Contents

Principal Component Analysis

> When using PCA for data reduction rather than inferential analysis, assumption of normality is not required (Tabachnick & Fidell 2001)

We will use principal component analysis to reduce the redundancy among the behavioural measurements scored from the open field (OF) and mirror image stimulation (MIS) trials and to identify the dominant axes of behavioural variation in the OF and MIS trials. Principal components are calculated separately for the OF and MIS behavioural measurements using a correlation matrix. The behavioural dataset used in this analysis is exactly the same as used in Taylor et al. (2012) so the principal component loadings and scores are also the same.

To evaluate the appropriateness of this analyses we will follow Budaev's advice (2010. Using Principal Components and Factor Analysis in Animal Behaviour Research: Caveats and Guidelines. Ethology 116: 472–480.). Budaev suggests some best practices for reporting PCA results that we will follow.

```
library(MASS) # MASS clashes with dplyr... so always load first
library(pander) # pander clashes with dplyr... so always load first
```

```
##
## Attaching package: 'pander'
##
## The following object is masked from 'package:knitr':
##
##     pandoc
```

```
library(foreach)
```

```
## foreach: simple, scalable parallel programming from Revolution Analytics
## Use Revolution R for scalability, fault tolerance and more.
## http://www.revolutionanalytics.com
```

```
library(doMC)
```

```
## Loading required package: iterators
## Loading required package: parallel
```

```
registerDoMC()
library(tidyr)
library(dplyr)
```

```
##
## Attaching package: 'dplyr'
##
## The following object is masked from 'package:MASS':
##
##     select
##
## The following objects are masked from 'package:stats':
##
##     filter, lag
##
## The following objects are masked from 'package:base':
##
##     intersect, setdiff, setequal, union
```

```
set.alignment('right', row.names = 'left')
library(mvnormtest)
library(psych)
library(ggplot2)
```

```
##
## Attaching package: 'ggplot2'
##
## The following object is masked from 'package:psych':
##
##     %+%
```

```
library(MCMCglmm)
```

```
## Loading required package: Matrix
## Loading required package: coda
## Loading required package: lattice
## Loading required package: ape
```

```
behav_data <- tbl_df(read.table(file = "data/behaviour.csv",
                                sep = ',',
                                header = TRUE,
                                stringsAsFactors = FALSE))
behav_data
```

```
## Source: local data frame [4,286 x 25]
##
##     ID Sex Grid Year julian         trial_id Obs docil handlevent_year
## 1    4   F   SU 2005    177               NA MRG    28              12
## 2  601   M   AG 2005    165               NA CLS    15               9
## 3  601   M   AG 2005    182 0.44270.2005.182 ADI    17              10
## 4  601   M   AG 2005    224 0.44270.2005.224 ADI     8              12
## 5    5   F   KL 2005    170 0.46255.2005.170 ADI    10              10
## 6    5   F   KL 2005    184               NA MAW    20              11
## 7    5   F   KL 2005    212 0.46255.2005.212 ADI    12              14
## 8    5   F   KL 2005    219               NA ADI    10              15
## 9  603   M   AG 2005    170               NA CLS    17               7
## 10 603   M   AG 2005    173 0.46342.2005.173 ADI    12               8
## .. ... ...  ...  ...    ...               ... ...   ...             ...
## Variables not shown: Study (chr), front (dbl), attack_rate (dbl), back
##   (dbl), ln_attack_latency (dbl), ln_approach_latency (dbl), hole_rate
##   (dbl), jump_rate (dbl), chew (dbl), still (dbl), hang (dbl), groom
##   (dbl), walk (dbl), fecal (dbl), trial_life (int), trial_year (int)
```

Mirror Image Stimulation PCA

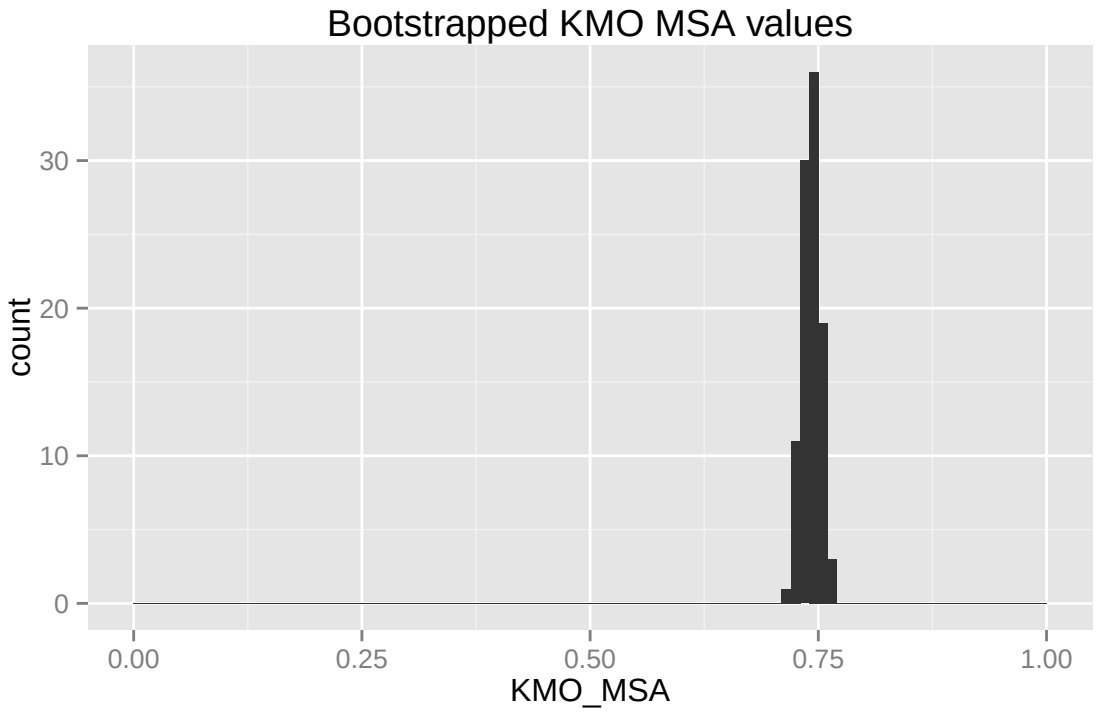
Budaev suggests using the Bartlett's test and the Kaiser–Meyer–Olkin (KMO) measure to assess sampling adequecy. Because the behaviour data contains multiple measures per individual we will first subsample the data, randomly choosing 1 record per individual. Bootstrap 100 times.

```
mis_data <-  behav_data %>%
  select(ID, trial_id, front, attack_rate, back, ln_attack_latency,
    ln_approach_latency)%>%
  filter(!is.na(front))

# Get only complete records of the MIS behaviours
mis_sub_data <- foreach(i = 1:100, .combine = 'rbind') %dopar% {
  mis_data %>%
  group_by(ID) %>%
  do(sample_n(., 1)) %>%
  mutate(itt = i)
}
save(mis_data, mis_sub_data, file = "data/analyses_data/mis_sub.Rdata")
```

Bartlett's test & KMO

```
load("data/analyses_data/mis_sub.Rdata")
n_trials <- mis_data %>% ungroup() %>% summarise(n = n())
mis_KMO_Bart <- mis_sub_data %>%
    group_by(itt) %>%
    select(-ID, -trial_id) %>%
    summarise(
      KMO_MSA = KMO(cbind(front, attack_rate, back,
        ln_attack_latency, ln_approach_latency))$MSA,
      cortest.bartlett = cortest.bartlett(R = cor(cbind(front,
```

```
        attack_rate, back, ln_attack_latency,
        ln_approach_latency)), n = n_trials$n)$chisq
      )

p <- ggplot(mis_KMO_Bart, aes(x = KMO_MSA))
p <- p + geom_histogram(binwidth = 0.01)
p + ggtitle("Bootstrapped KMO MSA values") + xlim(c(0,1))

## Warning: position_stack requires constant width: output may be incorrect
```

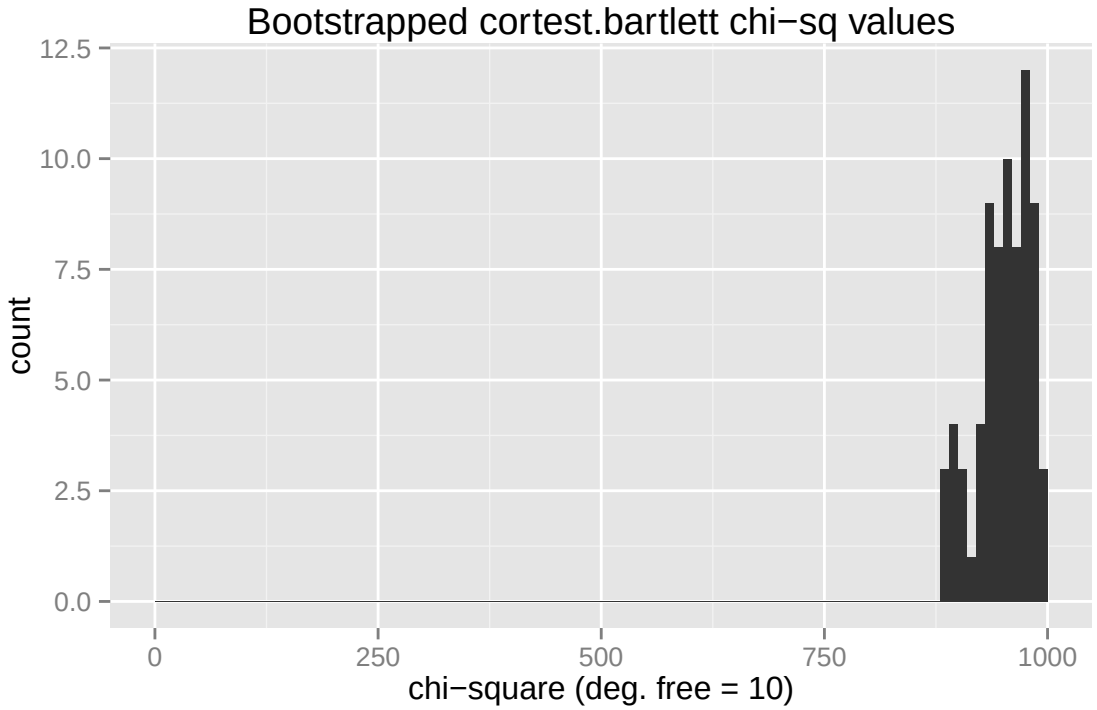



```
p <- ggplot(mis_KMO_Bart, aes(x = cortest.bartlett))
p <- p + geom_histogram(binwidth = 10)
p <- p + ggtitle("Bootstrapped cortest.bartlett chi-sq values")
p + xlim(c(0,1000)) + xlab("chi-square (deg. free = 10)")
```

The overall measure of sampling adequecy is fine (Measure of Sampling Adequacy = 0.7415). Bartletts test unsurprisingly rejects the hypotheses that all correlations are zero (P = 0).

Multivariate normality

```
load("data/analyses_data/mis_sub.Rdata")
mis_one <- mis_sub_data %>% filter(itt == 1)
mshapiro.test(t(as.matrix(mis_one[-c(1,2,8)])))
```




```
##
##  Shapiro-Wilk normality test
##
## data:  Z
## W = 0.5897, p-value < 2.2e-16
```

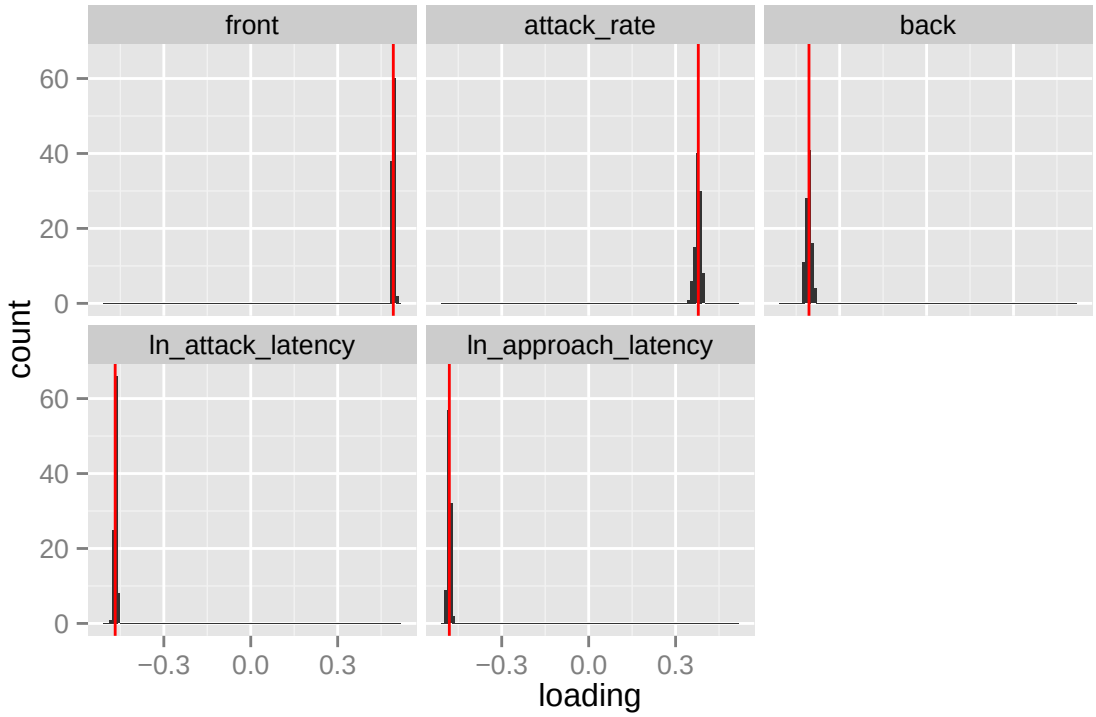
The data are not multi-normal. In our case this isn't a major problem because we are not performing any statistical tests alongside the PCA, we are just using the PCA to reduce the dimensionality of the data. We will note that the data are not multivariate normal.

Pseudo-replication

Because our data contains multiple records per individual we will check to be sure pesudo-replication isn't having a large effect on the PCA loadings.

```
load("data/analyses_data/mis_sub.Rdata")
# Calculate PCA for subsampled data
mis_pca <- foreach(i = 1:100, .combine = rbind) %do% {
  foo <- mis_sub_data %>% filter(itt == i)
  pc_loadings <- prcomp(foo[-c(1,2,8)], scale = TRUE)$rotation[ ,"PC1"]
  if(sign(pc_loadings["front"]) == -1) {pc_loadings <- pc_loadings * -1}
  pc_loadings
}

mis_pca <- gather(data.frame(mis_pca), front, attack_rate, back, ln_attack_latency, ln_approach_late
```

```r
# Calculate PCA for full dataset
mis_pca_full <- prcomp(mis_data[-c(1,2, 8)], scale = TRUE)

mis_pca_loadings <- data.frame(trait = dimnames(mis_pca_full$rotation)[[1]],
                         loading = mis_pca_full$rotation[ ,"PC1"])

if(sign(mis_pca_loadings$loading[mis_pca_loadings$trait == "front"]) == -1) {
  mis_pca_loadings$loading <- mis_pca_loadings$loading * -1
}

p <- ggplot(mis_pca, aes(x = loading))
p <- p + geom_histogram(binwidth = 0.01) + facet_wrap( ~ trait)
p + geom_vline(data = mis_pca_loadings,
       aes(xintercept = loading), color = 'red')
```



The loadings for PC 1 are nearly identical (subsampled to 1 trial per indidivual vs. full dataset). So we will continue with the loadings from the full dataset so that they are consistent with Taylor et al. 2012.

```r
#  Reverse sign if 'front' is negative. This way higher scores will always be
#  more aggressive. Sign of pc scores is arbitrary. Make sure to reverse scores
#  and loadings or else confusion!
if(mis_pca_full$rotation["front", "PC1"] < 0){
    mis_pca_full$rotation <- -1 * (mis_pca_full$rotation)
    mis_scores <- data.frame(-mis_pca_full$x)
    } else {
    mis_scores <- data.frame(mis_pca_full$x)
```

```
}

# reattach scores to trail.id (which was rowname after PCA)
mis_pca_scores <- data.frame(trial_id = mis_data$trial_id,
                   misPC1 = mis_scores$PC1,
                   misPC2 = mis_scores$PC2,
                   stringsAsFactors = FALSE)

mis_pca_summary <- rbind(mis_pca_full$rotation,
  StdDev = mis_pca_full$sdev,
  PropVar = mis_pca_full$sdev^2 / sum(mis_pca_full$sdev^2))
```

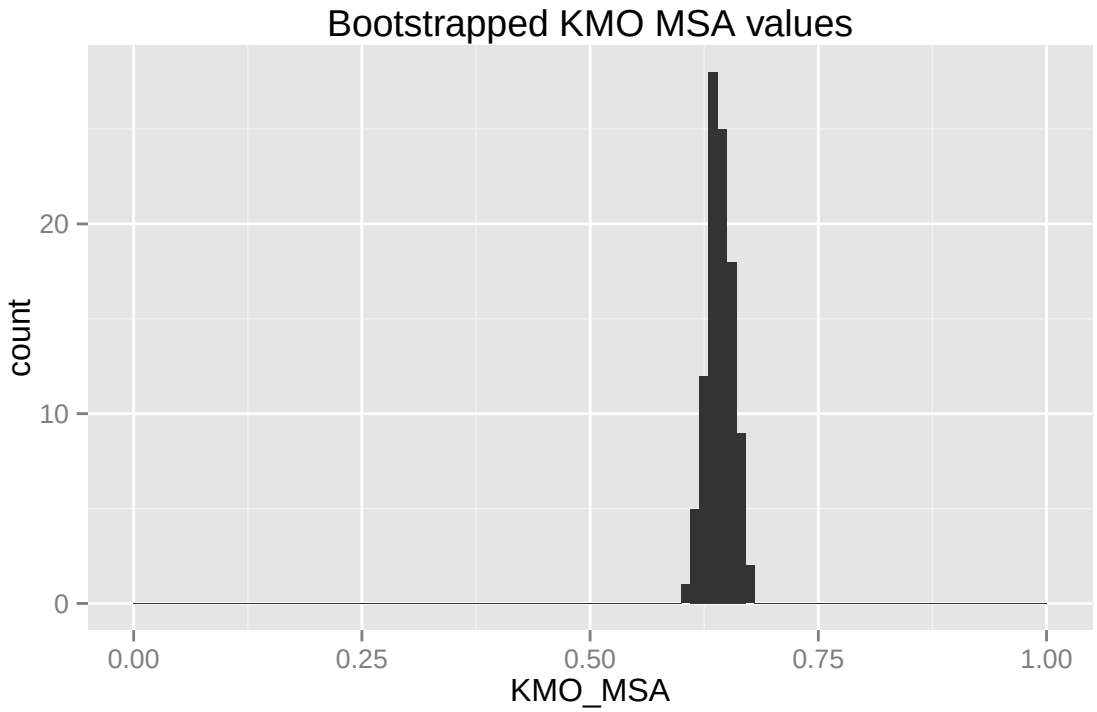
Open Field Arena PCA

Following same procedure as above, now for the open field behavioural measures.

```
# Get only complete records of the MIS behaviours
of_data <- behav_data %>%
             select(ID, trial_id, hole_rate,jump_rate, chew, still, hang,
              groom, walk, fecal) %>%
             filter(!is.na(hole_rate))

of_sub_data <- foreach(i = 1:100, .combine = 'rbind') %dopar% {
  of_data %>%
  group_by(ID) %>%
  do(sample_n(., 1)) %>%
  mutate(itt = i)
}
save(of_data, of_sub_data, file = "data/analyses_data/of_sub_data.RData")
```

Bartlett's test & KMO

```
load("data/analyses_data/of_sub_data.RData")
n_trials <- of_data %>% summarise(n = n())
of_KMO_Bart <- of_sub_data %>%
                group_by(itt) %>%
                select(-ID, -trial_id) %>%
                summarise(
                  KMO_MSA = KMO(cbind(hole_rate, jump_rate, chew, still, hang,
                    groom, walk, fecal))$MSA,
                  cortest.bartlett = cortest.bartlett(R = cor(cbind(hole_rate,
                    jump_rate, chew, still, hang, groom, walk, fecal)),
                    n = n_trials$n)$chisq
                  )

p <- ggplot(of_KMO_Bart, aes(x = KMO_MSA)) + geom_histogram(binwidth = 0.01)
p + ggtitle("Bootstrapped KMO MSA values") + xlim(c(0,1))

## Warning: position_stack requires constant width: output may be incorrect
```



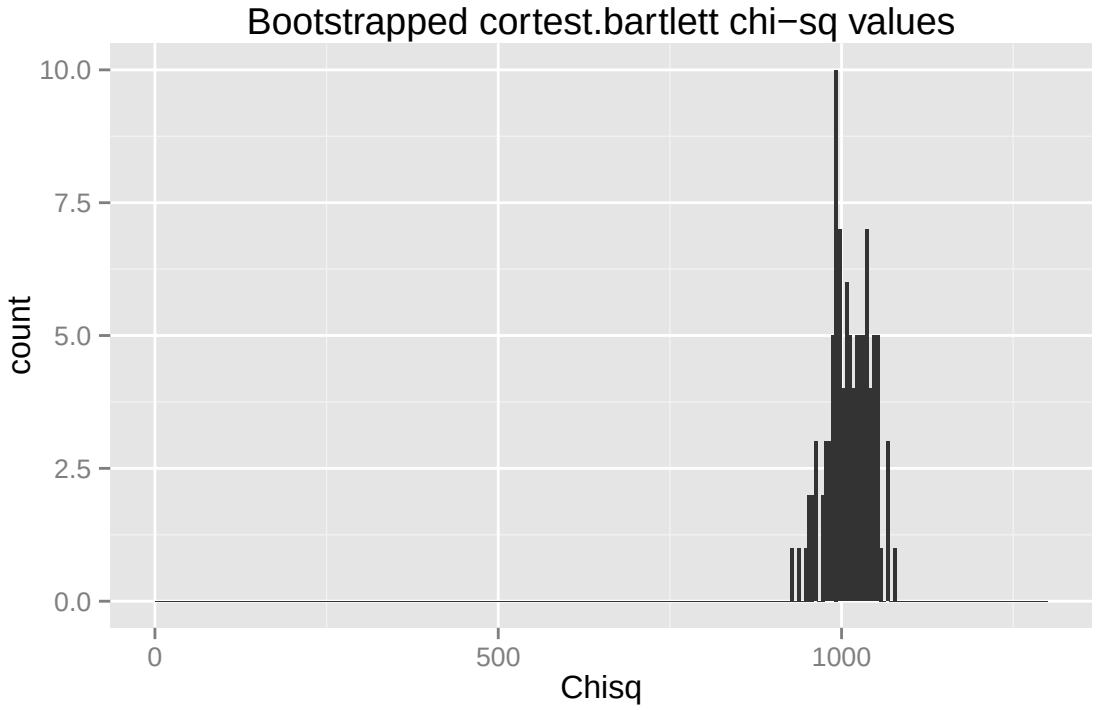

```
p <- ggplot(of_KMO_Bart, aes(x = cortest.bartlett))
p <- p + geom_histogram(binwidth = 5)
p <- p + ggtitle("Bootstrapped cortest.bartlett chi-sq values")
p + xlim(c(0,1300)) + xlab("Chisq")
```

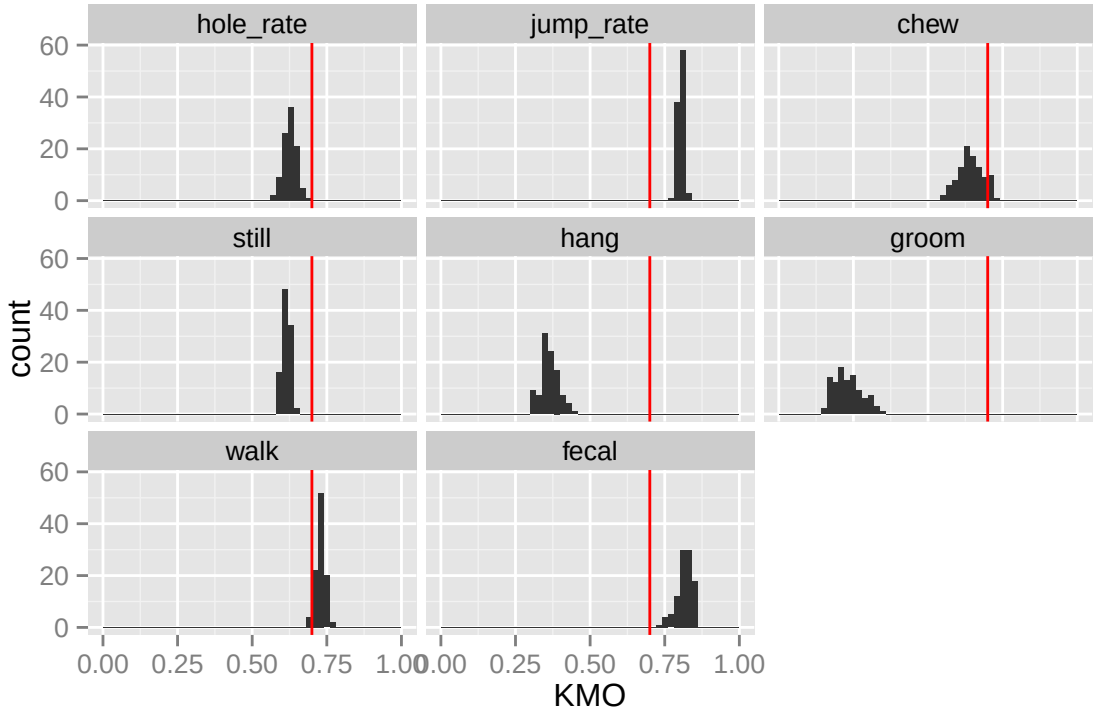
The overall measure of sampling adequecy is a bit low, maybe..., (MSA = 0.6419). Again, Bartlett's test rejects the hypotheses that all correlations are zero (P = 0).

Lets take a closer look at the KMO test.

```
load("data/analyses_data/of_sub_data.RData")
kmo_of <- foreach(i = 1:100, .combine = 'rbind') %do% {
  foo <- of_sub_data %>% ungroup() %>% filter(itt == i)
  KMO(foo[ ,c(-1,-2,-11)])$MSAi
}
kmo_of <- tbl_df(data.frame(kmo_of))

p <- ggplot(gather(kmo_of, hole_rate, jump_rate, chew, still, hang, groom, walk, fecal, key = "behav
p <- p + facet_wrap( ~ behav) + geom_histogram(binwidth = 0.02) + xlim(c(0,1))
p + geom_vline(xintercept = 0.7, color = 'red')
```

Looks like the low overall KMO index is driven by grooming and hanging. Both of which don't factor in very highly in the PCA loadings. I think therefore this is OK.


Bootstrapped cortest.bartlett chi−sq values
count
10.0
7.5
5.0
2.5
0.0
0
500
1000
Chisq


hole_rate
jump_rate
chew
still
hang
groom
walk
fecal
count
60
40
20
0
0.00
0.25
0.50
0.75
1.00
KMO

Multivariate normality

```
load("data/analyses_data/of_sub_data.RData")
of_one <- of_sub_data %>% filter(itt == 1)
mshapiro.test(t(as.matrix(of_one[-c(1,2,11)])))
```

```
##
##  Shapiro-Wilk normality test
##
## data:  Z
## W = 0.7185, p-value < 2.2e-16
```

The open field data are also not multi-normal (P = 2.4153 × 10-24).

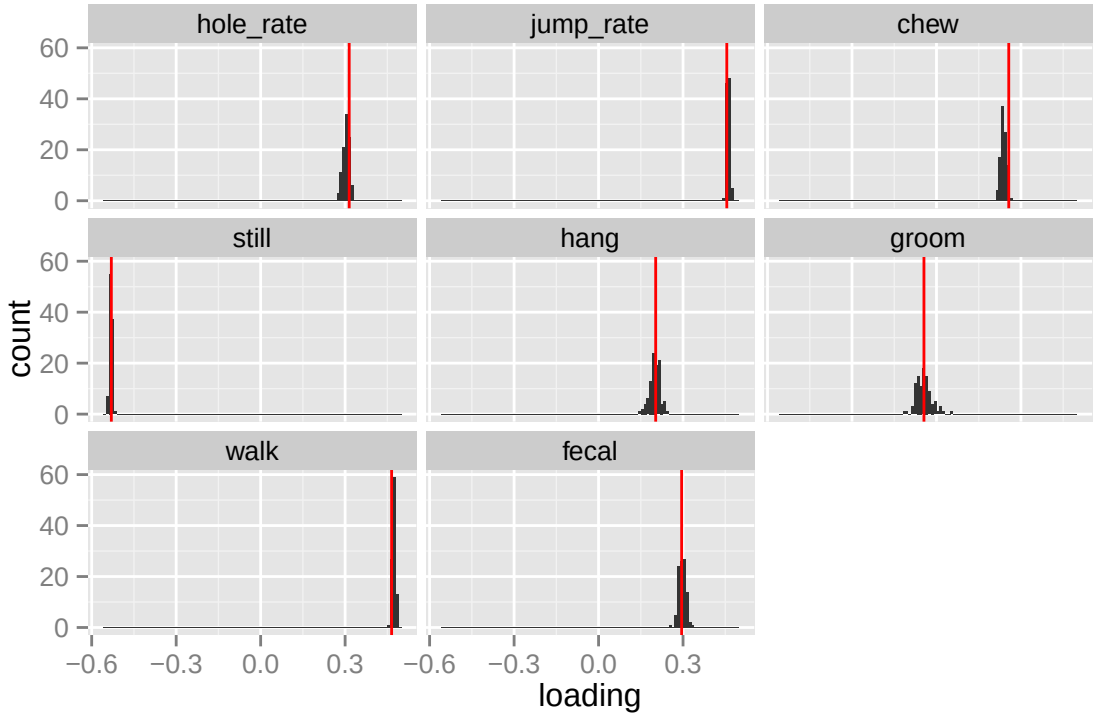
Pseudo-replication

Because our data contains multiple records per individual we will check to be sure pesudo-replication isn't having a large effect on the PCA loadings.

```
load("data/analyses_data/of_sub_data.RData")
# Calculate PCA for subsampled data
of_pca <- foreach(i = 1:100, .combine = rbind) %do% {
  foo <- of_sub_data %>% filter(itt == i)
  pc_loadings <- prcomp(foo[-c(1,2,11)], scale = TRUE)$rotation[ ,"PC1"]
  if(sign(pc_loadings["still"]) == 1) {pc_loadings <- pc_loadings * -1}
}

of_pca <- gather(data.frame(of_pca), hole_rate, jump_rate, chew, still, hang, groom, walk, fecal, ke

# Calculate PCA for full dataset
of_pca_full <- prcomp(of_data[-c(1,2,11)], scale = TRUE)

of_pca_loadings <- data.frame(trait = dimnames(of_pca_full$rotation)[[1]],
                        loading = of_pca_full$rotation[ ,"PC1"])

if(sign(of_pca_loadings$loading[of_pca_loadings$trait == "still"]) == 1) {
  of_pca_loadings$loading <- of_pca_loadings$loading * -1
}

p <- ggplot(of_pca, aes(x = loading)) + geom_histogram(binwidth = 0.01)
p <- p + facet_wrap( ~ trait)
p + geom_vline(data = of_pca_loadings, aes(xintercept = loading), color = 'red')
```

Once again the loadings for PC 1 are spot on the modes of the bootstrap distribution. So we will continue with the loadings from the full dataset so that they are consistent with Taylor et al. 2012. Interestingly, hang and groom have the largest bootstrap variance, and these were the triats identified by the KMO test as not being sampled adequately.

```
load("data/analyses_data/of_sub_data.RData")
of_pca_full <- prcomp(of_data[-c(1,2,11)], scale = TRUE)
```




```
#  If the pc coefficient for still is positive, then reverse sign of PC scores
#  so that high PC1 is more active.
if(of_pca_full$rotation["still", "PC1"] > 0){
    of_pca_full$rotation <- -1 * (of_pca_full$rotation)
    of_scores <- data.frame(-of_pca_full$x)
    } else {
    of_scores <- data.frame(of_pca_full$x)
}

#  Get scores
of_pca_scores <- data.frame(trial_id = of_data$trial_id, ofPC1 = of_scores$PC1,
  ofPC2 = of_scores$PC2, ofPC3 = of_scores$PC3, stringsAsFactors = FALSE)

of_pca_summary <- rbind(of_pca_full$rotation, StdDev = of_pca_full$sdev,
  PropVar = of_pca_full$sdev^2 / sum(of_pca_full$sdev^2))

# Save score data
save(mis_pca_summary, of_pca_summary,
  file = "data/analyses_data/pca.RData")
```

Merge PCA data

```
load("data/analyses_data/pca.RData")

# merge pc scores with rest of data by trial id.
```

```
pca_data <- left_join(behav_data, mis_pca_scores, by = "trial_id")
pca_data <- left_join(pca_data, of_pca_scores, by = "trial_id")

save(pca_data, mis_pca_summary, of_pca_summary,
  file = "data/analyses_data/pca.RData")


of_table <- c(of_pca_summary["walk",1], of_pca_summary["jump_rate",1],
              of_pca_summary["hole_rate",1], of_pca_summary["fecal",1],
              of_pca_summary["hang",1], of_pca_summary["chew",1],
              of_pca_summary["groom",1], of_pca_summary["still",1],
              of_pca_summary["StdDev",1], of_pca_summary["PropVar",1] * 100)
of_table <- format(of_table, nsmall = 2, digits = 0)

mis_table <- c(mis_pca_summary["front",1], mis_pca_summary["attack_rate",1],
               mis_pca_summary["back",1],
               mis_pca_summary["ln_attack_latency",1],
               mis_pca_summary["ln_approach_latency",1],
               mis_pca_summary["StdDev",1],
               mis_pca_summary["PropVar",1] * 100)
mis_table <-format(mis_table, nsmall = 2, digits = 0)


pca_table <- data.frame(check.names = FALSE,
  "OF Behaviour" = c("Walk", "Jump Rate", "Hole Rate", "No. Pellets", "Hang",
  "Chew", "Groom", "Still", "", "Std. Dev.", "% Total variance", "N records",
  "N individuals"),
  "OF PC1" = c(of_table[1:8], "", of_table[9:10], nrow(behav_data %>%
                filter(!is.na(still))), nrow(of_data %>% select(ID) %>%
                unique())),
  "MIS Behaviour" = c("Front", "Attack rate", "Back", "Attack latency",
  "Approach latency", rep("", 8)),
  "MIS PC1" = c(mis_table[1:5], rep("", 4), mis_table[6:7],
  nrow(behav_data %>% filter(!is.na(front))), nrow(mis_data %>%
  select(ID) %>% unique()))
  )
pandoc.table(pca_table)
```

OF Behaviour	OF PC1	MIS Behaviour	MIS PC1
Walk	0.47	Front	0.49
Jump Rate	0.46	Attack rate	0.38
Hole Rate	0.31	Back	-0.41
No. Pellets	0.30	Attack latency	-0.47
Hang	0.20	Approach latency	-0.48
Chew	0.26		
Groom	-0.04		
Still	-0.53		
Std. Dev.	1.67		1.67

OF Behaviour	OF PC1	MIS Behaviour	MIS PC1
% Total variance	34.73		55.79
N records	556		553
N individuals	365		364

```
# sample sizes
n_docil <- pca_data %>% filter(!is.na(docil)) %>%
            group_by(ID) %>% summarise(n = n())

n_oft <- pca_data %>% filter(!is.na(misPC1)) %>%
           group_by(ID) %>% summarise(n = n())

s_table <- data.frame(check.names = FALSE,
     Test = c("OF / MIS", "Handling"),
    "N trials" = c(sum(n_oft$n), sum(n_docil$n)),
    "N individuals" = c(length(unique(n_oft$ID)), length(unique(n_docil$ID))),
    "N > 1 trial"   = c(length(n_oft$ID[n_oft$n > 1]),
                        length(n_docil$ID[n_docil$n > 1]))
    )
pandoc.table(s_table)
```

Test	N trials	N individuals	N > 1 trial
OF / MIS	553	364	165
Handling	4227	869	621