

Sleep Bout Analysis

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```
bouts <- read.csv("../data/WT_McKO_combined_bouts.csv",
  fileEncoding = 'UTF-8-BOM')
bouts <- bouts %>% mutate(Time = mdy_hms(Time),
  Sleep.Stage = as.factor(Sleep.Stage),
  Genotype = as.factor(Genotype),
  Transmitter = as.factor(Transmitter))

bouts <- bouts %>% mutate(Hour = hour(Time))
bouts <- bouts %>% mutate(ZT = Time + hours(18))
bouts <- bouts %>% mutate(ZT = hour(ZT))

bouts <- bouts %>% dplyr::mutate(phase = as.factor(ifelse(ZT>11, "dark", "light")))

head(bouts)
```

##	Bout	Epoch	Time	Sleep.Stage	Count	Duration	Genotype
## 1	1	2216	2019-02-10 18:00:00	S	36	360	McKO
## 2	2	2252	2019-02-10 18:06:00	W	2	20	McKO
## 3	3	2254	2019-02-10 18:06:20	S	6	60	McKO
## 4	4	2260	2019-02-10 18:07:20	W	4	40	McKO
## 5	5	2264	2019-02-10 18:08:00	S	4	40	McKO
## 6	6	2268	2019-02-10 18:08:40	W	17	170	McKO

##	Transmitter	Day	Hour	ZT	phase
## 1	988823	1	18	12	dark
## 2	988823	1	18	12	dark
## 3	988823	1	18	12	dark
## 4	988823	1	18	12	dark
## 5	988823	1	18	12	dark
## 6	988823	1	18	12	dark

```
SumStats_daily <- bouts %>% group_by(Genotype, Transmitter, Sleep.Stage, Day) %>%
  summarise(n_bouts = n(),
    total_duration = sum(Duration),
    min_bout_length = min(Duration),
    mean_bout_length = mean(Duration),
    max_bout_length = max(Duration),
    sd_bout_length = sd(Duration))
```

'summarise()' has grouped output by 'Genotype', 'Transmitter', 'Sleep.Stage'. You can override using

```
head(SumStats_daily)
```

```
## # A tibble: 6 x 10
## # Groups:   Genotype, Transmitter, Sleep.Stage [2]
##   Genotype Transmitter Sleep.Stage Day n_bouts total_duration min_bout_length
##   <fct>    <fct>      <fct>    <int> <int>         <int>         <int>
## 1 McKO     1050240      P         1     138          4760          10
## 2 McKO     1050240      P         2     183          5340          10
## 3 McKO     1050240      P         3     229          4910          10
## 4 McKO     1050240      S         1     237          45220         10
## 5 McKO     1050240      S         2     257          45180         10
## 6 McKO     1050240      S         3     284          46290         10
## # ... with 3 more variables: mean_bout_length <dbl>, max_bout_length <int>,
## #   sd_bout_length <dbl>
```

```
SumStats_daily_n <- SumStats_daily %>% group_by(Genotype, Transmitter, Sleep.Stage) %>%
  summarise(mean_n_bouts_day = mean(n_bouts))
```

'summarise()' has grouped output by 'Genotype', 'Transmitter'. You can override using the '.groups' argument

```
head(SumStats_daily_n)
```

```
## # A tibble: 6 x 4
## # Groups:   Genotype, Transmitter [2]
##   Genotype Transmitter Sleep.Stage mean_n_bouts_day
##   <fct>    <fct>      <fct>         <dbl>
## 1 McKO     1050240      P             183.
## 2 McKO     1050240      S             259.
## 3 McKO     1050240      W             246.
## 4 McKO     1050240      X              6
## 5 McKO     1050241      P             283.
## 6 McKO     1050241      S             234
```

```
SumStats_avg <- bouts %>% group_by(Genotype, Transmitter, Sleep.Stage) %>%
  summarise(min_bout_length = min(Duration),
            mean_bout_length = mean(Duration),
            max_bout_length = max(Duration),
            sd_bout_length = sd(Duration))
```

'summarise()' has grouped output by 'Genotype', 'Transmitter'. You can override using the '.groups' argument

```
head(SumStats_avg)
```

```
## # A tibble: 6 x 7
## # Groups:   Genotype, Transmitter [2]
##   Genotype Transmitter Sleep.Stage min_bout_length mean_bout_length
##   <fct>    <fct>      <fct>         <int>         <dbl>
## 1 McKO     1050240      P             10             27.3
## 2 McKO     1050240      S             10             176.
## 3 McKO     1050240      W             10             145.
```

```
## 4 McKO      1050240      X                10                10
## 5 McKO      1050241      P                10               28.6
## 6 McKO      1050241      S                10              171.
## # ... with 2 more variables: max_bout_length <int>, sd_bout_length <dbl>
```

```
SumStats_phase <- bouts %>% group_by(Genotype, Transmitter, Sleep.Stage, phase) %>%
  summarise(n_bouts = n(),
            total_duration = sum(Duration),
            min_bout_length = min(Duration),
            mean_bout_length = mean(Duration),
            max_bout_length = max(Duration),
            sd_bout_length = sd(Duration))
```

'summarise()' has grouped output by 'Genotype', 'Transmitter', 'Sleep.Stage'. You can override using

```
head(SumStats_phase)
```

```
## # A tibble: 6 x 10
## # Groups:   Genotype, Transmitter, Sleep.Stage [3]
##   Genotype Transmitter Sleep.Stage phase n_bouts total_duration min_bout_length
##   <fct>    <fct>        <fct>    <fct>   <int>         <int>          <int>
## 1 McKO      1050240      P        dark     200         4750           10
## 2 McKO      1050240      P        light    350        10260           10
## 3 McKO      1050240      S        dark     271        47400           10
## 4 McKO      1050240      S        light    507        89290           10
## 5 McKO      1050240      W        dark     325        77640           10
## 6 McKO      1050240      W        light    414        29720           10
## # ... with 3 more variables: mean_bout_length <dbl>, max_bout_length <int>,
## #   sd_bout_length <dbl>
```

```
SumStats_phase_n <- SumStats_phase %>% group_by(Genotype, Transmitter, Sleep.Stage, phase) %>%
  summarise(mean_n_bouts_day = mean(n_bouts))
```

'summarise()' has grouped output by 'Genotype', 'Transmitter', 'Sleep.Stage'. You can override using

```
head(SumStats_phase_n)
```

```
## # A tibble: 6 x 5
## # Groups:   Genotype, Transmitter, Sleep.Stage [3]
##   Genotype Transmitter Sleep.Stage phase mean_n_bouts_day
##   <fct>    <fct>        <fct>    <fct>         <dbl>
## 1 McKO      1050240      P        dark           200
## 2 McKO      1050240      P        light          350
## 3 McKO      1050240      S        dark           271
## 4 McKO      1050240      S        light          507
## 5 McKO      1050240      W        dark           325
## 6 McKO      1050240      W        light          414
```

```
SumStats_avg %>% group_by(Genotype, Sleep.Stage) %>%
  summarise(n = n(),
            mean_bout_length = mean(mean_bout_length),
            min_bout_length = min(min_bout_length),
            max_bout_length = max(max_bout_length))
```

```
## 'summarise()' has grouped output by 'Genotype'. You can override using the '.groups' argument.
```

```
## # A tibble: 8 x 6
## # Groups:   Genotype [2]
##   Genotype Sleep.Stage      n mean_bout_length min_bout_length max_bout_length
##   <fct>    <fct>    <int>          <dbl>          <int>          <int>
## 1 McKO     P           9           26.8            10           250
## 2 McKO     S           9          142.            10          3920
## 3 McKO     W           9          95.2            10          6310
## 4 McKO     X           9           10             10           10
## 5 WT       P          10          30.9            10           350
## 6 WT       S          10          127.            10          4390
## 7 WT       W          10          100.            10          5230
## 8 WT       X          10           10             10           10
```

```
SumStats_phase_grps <- SumStats_phase %>% group_by(Genotype, Sleep.Stage, phase) %>%
  summarise(n = n(),
            avg_bout_length = mean(mean_bout_length),
            min_bout_length = min(min_bout_length),
            max_bout_length = max(max_bout_length),
            sd_bout_length = sd(mean_bout_length))
```

```
## 'summarise()' has grouped output by 'Genotype', 'Sleep.Stage'. You can override using the '.groups' argument.
```

```
SumStats_phase_grps
```

```
## # A tibble: 16 x 8
## # Groups:   Genotype, Sleep.Stage [8]
##   Genotype Sleep.Stage phase      n avg_bout_length min_bout_length
##   <fct>    <fct>    <fct> <int>          <dbl>          <int>
## 1 McKO     P        dark      9           20.5            10
## 2 McKO     P        light      9           31.0            10
## 3 McKO     S        dark      9          107.            10
## 4 McKO     S        light      9          190.            10
## 5 McKO     W        dark      9          120.            10
## 6 McKO     W        light      9           67.9            10
## 7 McKO     X        dark      9           10             10
## 8 McKO     X        light      9           10             10
## 9 WT       P        dark     10           25.8            10
## 10 WT      P        light     10           36.7            10
## 11 WT      S        dark     10          96.2            10
## 12 WT      S        light     10          164.            10
## 13 WT      W        dark     10          118.            10
## 14 WT      W        light     10           78.5            10
## 15 WT      X        dark     10           10             10
## 16 WT      X        light     10           10             10
## # ... with 2 more variables: max_bout_length <int>, sd_bout_length <dbl>
```

```
SumStats_phase_n_grps <- SumStats_phase_n %>% group_by(Genotype, Sleep.Stage, phase) %>%
  summarise(n = n(),
            mean_n_bouts_day = mean(mean_n_bouts_day))
```

```
## 'summarise()' has grouped output by 'Genotype', 'Sleep.Stage'. You can override using the '.groups' :
```

```
head(SumStats_phase_n_grps)
```

```
## # A tibble: 6 x 5
## # Groups:   Genotype, Sleep.Stage [3]
##   Genotype Sleep.Stage phase      n mean_n_bouts_day
##   <fct>     <fct>     <fct> <int>          <dbl>
## 1 McKO      P         dark     9           378.
## 2 McKO      P         light     9           418.
## 3 McKO      S         dark     9           881.
## 4 McKO      S         light     9           779.
## 5 McKO      W         dark     9          1005.
## 6 McKO      W         light     9           673.
```

```
lm_n_bouts <- lmer(mean_n_bouts_day ~ Sleep.Stage * phase * Genotype + (1 | Transmitter), data=SumStats,
#plot(lm_n_bouts, type=c("p", "smooth"), col.line=1)
#lattice::qqmath(lm_n_bouts)
anova(lm_n_bouts)
```

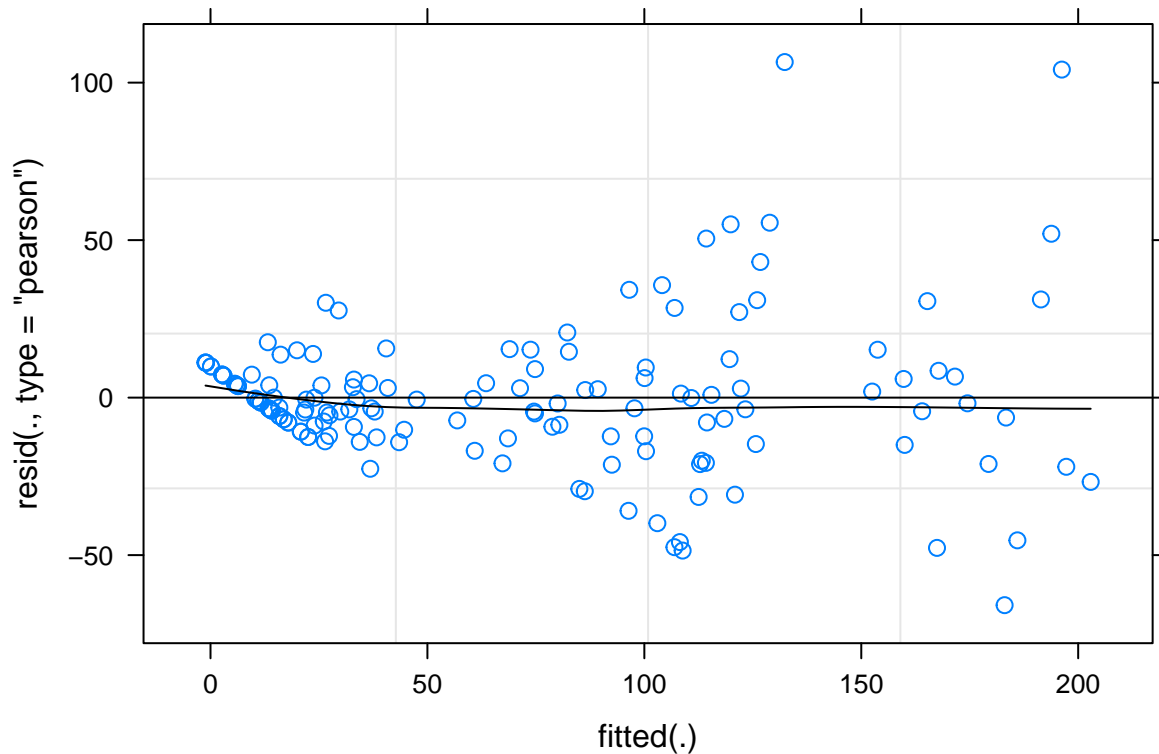
```
## Analysis of Variance Table
##               npar    Sum Sq Mean Sq F value
## Sleep.Stage      3 16786640 5595547 109.8216
## phase            1  453224   453224   8.8952
## Genotype          1     566     566   0.0111
## Sleep.Stage:phase  3  716742   238914   4.6891
## Sleep.Stage:Genotype 3    3057    1019   0.0200
## phase:Genotype     1    1379    1379   0.0271
## Sleep.Stage:phase:Genotype 3    5461    1820   0.0357
```

```
emmeans(lm_n_bouts, pairwise ~ Genotype | Sleep.Stage * phase)$contrasts
```

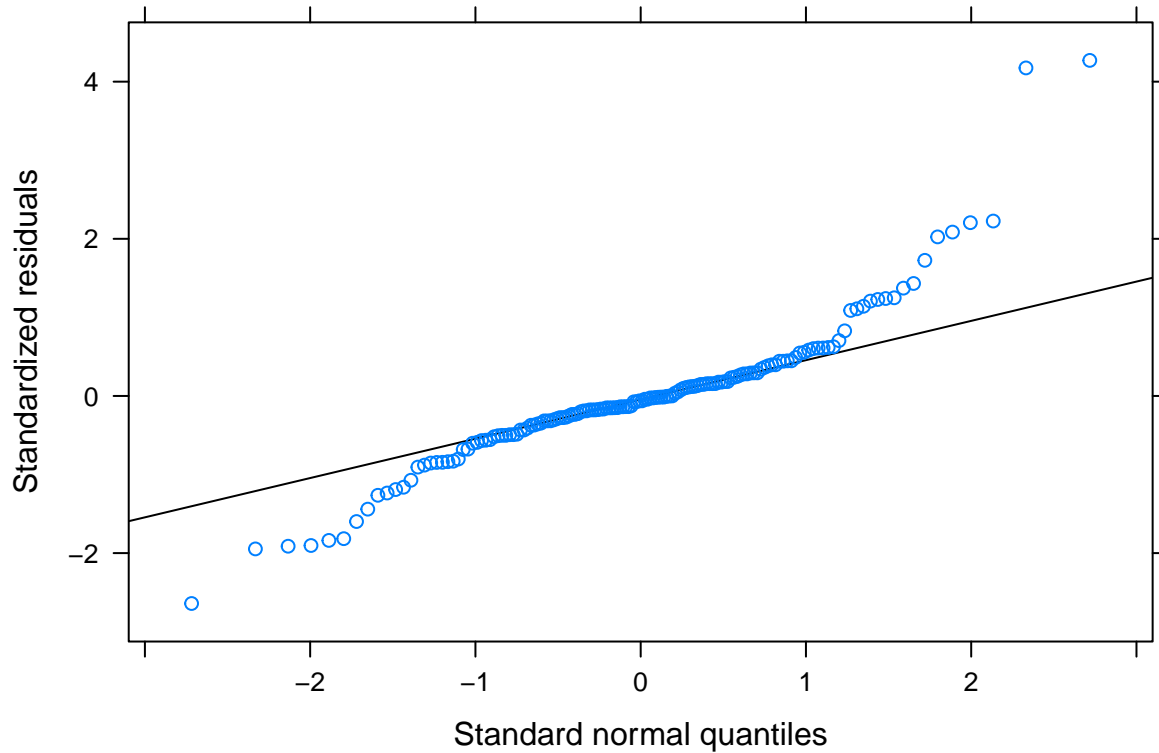
```
## Sleep.Stage = P, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT    -23.84 141 54.8 -0.169 0.8665
##
## Sleep.Stage = S, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT      8.97 141 54.8  0.064 0.9496
##
## Sleep.Stage = W, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT    -28.18 141 54.8 -0.200 0.8425
##
## Sleep.Stage = X, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT    -24.29 141 54.8 -0.172 0.8640
##
## Sleep.Stage = P, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT     20.09 141 54.8  0.142 0.8873
##
```

```
## Sleep.Stage = S, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT -14.33 141 54.8 -0.102 0.9195
##
## Sleep.Stage = W, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT -18.44 141 54.8 -0.131 0.8965
##
## Sleep.Stage = X, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT -6.40 141 54.8 -0.045 0.9640
##
## Degrees-of-freedom method: kenward-roger
```

```
lm_mean_length <- lmer(mean_bout_length ~ Sleep.Stage * phase * Genotype + (1 | Transmitter), data=SumS
plot(lm_mean_length, type=c("p","smooth"), col.line=1)
```



```
lattice::qqmath(lm_mean_length)
```



```
anova(lm_mean_length)
```

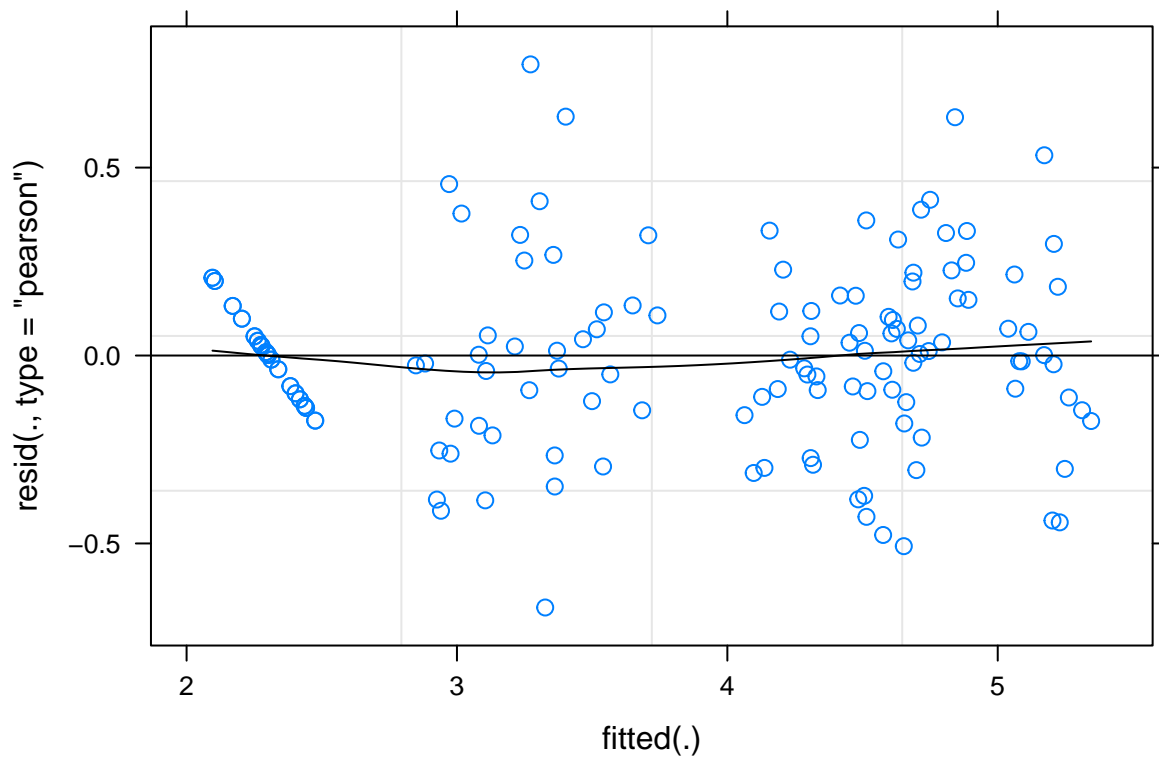
```
## Analysis of Variance Table
##               npar Sum Sq Mean Sq  F value
## Sleep.Stage      3 407882   135961  218.4922
## phase            1   3817     3817    6.1332
## Genotype          1     85       85    0.1372
## Sleep.Stage:phase  3  70113    23371   37.5577
## Sleep.Stage:Genotype 3   3678     1226    1.9700
## phase:Genotype     1      4        4    0.0065
## Sleep.Stage:phase:Genotype 3    923     308    0.4945
```

```
emmeans(lm_mean_length, pairwise ~ Genotype | Sleep.Stage * phase)$contrasts
```

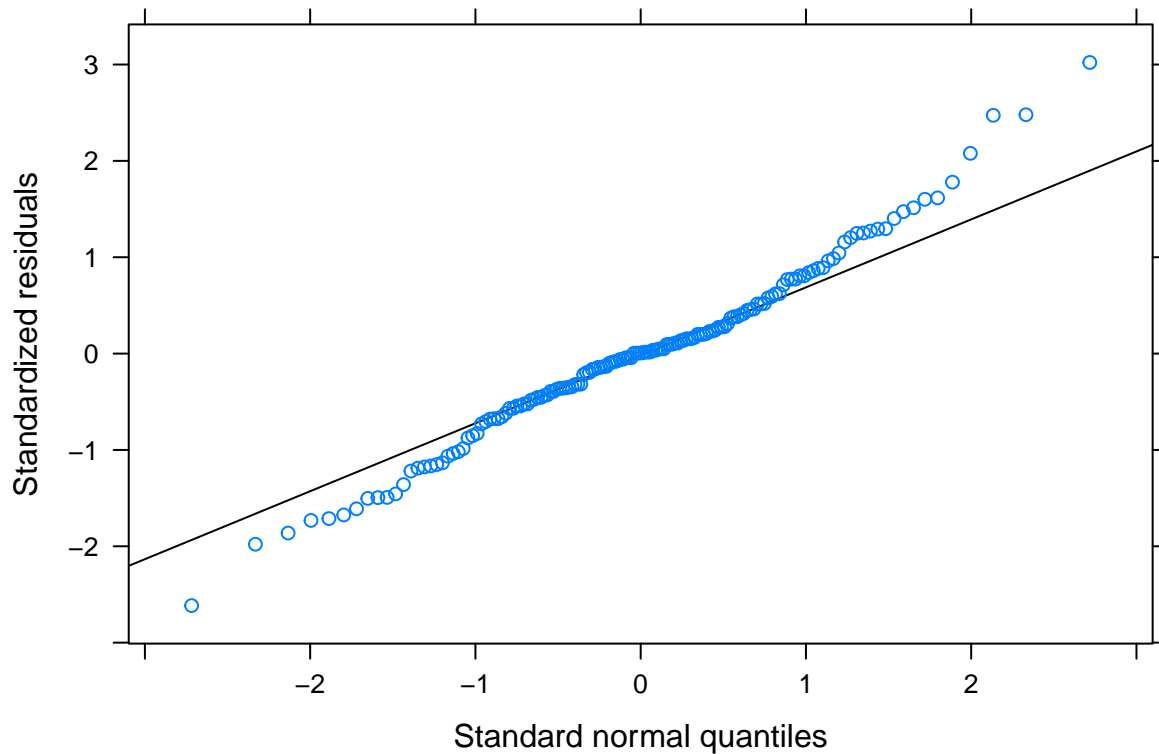
```
## Sleep.Stage = P, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT    -5.21 12.3 120 -0.422 0.6739
##
## Sleep.Stage = S, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT     11.22 12.3 120  0.908 0.3655
##
## Sleep.Stage = W, phase = dark:
## contrast estimate SE df t.ratio p.value
```

```
## McKO - WT      1.76 12.3 120   0.143  0.8869
##
## Sleep.Stage = X, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT      0.00 12.3 120   0.000  1.0000
##
## Sleep.Stage = P, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT     -5.76 12.3 120  -0.466  0.6420
##
## Sleep.Stage = S, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT     26.70 12.3 120   2.162  0.0326
##
## Sleep.Stage = W, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT    -10.55 12.3 120  -0.854  0.3947
##
## Sleep.Stage = X, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT      0.00 12.3 120   0.000  1.0000
##
## Degrees-of-freedom method: kenward-roger
```

```
lm_mean_length <- lmer(log(mean_bout_length) ~ Sleep.Stage * phase * Genotype + (1 | Transmitter), data=
plot(lm_mean_length, type=c("p","smooth"), col.line=1)
```




```
lattice::qqmath(lm_mean_length)
```



```
anova(lm_mean_length)
```

```
## Analysis of Variance Table
##               npar  Sum Sq Mean Sq  F value
## Sleep.Stage      3 155.645   51.882  789.4177
## phase            1   0.732    0.732   11.1352
## Genotype          1   0.014    0.014    0.2157
## Sleep.Stage:phase  3   5.978    1.993   30.3207
## Sleep.Stage:Genotype 3   0.420    0.140    2.1315
## phase:Genotype     1   0.016    0.016    0.2452
## Sleep.Stage:phase:Genotype 3   0.034    0.011    0.1736
```

```
emmeans(lm_mean_length, pairwise ~ Genotype | Sleep.Stage * phase)$contrasts
```

```
## Sleep.Stage = P, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT -0.15258 0.133 102 -1.146 0.2545
##
## Sleep.Stage = S, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT 0.10094 0.133 102 0.758 0.4501
##
```

```
## Sleep.Stage = W, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT -0.00471 0.133 102 -0.035 0.9719
##
## Sleep.Stage = X, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT 0.00000 0.133 102 0.000 1.0000
##
## Sleep.Stage = P, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT -0.20278 0.133 102 -1.523 0.1308
##
## Sleep.Stage = S, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT 0.12150 0.133 102 0.913 0.3636
##
## Sleep.Stage = W, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT -0.14005 0.133 102 -1.052 0.2953
##
## Sleep.Stage = X, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT 0.00000 0.133 102 0.000 1.0000
##
## Degrees-of-freedom method: kenward-roger
## Results are given on the log (not the response) scale.
```

```
lm_max_length <- lmer(max_bout_length ~ Sleep.Stage * phase * Genotype + (1 | Transmitter), data=SumSta
#plot(lm_max_length, type=c("p", "smooth"), col.line=1)
#lattice::qqmath(lm_max_length)
anova(lm_max_length)
```

```
## Analysis of Variance Table
##               npar    Sum Sq Mean Sq F value
## Sleep.Stage      3 88814079 29604693 63.6687
## phase            1  843042   843042  1.8131
## Genotype          1  170326   170326  0.3663
## Sleep.Stage:phase  3  7416195 2472065  5.3165
## Sleep.Stage:Genotype  3   244168    81389  0.1750
## phase:Genotype     1   192675   192675  0.4144
## Sleep.Stage:phase:Genotype  3   313821   104607  0.2250
```

```
emmeans(lm_max_length, pairwise ~ Genotype | Sleep.Stage * phase)$contrasts
```

```
## Sleep.Stage = P, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT -18.78 322 133 -0.058 0.9536
##
## Sleep.Stage = S, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT -425.78 322 133 -1.323 0.1881
##
## Sleep.Stage = W, phase = dark:
```

```
## contrast estimate SE df t.ratio p.value
## McKO - WT -162.78 322 133 -0.506 0.6139
##
## Sleep.Stage = X, phase = dark:
## contrast estimate SE df t.ratio p.value
## McKO - WT 0.00 322 133 0.000 1.0000
##
## Sleep.Stage = P, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT -46.56 322 133 -0.145 0.8852
##
## Sleep.Stage = S, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT 4.00 322 133 0.012 0.9901
##
## Sleep.Stage = W, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT 5.67 322 133 0.018 0.9860
##
## Sleep.Stage = X, phase = light:
## contrast estimate SE df t.ratio p.value
## McKO - WT 0.00 322 133 0.000 1.0000
##
## Degrees-of-freedom method: kenward-roger
```

```
sws_bouts <- SumStats_phase %>% filter(Sleep.Stage == "S" & phase == "light") %>%
  ggplot(aes(x=Genotype, y=mean_bout_length)) +
  stat_summary(fun = 'mean', geom="bar", width=0.75) +
  stat_summary(fun.data = mean_se, geom = "errorbar", width=.2) +
  geom_point() +
  ylab("Average SWS bout length in the light phase (s)") +
  theme_bw() +
  scale_y_continuous(expand = expansion(mult = c(0, 0.2)))

#ggsave(filename="../figures/sws_bouts.svg", plot=sws_bouts, height=4, width=3)
```