Supplemental code for Social Scaling of Stressor Impacts

Accompanying code for "Colony size buffers interactions between neonicotinoid exposure and cold stress in bumble bees"

#Queenright colony experiments

Load and clean data for queenright experiments

Load data

```
# Get colony demographic metadata
setwd("/Users/jamescrall/Dropbox/Work/_Research/Bees/Bumblebees/socialScalingOfStressors/Data/colonyInf
metadata <- read.csv("csvFiles/colonyMetaData.csv")</pre>
### Parse into separate treatments ### initialize
metadata$neonic <- 0
metadata$cold <- 0
metadata$neonic[str_detect(metadata$Treatment, "eonic")] <- 1</pre>
metadata$cold[str_detect(metadata$Treatment, "old")] <- 1</pre>
metadata$neonic <- as.numeric(metadata$neonic)</pre>
metadata$cold <- as.numeric(metadata$cold)</pre>
### Load behavioral tracking data
setwd("/Users/jamescrall/Dropbox/Work/_Research/Bees/Bumblebees/socialScalingOfStressors/Data/thermalRi
behData <- read.csv("behData_June2022.csv")</pre>
behData <- behData[complete.cases(behData[, c("porTimeMoving",</pre>
    "movingVelocity")]), ] ## Remove rows with missing data
# tracking.metadata <- read.csv('trackingMetadata.csv')</pre>
tag.data <- read.csv("/Users/jamescrall/Dropbox/Work/_Research/Bees/Bumblebees/socialScalingOfStressors
# Correct name for one misentified colony
tag.data$colony[tag.data$colony == "BB246"] <- "BG246"</pre>
### Add size at tagging to metadata by reading taglists
### replace nans with 'worker'
tag.data$caste[is.na(tag.data$caste)] <- "worker"</pre>
tag.data$caste[tag.data$caste == ""] <- "worker"</pre>
tag.data$caste[tag.data$caste == "Worker"] <- "worker"</pre>
tag.data$caste[tag.data$caste == "Queen"] <- "queen"</pre>
tag.data$caste[tag.data$caste == "Male"] <- "male"</pre>
# Add colony position to tag.data
for (i in 1:length(metadata[, 1])) {
    tag.data$thermalRigPosition[tag.data$colony == metadata$colonyID[i]] <- metadata$positionInThermalR
```

```
tag.data$unique_id <- paste(tag.data$block, tag.data$thermalRigPosition,</pre>
    tag.data$id, sep = " ")
for (i in 1:length(metadata[, 1])) {
    # Figure out how many bees in this colony
    metadata$colony.size.at.tagging[i] <- dim(subset(tag.data,</pre>
        colony == metadata$colonyID[i]))[1]
   metadata$new_queens[i] <- dim(subset(tag.data, colony ==</pre>
        metadata$colonyID[i] & caste == "queen" & founding_queen !=
        "Y"))[1]
   metadata$males[i] <- dim(subset(tag.data, colony == metadata$colonyID[i] &</pre>
        caste == "male"))[1]
}
metadata$reproductive <- as.factor((metadata$new_queens + metadata$males) >
reproductive.vs.size.model <- glm(reproductive ~ log10(colony.size.at.tagging),
   data = subset(metadata, colony.size.at.tagging > 0), family = "binomial")
summary(reproductive.vs.size.model)
##
## Call:
## glm(formula = reproductive ~ log10(colony.size.at.tagging), family = "binomial",
##
       data = subset(metadata, colony.size.at.tagging > 0))
##
## Deviance Residuals:
      Min
                     Median
                                           Max
                1Q
## -1.1884 -0.9461 -0.7742 1.3922
                                        1.6155
## Coefficients:
                                 Estimate Std. Error z value Pr(>|z|)
                                             0.7391 -1.950 0.0511 .
## (Intercept)
                                  -1.4415
## log10(colony.size.at.tagging)
                                   0.6478
                                              0.5418
                                                      1.196
                                                             0.2318
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## (Dispersion parameter for binomial family taken to be 1)
##
##
      Null deviance: 74.726 on 57 degrees of freedom
## Residual deviance: 73.239 on 56 degrees of freedom
## AIC: 77.239
##
## Number of Fisher Scoring iterations: 4
## Load thermal data
thermData <- read.csv("/Users/jamescrall/Dropbox/Work/_Research/Bees/Bumblebees/socialScalingOfStressor
thermData <- thermData[!is.na(thermData$block), ] #Remove some missing data
behData$colony.id <- paste(behData$block, behData$rigPosition,
    sep = " ")
behData$uniqueID <- paste(behData$colony.id, behData$id, sep = " ")
```

```
## Fill in metadata to individual behavioral and colony
## level thermal data frames
for (i in 1:length(metadata[, 1])) {
    # print(i)
    bd.ind <- behData$rigPosition == metadata$positionInThermalRig[i] &</pre>
        behData$block == metadata$experimentalBlock[i]
    if (sum(bd.ind) > 0) {
        behData$neonic[bd.ind] <- as.logical(metadata$neonic[i])</pre>
        behData$cold[bd.ind] <- as.logical(metadata$cold[i])</pre>
        behData$colony.size.at.tagging[bd.ind] <- metadata$colony.size.at.tagging[i]
        behData$species[bd.ind] <- metadata$Species[i]</pre>
        behData$experimental.day[bd.ind] <- floor(behData[bd.ind,</pre>
            ]$dateNum) - min(floor(behData[bd.ind, ]$dateNum)) +
        behData$time.from.start[bd.ind] <- behData[bd.ind, ]$dateNum -
            min(behData[bd.ind, ]$dateNum)
        worker.brood.ratio <- unique(behData$colony.size.at.tagging[bd.ind]/behData$numBrood[bd.ind])</pre>
        behData$worker.brood.ratio[bd.ind] <- worker.brood.ratio
        # Add reproductive state (binary)
        behData$reproductive[bd.ind] <- metadata$reproductive[i]</pre>
        # Identify first exposure
        rs <- behData$rampState[bd.ind]</pre>
        tfs <- behData$time.from.start[bd.ind]</pre>
        first.cold.time <- tfs[which(rs == 3)[1]]</pre>
        pre.exposure <- tfs < first.cold.time</pre>
        behData$pre.exposure[bd.ind] <- pre.exposure</pre>
        td.ind <- thermData$rigPosition == metadata$positionInThermalRig[i] &</pre>
            thermData$block == metadata$experimentalBlock[i]
        thermData$neonic[td.ind] <- as.logical(metadata$neonic[i])</pre>
        thermData$cold[td.ind] <- as.logical(metadata$cold[i])</pre>
        thermData$colony.size.at.tagging[td.ind] <- metadata$colony.size.at.tagging[i]
        thermData$species[td.ind] <- metadata$Species[i]</pre>
        thermData$experimental.day[td.ind] <- floor(thermData[td.ind,</pre>
            ]$dateNum) - min(floor(thermData[td.ind, ]$dateNum)) +
        thermData$worker.brood.ratio[td.ind] <- worker.brood.ratio</pre>
        # print(paste('no tracking data for colony ', i, ',
        # skipping', sep=''))
    }
}
# Remove missing values from thermData
thermData <- thermData[!is.na(thermData$block), ]</pre>
# Remove errant readings from thermData
thermData <- subset(thermData, (meanTempBrood_all - nestAirTemp) >
    -2)
# Add unique colony identifier to thermData
```

```
thermData$colony.id <- paste(thermData$block, thermData$rigPosition,</pre>
    sep = " ")
behData$tod.hours <- (behData$dateNum%%1) * 24  #Convert timestamp to daily hours
behData <- subset(behData, framesTracked > 10) #Select out low-quality tracking
# Variable conversion to factors
behData$id <- as.factor(behData$id)</pre>
behData$experimental.day <- as.factor(behData$experimental.day)
behData$neonic <- as.factor(behData$neonic)</pre>
behData$cold <- as.factor(behData$cold)</pre>
behData$trial <- paste(behData$colony.id, behData$date, sep = "_")</pre>
# Add marker for surface nesting species
behData$surfaceNesting <- behData$species == "griseocolis"
for (i in 1:length(tag.data$unique_id)) {
    behData$caste[behData$uniqueID == tag.data$unique_id[i]] <- tag.data$caste[i]
    behData$callow[behData$uniqueID == tag.data$unique_id[i]] <- tag.data$callow[i]
    behData$founding.queen[behData$uniqueID == tag.data$unique_id[i]] <- tag.data$founding_queen[i]
}
# create discrete rampState variable
behData$rampState_ch <- as.character(behData$rampState)</pre>
rmps <- as.character(seq(0, 5))
rep_names <- c("floating", "a_before", "ramp_down", "cold", "ramp_up",
    "b after")
for (i in 1:length(rmps)) {
    behData$rampState_ch[behData$rampState_ch == rmps[i]] <- rep_names[i]
behData$rampState_ch <- as.factor(behData$rampState_ch)</pre>
```

Perform principal components analysis on within-nest behavioral metrics

```
# Run principal components analysis on nest behavior
pc.vars <- c("distanceToCenterInstantaneous", "porTimeMoving",</pre>
    "movingVelocity", "medianDistancetoClosestBrood", "medianDistanceToClosestWaxpot",
    "medianDistanceToAllBrood", "medianDistanceToAllWaxpots",
    "degreeCentrality", "meanInteractionRate", "spatCorAll",
    "broodInfoRichness", "waxpotInfoRichness", "waxpotOccupancyRate",
    "broodOccupancyRate")
behData <- behData[complete.cases(behData[, pc.vars]), ]</pre>
pca.data.sub <- behData[, pc.vars]</pre>
# Variable transformation
pca.data.sub$porTimeMoving <- log10(1.05 - pca.data.sub$porTimeMoving)</pre>
lg.vars <- c("distanceToCenterInstantaneous", "medianDistancetoClosestBrood",</pre>
    "medianDistanceToClosestWaxpot", "medianDistanceToAllBrood",
    "medianDistanceToAllWaxpots", "degreeCentrality", "meanInteractionRate",
    "spatCorAll", "broodInfoRichness", "waxpotInfoRichness")
pca.data.sub[, lg.vars] <- log10(0.5 + pca.data.sub[, lg.vars])</pre>
```

```
nest.beh.pca <- prcomp(pca.data.sub, scale = TRUE, center = TRUE)</pre>
behData$PC1 <- nest.beh.pca$x[, 1]</pre>
behData$PC1 <- -behData$PC1 #NB Switch sign of PC1 to reflect 'spatial centrality' in other findings
behData$PC2 <- nest.beh.pca$x[, 2]
behData$PC3 <- nest.beh.pca$x[, 3]
# Subtract baseline PC1 for each colony
colonies <- unique(behData$colony.id)</pre>
baseline.pcs <- data.frame(matrix(nrow = length(colonies), ncol = 3))</pre>
colnames(baseline.pcs) <- c("colony", "pc1", "pc2")</pre>
for (i in 1:length(colonies)) {
    c.id <- behData$colony.id == colonies[i]</pre>
    first.ramp.time <- min(subset(behData, colony.id == colonies[i] &
        rampState > 0)$time.from.start)
    baseline.data <- subset(behData, colony.id == colonies[i] &</pre>
        rampState == 0 & time.from.start < first.ramp.time)</pre>
    if (length(baseline.data[, 1]) == 0) {
        print("no baseline data")
        print(i)
        print(colonies[i])
    mean.pc1 <- mean(baseline.data$PC1)</pre>
    behData$PC1.res[c.id] <- behData$PC1[c.id] - mean.pc1</pre>
    mean.pc2 <- mean(baseline.data$PC2)</pre>
    behData$PC2.res[c.id] <- behData$PC2[c.id] - mean.pc2
    behData$first.ramp.time <- first.ramp.time</pre>
}
# pc loadings visulization prep
pc.loadings <- -nest.beh.pca$rotation[, 1:2]</pre>
pc.loadings <- pc.loadings[order(pc.loadings[, 1]), ]</pre>
pc.loadings <- as.data.frame(pc.loadings)</pre>
pc.loadings$variable <- rownames(pc.loadings)</pre>
setwd(output_folder)
pdf("FigS2a.pdf")
ggplot(data = pc.loadings, aes(x = reorder(variable, +PC1), y = PC1,
    fill = PC1)) + geom_bar(stat = "identity") + coord_flip() +
    scale_fill_distiller(palette = "RdYlBu") + theme_minimal()
dev.off()
## pdf
pdf("FigS2b.pdf")
ggplot(data = pc.loadings, aes(x = reorder(variable, +PC1), y = PC2,
    fill = PC2)) + geom_bar(stat = "identity") + coord_flip() +
```

```
scale_fill_distiller(palette = "RdYlBu") + theme_minimal()
dev.off()

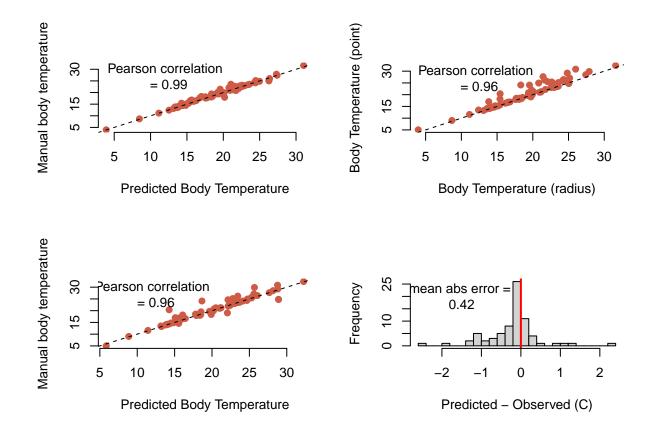
## pdf
## 2

behData$caste <- as.factor(behData$caste)
behData$uniqueID <- as.factor(behData$uniqueID)
behData$colony.id <- as.factor(behData$colony.id)</pre>
```

Body temperature validation

```
body_temp_ref_data <- read.csv("/Users/jamescrall/Dropbox/Work/_Research/Bees/Bumblebees/socialScaling0
temp_val_plots <- function(body_temp_ref_data) {</pre>
   par(mfcol = c(2, 2))
   plot(ground_truth_radius ~ predicted_radius, data = body_temp_ref_data,
        axes = FALSE, pch = 19, col = "coral3", xlab = ("Predicted Body Temperature"),
        ylab = "Manual body temperature")
   abline(a = 0, b = 1, lty = 2)
   axis(1)
   axis(2)
    cc = cor(body_temp_ref_data$ground_truth_radius, body_temp_ref_data$predicted_radius,
        method = "pearson", use = "complete.obs")
   text(12, 27, paste("Pearson correlation\n = ", signif(cc,
        2), sep = "")
   plot(ground_truth_pt ~ predicted_pt, data = body_temp_ref_data,
        axes = FALSE, pch = 19, col = "coral3", xlab = ("Predicted Body Temperature"),
        ylab = "Manual body temperature")
   abline(a = 0, b = 1, lty = 2)
   axis(1)
   axis(2)
    cc = cor(body_temp_ref_data$ground_truth_pt, body_temp_ref_data$predicted_pt,
        method = "pearson", use = "complete.obs")
   text(12, 27, paste("Pearson correlation\n = ", signif(cc,
        2), sep = ""))
   plot(ground_truth_pt ~ ground_truth_radius, data = body_temp_ref_data,
        axes = FALSE, pch = 19, col = "coral3", ylab = ("Body Temperature (point)"),
        xlab = "Body Temperature (radius)")
   abline(a = 0, b = 1, lty = 2)
    axis(1)
   axis(2)
    cc = cor(body_temp_ref_data$ground_truth_pt, body_temp_ref_data$ground_truth_radius,
        method = "pearson", use = "complete.obs")
   text(12, 27, paste("Pearson correlation\n = ", signif(cc,
        2), sep = ""))
   hist(body_temp_ref_data$predicted_radius - body_temp_ref_data$ground_truth_radius,
```

```
axes = FALSE, xlab = "Predicted - Observed (C)", ylab = "Frequency",
    breaks = 20, main = "")
axis(2)
abline(v = 0, lwd = 2, col = "red")
mean_error = mean(abs(body_temp_ref_data$predicted_radius -
    body_temp_ref_data$ground_truth_radius), na.rm = TRUE)
text(-1.5, 20, paste("mean abs error = \n", signif(mean_error,
    2), sep = ""))
axis(1)
}
# Plot inline
temp_val_plots(body_temp_ref_data)
```



```
setwd(output_folder)
# Export figure panel
pdf("FigS10.pdf")
temp_val_plots(body_temp_ref_data)
dev.off()
```

pdf ## 2

Aggregate behavioral effects

```
pc1.model <- lmer(PC1 ~ nestTemp + (1 | colony.id/uniqueID) +</pre>
    (1 | species) + (1 | block) + (1 | experimental.day), data = subset(behData,
   neonic == FALSE), REML = FALSE)
## Warning in checkConv(attr(opt, "derivs"), opt$par, ctrl = control$checkConv, :
## Model failed to converge with max|grad| = 0.00663284 (tol = 0.002, component 1)
pc2.model <- lmer(PC2 ~ nestTemp + (1 | colony.id/uniqueID) +</pre>
    (1 | species) + (1 | experimental.day), data = subset(behData,
    neonic == FALSE & rampState > 0), REML = FALSE)
## Warning in checkConv(attr(opt, "derivs"), opt$par, ctrl = control$checkConv, :
## Model failed to converge with max|grad| = 0.0039169 (tol = 0.002, component 1)
# NB removed 'block' random effect here to improve model
# convergence
summary(pc1.model)
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
     method [lmerModLmerTest]
## Formula: PC1 ~ nestTemp + (1 | colony.id/uniqueID) + (1 | species) + (1 |
       block) + (1 | experimental.day)
##
      Data: subset(behData, neonic == FALSE)
##
##
##
         AIC
                  BIC
                          logLik deviance df.resid
   256727.5 256800.0 -128355.7 256711.5
                                              63858
##
##
## Scaled residuals:
           1Q Median
##
      Min
                               ЗQ
                                      Max
## -4.4296 -0.5577 0.0821 0.6631 4.9196
##
## Random effects:
## Groups
                      Name
                                  Variance Std.Dev.
## uniqueID:colony.id (Intercept) 0.74655 0.8640
## colony.id
                      (Intercept) 1.32105 1.1494
## block
                       (Intercept) 0.29056 0.5390
## species
                       (Intercept) 0.15271 0.3908
## experimental.day
                      (Intercept) 0.01646 0.1283
## Residual
                                  3.16535 1.7791
## Number of obs: 63866, groups:
## uniqueID:colony.id, 716; colony.id, 21; block, 14; species, 4; experimental.day, 3
##
## Fixed effects:
                                            df t value Pr(>|t|)
##
                Estimate Std. Error
## (Intercept) 2.369e+00 4.019e-01 2.596e+00 5.894
                                                         0.0145 *
## nestTemp
            -7.552e-02 2.705e-03 6.233e+04 -27.920
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

```
##
## Correlation of Fixed Effects:
            (Intr)
## nestTemp -0.176
## optimizer (nloptwrap) convergence code: 0 (OK)
## Model failed to converge with max|grad| = 0.00663284 (tol = 0.002, component 1)
summary(pc2.model)
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
     method [lmerModLmerTest]
## Formula: PC2 ~ nestTemp + (1 | colony.id/uniqueID) + (1 | species) + (1 |
##
       experimental.day)
##
      Data: subset(behData, neonic == FALSE & rampState > 0)
##
##
        AIC
                       logLik deviance df.resid
##
   42091.6 42144.0 -21038.8 42077.6
## Scaled residuals:
      Min
               1Q Median
                                3Q
## -3.6740 -0.6812 -0.0525 0.6454 3.9710
## Random effects:
## Groups
                                   Variance Std.Dev.
                       Name
## uniqueID:colony.id (Intercept) 0.25828 0.5082
## colony.id
                       (Intercept) 0.39827 0.6311
## species
                       (Intercept) 0.30924 0.5561
## experimental.day
                       (Intercept) 0.02034 0.1426
## Residual
                                   1.29721 1.1390
## Number of obs: 13254, groups:
## uniqueID:colony.id, 692; colony.id, 21; species, 4; experimental.day, 3
##
## Fixed effects:
                 Estimate Std. Error
                                             df t value Pr(>|t|)
## (Intercept) 2.429e+00 3.559e-01 2.963e+00
                                                  6.826 0.00669 **
               -8.234e-02 2.976e-03 1.256e+04 -27.672 < 2e-16 ***
## nestTemp
## ---
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' 1
## Correlation of Fixed Effects:
            (Intr)
## nestTemp -0.197
## optimizer (nloptwrap) convergence code: 0 (OK)
## Model failed to converge with max|grad| = 0.0039169 (tol = 0.002, component 1)
Baseline model - test whether exposure has any effects pre-temperature stress
pc1.baseline <- lmer(PC1 ~ neonic * log10(colony.size.at.tagging) +</pre>
    (1 | block) + (1 | caste) + (1 | species/colony.id/uniqueID),
    data = subset(behData, rampState == 0 & time.from.start <</pre>
        first.ramp.time), REML = FALSE)
## Warning in checkConv(attr(opt, "derivs"), opt$par, ctrl = control$checkConv, :
## Model failed to converge with max|grad| = 0.00293511 (tol = 0.002, component 1)
```

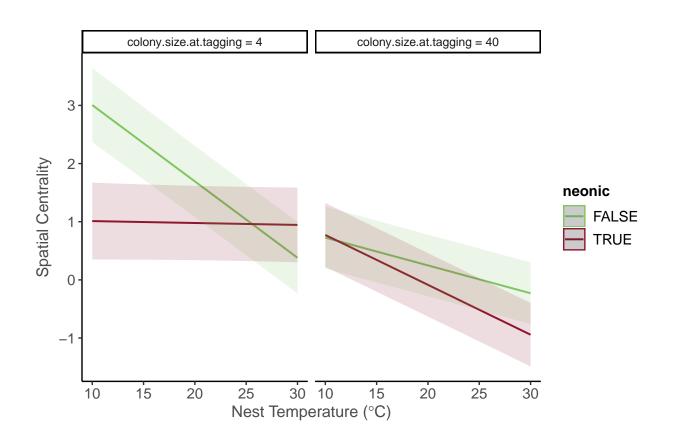
```
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
     method [lmerModLmerTest]
## Formula: PC1 ~ neonic * log10(colony.size.at.tagging) + (1 | block) +
       (1 | caste) + (1 | species/colony.id/uniqueID)
##
      Data: subset(behData, rampState == 0 & time.from.start < first.ramp.time)
##
##
                BIC
                       logLik deviance df.resid
## 184911.3 184998.8 -92445.7 184891.3
## Scaled residuals:
      Min
              1Q Median
                               30
                                       Max
## -4.4108 -0.5596 0.0657 0.6489 4.5172
##
## Random effects:
## Groups
                                             Variance Std.Dev.
                                Name
## uniqueID:(colony.id:species) (Intercept) 1.09448 1.0462
## colony.id:species
                                 (Intercept) 0.62475 0.7904
## block
                                 (Intercept) 0.43625 0.6605
## species
                                 (Intercept) 0.41058 0.6408
## caste
                                 (Intercept) 0.02557 0.1599
## Residual
                                             2.96743 1.7226
## Number of obs: 46316, groups:
## uniqueID:(colony.id:species), 1248; colony.id:species, 42; block, 15; species, 4; caste, 3
## Fixed effects:
                                            Estimate Std. Error
                                                                     df t value
## (Intercept)
                                              0.1772
                                                       0.8286 24.0020 0.214
## neonicTRUE
                                              1.7811
                                                         0.9681 36.6911
## log10(colony.size.at.tagging)
                                             -0.1442
                                                         0.5245 36.9546 -0.275
## neonicTRUE:log10(colony.size.at.tagging)
                                            -1.5602
                                                         0.6802 32.8656 -2.294
                                            Pr(>|t|)
## (Intercept)
                                              0.8325
## neonicTRUE
                                              0.0739 .
## log10(colony.size.at.tagging)
                                              0.7850
## neonicTRUE:log10(colony.size.at.tagging)
                                              0.0283 *
## ---
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' 1
##
## Correlation of Fixed Effects:
               (Intr) nnTRUE 110(...
##
## neonicTRUE -0.482
## lg10(cl...) -0.840 0.455
## nTRUE:10(.. 0.476 -0.955 -0.505
## optimizer (nloptwrap) convergence code: 0 (OK)
## Model failed to converge with max|grad| = 0.00293511 (tol = 0.002, component 1)
```

Active response model - effects of treatment during active cooling

```
pc1.active.response.model <- lmer(PC1 ~ log10(colony.size.at.tagging) *</pre>
    neonic * nestTemp + (1 | experimental.day) + (1 | block) +
    (1 | species) + (1 | colony.id/uniqueID) + (1 | caste), data = subset(behData,
    rampState %in% c(1, 2, 3) & caste %in% c("worker", "queen")))
summary(pc1.active.response.model)
## Linear mixed model fit by REML. t-tests use Satterthwaite's method [
## lmerModLmerTest]
## Formula: PC1 ~ log10(colony.size.at.tagging) * neonic * nestTemp + (1 |
       experimental.day) + (1 | block) + (1 | species) + (1 | colony.id/uniqueID) +
##
##
      Data: subset(behData, rampState %in% c(1, 2, 3) & caste %in% c("worker",
##
##
       "queen"))
##
## REML criterion at convergence: 49388.1
##
## Scaled residuals:
##
       Min
                1Q Median
                                3Q
                                       Max
## -4.7714 -0.5426 0.0700 0.6337
                                   3.4624
##
## Random effects:
## Groups
                                   Variance Std.Dev.
                       Name
## uniqueID:colony.id (Intercept) 1.4624
                                            1.2093
## colony.id
                                            0.9317
                       (Intercept) 0.8682
## block
                       (Intercept) 0.7168
                                            0.8466
## species
                       (Intercept) 1.1039
                                            1.0507
## experimental.day
                                            0.5640
                       (Intercept) 0.3181
## caste
                       (Intercept) 0.1179
                                            0.3433
## Residual
                                   2.5717
                                            1.6036
## Number of obs: 12509, groups:
## uniqueID:colony.id, 1084; colony.id, 42; block, 15; species, 4; experimental.day, 3; caste, 2
## Fixed effects:
                                                        Estimate Std. Error
## (Intercept)
                                                                   1.37028
                                                        6.19372
## log10(colony.size.at.tagging)
                                                        -3.11416
                                                                    0.78877
## neonicTRUE
                                                        -5.50255
                                                                    1.58819
## nestTemp
                                                        -0.18153
                                                                    0.03044
## log10(colony.size.at.tagging):neonicTRUE
                                                        3.70027
                                                                    1.08343
## log10(colony.size.at.tagging):nestTemp
                                                        0.08347
                                                                    0.01856
## neonicTRUE:nestTemp
                                                        0.22789
                                                                    0.04364
## log10(colony.size.at.tagging):neonicTRUE:nestTemp
                                                        -0.16594
                                                                    0.02801
##
                                                              df t value Pr(>|t|)
## (Intercept)
                                                        32.30369
                                                                 4.520 7.83e-05
## log10(colony.size.at.tagging)
                                                        62.97617 -3.948 0.000201
                                                        86.50730 -3.465 0.000828
## neonicTRUE
## nestTemp
                                                      8174.76380 -5.964 2.56e-09
## log10(colony.size.at.tagging):neonicTRUE
                                                                 3.415 0.001060
                                                        70.52601
## log10(colony.size.at.tagging):nestTemp
                                                     8789.86681
                                                                  4.497 6.99e-06
## neonicTRUE:nestTemp
                                                     8336.95162
                                                                 5.221 1.82e-07
## log10(colony.size.at.tagging):neonicTRUE:nestTemp 9189.52748 -5.924 3.26e-09
```

##

```
## (Intercept)
## log10(colony.size.at.tagging)
## neonicTRUE
## nestTemp
## log10(colony.size.at.tagging):neonicTRUE
## log10(colony.size.at.tagging):nestTemp
## neonicTRUE:nestTemp
## log10(colony.size.at.tagging):neonicTRUE:nestTemp ***
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' ' 1
## Correlation of Fixed Effects:
                  (Intr) lg10(...) nnTRUE nstTmp lg10(...):TRUE lg10(...):T nTRUE:
## lg10(cl...)
                  -0.820
## neonicTRUE
                  -0.517 0.536
## nestTemp
                  -0.532 0.554
                                    0.471
## lg10(...):TRUE 0.500 -0.559
                                   -0.962 -0.417
## lg10(...):T
                   0.524 - 0.567
                                   -0.462 -0.980 0.426
## nncTRUE:nsT
                   0.372 -0.386
                                   -0.662 -0.697 0.609
                                                                 0.683
## 110(...):TRUE: -0.349 0.376
                                    0.647 0.650 -0.627
                                                                -0.663
                                                                            -0.973
plot_model(pc1.active.response.model, type = "pred", terms = c("nestTemp",
    "neonic", "colony.size.at.tagging [4, 40]"), ci.lvl = 0.5,
    colors = list(ctrl_color, imid_color), title = "", axis.title = c(expression(paste("Nest Temperatur
        degree, "C)", sep = "")), "Spatial Centrality"))
```



```
# Export figure panel
setwd(output_folder)
pdf("Fig3 ab.pdf")
plot_model(pc1.active.response.model, type = "pred", terms = c("nestTemp",
    "neonic", "colony.size.at.tagging [4, 40]"), ci.lvl = 0.5,
    colors = list(ctrl_color, imid_color), title = "", axis.title = c(expression(paste("Nest Temperatur
        degree, "C)", sep = "")), "Spatial Centrality (PC1)"),
    show.legend = F)
dev.off()
## pdf
##
```

Recovery model - effects of treatment on behavioral recovery after cold exposure

```
sub.dat <- subset(behData, rampState %in% c(1, 5) & cold == TRUE)</pre>
sub.dat$rampState <- as.factor(sub.dat$rampState)</pre>
pc1.recovery.model <- lmer(PC1 ~ rampState_ch * neonic * log10(colony.size.at.tagging) +</pre>
    (1 | colony.id/uniqueID), data = sub.dat, REML = FALSE)
summary(pc1.recovery.model)
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
     method [lmerModLmerTest]
## Formula: PC1 ~ rampState_ch * neonic * log10(colony.size.at.tagging) +
       (1 | colony.id/uniqueID)
##
##
      Data: sub.dat
##
##
        AIC
                 BIC
                       logLik deviance df.resid
   13525.0 13592.1 -6751.5 13503.0
##
##
## Scaled residuals:
            1Q Median
##
       Min
                                ЗQ
                                       Max
## -4.3031 -0.5517 0.0608 0.6219 3.1433
##
## Random effects:
## Groups
                       Name
                                   Variance Std.Dev.
## uniqueID:colony.id (Intercept) 1.311
                                            1.145
## colony.id
                       (Intercept) 1.534
                                            1.239
## Residual
                                   2.858
                                             1.691
## Number of obs: 3281, groups: uniqueID:colony.id, 560; colony.id, 21
##
## Fixed effects:
##
                                                                  Estimate
## (Intercept)
                                                                    1.5541
## rampState_chb_after
                                                                    1.8789
## neonicTRUE
                                                                   -0.1569
## log10(colony.size.at.tagging)
                                                                   -0.5644
## rampState_chb_after:neonicTRUE
                                                                   -3.5832
## rampState_chb_after:log10(colony.size.at.tagging)
                                                                   -0.9422
## neonicTRUE:log10(colony.size.at.tagging)
                                                                   -0.7793
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging)
```

2.3034

```
Std. Error
##
## (Intercept)
                                                                    1.4934
## rampState chb after
                                                                    0.4796
## neonicTRUE
                                                                    2.0750
## log10(colony.size.at.tagging)
                                                                    1.0614
## rampState chb after:neonicTRUE
                                                                    0.6491
## rampState chb after:log10(colony.size.at.tagging)
                                                                    0.2953
## neonicTRUE:log10(colony.size.at.tagging)
                                                                    1.4793
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging)
                                                                    0.4011
##
                                                                       df t value
## (Intercept)
                                                                  25.0239
                                                                            1.041
## rampState_chb_after
                                                                2836.3541
                                                                            3.917
## neonicTRUE
                                                                  24.1768 -0.076
## log10(colony.size.at.tagging)
                                                                  22.9009 -0.532
## rampState_chb_after:neonicTRUE
                                                                2879.7773 -5.520
## rampState_chb_after:log10(colony.size.at.tagging)
                                                                2844.3204 -3.191
## neonicTRUE:log10(colony.size.at.tagging)
                                                                  22.3783 -0.527
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging) 2905.3936
                                                                            5.743
                                                                Pr(>|t|)
## (Intercept)
                                                                 0.30801
## rampState_chb_after
                                                                9.17e-05 ***
## neonicTRUE
                                                                 0.94034
## log10(colony.size.at.tagging)
                                                                 0.60002
## rampState chb after:neonicTRUE
                                                                3.69e-08 ***
## rampState_chb_after:log10(colony.size.at.tagging)
                                                                 0.00144 **
## neonicTRUE:log10(colony.size.at.tagging)
                                                                 0.60350
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging) 1.03e-08 ***
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' ' 1
## Correlation of Fixed Effects:
##
               (Intr) rmpS_ nnTRUE 110(.. rmS__:TRUE rS__:1 nTRUE:
## rmpStt_chb_ -0.217
## neonicTRUE -0.720 0.156
## lg10(cl...) -0.960 0.180 0.691
## rmpS__:TRUE   0.160 -0.739 -0.219 -0.133
## rS :10(... 0.211 -0.982 -0.152 -0.181 0.726
## nTRUE:10(.. 0.689 -0.129 -0.960 -0.718 0.183
                                                       0.130
## rS_:TRUE:1 -0.155 0.723 0.213 0.133 -0.977
                                                      -0.736 -0.186
pc2.recovery.model <- lmer(PC2 ~ rampState_ch * neonic * colony.size.at.tagging +
    (1 | colony.id/uniqueID), data = sub.dat, REML = FALSE)
summary(pc2.recovery.model)
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
    method [lmerModLmerTest]
## Formula:
## PC2 ~ rampState_ch * neonic * colony.size.at.tagging + (1 | colony.id/uniqueID)
##
      Data: sub.dat
##
##
        AIC
                 BIC logLik deviance df.resid
  10348.6 10415.6 -5163.3 10326.6
##
                                           3270
## Scaled residuals:
```

```
1Q Median
                               3Q
## -3.0779 -0.6323 -0.0433 0.6046 4.1242
##
## Random effects:
## Groups
                       Name
                                   Variance Std.Dev.
## uniqueID:colony.id (Intercept) 0.3291
                                           0.5737
## colony.id
                                            0.5515
                       (Intercept) 0.3041
## Residual
                                   1.1444
                                            1.0698
## Number of obs: 3281, groups: uniqueID:colony.id, 560; colony.id, 21
##
## Fixed effects:
##
                                                           Estimate Std. Error
## (Intercept)
                                                          5.663e-01 3.226e-01
## rampState_chb_after
                                                         -6.916e-01 1.301e-01
## neonicTRUE
                                                          3.163e-01 4.221e-01
## colony.size.at.tagging
                                                         -1.253e-02 8.076e-03
## rampState_chb_after:neonicTRUE
                                                          3.090e-01 1.724e-01
## rampState_chb_after:colony.size.at.tagging
                                                          8.203e-03 2.467e-03
                                                          1.073e-02 9.989e-03
## neonicTRUE:colony.size.at.tagging
## rampState_chb_after:neonicTRUE:colony.size.at.tagging -6.766e-03 3.056e-03
##
                                                                 df t value
## (Intercept)
                                                          2.789e+01
                                                                      1.755
                                                          2.952e+03 -5.316
## rampState_chb_after
## neonicTRUE
                                                          2.800e+01
                                                                      0.749
## colony.size.at.tagging
                                                          2.221e+01 -1.552
## rampState_chb_after:neonicTRUE
                                                          2.970e+03
                                                                     1.792
## rampState_chb_after:colony.size.at.tagging
                                                                      3.326
                                                          2.978e+03
## neonicTRUE:colony.size.at.tagging
                                                          2.239e+01
                                                                      1.075
## rampState_chb_after:neonicTRUE:colony.size.at.tagging 3.008e+03 -2.214
##
                                                         Pr(>|t|)
## (Intercept)
                                                         0.090162 .
## rampState_chb_after
                                                         1.14e-07 ***
## neonicTRUE
                                                         0.459943
## colony.size.at.tagging
                                                         0.134858
## rampState_chb_after:neonicTRUE
                                                         0.073178 .
## rampState_chb_after:colony.size.at.tagging
                                                         0.000893 ***
## neonicTRUE:colony.size.at.tagging
                                                         0.293959
## rampState_chb_after:neonicTRUE:colony.size.at.tagging 0.026905 *
## ---
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' 1
## Correlation of Fixed Effects:
               (Intr) rmpS_ nnTRUE cln... rmS_:TRUE rS_:. nTRUE:
## rmpStt_chb_ -0.278
## neonicTRUE -0.764 0.213
## clny.sz.t.t -0.806 0.175 0.616
## rmpS__:TRUE   0.210 -0.755 -0.291 -0.132
## rmpSt__:... 0.242 -0.899 -0.185 -0.183 0.679
## nncTRUE:... 0.652 -0.142 -0.776 -0.809 0.178
                                                       0.148
## rS_:TRUE:. -0.195  0.726  0.244  0.147  -0.865
                                                      -0.807 -0.197
# Export panels
setwd(output folder)
pdf("Fig3_cd.pdf")
```

#Worker group models Characterize treatment effects by initial behavioral groups (bees that are on vs. off the nest at beginning of trials)

```
### look at effects during ramp down by whether bees were
### already on the nest
sub.dat <- subset(behData, rampState %in% c(1, 2, 3) & cold ==</pre>
    TRUE) #Subset to data in initial cold exposure
colony.size.cuttoff <- 30</pre>
# Define workers that were on nest at ambient temperatures
sd <- subset(sub.dat, rampState < 2 & nestTemp > 23)
tmp <- tapply(sd$medianDistancetoClosestBrood, sd$uniqueID, median,</pre>
    na.rm = TRUE) #Get averages in this time period for all bees
incubator.list <- names(tmp)[which(tmp < pixel_conversion)]</pre>
non.incubator.list <- names(tmp)[which(tmp > pixel_conversion *
    2)]
# Model responses with 'incubator' as an explicit
# interaction term
sub.dat$incubator <- NA</pre>
sub.dat$incubator[sub.dat$uniqueID %in% incubator.list] <- "inc"</pre>
sub.dat$incubator[sub.dat$uniqueID %in% non.incubator.list] <- "ninc"</pre>
pc1.response.model.by.incubator <- lmer(PC1 ~ nestTemp * neonic *</pre>
    incubator + (1 | colony.id/uniqueID), data = subset(sub.dat,
    caste == "worker" & incubator %in% c("inc", "ninc") & colony.size.at.tagging <</pre>
        colony.size.cuttoff), REML = FALSE)
summary(pc1.response.model.by.incubator)
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
```

```
method [lmerModLmerTest]
## Formula: PC1 ~ nestTemp * neonic * incubator + (1 | colony.id/uniqueID)
     Data: subset(sub.dat, caste == "worker" & incubator %in% c("inc", "ninc") &
##
       colony.size.at.tagging < colony.size.cuttoff)</pre>
##
##
        AIC
                 BIC
                     logLik deviance df.resid
     5558.0 5616.2 -2768.0
##
                                5536.0
                                           1454
##
## Scaled residuals:
       Min
                1Q Median
                                3Q
                                       Max
## -4.8545 -0.4826 0.0250 0.5353 3.5859
##
```

```
## Random effects:
                                  Variance Std.Dev.
## Groups
                      Name
## uniqueID:colony.id (Intercept) 1.2910
                                           1.1362
## colony.id
                       (Intercept) 0.3442
                                           0.5867
## Residual
                                  2.2086
                                            1.4862
## Number of obs: 1465, groups: uniqueID:colony.id, 101; colony.id, 13
## Fixed effects:
##
                                      Estimate Std. Error
                                                                  df t value
## (Intercept)
                                     3.516e+00 5.224e-01 4.187e+01
                                                                      6.731
## nestTemp
                                    -5.559e-02 1.755e-02 1.372e+03 -3.168
## neonicTRUE
                                    -2.911e-01 6.644e-01 3.523e+01 -0.438
## incubatorninc
                                    -4.336e-02 7.086e-01 3.859e+02 -0.061
## nestTemp:neonicTRUE
                                     7.875e-03 2.182e-02 1.371e+03
                                                                      0.361
## nestTemp:incubatorninc
                                    -1.207e-01 2.917e-02 1.405e+03 -4.136
## neonicTRUE:incubatorninc
                                    -3.829e+00 9.483e-01 3.316e+02 -4.038
## nestTemp:neonicTRUE:incubatorninc 1.064e-01 3.730e-02 1.426e+03
                                                                       2.851
##
                                    Pr(>|t|)
## (Intercept)
                                    3.58e-08 ***
## nestTemp
                                     0.00157 **
## neonicTRUE
                                     0.66401
## incubatorninc
                                     0.95124
## nestTemp:neonicTRUE
                                     0.71828
## nestTemp:incubatorninc
                                    3.74e-05 ***
## neonicTRUE:incubatorninc
                                    6.71e-05 ***
## nestTemp:neonicTRUE:incubatorninc 0.00442 **
## ---
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' 1
## Correlation of Fixed Effects:
##
               (Intr) nstTmp nnTRUE incbtr nsT:TRUE nstTm: nTRUE:
## nestTemp
               -0.613
## neonicTRUE -0.786 0.482
## incubatrnnc -0.559 0.449 0.440
## nstTmp:TRUE 0.493 -0.804 -0.597 -0.361
## nstTmp:ncbt 0.363 -0.601 -0.285 -0.750 0.483
## nncTRUE:ncb 0.418 -0.335 -0.537 -0.747 0.417
                                                    0.560
## nstTm:TRUE: -0.284 0.470 0.344 0.587 -0.585
                                                   -0.782 -0.743
setwd("/Users/jamescrall/Dropbox/Work/ Writing/2022 SocialScaling/Figures")
pdf("Fig5A.pdf")
dat <- subset(sub.dat, rampState == 3 & bodyTemp localAv > 10 &
    colony.size.at.tagging < colony.size.cuttoff)</pre>
beeswarm(bodyTemp_localAv ~ incubator, data = dat, pch = 19,
    cex = 0.75, col = c("darkgoldenrod2", "mediumpurple3"), axes = FALSE,
    ann = FALSE)
bxplot(bodyTemp_localAv ~ incubator, data = dat, add = TRUE,
axis(2)
dev.off()
## pdf
```

##

```
pdf("Fig5B.pdf")
plot_model(pc1.response.model.by.incubator, type = "pred", terms = c("nestTemp",
    "neonic", "incubator"), ci.lvl = 0.5, colors = list(ctrl_color,
    imid_color), axis.title = c(expression(paste("Nest Temperature (",
    degree, "C)", sep = "")), "Spatial Centrality (PC1)"), show.legend = F,
    title = "")
dev.off()
## pdf
##
# Subset data
sub.dat <- subset(behData, rampState %in% c(1, 5) & cold == TRUE)</pre>
sub.dat$rampState <- as.factor(sub.dat$rampState)</pre>
# Separate model for incubators (bees on nest)
pc1.recovery.model.incubators <- lmer(PC1 ~ rampState_ch * neonic *</pre>
    log10(colony.size.at.tagging) + experimental.day + (1 | uniqueID) +
    (1 | colony.id), data = subset(sub.dat, caste == "worker" &
    uniqueID %in% incubator.list), REML = FALSE)
summary(pc1.recovery.model.incubators)
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
     method [lmerModLmerTest]
## Formula: PC1 ~ rampState_ch * neonic * log10(colony.size.at.tagging) +
##
       experimental.day + (1 | uniqueID) + (1 | colony.id)
      Data: subset(sub.dat, caste == "worker" & uniqueID %in% incubator.list)
##
##
##
        AIC
                       logLik deviance df.resid
##
     3681.4
             3744.3 -1827.7
                              3655.4
##
## Scaled residuals:
               1Q Median
                                30
       Min
## -4.0867 -0.4323 0.1868 0.6505 2.5209
## Random effects:
## Groups
             Name
                          Variance Std.Dev.
## uniqueID (Intercept) 0.2770 0.5263
## colony.id (Intercept) 0.2106
                                 0.4589
## Residual
                          2.6034
                                  1.6135
## Number of obs: 939, groups: uniqueID, 151; colony.id, 15
##
## Fixed effects:
##
                                                                  Estimate
                                                                   3.07585
## (Intercept)
## rampState_chb_after
                                                                  -0.03334
## neonicTRUE
                                                                  -0.49768
## log10(colony.size.at.tagging)
                                                                  -0.97400
## experimental.day2
                                                                   0.32133
## experimental.day3
                                                                   0.31286
## rampState_chb_after:neonicTRUE
                                                                   0.40138
## rampState chb after:log10(colony.size.at.tagging)
                                                                  -0.18455
## neonicTRUE:log10(colony.size.at.tagging)
                                                                   0.23139
```

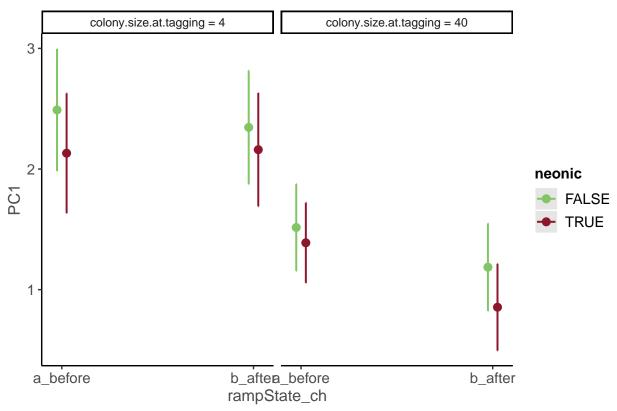
```
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging)
                                                                 -0.37846
##
                                                                Std. Error
## (Intercept)
                                                                    1.04321
                                                                    0.77982
## rampState_chb_after
## neonicTRUE
                                                                    1.27024
## log10(colony.size.at.tagging)
                                                                    0.60943
## experimental.day2
                                                                    0.47598
## experimental.day3
                                                                    0.49287
## rampState_chb_after:neonicTRUE
                                                                    1.04133
## rampState_chb_after:log10(colony.size.at.tagging)
                                                                    0.47605
## neonicTRUE:log10(colony.size.at.tagging)
                                                                    0.84266
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging)
                                                                    0.64992
                                                                        df t value
## (Intercept)
                                                                  32.12358
                                                                             2.948
## rampState_chb_after
                                                                859.05955 -0.043
## neonicTRUE
                                                                  14.59760
                                                                            -0.392
## log10(colony.size.at.tagging)
                                                                  13.26451 -1.598
## experimental.day2
                                                                796.08981
                                                                            0.675
## experimental.day3
                                                                780.81870
                                                                             0.635
## rampState_chb_after:neonicTRUE
                                                                865.26567
                                                                             0.385
## rampState_chb_after:log10(colony.size.at.tagging)
                                                                863.94173 -0.388
## neonicTRUE:log10(colony.size.at.tagging)
                                                                 10.36874
                                                                             0.275
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging) 867.44245 -0.582
                                                                Pr(>|t|)
## (Intercept)
                                                                 0.00591 **
## rampState_chb_after
                                                                  0.96591
## neonicTRUE
                                                                  0.70086
## log10(colony.size.at.tagging)
                                                                  0.13353
## experimental.day2
                                                                  0.49981
## experimental.day3
                                                                  0.52576
## rampState_chb_after:neonicTRUE
                                                                  0.70000
## rampState_chb_after:log10(colony.size.at.tagging)
                                                                  0.69836
## neonicTRUE:log10(colony.size.at.tagging)
                                                                  0.78902
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging)
                                                                 0.56050
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' ' 1
## Correlation of Fixed Effects:
               (Intr) rmpS_ nnTRUE 110(.. expr.2 expr.3 rmS_:TRUE rS_:1 nTRUE:
##
## rmpStt_chb_ -0.477
## neonicTRUE -0.625 0.391
## lg10(cl...) -0.862 0.478 0.715
## exprmntl.d2 -0.437 -0.024 -0.081 -0.017
## exprmntl.d3 -0.437 0.013 -0.087 -0.017 0.966
## rmpS__:TRUE   0.328 -0.755 -0.502 -0.359   0.090   0.051
## rS__:10(... 0.462 -0.982 -0.375 -0.473 0.021 -0.024 0.743
## nTRUE:10(.. 0.574 -0.340 -0.963 -0.725 0.118 0.129 0.448
                                                                      0.334
## rS_:TRUE:1 -0.285 0.726 0.484 0.349 -0.140 -0.097 -0.974
                                                                    -0.739 - 0.456
pc1.recovery.model.non.incubators <- lmer(PC1 ~ rampState_ch *</pre>
   neonic * log10(colony.size.at.tagging) + experimental.day +
    (1 | uniqueID) + (1 | colony.id), data = subset(sub.dat,
    caste == "worker" & uniqueID %in% non.incubator.list), REML = FALSE)
summary(pc1.recovery.model.non.incubators)
```

```
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
    method [lmerModLmerTest]
## Formula: PC1 ~ rampState_ch * neonic * log10(colony.size.at.tagging) +
       experimental.day + (1 | uniqueID) + (1 | colony.id)
      Data: subset(sub.dat, caste == "worker" & uniqueID %in% non.incubator.list)
##
##
                       logLik deviance df.resid
##
        AIC
     3457.5
              3519.0 -1715.8 3431.5
##
##
## Scaled residuals:
        Min
                  1Q
                       Median
                                    3Q
                                            Max
## -2.73891 -0.71961 -0.05607 0.57924 3.14171
## Random effects:
## Groups
                          Variance Std.Dev.
              Name
## uniqueID (Intercept) 1.1973
                                   1.0942
                                   0.3887
## colony.id (Intercept) 0.1511
## Residual
                          2.8797
                                   1.6970
## Number of obs: 836, groups: uniqueID, 144; colony.id, 15
## Fixed effects:
##
                                                                 Estimate
## (Intercept)
                                                                   0.3028
## rampState chb after
                                                                   2.4213
## neonicTRUE
                                                                  -1.1893
## log10(colony.size.at.tagging)
                                                                  -0.5204
## experimental.day2
                                                                  -0.7614
## experimental.day3
                                                                  -0.9498
## rampState_chb_after:neonicTRUE
                                                                  -6.4569
## rampState_chb_after:log10(colony.size.at.tagging)
                                                                  -0.5644
## neonicTRUE:log10(colony.size.at.tagging)
                                                                   0.0589
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging)
                                                                   3.8719
##
                                                                 Std. Error
## (Intercept)
                                                                     1.4757
## rampState_chb_after
                                                                     1.1863
## neonicTRUE
                                                                     1.8935
## log10(colony.size.at.tagging)
                                                                     0.9297
## experimental.day2
                                                                     0.3654
## experimental.day3
                                                                     0.3990
## rampState_chb_after:neonicTRUE
                                                                     1.4472
## rampState_chb_after:log10(colony.size.at.tagging)
                                                                     0.7546
## neonicTRUE:log10(colony.size.at.tagging)
                                                                     1.1963
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging)
                                                                     0.9026
##
                                                                       df t value
## (Intercept)
                                                                            0.205
                                                                  23.9353
## rampState_chb_after
                                                                 747.4004
                                                                            2.041
## neonicTRUE
                                                                  15.9790 -0.628
## log10(colony.size.at.tagging)
                                                                  15.2236 -0.560
## experimental.day2
                                                                 658.6795
                                                                          -2.084
## experimental.day3
                                                                 637.0708
                                                                           -2.381
## rampState_chb_after:neonicTRUE
                                                                 745.7581
                                                                           -4.462
## rampState chb after:log10(colony.size.at.tagging)
                                                                 748.9345 -0.748
## neonicTRUE:log10(colony.size.at.tagging)
                                                                  11.3623
                                                                           0.049
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging) 748.4433
                                                                            4.290
```

```
##
                                                                 Pr(>|t|)
## (Intercept)
                                                                   0.8392
                                                                   0.0416 *
## rampState chb after
## neonicTRUE
                                                                   0.5388
## log10(colony.size.at.tagging)
                                                                   0.5838
## experimental.day2
                                                                   0.0376 *
## experimental.day3
                                                                   0.0176 *
## rampState_chb_after:neonicTRUE
                                                                 9.38e-06 ***
## rampState_chb_after:log10(colony.size.at.tagging)
                                                                   0.4547
## neonicTRUE:log10(colony.size.at.tagging)
                                                                   0.9616
## rampState_chb_after:neonicTRUE:log10(colony.size.at.tagging) 2.02e-05 ***
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' ' 1
##
## Correlation of Fixed Effects:
##
               (Intr) rmpS_ nnTRUE 110(.. expr.2 expr.3 rmS_:TRUE rS_:1 nTRUE:
## rmpStt_chb_ -0.478
## neonicTRUE -0.730 0.373
## lg10(cl...) -0.950 0.468 0.753
## exprmntl.d2 -0.213 -0.050 -0.067 -0.029
## exprmntl.d3 -0.193 -0.012 -0.119 -0.046 0.907
## rmpS :TRUE 0.382 -0.815 -0.488 -0.389 0.088 0.088
## rS_:10(... 0.459 -0.983 -0.355 -0.462 0.044 -0.002 0.800
## nTRUE:10(.. 0.724 -0.359 -0.979 -0.784 0.088 0.142 0.463
                                                                      0.351
## rS :TRUE:1 -0.367 0.820 0.466 0.391 -0.113 -0.097 -0.982
                                                                     -0.832 - 0.456
# Explicit model for incubator interaction effect, subset
# to small colonies
sub.dat$incubator <- NA</pre>
sub.dat$incubator[sub.dat$uniqueID %in% incubator.list] <- "inc"</pre>
sub.dat$incubator[sub.dat$uniqueID %in% non.incubator.list] <- "ninc"</pre>
pc1.recovery.model.by.incubator <- lmer(PC1 ~ rampState_ch *</pre>
    neonic * incubator + (1 | colony.id/uniqueID), data = subset(sub.dat,
    caste == "worker" & incubator %in% c("inc", "ninc") & colony.size.at.tagging <</pre>
        colony.size.cuttoff), REML = FALSE)
summary(pc1.recovery.model.by.incubator)
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
    method [lmerModLmerTest]
## Formula: PC1 ~ rampState_ch * neonic * incubator + (1 | colony.id/uniqueID)
      Data: subset(sub.dat, caste == "worker" & incubator %in% c("inc", "ninc") &
##
       colony.size.at.tagging < colony.size.cuttoff)</pre>
##
##
        AIC
                       logLik deviance df.resid
     2965.8
              3016.2 -1471.9
##
                                2943.8
                                            714
## Scaled residuals:
               10 Median
                                3Q
## -3.8747 -0.5014 0.0561 0.5411 2.9187
## Random effects:
                                   Variance Std.Dev.
## Groups
                       Name
## uniqueID:colony.id (Intercept) 0.7474
```

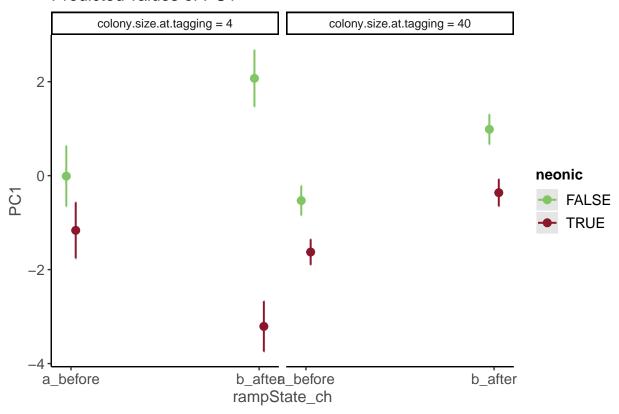
```
## colony.id
                       (Intercept) 0.6445
                                            0.8028
## Residual
                                   2.8696
                                            1.6940
## Number of obs: 725, groups: uniqueID:colony.id, 101; colony.id, 13
## Fixed effects:
##
                                                Estimate Std. Error
                                                                          df
## (Intercept)
                                                            0.4881 20.4189
                                                  2.4337
## rampState_chb_after
                                                 -0.4278
                                                             0.2958 645.9634
## neonicTRUE
                                                 -0.2821
                                                             0.6471 18.0846
## incubatorninc
                                                 -3.6775
                                                             0.5274 190.8373
## rampState_chb_after:neonicTRUE
                                                  0.1318
                                                             0.3914 650.7818
## rampState_chb_after:incubatorninc
                                                  1.9429
                                                             0.4659 656.6091
## neonicTRUE:incubatorninc
                                                 -0.8337
                                                             0.7123 176.6817
## rampState_chb_after:neonicTRUE:incubatorninc -1.4016
                                                             0.5824 656.0326
##
                                                t value Pr(>|t|)
## (Intercept)
                                                  4.986 6.69e-05 ***
## rampState_chb_after
                                                 -1.446
                                                          0.1486
## neonicTRUE
                                                 -0.436
                                                          0.6680
## incubatorninc
                                                 -6.972 4.96e-11 ***
## rampState chb after:neonicTRUE
                                                  0.337
                                                          0.7364
                                                  4.170 3.45e-05 ***
## rampState_chb_after:incubatorninc
## neonicTRUE:incubatorninc
                                                 -1.170
                                                          0.2434
## rampState_chb_after:neonicTRUE:incubatorninc -2.407
                                                          0.0164 *
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## Correlation of Fixed Effects:
               (Intr) rmpS__ nnTRUE incbtr rmS__:TRUE rmS__: nTRUE:
## rmpStt_chb_ -0.408
## neonicTRUE -0.754 0.308
## incubatrnnc -0.489 0.375 0.369
## rmpS__:TRUE   0.309 -0.756 -0.400 -0.283
## rmpStt_ch_: 0.242 -0.633 -0.182 -0.562 0.478
## nncTRUE:ncb 0.362 -0.277 -0.504 -0.740 0.365
                                                       0.416
## rmS__:TRUE: -0.193  0.506  0.253  0.450 -0.671
                                                      -0.800 -0.524
# Plot models
plot_model(pc1.recovery.model.incubators, type = "pred", terms = c("rampState_ch",
    "neonic", "colony.size.at.tagging [4, 40]]"), ci.lvl = 0.5,
   colors = list(ctrl_color, imid_color))
```

Predicted values of PC1



```
plot_model(pc1.recovery.model.non.incubators, type = "pred",
    terms = c("rampState_ch", "neonic", "colony.size.at.tagging [4, 40]]"),
    ci.lvl = 0.5, colors = list(ctrl_color, imid_color))
```

Predicted values of PC1



"Spatial Centrality (PC1)"), show.legend = F, title = "Off-nest workers")

"Spatial Centrality (PC1)"), show.legend = F, title = "On-nest workers")

ci.lvl = 0.5, colors = list(ctrl_color, imid_color), axis.title = c("Observation Period",

terms = c("rampState_ch", "neonic", "colony.size.at.tagging [4, 40]]"),

b <- plot_model(pc1.recovery.model.non.incubators, type = "pred",</pre>

grid.arrange(a, b, nrow = 1)

dev.off()

```
## pdf
## 2
```

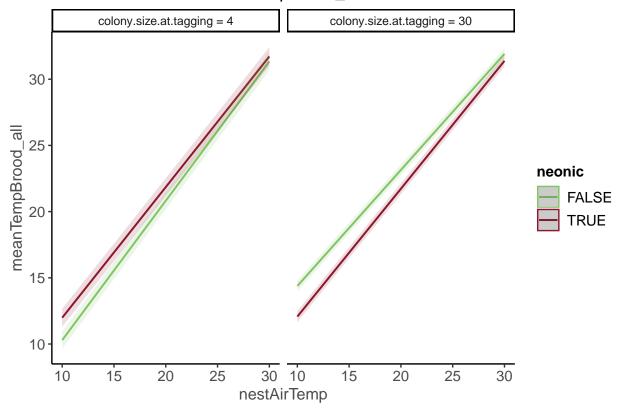
Brood surface temperature analysis

```
thermData$neonic <- as.factor(thermData$neonic) #Convert to factor
dat <- subset(thermData, tempRampState %in% c(1, 2, 3) & cold ==</pre>
brood.temp.model <- lmer(meanTempBrood_all ~ nestAirTemp * neonic *</pre>
    log10(colony.size.at.tagging) + (1 | species/colony.id) +
    (1 | experimental.day), data = dat, REML = FALSE)
rem.ind <- which(abs(residuals(brood.temp.model)) > 4) #Remove two outliers
dat.no <- dat[-rem.ind, ]</pre>
brood.temp.modelno.outliers <- lmer(meanTempBrood_all ~ nestAirTemp *</pre>
    neonic * log10(colony.size.at.tagging) + (1 | species/colony.id) +
    (1 | block), data = dat.no)
# No qualitative effect of removing outliers - retain
# original model
# Recovery model
dat <- subset(thermData, tempRampState %in% c(1, 5) & cold ==</pre>
    TRUE)
dat$tempRampState <- as.factor(dat$tempRampState)</pre>
brood.temp.recovery.model <- lmer(meanTempBrood_all ~ tempRampState *</pre>
    neonic * log10(colony.size.at.tagging) + (1 | colony.id),
    data = dat)
# Control model for comparison
dat <- subset(thermData, tempRampState %in% c(1, 5) & cold ==</pre>
    FALSE)
dat$tempRampState <- as.factor(dat$tempRampState)</pre>
brood.temp.recovery.model.ctl <- lmer(meanTempBrood_all ~ tempRampState +</pre>
    neonic + log10(colony.size.at.tagging) + (1 | colony.id),
    data = dat)
summary(brood.temp.model)
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
     method [lmerModLmerTest]
## Formula:
## meanTempBrood_all ~ nestAirTemp * neonic * log10(colony.size.at.tagging) +
##
       (1 | species/colony.id) + (1 | experimental.day)
##
      Data: dat
##
##
        AIC
                 BIC logLik deviance df.resid
##
     1946.6
              2000.0 -961.3
                                1922.6
##
```

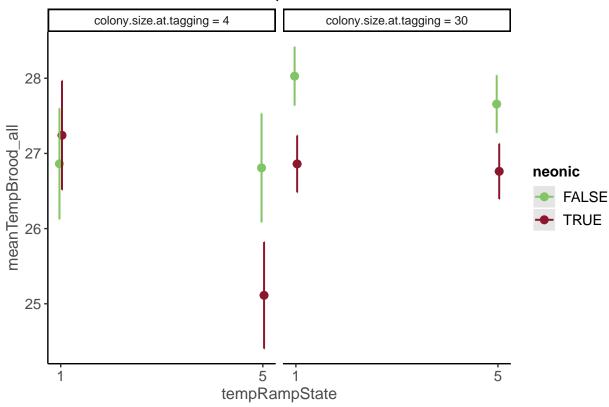
```
## Scaled residuals:
##
      Min
           1Q Median
                                30
                                       Max
## -5.2071 -0.5925 -0.0430 0.5243 4.4022
## Random effects:
## Groups
                                  Variance Std.Dev.
                      Name
## colony.id:species (Intercept) 1.5439
                                           1.2425
## species
                      (Intercept) 0.3092
                                           0.5561
## experimental.day (Intercept) 0.1293
                                           0.3595
## Residual
                                  1.0563
                                           1.0278
## Number of obs: 636, groups:
## colony.id:species, 20; species, 4; experimental.day, 3
## Fixed effects:
##
                                                         Estimate Std. Error
## (Intercept)
                                                         -4.24856
                                                                     1.70365
                                                          1.17337
                                                                     0.04667
## nestAirTemp
## neonicTRUE
                                                          6.16911
                                                                     2.53017
## log10(colony.size.at.tagging)
                                                          6.67527
                                                                     1.17965
## nestAirTemp:neonicTRUE
                                                         -0.17242
                                                                     0.06885
## nestAirTemp:log10(colony.size.at.tagging)
                                                         -0.20074
                                                                     0.03345
## neonicTRUE:log10(colony.size.at.tagging)
                                                         -6.34302
                                                                     1.85403
## nestAirTemp:neonicTRUE:log10(colony.size.at.tagging)
                                                          0.17696
                                                                     0.05189
                                                               df t value Pr(>|t|)
                                                         35.62000 -2.494 0.017423
## (Intercept)
## nestAirTemp
                                                        615.47005 25.140 < 2e-16
## neonicTRUE
                                                         33.68491
                                                                    2.438 0.020191
## log10(colony.size.at.tagging)
                                                                    5.659 3.34e-06
                                                         30.73865
## nestAirTemp:neonicTRUE
                                                        615.90154 -2.504 0.012521
## nestAirTemp:log10(colony.size.at.tagging)
                                                        615.17944 -6.001 3.34e-09
## neonicTRUE:log10(colony.size.at.tagging)
                                                         35.82445 -3.421 0.001573
## nestAirTemp:neonicTRUE:log10(colony.size.at.tagging) 616.27945
                                                                    3.410 0.000692
##
## (Intercept)
## nestAirTemp
## neonicTRUE
## log10(colony.size.at.tagging)
## nestAirTemp:neonicTRUE
## nestAirTemp:log10(colony.size.at.tagging)
## neonicTRUE:log10(colony.size.at.tagging)
## nestAirTemp:neonicTRUE:log10(colony.size.at.tagging) ***
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Correlation of Fixed Effects:
               (Intr) nstArT nnTRUE 110(.. nsAT:TRUE nAT:10 nTRUE:
##
## nestAirTemp -0.480
## neonicTRUE -0.673 0.323
## lg10(cl...) -0.931 0.474 0.630
## nstArT:TRUE 0.327 -0.676 -0.483 -0.321
## nAT:10(...) 0.458 -0.954 -0.308 -0.497 0.645
## nTRUE:10(.. 0.636 -0.301 -0.963 -0.649 0.479
                                                      0.315
## nAT:TRUE:10 -0.297 0.612 0.465 0.320 -0.960
                                                     -0.642 -0.500
```

```
## Linear mixed model fit by REML. t-tests use Satterthwaite's method [
## lmerModLmerTest]
## Formula:
## meanTempBrood_all ~ tempRampState * neonic * log10(colony.size.at.tagging) +
       (1 | colony.id)
##
##
      Data: dat
## REML criterion at convergence: 614.7
## Scaled residuals:
      Min
              10 Median
                                30
                                       Max
## -4.0748 -0.5206 0.0313 0.4608 3.3561
##
## Random effects:
                          Variance Std.Dev.
## Groups Name
## colony.id (Intercept) 2.605
                                   1.6140
## Residual
                          0.701
                                   0.8373
## Number of obs: 222, groups: colony.id, 21
##
## Fixed effects:
##
                                                           Estimate Std. Error
## (Intercept)
                                                            26.0603
                                                                       1.8385
                                                                        0.6470
## tempRampState5
                                                             0.1631
## neonicTRUE
                                                             1.4439
                                                                        2.5934
## log10(colony.size.at.tagging)
                                                             1.3320
                                                                        1.3388
## tempRampState5:neonicTRUE
                                                            -3.6901
                                                                        0.9158
## tempRampState5:log10(colony.size.at.tagging)
                                                            -0.3614
                                                                        0.4792
## neonicTRUE:log10(colony.size.at.tagging)
                                                            -1.7679
                                                                        1.8971
## tempRampState5:neonicTRUE:log10(colony.size.at.tagging)
                                                             2.6825
                                                                        0.6908
##
                                                                 df t value
## (Intercept)
                                                            18.0818 14.175
## tempRampState5
                                                           198.2826
                                                                     0.252
## neonicTRUE
                                                            18.4717
                                                                      0.557
## log10(colony.size.at.tagging)
                                                            18.2007
                                                                      0.995
## tempRampState5:neonicTRUE
                                                           198.2539 -4.029
## tempRampState5:log10(colony.size.at.tagging)
                                                                     -0.754
                                                           198.6021
## neonicTRUE:log10(colony.size.at.tagging)
                                                            18.8889
                                                                     -0.932
## tempRampState5:neonicTRUE:log10(colony.size.at.tagging) 198.4540
                                                                      3.883
                                                           Pr(>|t|)
                                                           3.10e-11 ***
## (Intercept)
## tempRampState5
                                                            0.80120
                                                            0.58438
## neonicTRUE
## log10(colony.size.at.tagging)
                                                            0.33280
## tempRampState5:neonicTRUE
                                                           7.97e-05 ***
## tempRampState5:log10(colony.size.at.tagging)
                                                            0.45165
## neonicTRUE:log10(colony.size.at.tagging)
                                                            0.36317
## tempRampState5:neonicTRUE:log10(colony.size.at.tagging) 0.00014 ***
## ---
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' 1
##
## Correlation of Fixed Effects:
```

Predicted values of meanTempBrood_all



Predicted values of meanTempBrood_all



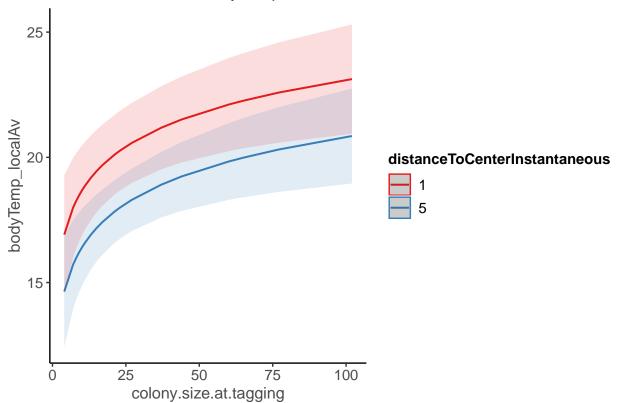
```
# Export panels
setwd(output_folder)
pdf("FigS5.pdf")
plot_model(brood.temp.model, type = "pred", terms = c("nestAirTemp",
    "neonic", "colony.size.at.tagging [4, 40]"), colors = list(ctrl_color,
    imid_color), axis.title = c("Nest Temperature (CO", "Spatial Centrality (PC1)"),
    show.legend = F, title = "Brood temperature during cold exposure")
dev.off()
## pdf
##
pdf("FigS6.pdf")
plot_model(brood.temp.recovery.model, type = "pred", terms = c("tempRampState",
    "neonic", "colony.size.at.tagging [4, 40]"), colors = list(ctrl_color,
    imid_color), axis.title = c("Observation Period", "Spatial Centrality (PC1)"),
    show.legend = F, title = "Brood temperature during recovery")
dev.off()
## pdf
```

##

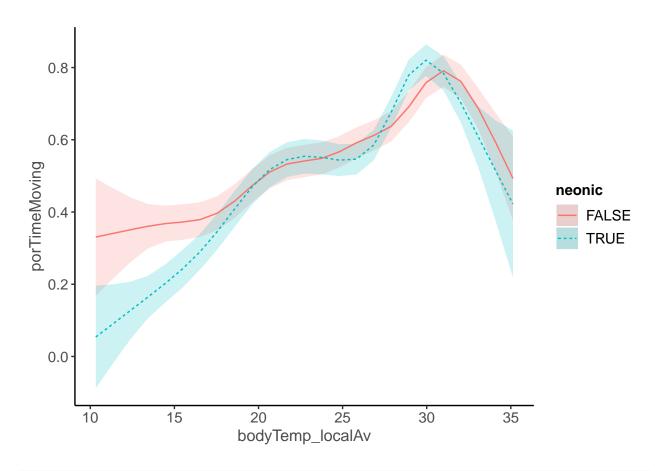
2

Proximate behavioral mechanisms

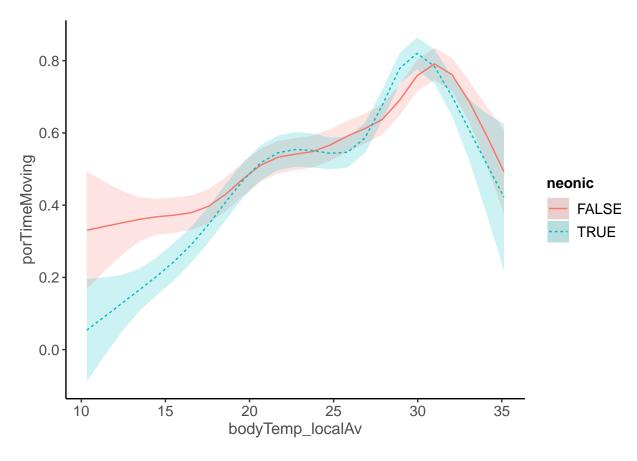
Predicted values of bodyTemp_localAv



```
# Overall activity by
activity.body.temp.model <- bam(porTimeMoving ~ s(colony.id,
    bs = "re") + s(uniqueID, bs = "re") + s(bodyTemp_localAv,
    by = neonic), data = subset(behData, bodyTemp_localAv > 10 &
    rampState > 0))
plot_smooths(activity.body.temp.model, bodyTemp_localAv, neonic)
```



```
pc1.body.temp.model <- bam(PC1 ~ s(colony.id, bs = "re") + s(uniqueID,
    bs = "re") + s(bodyTemp_localAv, by = neonic), data = subset(behData,
    bodyTemp_localAv > 10 & rampState > 0))
plot_smooths(activity.body.temp.model, bodyTemp_localAv, neonic)
```



```
# Activity state switching vs body temperature
pai.body.temp.model <- bam(Pai ~ s(colony.id, bs = "re") + s(uniqueID,
    bs = "re") + s(bodyTemp_localAv, by = neonic), data = subset(behData,
    bodyTemp_localAv > 10 & rampState %in% c(1, 2, 3)))
plot_smooths(pai.body.temp.model, bodyTemp_localAv, neonic)
```

```
0.4

0.2

0.2

neonic
FALSE
TRUE

TRUE
```

```
### Figure 3
pdf("Fig4_a.pdf")
# plot_smooths(body.temp.by.dist.model,
# distanceToCenterInstantaneous, small.colony)
plot_model(temp.model, type = "pred", terms = c("colony.size.at.tagging",
    "distanceToCenterInstantaneous[1,5]"))
dev.off()
## pdf
##
pdf("Fig4_b.pdf")
plot_smooths(pai.body.temp.model, bodyTemp_localAv, neonic)
dev.off()
## pdf
##
pdf("Fig4_c.pdf")
plot_smooths(activity.body.temp.model, bodyTemp_localAv, neonic)
dev.off()
```

pdf ## 2

```
# Colony size vs. activity
colony.sizes <- aggregate(behData$colony.size.at.tagging, list(behData$colony.id),</pre>
   FUN = mean)[, 2]
mean.activity <- aggregate(behData$porTimeMoving, list(behData$colony.id),</pre>
   FUN = mean)[, 2]
activity.size.model <- gam(mean.activity ~ s(colony.sizes))</pre>
summary(activity.size.model)
##
## Family: gaussian
## Link function: identity
##
## Formula:
## mean.activity ~ s(colony.sizes)
## Parametric coefficients:
              Estimate Std. Error t value Pr(>|t|)
## (Intercept) 0.70759
                          0.01701 41.59 <2e-16 ***
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' ' 1
## Approximate significance of smooth terms:
##
                   edf Ref.df
                                  F p-value
## s(colony.sizes)
                   1
                           1 4.744 0.0354 *
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## R-sq.(adj) = 0.0837 Deviance explained = 10.6%
## GCV = 0.012762 Scale est. = 0.012154 n = 42
# Colony size vs. interaction rate
colony.sizes <- aggregate(behData$colony.size.at.tagging, list(behData$colony.id),</pre>
   FUN = mean)[, 2]
interaction.rate <- aggregate(behData$meanInteractionRate * behData$colony.size.at.tagging,
   list(behData$colony.id), FUN = mean)[, 2]
interaction.size.model <- gam(interaction.rate ~ s(colony.sizes))</pre>
summary(interaction.size.model)
##
## Family: gaussian
## Link function: identity
## Formula:
## interaction.rate ~ s(colony.sizes)
## Parametric coefficients:
              Estimate Std. Error t value Pr(>|t|)
## (Intercept) 0.46364
                          0.02902 15.98 <2e-16 ***
## ---
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' 1
## Approximate significance of smooth terms:
##
                   edf Ref.df
                                  F p-value
```

```
## s(colony.sizes)
                           1 79.16 <2e-16 ***
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
## R-sq.(adj) = 0.656
                        Deviance explained = 66.4%
## GCV = 0.037141 Scale est. = 0.035373 n = 42
setwd(output_folder)
png("FigS8.png", width = 640, height = 300)
a <- plot_smooths(activity.size.model, colony.sizes) + xlab("Colony Size") +
   ylab("Mean Activity")
b <- plot_smooths(interaction.size.model, colony.sizes) + xlab("Colony Size") +
   ylab("Interaction Rate (interactions/frame)")
grid.arrange(a, b, nrow = 1)
dev.off()
## pdf
##
    2
```

Microcolony size-manipulation experiments

Load and clean data

```
## 1 0
## 2 1
                          6.518312
                                                          0
                                                                         8.073369
## 3 2
                          1.622784
                                                          0
                                                                         3.783518
                                                         73
## 4 3
                          0.817224
                                                                         4.001826
## 5 4
                          1.681611
                                                          0
                                                                         4.743056
## 6 5
                          6.861974
                                                          0
                                                                         6.188042
     total_social_interactions countable_interaction_frames activity
## 1
                             10
                                                            22
## 2
                                                                       0
                              0
                                                            18
                                                                       0
## 3
                             67
                                                            153
                              67
                                                            170
                                                                       0
## 4
                              0
                                                                       0
## 5
                                                            44
## 6
                              0
     speed_when_moving distance_to_social_center frames_tracked tag_ids
## 1
                                         0.5888912
                                                                  5
                     NΑ
## 2
                     NA
                                         7.6145221
                                                                  6
                                                                         17
## 3
                     NA
                                                                 62
                                                                         22
                                         0.5811764
## 4
                     NA
                                         0.4717314
                                                                 73
                                                                         33
## 5
                     NA
                                         3.3990098
                                                                 14
                                                                         36
```

```
## 6
                    NA
                                        5.5870597
                                                                        37
##
                                video file block colony
                                                                   timestamp
## 1 microcol 1-2021-09-30 15-42-01.mjpeg
                                              1 1 2021-09-30 15-42-01
## 2 microcol_1-2021-09-30_15-42-01.mjpeg
                                                       1 2021-09-30_15-42-01
                                                1
                                                    1 2021-09-30_15-42-01
1 2021-09-30_15-42-01
## 3 microcol_1-2021-09-30_15-42-01.mjpeg
                                               1
## 4 microcol 1-2021-09-30 15-42-01.mjpeg
                                              1
## 5 microcol 1-2021-09-30 15-42-01.mjpeg
                                                     1 2021-09-30 15-42-01
                                               1
                                               1
## 6 microcol 1-2021-09-30 15-42-01.mjpeg
                                                     1 2021-09-30 15-42-01
##
     time num nest temp
## 1 272.6542
                16.583
## 2 272.6542
                 16.583
## 3 272.6542
               16.583
               16.583
## 4 272.6542
## 5 272.6542 16.583
## 6 272.6542
               16.583
### Add metadata to data
for (i in 1:length(metadata[, 1])) {
    idx = mc_data$block == metadata$block[i] & mc_data$colony ==
        metadata$colony_number[i]
    mc_data$treatment[idx] <- metadata$treatment[i]</pre>
    mc_data$colony_size[idx] <- metadata$colony_size[i]</pre>
    mc_data$source_colony[idx] <- metadata$source_colony[i]</pre>
}
# Convert to factors
mc data$block <- as.factor(mc data$block)</pre>
mc_data$colony_size <- as.factor((mc_data$colony_size))</pre>
mc_data$source_colony <- as.factor(mc_data$source_colony)</pre>
mc_data$unique_colony <- paste(mc_data$block, mc_data$colony,</pre>
    sep = "_")
mc_data$tag_ids <- as.factor(mc_data$tag_ids)</pre>
mc_data$unique.id <- paste(mc_data$tag_ids, mc_data$unique_colony,</pre>
    sep = " ")
mc_data$unique.id <- as.factor(mc_data$unique.id)</pre>
### Clean up mc_data Clear out faulty temperature readings
mc_data$nest_temp[mc_data$nest_temp < 5 | mc_data$nest_temp >
    35] <- NaN
# Log transform speed
mc_data$speed_when_moving <- log10(mc_data$speed_when_moving)</pre>
# Square root transform brood interactions
mc_data$num_brood_interactions <- sqrt(mc_data$num_brood_interactions)</pre>
# hist(mc_data$mean_dist_other_bees)
# hist(mc_data$mean_min_distance_to_brood, breaks = 100)
# hist(mc_data$distance_to_social_center)
# hist(mc_data$num_brood_interactions^(1/2))
# hist(mc_data$activity)
# hist(log10(1+mc_data$total_social_interactions))
# hist(log10(mc_data$speed_when_moving))
# hist(mc_data$frames_tracked)
```

```
# Get tag lists from experiments
mc_taglist <- read.csv("/Users/jamescrall/Dropbox/Work/_Research/Bees/Bumblebees/socialScalingOfStresso
# Convert to factors
attach(mc_taglist)
valid.ids <- paste(tag.ID, block, colony, sep = "_")</pre>
detach(mc_taglist)
mc_data <- subset(mc_data, unique.id %in% valid.ids & frames_tracked >
dim(mc_data) #Size of data -> # of unique observations
## [1] 106164
                  22
length(unique(mc_data$unique.id)) #unique bees
## [1] 237
# Remove two sets of erroneous data
rm.ind.1 <- mc_data$unique_colony == "1_12" & mc_data$time_num <
    270 #Some erroneous timestamps
rm.ind.2 <- mc_data$unique_colony == "2_3" & mc_data$time_num >
    280 #Period of artifically high readings, removing
rm.ind <- rm.ind.1 | rm.ind.2
mc_data <- mc_data[!rm.ind, ]</pre>
# Remove some abnormally high temp readings (a few hundred)
# mc_data <- subset(mc_data, nest_temp < 28) Separate pre</pre>
# and post
cols <- unique(mc_data$unique_colony)</pre>
rampThresh <- 20
define_ramp_states <- function(sd, plt = FALSE) {</pre>
    minT <- min(sd$nest_temp, na.rm = TRUE)</pre>
    minT_time <- sd$time_num[sd$nest_temp == minT][1]</pre>
    sd$rampState[sd$nest_temp > 20 & sd$time_num < minT_time] <- "pre"</pre>
    sd$rampState[sd$nest_temp > 20 & sd$time_num > minT_time] <- "post"
    sd$rampState[sd$nest_temp <= 20] <- "ramp"</pre>
    sd$cl[sd$ramp == "pre"] <- "coral1"</pre>
    sd$cl[sd$ramp == "ramp"] <- "cadetblue"</pre>
    sd$cl[sd$ramp == "post"] <- "coral4"</pre>
    plt_data <- subset(sd, rampState %in% c("pre", "ramp", "post"))</pre>
    if (plt == TRUE) {
        plot(nest_temp ~ time_num, data = plt_data, col = plt_data$cl,
            pch = 19, cex = 0.5)
        title(unique(sd$unique_colony))
    }
    return(sd$rampState)
}
for (i in 1:length(cols)) {
```

```
cur.ind <- mc_data$unique_colony == cols[i]
  sd <- mc_data[cur.ind, ]
  rampState <- define_ramp_states(sd)
  mc_data$rampState[cur.ind] <- rampState
}

# mc_data <- subset(mc_data, rampState %in% c('pre',
# 'ramp'))</pre>
```

PCA analysis

```
spat.vars <- c("mean_dist_other_bees", "mean_min_distance_to_brood",
    "distance_to_social_center", "total_social_interactions",
    "num_brood_interactions")

pc.dat <- mc_data[, spat.vars]
cmplt_ind <- complete.cases(pc.dat)
pc.dat <- pc.dat[cmplt_ind, ]
mc_data <- mc_data[cmplt_ind, ]
mc_nest.beh.pca <- prcomp(pc.dat, scale = TRUE, center = TRUE)
mc_data$pc1 <- -mc.nest.beh.pca$x[, 1] #Inverse sign for consistency with other analysis, so higher pc
head(mc_data)</pre>
## X mean_min_distance_to_brood num_brood_interactions mean_dist_other_bees
```

```
## 3
                           1.6227837
                                                    0.000000
       2
                                                                          3.783518
## 4
       3
                           0.8172240
                                                    8.544004
                                                                          4.001826
## 8
       7
                           2.7178458
                                                    0.000000
                                                                          5.169691
## 11 10
                           0.9112662
                                                    8.774964
                                                                          4.162098
## 12 11
                                                    8.831761
                           1.1629358
                                                                          4.142163
## 14 13
                           3.9095601
                                                    0.000000
                                                                          5.309894
##
      total_social_interactions countable_interaction_frames activity
## 3
                              67
                                                           153 0.0000000
## 4
                              67
                                                           170 0.0000000
## 8
                               0
                                                           170 0.0000000
## 11
                              17
                                                           108 0.8409091
                              20
## 12
                                                           140 0.7391304
## 14
                               0
                                                           140 0.0000000
##
      speed_when_moving distance_to_social_center frames_tracked tag_ids
## 3
                     NA
                                          0.5811764
                                                                 62
                                                                         22
## 4
                     NA
                                         0.4717314
                                                                 73
                                                                         33
## 8
                     NA
                                          1.9373203
                                                                 73
                                                                         47
## 11
             -0.2440059
                                          2.2086506
                                                                 47
                                                                         22
## 12
             -0.6191297
                                          0.7177818
                                                                 72
                                                                         33
## 14
                                         2.1745005
                                                                 74
                                                                         47
                     NΑ
##
                                 video file block colony
                                                                     timestamp
                                                        1 2021-09-30_15-42-01
## 3 microcol_1-2021-09-30_15-42-01.mjpeg
                                                 1
## 4 microcol_1-2021-09-30_15-42-01.mjpeg
                                                 1
                                                        1 2021-09-30 15-42-01
## 8 microcol_1-2021-09-30_15-42-01.mjpeg
                                                 1
                                                        1 2021-09-30_15-42-01
## 11 microcol_1-2021-09-30_12-42-01.mjpeg
                                                        1 2021-09-30 12-42-01
                                                 1
## 12 microcol_1-2021-09-30_12-42-01.mjpeg
                                                        1 2021-09-30_12-42-01
                                                 1
```

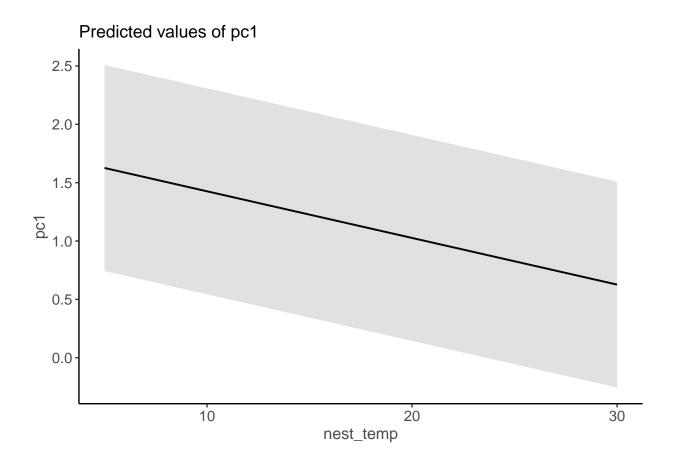
```
## 14 microcol_1-2021-09-30_12-42-01.mjpeg
                                              1 1 2021-09-30 12-42-01
##
      time_num nest_temp treatment colony_size source_colony unique_colony
## 3 272.6542
                 16.583
                           neonic
                                            4
                                                           3
                                                                       1 1
## 4 272.6542
                 16.583
                                                           3
                                                                       1_1
                           neonic
                                             4
## 8 272.6542
                 16.583
                           neonic
                                             4
                                                           3
                                                                       1_1
## 11 272.5292
                                             4
                                                           3
                 22.529
                           neonic
                                                                       1 1
## 12 272.5292
                 22.529
                                                           3
                           neonic
                                             4
                                                                       1 1
## 14 272.5292
                 22.529
                            neonic
                                             4
                                                           3
                                                                       1 1
##
      unique.id rampState
                                pc1
## 3
        22_1_1
                    ramp 2.0437801
## 4
        33_1_1
                    ramp 2.8073377
        47_1_1
## 8
                    ramp 0.7142319
## 11
        22_1_1
                    pre 1.9910808
                     pre 2.2503496
## 12
        33_1_1
## 14
        47_1_1
                     pre 0.4409615
# plot(pc1~nest_temp, data = mc_data, pch = 19, cex = 0.1,
# col=rgb(0.6,0.2, 0.3, 0.1))
##Models Statistical models
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
     method [lmerModLmerTest]
## Formula: pc1 ~ nest_temp + (1 | block/colony/tag_ids) + (1 | source_colony)
      Data: subset(mc_data, treatment == "control")
##
##
##
                 BIC
                      logLik deviance df.resid
        ATC
## 164423.5 164485.1 -82204.8 164409.5
##
## Scaled residuals:
               1Q Median
##
      Min
                                3Q
                                       Max
## -4.3389 -0.6082 0.0115 0.6053 4.3088
##
## Random effects:
## Groups
                           Name
                                       Variance Std.Dev.
## tag_ids:(colony:block) (Intercept) 1.0101
                                                1.0050
## colony:block
                           (Intercept) 0.1925
                                                0.4388
## block
                           (Intercept) 3.7515
                                                1.9369
## source_colony
                           (Intercept) 1.2065
                                                1.0984
## Residual
                                       1.6743
                                                1.2939
## Number of obs: 48836, groups:
## tag_ids:(colony:block), 115; colony:block, 15; block, 3; source_colony, 3
##
## Fixed effects:
                 Estimate Std. Error
                                             df t value Pr(>|t|)
## (Intercept) 1.826e+00 1.308e+00 3.579e+00
                                                  1.395
                                                           0.243
## nest_temp
             -3.998e-02 1.350e-03 4.874e+04 -29.608
                                                          <2e-16 ***
## ---
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' 1
## Correlation of Fixed Effects:
##
             (Intr)
```

nest_temp -0.020

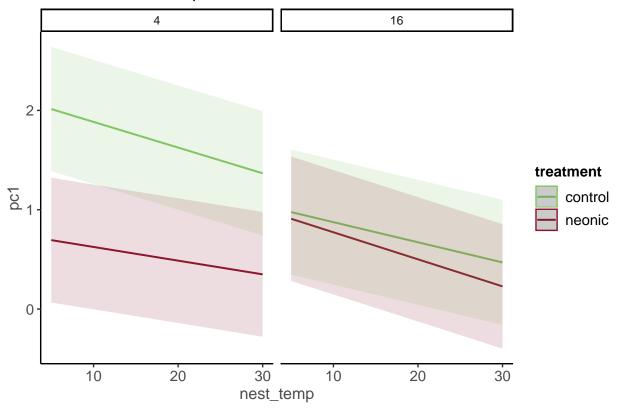
```
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
    method [lmerModLmerTest]
## Formula: pc1 ~ nest temp * treatment * colony size + (1 | block) + (1 |
       colony/unique.id) + (1 | source_colony) + (1 | rampState)
##
      Data: subset(mc_data, rampState %in% c("pre", "ramp"))
##
                      logLik deviance df.resid
                BIC
## 127522.8 127643.6 -63747.4 127494.8
##
## Scaled residuals:
      Min
               1Q Median
                                3Q
                                      Max
## -5.1534 -0.5489 -0.0097 0.5379 4.9599
## Random effects:
## Groups
                                Variance Std.Dev.
                    Name
## unique.id:colony (Intercept) 1.19429 1.0928
## colony
                    (Intercept) 0.16517 0.4064
## source_colony
                     (Intercept) 0.57625 0.7591
## block
                     (Intercept) 1.77078 1.3307
## rampState
                     (Intercept) 0.01119 0.1058
## Residual
                                 1.24396 1.1153
## Number of obs: 41316, groups:
## unique.id:colony, 235; colony, 13; source_colony, 3; block, 3; rampState, 2
## Fixed effects:
                                            Estimate Std. Error
## (Intercept)
                                            2.143e+00 9.285e-01 4.133e+00
## nest_temp
                                           -2.587e-02 3.509e-03 4.481e+03
## treatmentneonic
                                          -1.380e+00 2.777e-01 2.335e+02
                                          -1.065e+00 3.077e-01 1.154e+02
## colony_size16
## nest_temp:treatmentneonic
                                           1.209e-02 3.624e-03 4.112e+04
## nest_temp:colony_size16
                                           5.604e-03 3.262e-03 4.111e+04
## treatmentneonic:colony_size16
                                           1.348e+00 3.825e-01 1.403e+02
## nest_temp:treatmentneonic:colony_size16 -1.905e-02 4.470e-03 4.112e+04
                                          t value Pr(>|t|)
## (Intercept)
                                            2.308 0.080087
## nest temp
                                            -7.372 1.99e-13 ***
## treatmentneonic
                                            -4.969 1.30e-06 ***
## colony size16
                                            -3.462 0.000752 ***
                                            3.335 0.000854 ***
## nest_temp:treatmentneonic
## nest_temp:colony_size16
                                            1.718 0.085771 .
## treatmentneonic:colony_size16
                                            3.523 0.000576 ***
## nest_temp:treatmentneonic:colony_size16 -4.262 2.03e-05 ***
## ---
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' ' 1
##
## Correlation of Fixed Effects:
##
               (Intr) nst_tm trtmnt cln_16 nst_t: n_:_16 tr:_16
## nest_temp
              -0.065
## treatmntnnc -0.136 0.128
## colony_sz16 -0.150 0.118 0.533
## nst_tmp:trt 0.036 -0.567 -0.221 -0.109
## nst_tmp:_16  0.040 -0.636 -0.134 -0.180  0.600
## trtmntn:_16  0.108 -0.094 -0.722 -0.742  0.161  0.144
```

```
## boundary (singular) fit: see help('isSingular')
## Warning: Model failed to converge with 1 negative eigenvalue: -3.4e+00
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
    method [lmerModLmerTest]
## Formula: pc1 ~ treatment * nest_temp + (1 | block) + (1 | colony/tag_ids) +
      (1 | source_colony)
     Data: subset(mc_data, colony_size == "16" & rampState %in% c("pre",
##
##
      "ramp"))
##
##
       AIC
                BIC logLik deviance df.resid
##
   88857.3 88931.2 -44419.7 88839.3
                                        26961
##
## Scaled residuals:
##
      Min
               1Q Median
                              30
                                     Max
## -4.8269 -0.5958 -0.0280 0.5949
                                  4.4549
##
## Random effects:
## Groups
                             Variance Std.Dev.
                  Name
## tag_ids:colony (Intercept) 1.231e+00 1.1094960
## colony
                  (Intercept) 2.128e+00 1.4587787
## block
                  (Intercept) 4.523e-01 0.6725650
## source_colony (Intercept) 1.293e-08 0.0001137
## Residual
                              1.536e+00 1.2394476
## Number of obs: 26970, groups:
## tag_ids:colony, 139; colony, 7; block, 3; source_colony, 2
##
## Fixed effects:
##
                              Estimate Std. Error
                                                         df t value Pr(>|t|)
                             1.570e+00 6.835e-01 8.767e+00
                                                              2.298
                                                                     0.0479 *
## (Intercept)
## treatmentneonic
                            -1.761e+00 9.689e-02 2.359e+04 -18.174
                                                                     <2e-16 ***
                            -3.349e-02 2.088e-03 2.685e+04 -16.034
## nest temp
                                                                     <2e-16 ***
## treatmentneonic:nest_temp -5.245e-03 2.904e-03 2.686e+04 -1.806
                                                                     0.0709 .
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Correlation of Fixed Effects:
##
              (Intr) trtmnt nst tm
## treatmntnnc -0.067
## nest_temp
             -0.052 0.378
## trtmntnnc:_ 0.038 -0.531 -0.719
## optimizer (nloptwrap) convergence code: 0 (OK)
## boundary (singular) fit: see help('isSingular')
## Linear mixed model fit by maximum likelihood . t-tests use Satterthwaite's
    method [lmerModLmerTest]
## Formula: pc1 ~ treatment * nest_temp + (1 | block) + (1 | colony/tag_ids) +
##
       (1 | source_colony)
##
     Data: subset(mc_data, colony_size == "4" & rampState %in% c("pre",
##
      "ramp"))
```

```
##
##
       AIC
                BIC logLik deviance df.resid
   39842.9 39911.1 -19912.5 39824.9
##
##
## Scaled residuals:
##
      Min 1Q Median
                               3Q
                                     Max
## -5.5019 -0.5376 0.0238 0.5764 4.6610
##
## Random effects:
## Groups
                              Variance Std.Dev.
                  Name
## tag_ids:colony (Intercept) 0.5426
                                     0.7366
                  (Intercept) 0.4876
                                      0.6983
## colony
## source_colony (Intercept) 2.2853
                                      1.5117
## block
                  (Intercept) 5.5371
                                      2.3531
## Residual
                              0.9133
                                      0.9557
## Number of obs: 14346, groups:
## tag_ids:colony, 79; colony, 13; source_colony, 3; block, 3
##
## Fixed effects:
                              Estimate Std. Error
                                                         df t value Pr(>|t|)
##
## (Intercept)
                            2.418e+00 1.640e+00 4.664e+00
                                                             1.474
                                                                       0.205
## treatmentneonic
                            -1.244e+00 1.886e-01 9.467e+01 -6.596 2.40e-09 ***
                            -3.855e-02 2.281e-03 1.428e+04 -16.906 < 2e-16 ***
## nest_temp
## treatmentneonic:nest_temp 1.248e-02 3.105e-03 1.428e+04
                                                             4.020 5.85e-05 ***
## ---
## Signif. codes: 0 '*** 0.001 '** 0.01 '* 0.05 '.' 0.1 ' ' 1
## Correlation of Fixed Effects:
##
              (Intr) trtmnt nst_tm
## treatmntnnc -0.074
## nest_temp -0.023 0.209
## trtmntnnc:_ 0.017 -0.280 -0.735
## $nest_temp
```



Predicted values of pc1



```
## pdf
## 2

## $nest_temp

## pdf
## 2
```

Colony demography

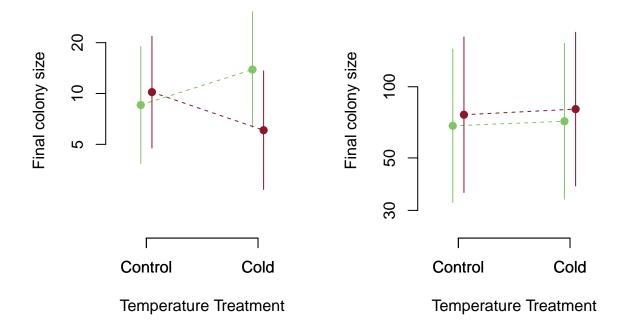
Load and clean data

Growth models

```
gen.model <- glmer(total ~ log10(colony.size.at.tagging) * neonic *
   cold + growth.length + (1 | experimentalBlock) + (1 | Species),
   data = subset(demog.data, colony.size.at.tagging > 2 & alive.at.tagging ==
        1 & Species != "perplexus"), family = "poisson")
```

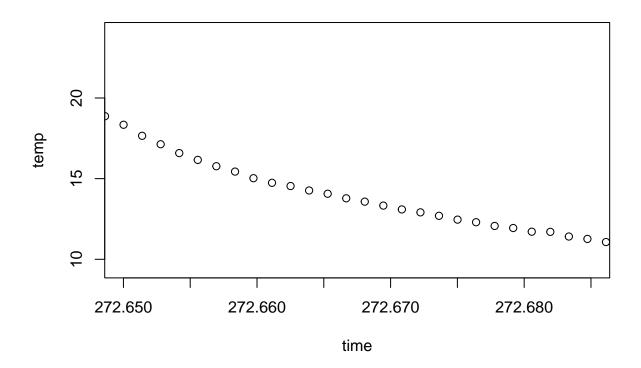
```
## Warning in checkConv(attr(opt, "derivs"), opt$par, ctrl = control$checkConv, :
## Model failed to converge with max|grad| = 0.00922731 (tol = 0.002, component 1)
```

```
gen.model.exclude.large.colonies <- glmer(total ~ log10(colony.size.at.tagging) *</pre>
   neonic * cold + growth.length + (1 | experimentalBlock) +
    (1 | Species), data = subset(demog.data, colony.size.at.tagging >
    2 & colony.size.at.tagging < 30 & alive.at.tagging == 1 &
   Species != "perplexus"), family = "poisson")
gr.model <- lmer(growth.rate ~ log10(colony.size.at.tagging) *</pre>
   neonic * cold + (1 | experimentalBlock) + (1 | Species),
   data = subset(demog.data, colony.size.at.tagging > 2 & colony.size.at.tagging <</pre>
        30 & alive.at.tagging == 1 & Species != "perplexus"))
# ineline plot
par(mfcol = c(1, 2))
out <- plot_model(gen.model.exclude.large.colonies, type = "pred",
   terms = c("cold", "neonic", "colony.size.at.tagging [4]"),
   transform = "exp")
custom_plot(out, c(0.2, 1.6), c(5, 10, 20))
## [1] 0.2 1.6
## [1] 5 10 20
## [1] 0.9319349 1.0082704 1.1418005 0.7827807
out <- plot_model(gen.model.exclude.large.colonies, type = "pred",</pre>
   terms = c("cold", "neonic", "colony.size.at.tagging [30]"),
   transform = "exp")
custom_plot(out, c(1.4, 2.4), c(30, 50, 100))
## [1] 1.4 2.4
## [1] 30 50 100
## [1] 1.834809 1.881678 1.854045 1.905451
```



```
# Figure 2: growth
setwd(output_folder)
pdf("Fig2.pdf", width = 4, height = 8)
par(mfcol = c(2, 1))
out <- plot_model(gen.model.exclude.large.colonies, type = "pred",</pre>
    terms = c("cold", "neonic", "colony.size.at.tagging [4]"),
    transform = "exp")
custom_plot(out, c(0.2, 1.6), c(5, 10, 20))
## [1] 0.2 1.6
## [1] 5 10 20
## [1] 0.9319349 1.0082704 1.1418005 0.7827807
out <- plot_model(gen.model.exclude.large.colonies, type = "pred",</pre>
    terms = c("cold", "neonic", "colony.size.at.tagging [30]"),
    transform = "exp")
custom_plot(out, c(1.4, 2.4), c(30, 50, 100))
## [1] 1.4 2.4
## [1] 30 50 100
## [1] 1.834809 1.881678 1.854045 1.905451
dev.off()
```

```
## pdf
## 2
```



[1] 0.14156

```
# Confirm achieved nest temperature in manipulations above
tmp <- subset(behData, cold == TRUE & rampState == 3) #Create a subset with data in cold-exposed colon
# Nest temperatures
nest.tmps <- aggregate(tmp$nestTemp, by = list(tmp$date), FUN = mean)
colnames(nest.tmps) <- c("timestamp", "nest_temp")
mean(nest.tmps$nest_temp)</pre>
```

[1] 14.52631

```
median(nest.tmps$nest_temp)
```

[1] 14.3125

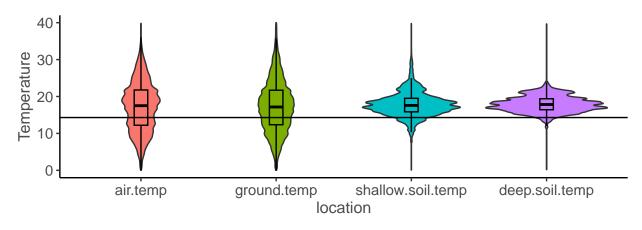
```
# Chamber temps
ch.tmps <- aggregate(tmp$chamberTemp, by = list(tmp$date), FUN = mean)
colnames(ch.tmps) <- c("timestamp", "nest_temp")
mean(ch.tmps$nest_temp)</pre>
```

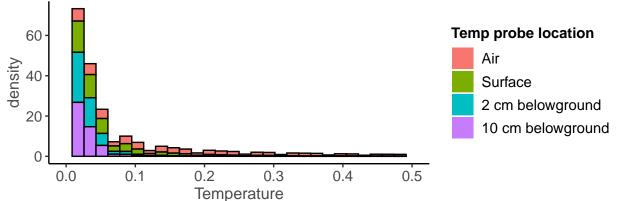
```
## [1] 10.14476
median(ch.tmps$nest_temp)
## [1] 9.75
setwd("/Users/jamescrall/Dropbox/Work/_Research/Bees/Bumblebees/socialScalingOfStressors/Data/AppletonT
filelist <- list.files(pattern = ".csv")</pre>
par(mfcol = c(4, 1))
for (i in 1:length(filelist)) {
    tempdata <- read.csv(filelist[i], skip = 1)</pre>
    tempdata <- tempdata[, 1:6]
    colnames(tempdata) <- c("Num", "Time", "air.temp", "ground.temp",</pre>
        "shallow.soil.temp", "deep.soil.temp")
    tempdata$air.temp.diff <- NaN</pre>
    tempdata$air.temp.diff[2:length(tempdata[, 1])] <- diff(tempdata$air.temp)</pre>
    tempdata$ground.temp.diff <- NaN</pre>
    tempdata$ground.temp.diff[2:length(tempdata[, 1])] <- diff(tempdata$ground.temp)</pre>
    tempdata$shallow.soil.temp.diff <- NaN
    tempdata$shallow.soil.temp.diff[2:length(tempdata[, 1])] <- diff(tempdata$shallow.soil.temp)
    tempdata$deep.soil.temp.diff <- NaN</pre>
    tempdata$deep.soil.temp.diff[2:length(tempdata[, 1])] <- diff(tempdata$deep.soil.temp)</pre>
    tempdata$unit <- i
    if (i == 1) {
        m.dat <- tempdata</pre>
    } else {
        m.dat <- rbind(m.dat, tempdata)</pre>
    }
}
temp.data <- gather(m.dat[, c("Time", "air.temp", "ground.temp",</pre>
    "shallow.soil.temp", "deep.soil.temp", "unit")], key = "location",
    value = "Temperature", air.temp, ground.temp, shallow.soil.temp,
    deep.soil.temp)
## Single colony
diff.data <- gather(m.dat[, c("Time", "air.temp.diff", "ground.temp.diff",</pre>
    "shallow.soil.temp.diff", "deep.soil.temp.diff", "unit")],
    key = "location", value = "Temperature", air.temp.diff, ground.temp.diff,
    shallow.soil.temp.diff, deep.soil.temp.diff)
# Clean out errant readings and nans
temp.data$Temperature[temp.data$Temperature < -10 | temp.data$Temperature >
diff.data$Temperature[diff.data$Temperature < -5 | diff.data$Temperature >
    5] <- NA
temp.data$location <- factor(temp.data$location, levels = c("air.temp",</pre>
```

"ground.temp", "shallow.soil.temp", "deep.soil.temp"))

diff.data\$location <- factor(diff.data\$location, levels = c("air.temp.diff",</pre> "ground.temp.diff", "shallow.soil.temp.diff", "deep.soil.temp.diff"))

```
diff.data$Temperature <- abs(diff.data$Temperature)</pre>
# hist(temp.data$Temperature) hist(diff.data$Temperature)
setwd(output_folder)
pdf("FigS7.pdf")
par(mfcol = c(2, 1))
p1 <- ggplot(temp.data, aes(x = location, y = Temperature, fill = location)) +
   geom violin() + geom boxplot(width = 0.1, outlier.shape = NA) +
   ylim(0, 40) + geom_hline(yintercept = 14.3) + scale_fill_discrete(labels = c("Air",
    "Surface", "2 cm belowground", "10 cm belowground"), name = "Temp probe location") +
    theme(legend.position = "none")
p2 \leftarrow ggplot(diff.data, aes(x = Temperature, fill = location)) +
    geom_histogram(aes(y = ..density..)) + xlim(0, 0.5) + scale_fill_discrete(labels = c("Air",
    "Surface", "2 cm belowground", "10 cm belowground"), name = "Temp probe location")
grid.arrange(p1, p2)
## Warning: Removed 12637 rows containing non-finite values ('stat_ydensity()').
## Warning: Removed 12637 rows containing non-finite values ('stat boxplot()').
## Warning: The dot-dot notation ('..density..') was deprecated in ggplot2 3.4.0.
## i Please use 'after_stat(density)' instead.
## 'stat_bin()' using 'bins = 30'. Pick better value with 'binwidth'.
## Warning: Removed 100721 rows containing non-finite values ('stat_bin()').
## Warning: Removed 8 rows containing missing values ('geom_bar()').
dev.off()
## pdf
# Plot for console
grid.arrange(p1, p2)
## Warning: Removed 12637 rows containing non-finite values ('stat_ydensity()').
## Warning: Removed 12637 rows containing non-finite values ('stat boxplot()').
## 'stat_bin()' using 'bins = 30'. Pick better value with 'binwidth'.
## Warning: Removed 100721 rows containing non-finite values ('stat_bin()').
## Warning: Removed 8 rows containing missing values ('geom_bar()').
```





```
sm <- diff.data %>%
    group_by(location) %>%
    summarize(quant50 = quantile(Temperature, probs = 0.5, na.rm = TRUE),
        quant95 = quantile(Temperature, probs = 0.95, na.rm = TRUE),
        quant99 = quantile(Temperature, probs = 0.99, na.rm = TRUE))
print(sm)
```

```
## # A tibble: 4 x 4
##
     location
                             quant50 quant95 quant99
##
     <fct>
                               <dbl>
                                       <dbl>
                                               <dbl>
                                               2.64
## 1 air.temp.diff
                              0.220
                                      1.58
## 2 ground.temp.diff
                              0.0400
                                               1.82
                                     0.460
## 3 shallow.soil.temp.diff
                             0.0200
                                     0.0800
                                               0.190
## 4 deep.soil.temp.diff
                              0.0200
                                     0.0700
                                               0.130
```

```
# Get range of dates
dates <- unique(parse_date_time(m.dat$Time, "mdy HMS"))
min(dates)</pre>
```

[1] "2019-08-08 05:54:02 UTC"

```
max(dates)
```

[1] "2019-10-02 12:59:35 UTC"

difftime(min(dates), max(dates))

Time difference of -55.29552 days