753
```

```
anova(He_model, He_model_null) # no significant difference
```

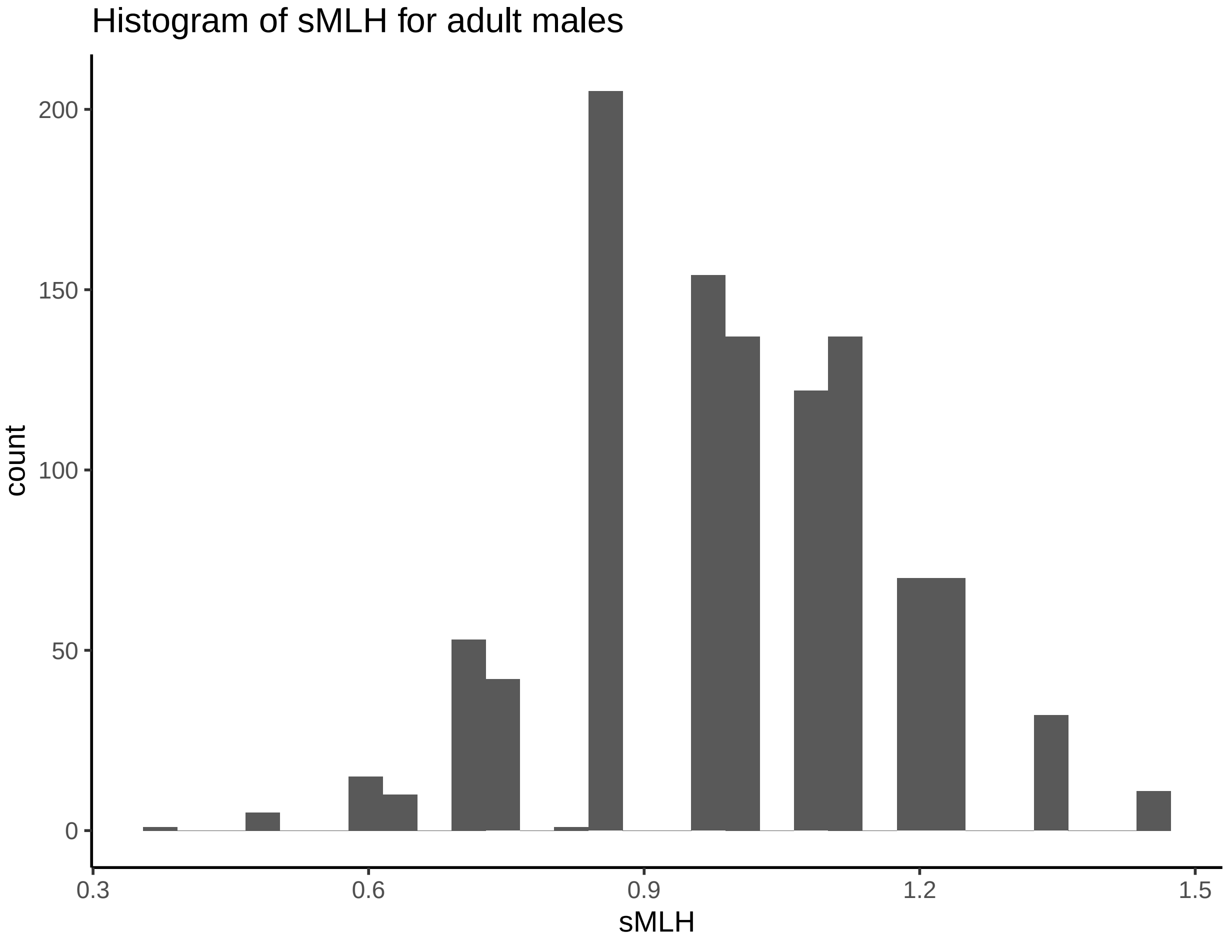
```
## Data: subset(measures.all, locus != "L13")
## Models:
## He_model_null: Hexp ~ (1 | locus) + (1 | pop)
## He_model: Hexp ~ hunt + (1 | locus) + (1 | pop)
##               npar     AIC     BIC logLik deviance Chisq Df Pr(>Chisq)
## He_model_null    4 -263.00 -251.12 135.50  -271.00
## He_model         5 -261.48 -246.63 135.74  -271.48 0.478  1     0.4893
```

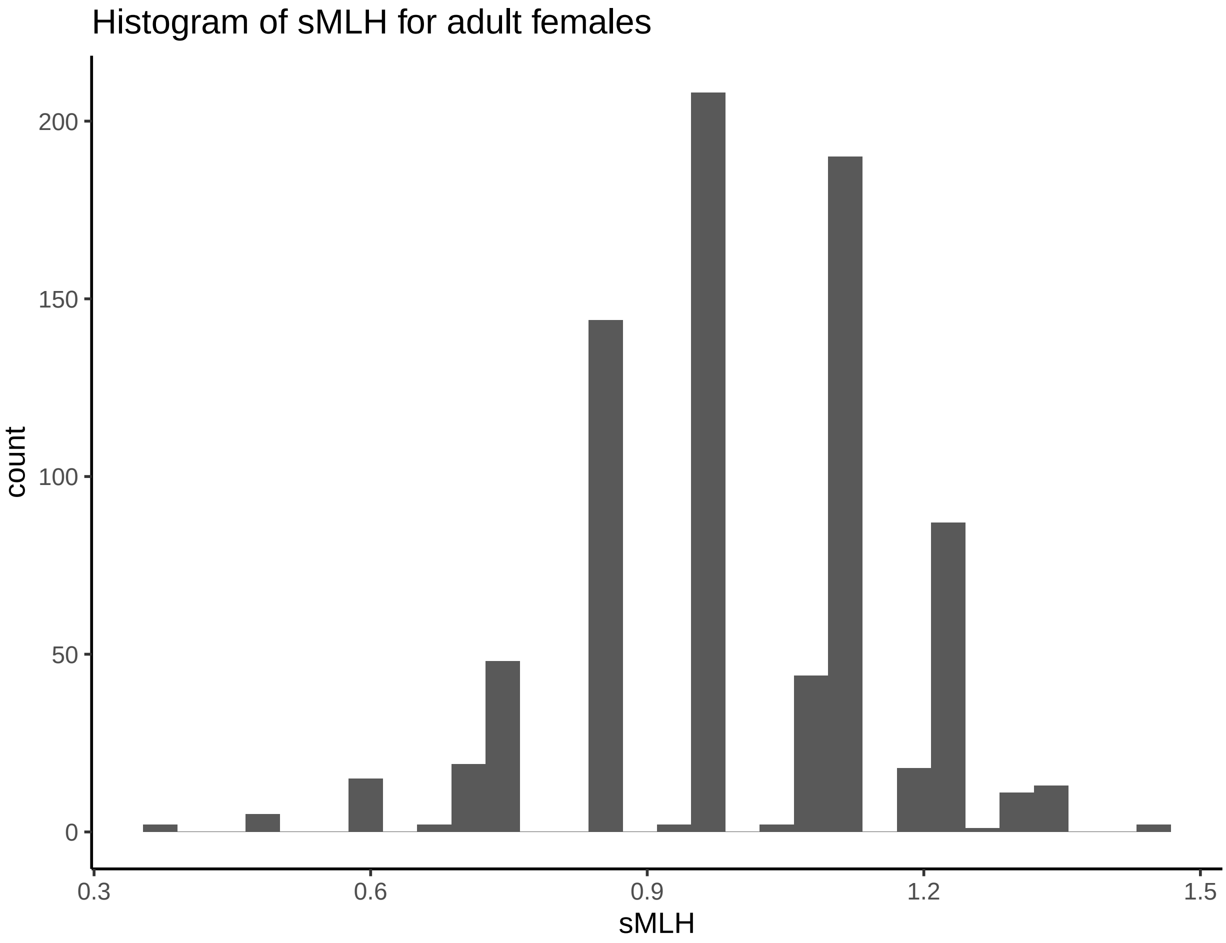
```
anova(Ar_model, Ar_model_null) # no significant difference
```

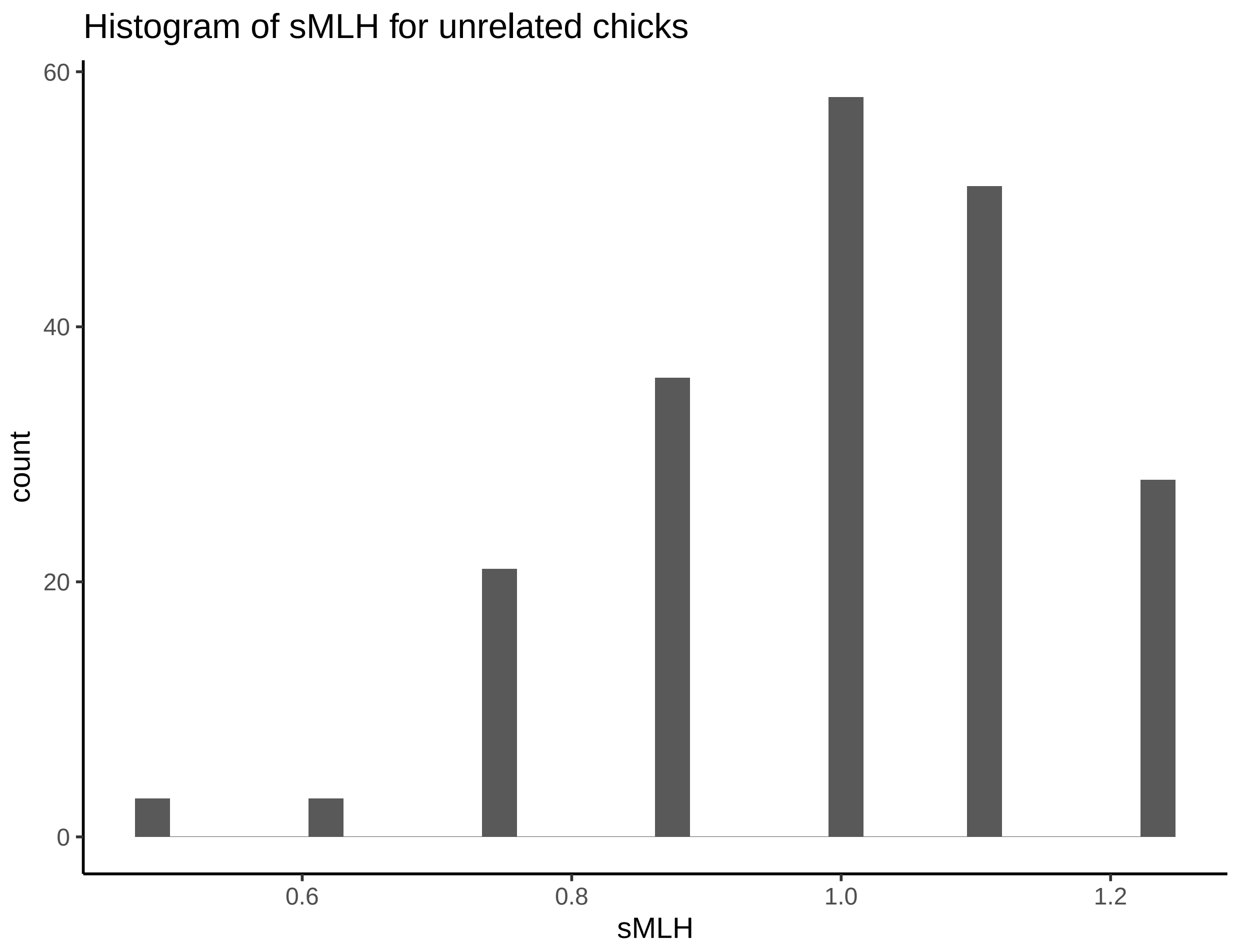
```
## Data: subset(measures.all, locus != "L13")
## Models:
## Ar_model_null: Ar ~ (1 | locus) + (1 | pop)
## Ar_model: Ar ~ hunt + (1 | locus) + (1 | pop)
##               npar     AIC     BIC logLik deviance Chisq Df Pr(>Chisq)
## Ar_model_null    4 -489.23 -477.38 248.61  -497.23
## Ar_model         5 -487.23 -472.42 248.62  -497.23 0.006  1     0.9384
```

Calculating and modelling sMLH

To quantify inbreeding levels, we calculated sMLH using the inbreedR package. Then, to understand the effects of hunting on inbreeding, we built a mixed-model and investigated the fit of the models.




Histogram of sMLH for adult females
count
200
150
100
50
0
0.3
0.6
0.9
1.2
1.5
sMLH



```
### Setup models ####
sMLH.all.no.na <- subset(sMLH.all, !is.na(density))
#exclude those without density otherwise lrt won't work

sMLH.model.all.lmer <- lmerTest::lmer(sMLH ~
                                        hunt*density + sex + age+ (1|pop),
                                      data = sMLH.all.no.na)
sMLH.model.all.lmer.noint <- lmerTest::lmer(sMLH ~
                                              hunt + sex + age+ density + (1|pop),
                                            data = sMLH.all.no.na)
sMLH.model.all.lmer.null <- lmerTest::lmer(sMLH ~
                                             density+ sex + age+ (1|pop),
                                           data = sMLH.all.no.na)

anova(sMLH.model.all.lmer.null, sMLH.model.all.lmer)
```

```
## Data: sMLH.all.no.na
## Models:
## sMLH.model.all.lmer.null: sMLH ~ density + sex + age + (1 | pop)
## sMLH.model.all.lmer: sMLH ~ hunt * density + sex + age + (1 | pop)
##                          npar     AIC     BIC logLik deviance Chisq Df
## sMLH.model.all.lmer.null    6 -1321.8 -1288.4 666.91  -1333.8
## sMLH.model.all.lmer         8 -1320.0 -1275.4 667.99  -1336.0 2.159  2
##                           Pr(>Chisq)
## sMLH.model.all.lmer.null
## sMLH.model.all.lmer          0.3398
```

```
anova(sMLH.model.all.lmer.null, sMLH.model.all.lmer.noint)
```

```
## Data: sMLH.all.no.na
## Models:
## sMLH.model.all.lmer.null: sMLH ~ density + sex + age + (1 | pop)
## sMLH.model.all.lmer.noint: sMLH ~ hunt + sex + age + density + (1 | pop)
##                            npar     AIC     BIC logLik deviance  Chisq Df
## sMLH.model.all.lmer.null      6 -1321.8 -1288.4 666.91  -1333.8
## sMLH.model.all.lmer.noint     7 -1321.7 -1282.6 667.83  -1335.7 1.8443  1
##                            Pr(>Chisq)
## sMLH.model.all.lmer.null
## sMLH.model.all.lmer.noint     0.1744
```

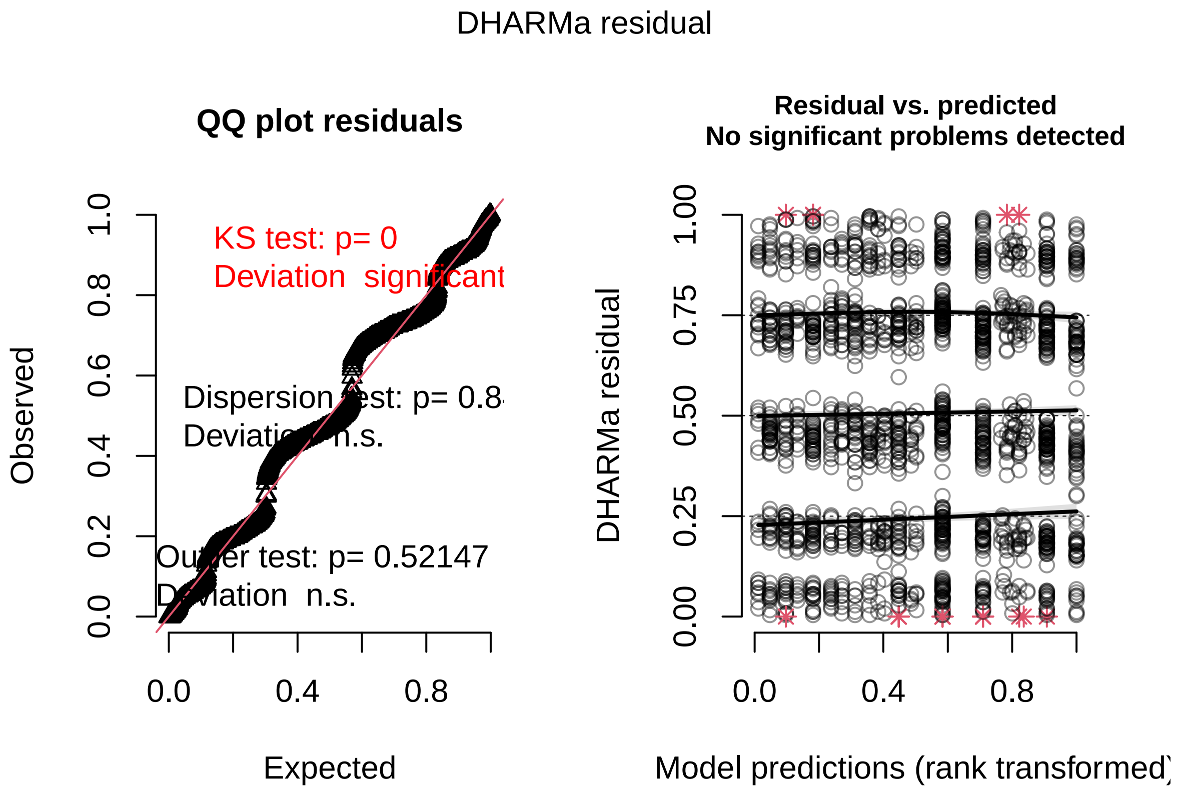
```
coef(summary(sMLH.model.all.lmer))
```

```
##                         Estimate  Std. Error        df    t value      Pr(>|t|)
## (Intercept)          0.980353176 0.015753847  198.1223 62.2294471 5.288112e-132
## hunthunted          -0.005409208 0.031355077  125.7485 -0.1725146  8.633104e-01
## density              0.001509275 0.001381680  967.5943  1.0923474  2.749524e-01
## sexm                -0.001392771 0.007890709 1771.8702 -0.1765077  8.599153e-01
## agechick             0.014933141 0.018091870 1930.0150  0.8254061  4.092431e-01
## hunthunted:density   0.001516963 0.002583694  415.7640  0.5871295  5.574354e-01
```

```
VarCorr(sMLH.model.all.lmer)
```

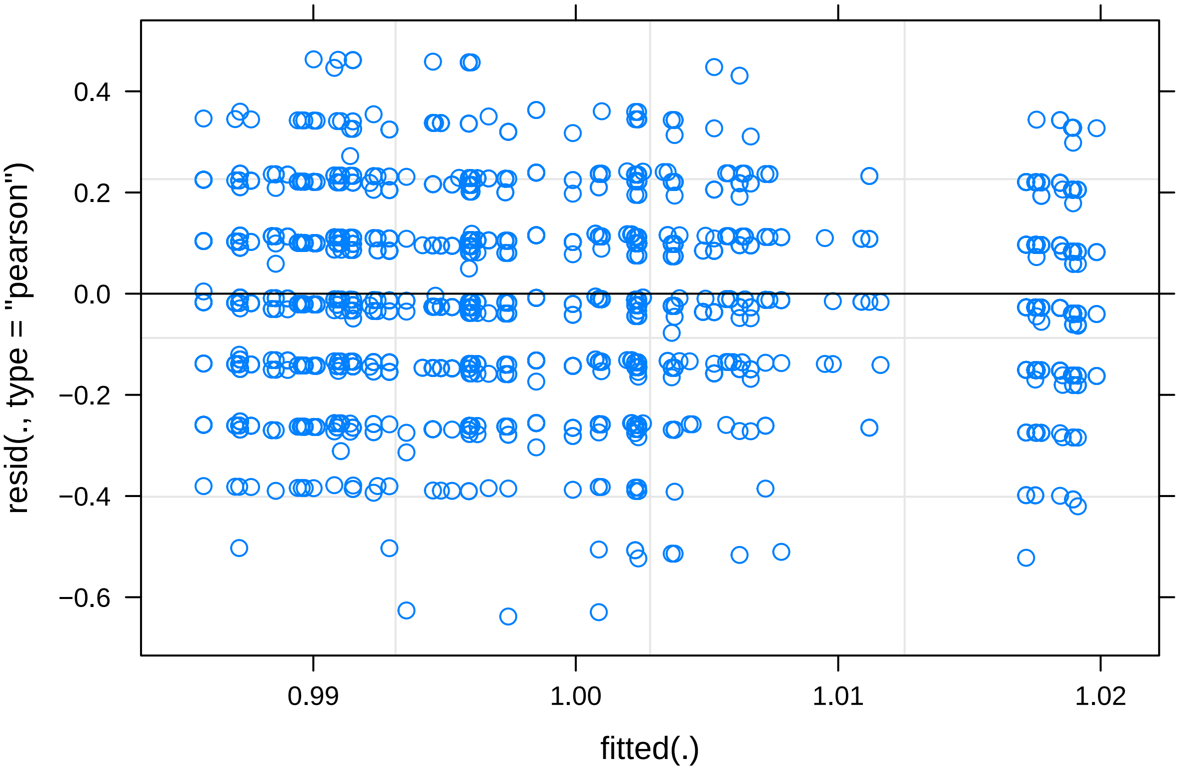
```
##  Groups   Name        Std.Dev.
##  pop      (Intercept) 0.0039595
##  Residual             0.1718672
```

```
simulateResiduals(fittedModel = sMLH.model.all.lmer, plot = T)
```



```
## Object of Class DHARMa with simulated residuals based on 250 simulations with refit = FALSE . See ?DI
##
## Scaled residual values: 0.188 0.76 0.756 0.048 0.468 0.076 0.748 0.732 0.884 0.744 0.884 0.436 0.988
```

```
plot(sMLH.model.all.lmer)
```



```
r.squaredGLMM(sMLH.model.all.lmer)
```

```
##              R2m         R2c
## [1,] 0.003327348 0.003856053
```

```
icc(model = sMLH.model.all.lmer, by_group = TRUE)
```

```
## # ICC by Group
## 
## Group |       ICC
## -----------------
## pop   | 5.305e-04
```

```
compare_performance(sMLH.model.all.lmer.null, sMLH.model.all.lmer, rank = T)
```

```
## # Comparison of Model Performance Indices
## 
## Name                     |           Model | R2 (cond.) | R2 (marg.) |       ICC |  RMSE | Sigma | A
## ----------------------------------------------------------------------------------------------------
## sMLH.model.all.lmer      | lmerModLmerTest |      0.004 |      0.003 | 5.305e-04 | 0.172 | 0.172 |
## sMLH.model.all.lmer.null | lmerModLmerTest |      0.003 |      0.002 | 5.137e-04 | 0.172 | 0.172 |
compare_performance(sMLH.model.all.lmer.null, sMLH.model.all.lmer.noint, rank = T)
```

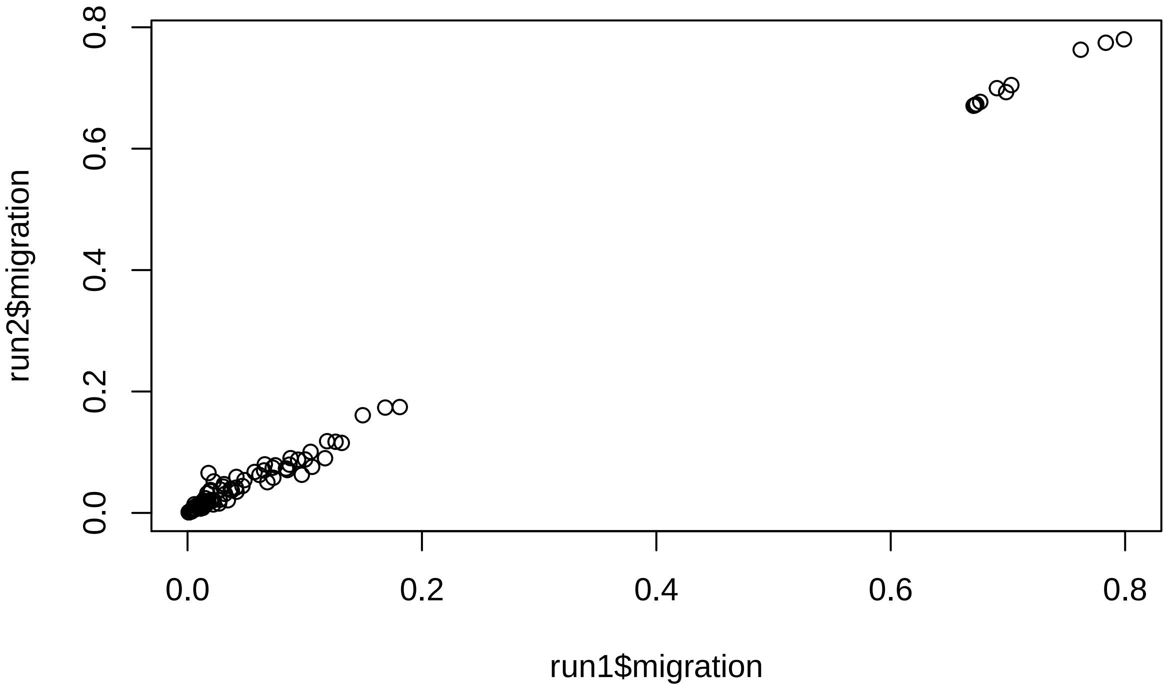
```
## # Comparison of Model Performance Indices
##
## Name                         |           Model | R2 (cond.) | R2 (marg.) |       ICC |  RMSE | Sigma |
## ----------------------------------------------------------------------------------------------------
## sMLH.model.all.lmer.noint | lmerModLmerTest |      0.004 |      0.003 | 3.198e-04 | 0.172 | 0.172 |
## sMLH.model.all.lmer.null  | lmerModLmerTest |      0.003 |      0.002 | 5.137e-04 | 0.172 | 0.172 |
```

Migration models

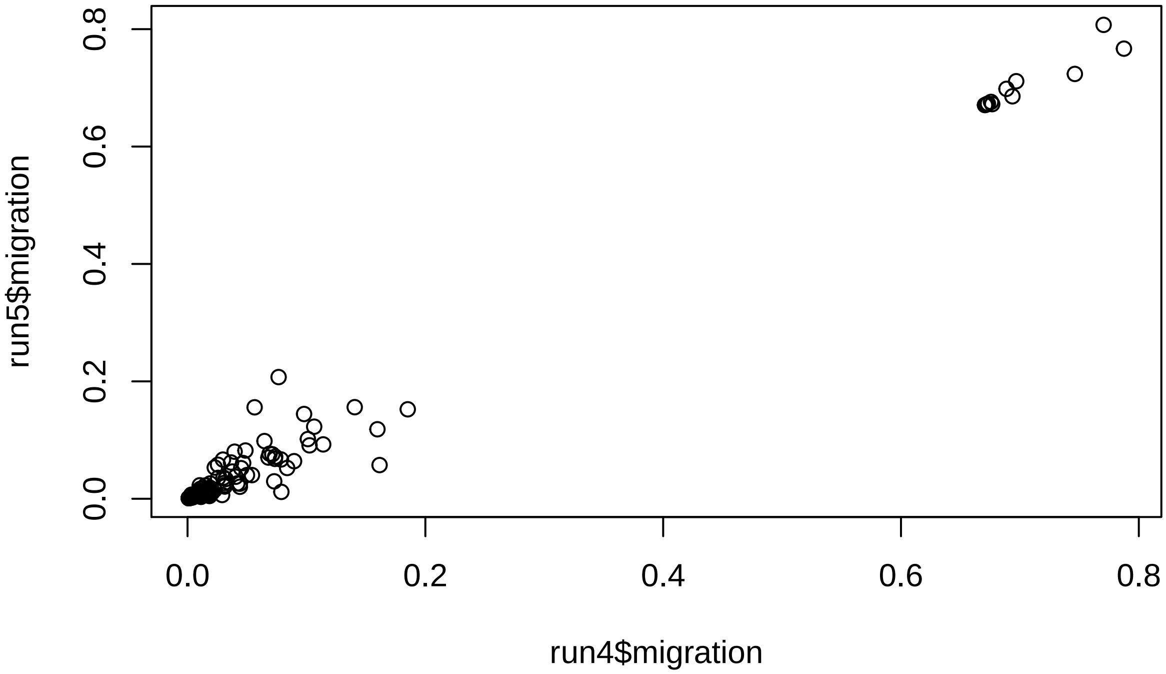
Next, we investigated patterns of dispersion and how these are affected by hunting. We used BA3 to calculate migration directions and rates, followed by building mixed models to estimate the effect of hunting on migration.

```
#### Running BA3 ####

#install BA3, run through command line/terminal

#5 runs with 5 different random seeds
system("~/bin/BA3/BA3MSAT -v -t -g -u -a 0.30 -f 0.40 -s 65323 -i 10000000
       -b 1000000 -n 500 -o run1_nohwe.txt
       analyses/migrationanalysis/data_all_ba3_allloci.txt") #repeat for another 4 runs
```

```
#### Compare runs ####
plot(run1$migration, run2$migration)
```

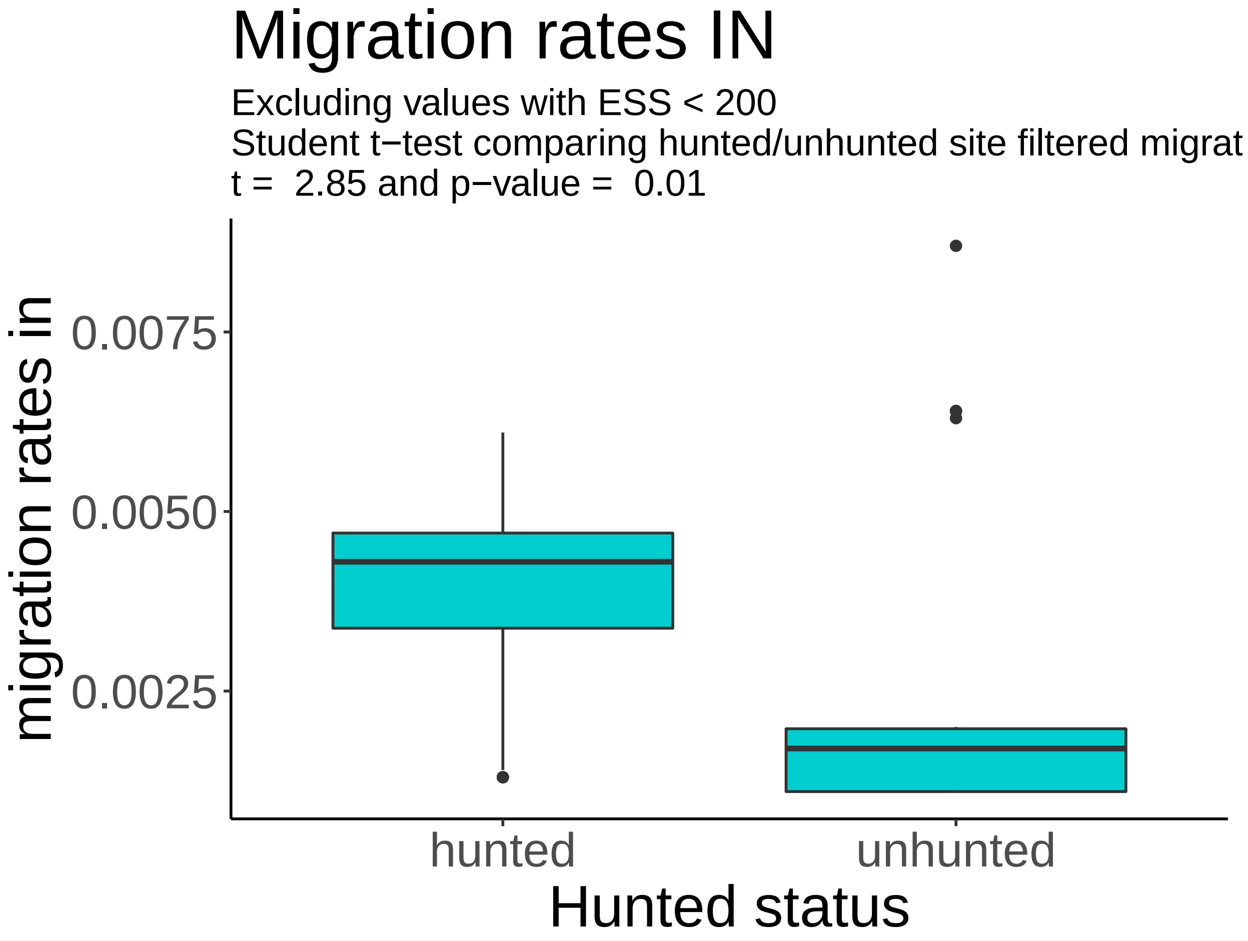


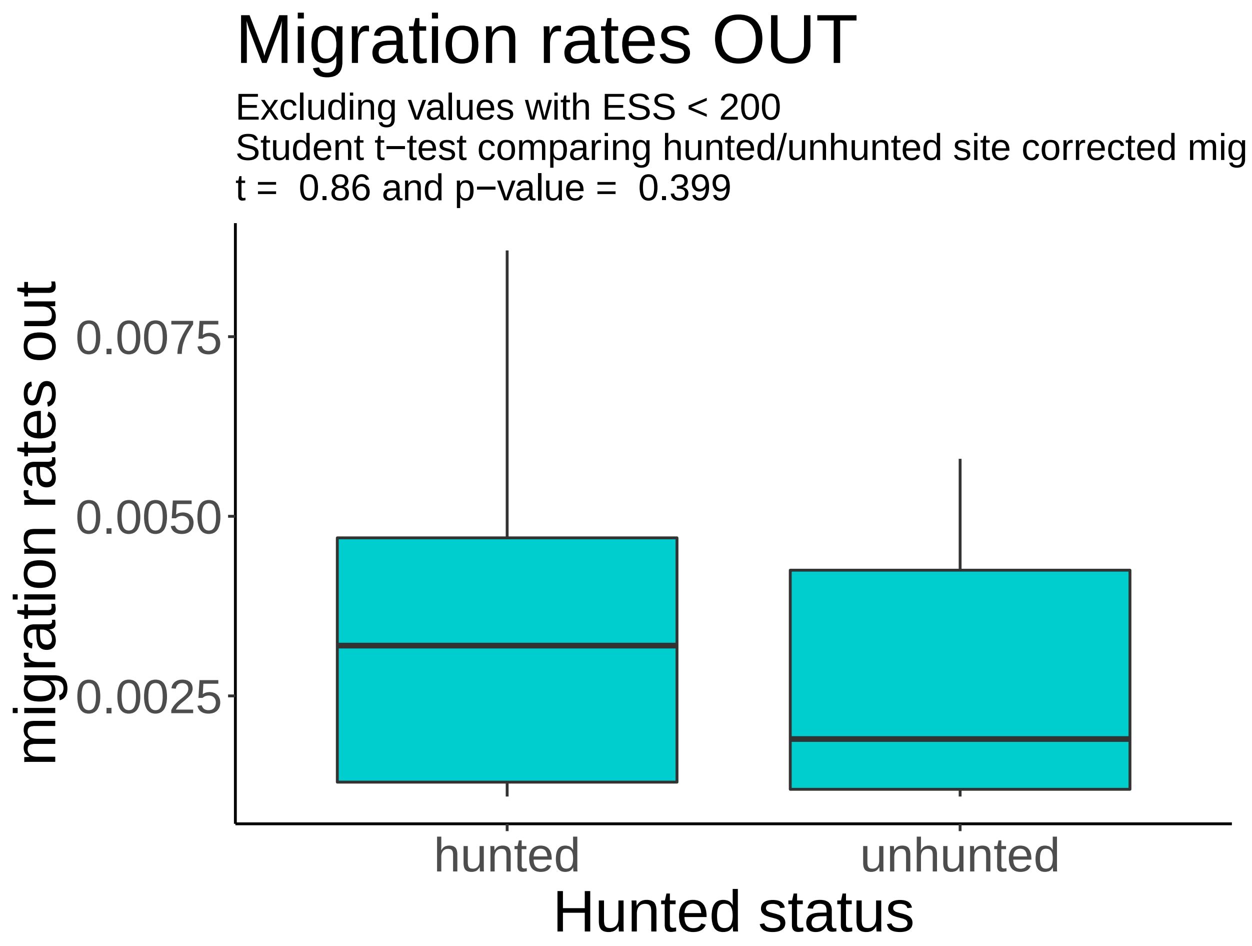
```
# plot(run1$migration, run3$migration) #hide these in RMarkdown
# plot(run1$migration, run4$migration) plot(run1$migration, run5$migration)
# plot(run2$migration, run3$migration) plot(run2$migration, run4$migration)
# plot(run2$migration, run5$migration) plot(run3$migration, run4$migration)
# plot(run3$migration, run5$migration)
plot(run4$migration, run5$migration)
```



```
# all runs correspond

## Going to pick run 5 for both
```

```
##   m_in m_out migration migration_SE    ESS migration_ESSc    pop_in hunt_in
## 1    1     1    0.6856       0.0122    9.2             NA Koskenpää  hunted
## 2    1     2    0.0097       0.0080   38.6             NA Koskenpää  hunted
## 3    1     3    0.0013       0.0013 1765.1         0.0013 Koskenpää  hunted
## 4    1     4    0.0169       0.0152   17.5             NA Koskenpää  hunted
## 5    1     5    0.2073       0.0457    6.1             NA Koskenpää  hunted
## 6    1     6    0.0020       0.0025   19.3             NA Koskenpää  hunted
##     pop_out hunt_out Distance
## 1 Koskenpää   hunted       NA
## 2 Kummunsuo unhunted 41.01050
## 3 Lauttasuo   hunted 54.91027
## 4  Lehtusuo unhunted 27.19093
## 5    Nyrölä unhunted 34.75899
## 6   Palosuo   hunted 38.39597
```


Migration rates IN
Excluding values with ESS < 200
Student t−test comparing hunted/unhunted site filtered migrat
t = 2.85 and p−value = 0.01
migration rates in
0.0075
0.0050
0.0025
hunted
unhunted
Hunted status



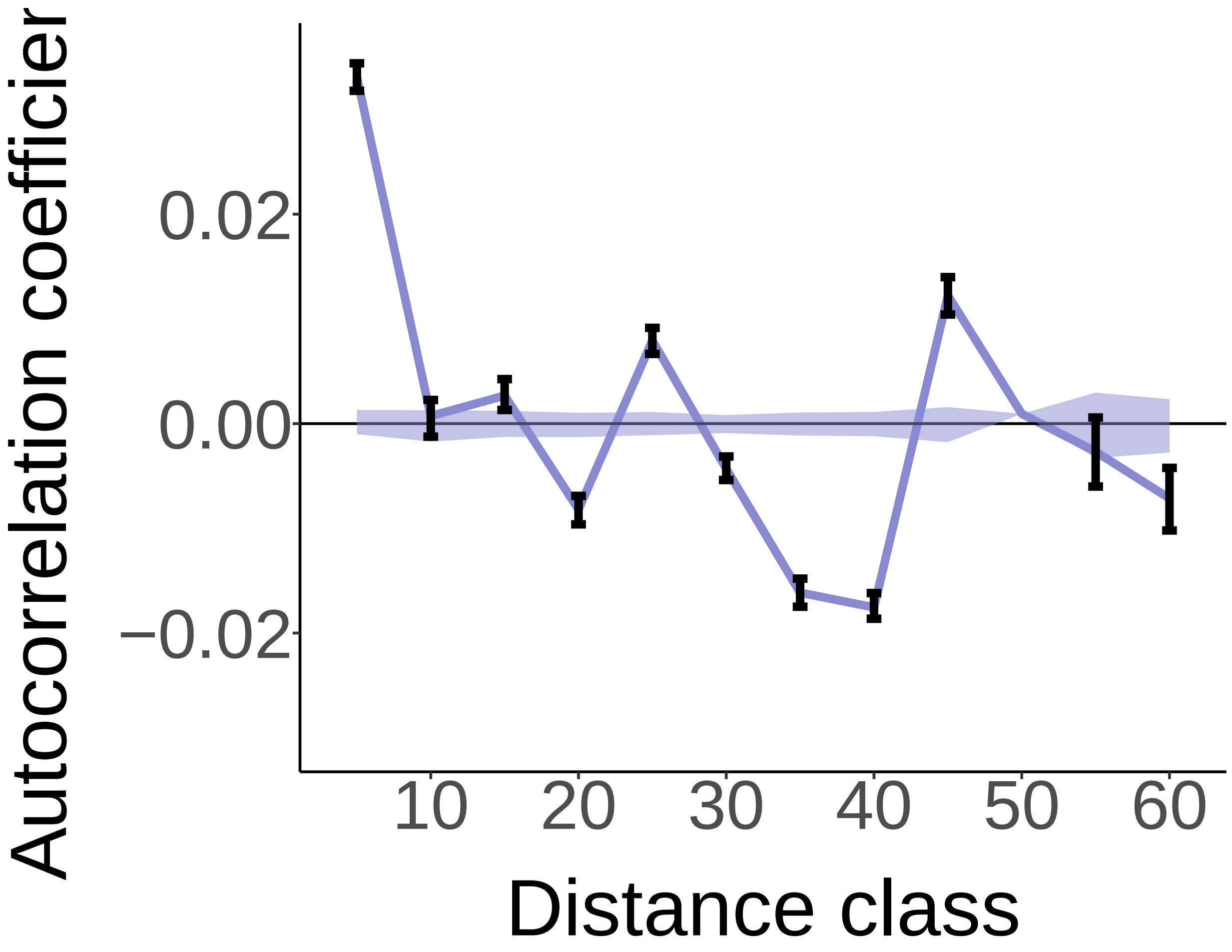
```
#### Modelling migration ####

#change levels hunted/unhunted
run5_clean$hunt_in <- relevel(as.factor(run5_clean$hunt_in), ref = "unhunted")
run5_clean$hunt_out <- relevel(as.factor(run5_clean$hunt_out), ref = "unhunted")

#immigration

model.in <- glmmTMB(migration_ESSc~ hunt_in + Distance +  (1|pop_out) + (1|pop_in),
                         data = run5_clean,
                         family = Gamma(link = "log"))

model.in.null <- glmmTMB(migration_ESSc~ Distance +  (1|pop_out) + (1|pop_in),
                         data = run5_clean,
                         family = Gamma(link = "log"))

anova(model.in, model.in.null)

summary(model.in)
simulateResiduals(fittedModel = model.in, plot = T)

#out
```

```
model.out <- glmmTMB(migration_ESSc~ hunt_out + Distance + (1|pop_out) + (1|pop_in),
                          data = run5_clean,
                          family = Gamma(link = "log"))
model.out.null <- glmmTMB(migration_ESSc~ Distance + (1|pop_out) + (1|pop_in),
                          data = run5_clean,
                          family = Gamma(link = "log"))

anova(model.out, model.out.null)

summary(model.out)
simulateResiduals(fittedModel = model.out, plot = T)

compare_performance(model.in, model.in.null, rank=T)
compare_performance(model.out, model.out.null, rank=T)
```

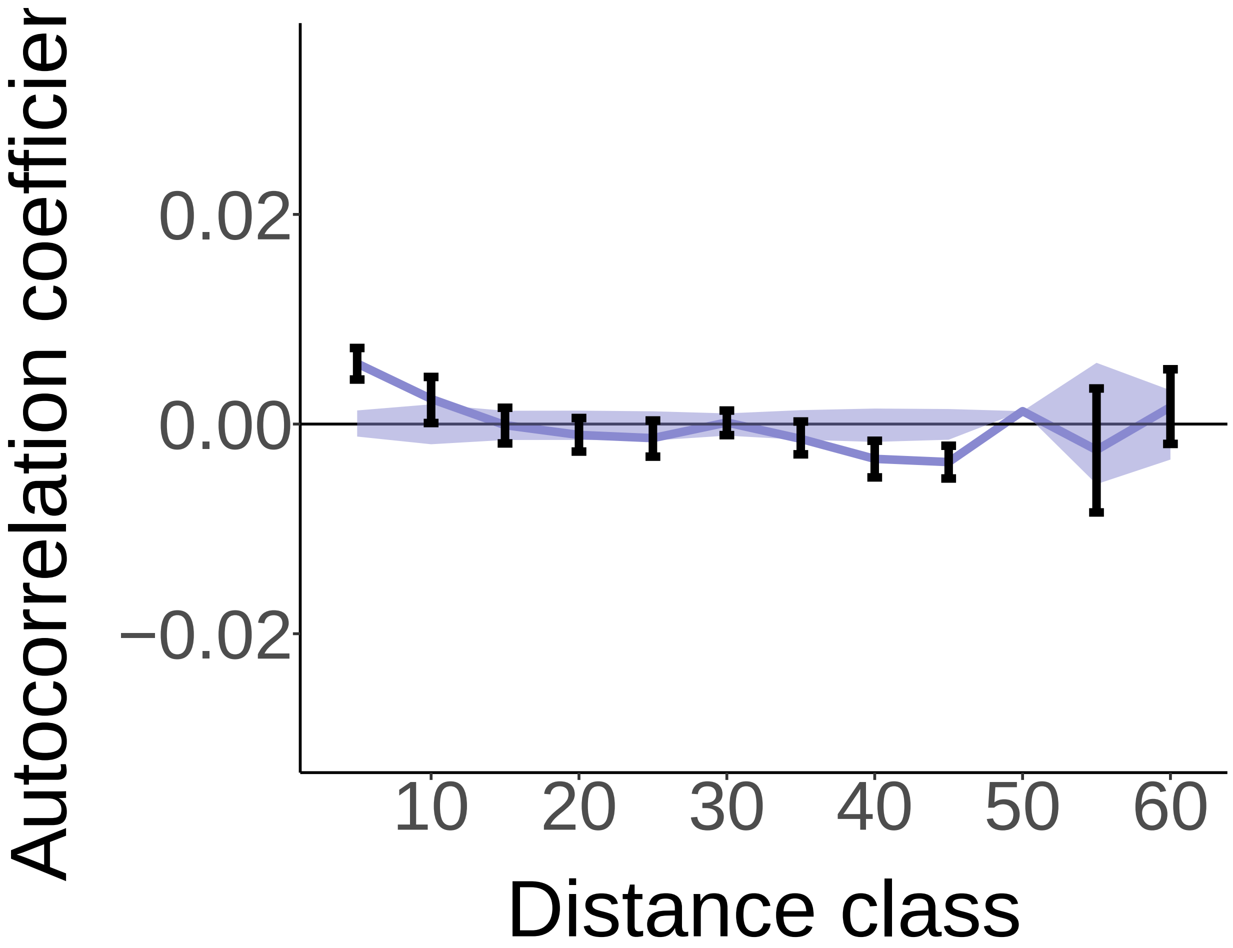
Other plots included in the manuscript

Figure 1 was created with qGIS, for barcharts of STRUCTURE output as in Supplementary Figure 2, please refer to R script 6.CreatingPlots.R

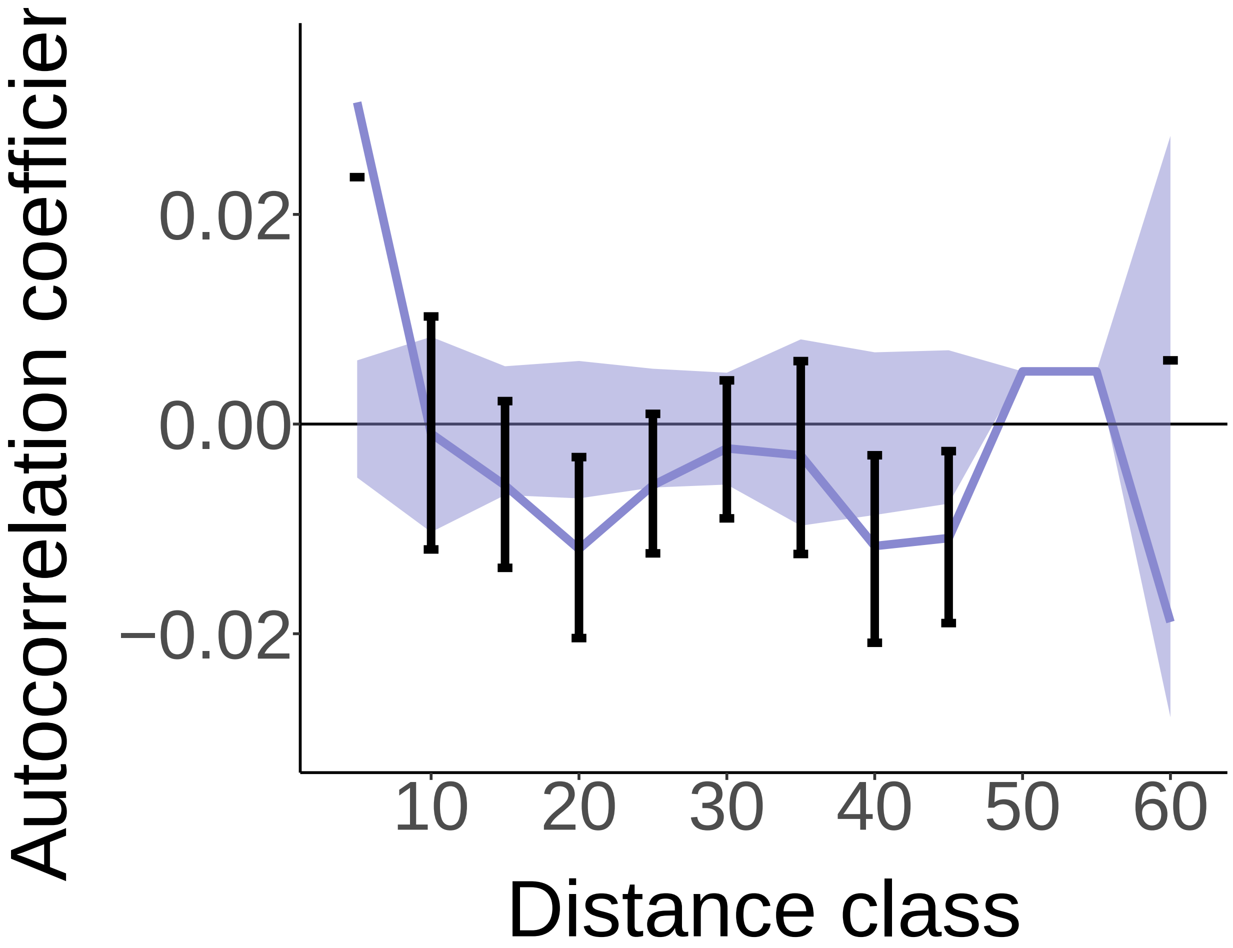
```
### Figure 2 - correlograms
spatial <- read_excel("tables/SpatialAutocor_04.10.22.xlsx", sheet = "ForR")

### Males - spatial
spatial %>% filter(Who == "Male") %>% ggplot(aes(x = What)) +
  geom_line(aes(y = r), col = "#8989D0", size = 1.5) + theme_classic() +
  geom_hline(yintercept = 0, col = "black")+
  geom_ribbon(aes(ymax = U, ymin = L), fill = "#8989D0", alpha = 0.5)+
  geom_errorbar(aes(y = r, ymin = r-Le, ymax = r+Ue), width=1,
              size=1.5, color="black", stat = "identity")+
  ylim(-0.03, 0.035) +
  xlab("Distance class") + ylab("Autocorrelation coefficient r")+
  scale_x_continuous(breaks = c(seq(0, 60, by = 10)), limits=c(4,61))+
  theme(text = element_text(size = 14),
        plot.title = element_text(size = 38),
        axis.text.x = element_text(size = 26, margin = margin(b = 10)),
        axis.text.y = element_text(size = 26, margin = margin(l = 10)),
        axis.title.x = element_text(size = 30),
        axis.title.y = element_text(size = 30))
```



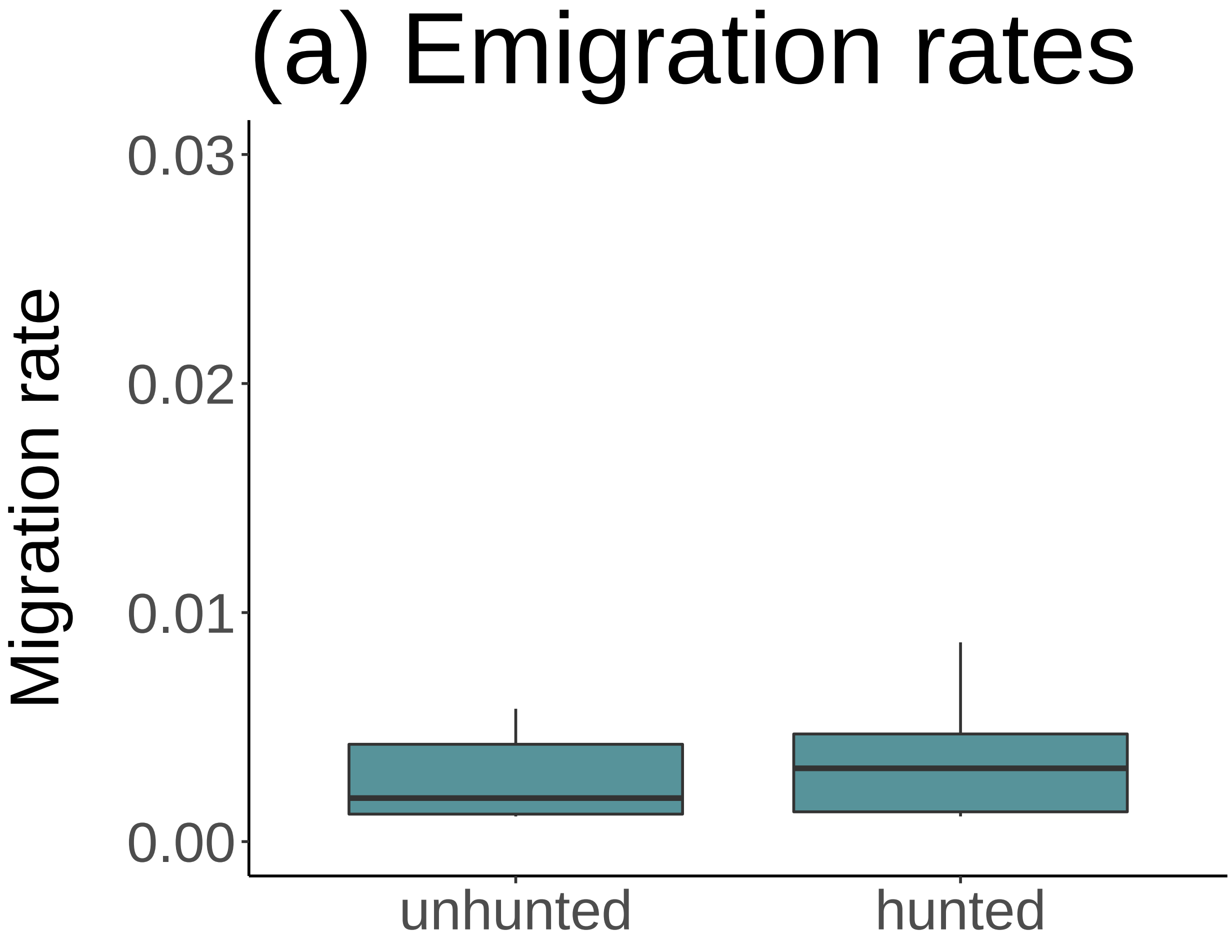
```
### Females - spatial

spatial %>% filter(Who == "Females") %>% ggplot(aes(x = What)) +
  geom_line(aes(y = r), col = "#8989D0", size = 1.5) + theme_classic() +
  geom_hline(yintercept = 0, col = "black")+
  geom_ribbon(aes(ymax = U, ymin = L), fill = "#8989D0", alpha = 0.5)+
  geom_errorbar(aes(y = r, ymin = r-Le, ymax = r+Ue), width=1, size=1.5,
               color="black", stat = "identity")+
  ylim(-0.03, 0.035) +
  xlab("Distance class") + ylab("Autocorrelation coefficient r")+
  scale_x_continuous(breaks = c(seq(0, 60, by = 10)), limits=c(4,61))+
  theme(text = element_text(size = 14),
        plot.title = element_text(size = 38),
        axis.text.x = element_text(size = 26, margin = margin(b = 10)),
        axis.text.y = element_text(size = 26, margin = margin(l = 10)),
        axis.title.x = element_text(size = 30),
        axis.title.y = element_text(size = 30))
```



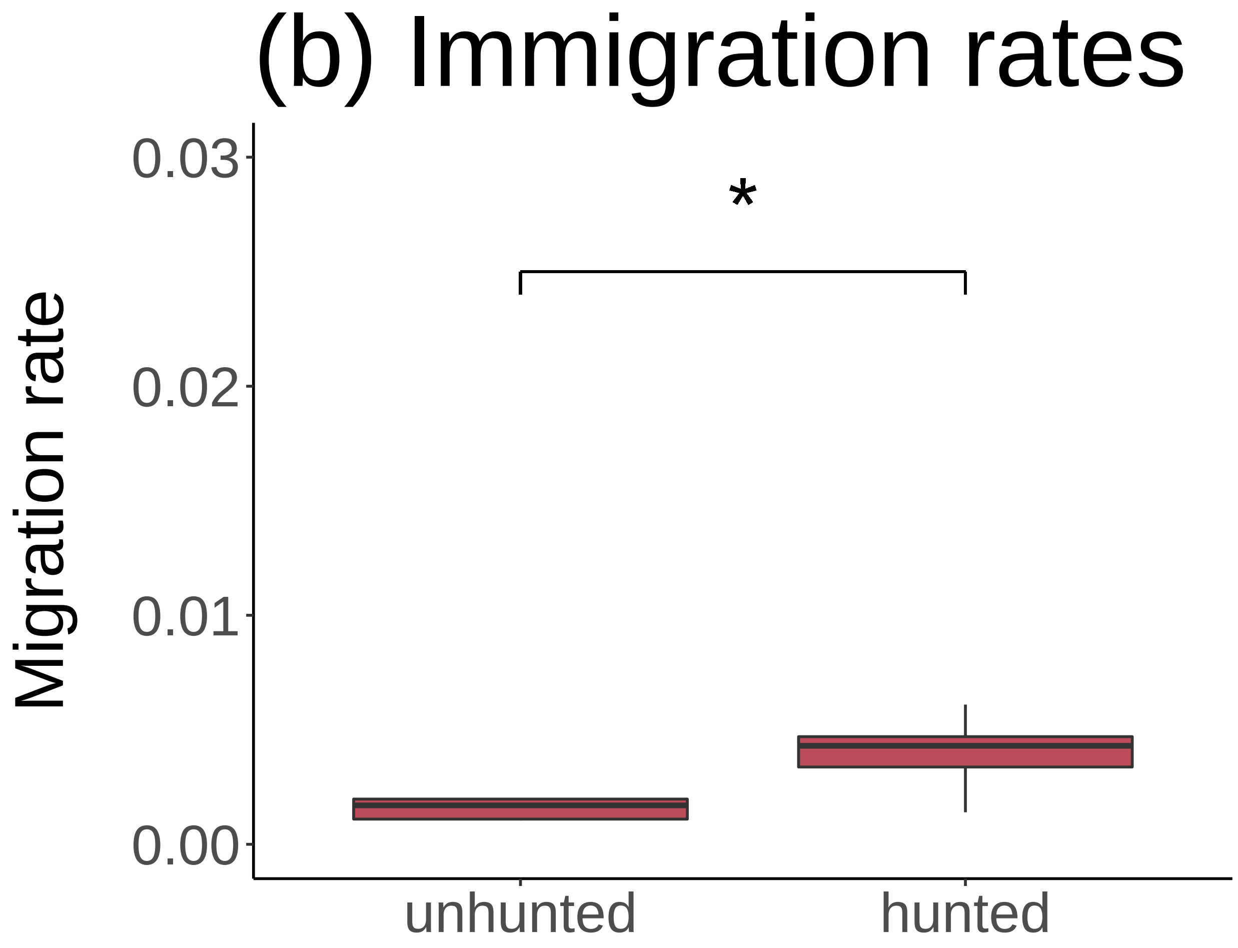
```
### Chicks - spatial
spatial %>% filter(Who == "Unrelated_chicks") %>% ggplot(aes(x = What)) +
  geom_line(aes(y = r), col = "#8989D0", size = 1.5) + theme_classic() +
  geom_hline(yintercept = 0, col = "black")+
  geom_ribbon(aes(ymax = U, ymin = L), fill = "#8989D0", alpha = 0.5)+
  geom_errorbar(aes(y = r, ymin = r-Le, ymax = r+Ue), width=1, size=1.5,
                color="black", stat = "identity")+
  ylim(-0.03, 0.035) +
  xlab("Distance class") + ylab("Autocorrelation coefficient r")+
  scale_x_continuous(breaks = c(seq(0, 60, by = 10)), limits=c(4,61))+
  theme(text = element_text(size = 14),
        plot.title = element_text(size = 38),
        axis.text.x = element_text(size = 26, margin = margin(b = 10)),
        axis.text.y = element_text(size = 26, margin = margin(l = 10)),
        axis.title.x = element_text(size = 30),
        axis.title.y = element_text(size = 30))
```



```
### Figure 3: boxplots migration rates ####
run5_clean <- read.csv("analyses/migrationanalysis/run5_clean.csv")

#change levels
run5_clean$hunt_in <- relevel(as.factor(run5_clean$hunt_in), ref = "unhunted")
run5_clean$hunt_out <- relevel(as.factor(run5_clean$hunt_out), ref = "unhunted")

#first, exclude the 'non-migration rates' which are those where pop in = pop out
run5_clean <- subset(run5_clean, m_in != m_out)

# plot emigration rates
ggplot(run5_clean, aes(x = hunt_out, y = migration_ESSc, fill = "#57939a")) +
  geom_boxplot(outlier.shape = NA, aes(middle = mean(migration_ESSc))) + ylim(0, 0.03)+
  labs(title = "(a) Emigration rates") +
  ylab("Migration rate")+
  theme(text = element_text(size = 26),
        legend.text = element_text(size = 28),
        legend.title = element_text(size = 28),
        legend.key.size = unit(1, 'cm'),
        plot.title = element_text(size = 38),
        axis.title.x = element_blank(),
        axis.title.y = element_text(margin = margin(t = 0, r = 20, b = 0, l = 0)),
        legend.position = "none")+
  scale_fill_manual(values=c("#57939a"))
```



```
#plot immigration rates

ggplot(run5_clean, aes(x = hunt_in, y = migration_ESSc, fill = "#be4d5a")) +
  geom_boxplot(outlier.shape = NA, aes(middle = mean(migration_ESSc))) + ylim(0, 0.03)+
  labs(title = "(b) Immigration rates") +
  ylab("Migration rate")+
  theme(text = element_text(size = 26),
        legend.text = element_text(size = 28),
        legend.title = element_text(size = 28),
        legend.key.size = unit(1, 'cm'),
        plot.title = element_text(size = 38),
        axis.title.x = element_blank(),
        axis.title.y = element_text(margin = margin(t = 0, r = 20, b = 0, l = 0)),
        legend.position = "none")+
  scale_fill_manual(values = c("#be4d5a"))+geom_text(x = 1.5, y = 0.028, label = "*", size = 10) + geom_
  geom_segment(x=1, xend = 1, y = 0.025, yend = 0.024)+geom_segment(x=2, xend = 2, y = 0.025, yend = 0.(
```



See below session information including package versions.

```
session_info()
```

```
## - Session info ---------------------------------------------------------------
##  setting  value
##  version  R version 4.0.1 (2020-06-06)
##  os       Windows 10 x64
##  system   x86_64, mingw32
##  ui       RTerm
##  language (EN)
##  collate  English_United Kingdom.1252
##  ctype    English_United Kingdom.1252
##  tz       Europe/Berlin
##  date     2022-11-02
##
## - Packages -------------------------------------------------------------------
##  ! package           * version    date       lib
##    ade4              * 1.7-18     2021-09-16 [1]
##    adegenet          * 2.1.5      2021-10-09 [1]
##    ape               * 5.5        2021-04-25 [1]
##    assertthat          0.2.1      2019-03-21 [1]
##    backports           1.2.1      2020-12-09 [1]
##    boot                1.3-28     2021-05-03 [1]
##    broom               0.7.9      2021-07-27 [1]
##    cachem              1.0.6      2021-08-19 [1]
##    callr               3.7.0      2021-04-20 [1]
##    cellranger          1.1.0      2016-07-27 [1]
##    cli                 3.0.1      2021-07-17 [1]
##    cluster             2.1.2      2021-04-17 [1]
##    coda                0.19-4     2020-09-30 [1]
##    codetools           0.2-18     2020-11-04 [1]
##    colorspace          2.0-2      2021-06-24 [1]
##    combinat            0.0-8      2012-10-29 [1]
##    crayon              1.4.2      2021-10-29 [1]
##    data.table        * 1.14.0     2021-02-21 [1]
##    DBI                 1.1.1      2021-01-15 [1]
##    dbplyr              2.1.1      2021-04-06 [1]
##    desc                1.4.0      2021-09-28 [1]
##    devtools          * 2.4.2      2021-06-07 [1]
##    DHARMa            * 0.4.5      2022-01-16 [1]
##    digest              0.6.27     2020-10-24 [1]
##    doParallel          1.0.17     2022-02-07 [1]
##    dplyr             * 1.0.7      2021-06-18 [1]
##    ellipsis            0.3.2      2021-04-29 [1]
##    emmeans             1.7.3      2022-03-27 [1]
##    estimability        1.3        2018-02-11 [1]
##    evaluate            0.17       2022-10-07 [1]
##    extrafont         * 0.17       2014-12-08 [1]
##    extrafontdb         1.0        2012-06-11 [1]
##    fansi               0.5.0      2021-05-25 [1]
##    farver              2.1.0      2021-02-28 [1]
##    fastmap             1.1.0      2021-01-25 [1]
##    forcats           * 0.5.1      2021-01-27 [1]
##    foreach             1.5.1      2020-10-15 [1]
##    formatR             1.12       2022-03-31 [1]
##    fs                  1.5.0      2020-07-31 [1]
##    gap                 1.2.3-1    2021-04-21 [1]
##    generics            0.1.1      2021-10-25 [1]
##    ggplot2           * 3.3.6      2022-05-03 [1]
##    glmmTMB           * 1.1.2.3    2021-09-20 [1]
##    glue                1.4.2      2020-08-27 [1]
##    gridExtra         * 2.3        2017-09-09 [1]
##    gtable              0.3.0      2019-03-25 [1]
##    haven               2.4.3      2021-08-04 [1]
##    hierfstat         * 0.5-7      2020-07-20 [1]
##    highr               0.9        2021-04-16 [1]
##    hms                 1.1.1      2021-09-26 [1]
##    htmltools           0.5.2      2021-08-25 [1]
##    httpuv              1.6.3      2021-09-09 [1]
##    httr                1.4.2      2020-07-20 [1]
##    igraph              1.2.6      2020-10-06 [1]
##    inbreedR          * 0.3.2      2016-09-09 [1]
##    insight             0.17.0     2022-03-29 [1]
##    iterators           1.0.13     2020-10-15 [1]
##    jsonlite            1.7.2      2020-12-09 [1]
##    knitr               1.40       2022-08-24 [1]
##    label.switching     1.8        2019-07-01 [1]
##    labeling            0.4.2      2020-10-20 [1]
##    later               1.3.0      2021-08-18 [1]
##    lattice             0.20-41    2020-04-02 [2]
##    lifecycle           1.0.1      2021-09-24 [1]
##    lme4              * 1.1-27.1   2021-06-22 [1]
##    lmerTest          * 3.1-3      2020-10-23 [1]
##    lpSolve             5.6.15     2020-01-24 [1]
##    lubridate           1.8.0      2021-10-07 [1]
##    magrittr            2.0.1      2020-11-17 [1]
##    MASS                7.3-54     2021-05-03 [1]
##    Matrix            * 1.3-4      2021-06-01 [1]
##    memoise             2.0.0      2021-01-26 [1]
##    mgcv                1.8-37     2021-09-23 [1]
##    mime                0.12       2021-09-28 [1]
##    minqa               1.2.4      2014-10-09 [1]
##    modelr              0.1.8      2020-05-19 [1]
##    multcomp            1.4-17     2021-04-29 [1]
##    MuMIn             * 1.46.0     2022-02-24 [1]
##    munsell             0.5.0      2018-06-12 [1]
##    mvtnorm             1.1-3      2021-10-08 [1]
##    nlme                3.1-153    2021-09-07 [1]
##    nloptr              1.2.2.2    2020-07-02 [1]
##    numDeriv            2016.8-1.1 2019-06-06 [1]
##    ParallelStructure * 1.0        2018-05-11 [1]
##    pegas             * 1.0-1      2021-05-17 [1]
##    performance       * 0.8.0      2021-10-01 [1]
##    permute             0.9-5      2019-03-12 [1]
##    pillar              1.6.4      2021-10-18 [1]
##    pkgbuild            1.2.0      2020-12-15 [1]
##    pkgconfig           2.0.3      2019-09-22 [1]
##    pkgload             1.2.2      2021-09-11 [1]
##    plot.matrix       * 1.6        2021-04-26 [1]
##    plyr                1.8.6      2020-03-03 [1]
##    pophelper         * 2.3.1      2021-10-13 [1]
##    prettyunits         1.1.1      2020-01-24 [1]
##    processx            3.5.2      2021-04-30 [1]
##    promises            1.2.0.1    2021-02-11 [1]
##    ps                  1.6.0      2021-02-28 [1]
##    purrr             * 0.3.4      2020-04-17 [1]
##    qgam                1.3.4      2021-11-22 [1]
##    R6                  2.5.1      2021-08-19 [1]
##    RColorBrewer      * 1.1-2      2014-12-07 [1]
##    Rcpp                1.0.7      2021-07-07 [1]
##    readr             * 2.1.2      2022-01-30 [1]
##    readxl            * 1.3.1      2019-03-13 [1]
##    remotes             2.4.1      2021-09-29 [1]
##    reprex              2.0.1      2021-08-05 [1]
##    reshape2            1.4.4      2020-04-09 [1]
##    rlang               0.4.11     2021-04-30 [1]
##    rmarkdown           2.17       2022-10-07 [1]
##    rprojroot           2.0.2      2020-11-15 [1]
##    rstudioapi          0.13       2020-11-12 [1]
##    Rttf2pt1            1.3.9      2021-07-22 [1]
##    rvest               1.0.1      2021-07-26 [1]
##    sandwich            3.0-1      2021-05-18 [1]
##    scales              1.1.1      2020-05-11 [1]
##    seqinr              4.2-8      2021-06-09 [1]
##    sessioninfo         1.1.1      2018-11-05 [1]
##    shiny               1.7.1      2021-10-02 [1]
##    stringi             1.7.4      2021-08-25 [1]
##    stringr           * 1.4.0      2019-02-10 [1]
##    survival            3.1-12     2020-04-10 [2]
##    testthat            3.1.0      2021-10-04 [1]
##    TH.data             1.1-0      2021-09-27 [1]
##    tibble            * 3.1.4      2021-08-25 [1]
##    tidyr             * 1.1.4      2021-09-27 [1]
##    tidyselect          1.1.1      2021-04-30 [1]
##    tidyverse         * 1.3.1      2021-04-15 [1]
##  D TMB                 1.7.22     2021-09-28 [1]
##    tzdb                0.1.2      2021-07-20 [1]
##    usethis           * 2.0.1      2021-02-10 [1]
##    utf8                1.2.2      2021-07-24 [1]
##    vctrs               0.3.8      2021-04-29 [1]
##    vegan               2.5-7      2020-11-28 [1]
##    withr               2.4.2      2021-04-18 [1]
##    xfun                0.33       2022-09-12 [1]
##    xml2                1.3.2      2020-04-23 [1]
##    xtable              1.8-4      2019-04-21 [1]
##    yaml                2.2.1      2020-02-01 [1]
##    zoo                 1.8-9      2021-03-09 [1]
##  source
##  CRAN (R 4.0.5)
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##  Github (royfrancis/pophelper@3807852)
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##
## [1] C:/Users/rchen2/Documents/R/win-library/4.0
## [2] C:/Program Files/R/R-4.0.1/library
##
##  D -- DLL MD5 mismatch, broken installation.
```