1. Mutational Spectrum of Actinopterygii versus temperature and longevity (Time of maturation):
   a. **All nominally significant spearman rank correlations between frequencies of 4 transitions and temperature:**

   N = 128
   - data: TemperMut$T_C and TemperMut$Temperature

   S = 474663, p-value = 3.321e-05
   alternative hypothesis: true rho is not equal to 0
   sample estimates:
       rho
   -0.3581037
   - data: TemperMut$A_G and TemperMut$Temperature

   S = 256954, p-value = 0.002522
   alternative hypothesis: true rho is not equal to 0
   sample estimates:
       rho
   0.2648037
   - data: allparameters$AGdividedTC and allparameters$Temperature

   S = 179782, p-value = 1.654e-07
   alternative hypothesis: true rho is not equal to 0
   sample estimates:
     rho
   0.447675
   - **other transitions give p-value >0,1**

   ---

   b. **PICs:**

   Coefficients:
   Estimate Std. Error t value Pr(>|t|)
   (Intercept)   -0.961202 2.371449 -0.405   0.687
   pic(data$Temperature, tree_pruned) -0.005479 0.091820 -0.060   0.953
   pic(data$Tm, tree_pruned)   0.110200 0.127524   0.864   0.392

   ---

   c. **Multiple models between temperature and fractions of $A_H>G_H + T_H>C_H$:**

   N=128
   - lm(formula = Temperature ~ scale(T_C) + scale(A_G), data = allparameters)

   (Intercept) 17.2395   0.6029 28.594 < 2e-16 ***
   scale(A_G)   1.6769   0.6097 2.750 0.006835 **
   scale(T_C) -2.4375   0.6097 -3.998 0.000109 ***
   Residual standard error: 6.821 on 125 degrees of freedom
   Multiple R-squared: 0.1753,   Adjusted R-squared: 0.1621
   F-statistic: 13.28 on 2 and 125 DF, p-value: 5.869e-06
   N=65
   - lm(formula = Temperature ~ scale(T_C) + scale(A_G), data = allparameters)

   (Intercept) 15.9015   0.8532 18.638 < 2e-16 ***
   scale(A_G)   2.9136   0.8660 3.364 0.00132 **
   scale(T_C) -1.5153   0.8660 -1.750 0.08511 .
   Residual standard error: 6.879 on 62 degrees of freedom
   Multiple R-squared: 0.2053,   Adjusted R-squared: 0.1796
   F-statistic: 8.007 on 2 and 62 DF, p-value: 0.0008065
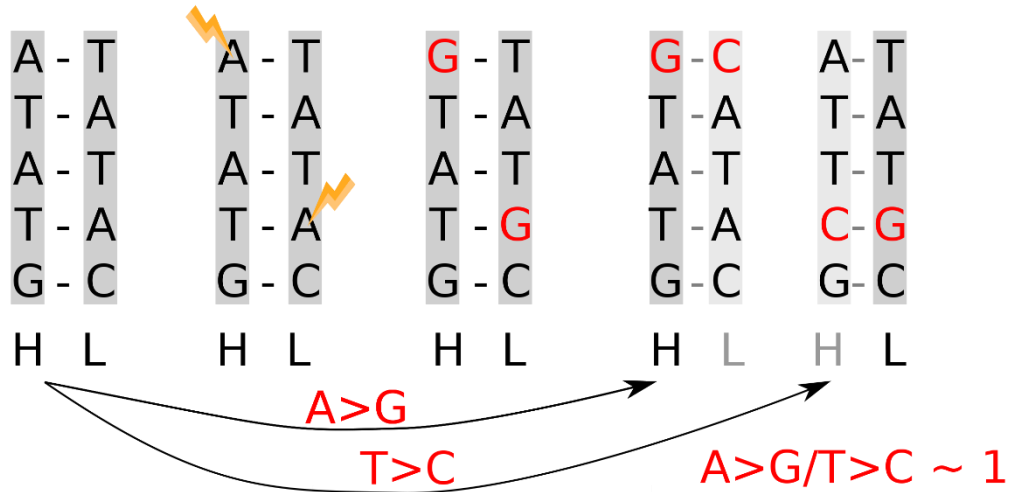
   ---

d. Asymmetry of equivalent substitutions traces chemical damage:

equivalent substitutions => (a)symmetry of mutagenesis

1) symmetrical mutagenesis:

the same probability of A>G on both heavy ('H') and light ('L') strands =>
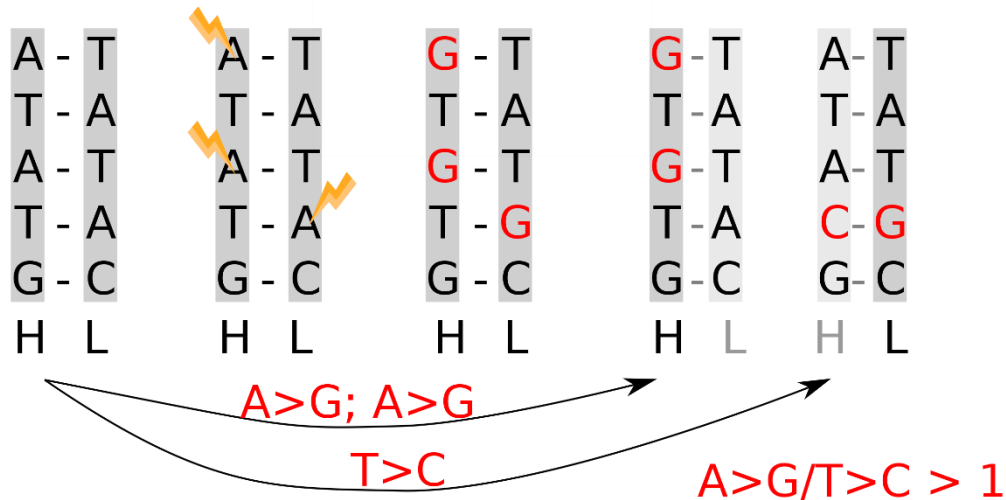fractions of equivalent substitutions are similar

```
A - T      A - T      G - T      G - C      A - T
T - A      T - A      T - A      T - A      T - A
A - T      A - T      A - T      A - T      T - T
T - A      T - A      T - G      T - A      C - G
G - C      G - C      G - C      G - C      G - C
H   L      H   L      H   L      H   L      H   L
```

A>G
T>C   A>G/T>C ~ 1

2) asymmetrical mutagenesis

A>G mainly occurs on heavy strand =>
fractions of equivalent substitutions are different

```
A - T      A - T      G - T      G - T      A - T
T - A      T - A      T - A      T - A      T - A
A - T      A - T      G - T      G - T      A - T
T - A      T - A      T - G      T - A      C - G
G - C      G - C      G - C      G - C      G - C
H   L      H   L      H   L      H   L      H   L
```

A>G; A>G
T>C   A>G/T>C > 1

If a mutagenic process introduces for example mutation A>G on one chain it will be equivalent to T>C on opposite chain and thus symmetrical mutagenesis would lead to similar probabilities of A>G and T>C on any chain. However, probabilities are not equal with AH>GH being significantly higher than TH>CH (Fig 1A). This means that a mutagen, such as a high temperature, affects predominantly heavy chain, which stays single-stranded during mtDNA replication, introducing AH>GH substitutions and the increased temperature is associated with even more elevated asymmetry: AH>GH /TH>CH.

We interpret their increased asymmetry of AH>GH /TH>CH in warm-water species as increased chemical damage of the heavy chain of mtDNA. The logic behind this is following: both these substitutions AH>GH and TH>CH are equivalent, in terms that they are complementary to each other on opposite strands: AH>GH correspond to TL>CL, while TH>CH correspond to AL>GL (see Supplementary Mat.); if mtDNA is mutated mainly on a heavy strand (AH>GH will occur mostly on a heavy strand) the ratio of AH>GH /TH>CH will be more than 1.

e. All multiple models between fractions of A$_H$>G$_H$ and T$_H$>C$_H$ and temperature + longevity:

```
N=65
    •   lm(formula = A_G ~ scale(Temperature) * scale(Tm), data = allparameters)
 (Intercept)              0.131507  0.008055  16.325  < 2e-16 ***
scale(Temperature)        0.024866  0.008083   3.076  0.00313 **
scale(Tm)                -0.008903  0.008722  -1.021  0.31142
scale(Temperature):scale(Tm) -0.000811  0.009005  -0.090  0.92854
Residual standard error: 0.062 on 61 degrees of freedom
Multiple R-squared: 0.1815,          Adjusted R-squared: 0.1412
F-statistic: 4.508 on 3 and 61 DF, p-value: 0.006387
    •   lm(formula = A_G ~ scale(Temperature) + scale(Tm), data = allparameters)
 (Intercept)        0.131723  0.007628  17.269  < 2e-16 ***
scale(Temperature) 0.024931  0.007985   3.122  0.00273 **
scale(Tm)         -0.008600  0.007985  -1.077  0.28564
Residual standard error: 0.0615 on 62 degrees of freedom
Multiple R-squared: 0.1814,          Adjusted R-squared: 0.1549
F-statistic: 6.867 on 2 and 62 DF, p-value: 0.002023
    •   lm(formula = A_G ~ scale(Temperature), data = allparameters)
(Intercept)       0.131723  0.007637  17.247  < 2e-16 ***
scale(Temperature) 0.027259 0.007697  3.542 0.000755 ***
Residual standard error: 0.06158 on 63 degrees of freedom
Multiple R-squared: 0.166,           Adjusted R-squared: 0.1528
F-statistic: 12.54 on 1 and 63 DF, p-value: 0.0007547
    •   lm(formula = T_C ~ scale(Temperature) + scale(Tm), data = allparameters)
 (Intercept)          0.068819  0.006021  11.430  <2e-16 ***
scale(Temperature ) -0.014254  0.006303  -2.261  0.0273 *
scale(Tm)           -0.007483  0.006303  -1.187  0.2397
Residual standard error: 0.04854 on 62 degrees of freedom
Multiple R-squared: 0.08108, Adjusted R-squared: 0.05144
F-statistic: 2.735 on 2 and 62 DF, p-value: 0.07271
    •   lm(formula = T_C ~ scale(Temperature), data = allparameters)
(Intercept)        0.068819  0.006041  11.393  <2e-16 ***
scale(Temperature) -0.012228 0.006088 -2.009  0.0489 *
Residual standard error: 0.0487 on 63 degrees of freedom
Multiple R-squared: 0.06019, Adjusted R-squared: 0.04527
F-statistic: 4.035 on 1 and 63 DF, p-value: 0.04886
```

f. All multiple models between A$_H$>G$_H$: T$_H$>C$_H$ and temperature + longevity:

```
N=123 (no NULL in A_G and T_C)
    •   lm(formula = log2(AGdivTC) ~ scale(Temperature), data = allparameters)
(Intercept)        1.2664    0.1194  10.607  < 2e-16 ***
scale(Temperature) 0.5674    0.1199   4.733 6.06e-06 ***
Residual standard error: 1.324 on 121 degrees of freedom
Multiple R-squared: 0.1562, Adjusted R-squared: 0.1492
F-statistic: 22.4 on 1 and 121 DF, p-value: 6.056e-06
    •   lm(formula = Temperature ~ scale(AGdivTC), data = allparameters)
(Intercept)   17.2793    0.6683 25.855  <2e-16 ***
scale(TCdivAG) 1.2923    0.6711  1.926  0.0565 .
Residual standard error: 7.412 on 121 degrees of freedom
Multiple R-squared: 0.02974,         Adjusted R-squared: 0.02172
F-statistic: 3.709 on 1 and 121 DF, p-value: 0.05648
N=62 (no NULL)
    •   lm(formula = Temperature ~ scale(AGdivTC), data = allparameters)
 (Intercept)    16.0129    0.9507 16.844  <2e-16 ***
scale(TCdivAG)  1.8008    0.9584  1.879  0.0651 .
Residual standard error: 7.486 on 60 degrees of freedom
Multiple R-squared: 0.05557,         Adjusted R-squared: 0.03983
F-statistic: 3.53 on 1 and 60 DF, p-value: 0.06512
    •   lm(formula = log2(AGdivTC) ~ scale(Temperature) + scale(Tm),
   data = allparameters)
(Intercept)        1.11377   0.16982   6.559 1.51e-08 ***
scale(Temperature) 0.58133   0.17790   3.268  0.00181 **
scale(Tm)          0.03231   0.17790   0.182  0.85649
Residual standard error: 1.337 on 59 degrees of freedom
Multiple R-squared: 0.1597, Adjusted R-squared: 0.1313
F-statistic: 5.608 on 2 and 59 DF, p-value: 0.005891
```

g. All nominally significant spearman rank correlations between fractions of 4 transitions and longevity:

```
N=106
    •   data: MATUTmmut$C_G and MATUTmmut$Tm
S = 242544, p-value = 0.0222
alternative hypothesis: true rho is not equal to 0
sample estimates:
    rho
-0.2219755
```

2. Whole genomes of Actinipterygii versus temperature and longevity (Time of maturation):
   a. **All nominally significant spearman rank correlations between fractions of 4 nucleotides and temperature:**

   - data: log2(SynNuc$Temperature) and SynNuc$FrT
   S = 5335566, p-value = 0.03301
   alternative hypothesis: true rho is not equal to 0
   sample estimates:
       rho
   0.1172215
   - data: log2(SynNuc$Temperature) and SynNuc$FrA
   S = 6966996, p-value = 0.005369
   alternative hypothesis: true rho is not equal to 0
   sample estimates:
       rho
   -0.1527014
   - data: log2(SynNuc$Temperature) and SynNuc$FrC
   S = 7546431, p-value = 4.711e-06
   alternative hypothesis: true rho is not equal to 0
   sample estimates:
       rho
   -0.2485698
   - data: log2(SynNuc$Temperature) and SynNuc$FrG
   S = 5248321, p-value = 0.01655
   alternative hypothesis: true rho is not equal to 0
   sample estimates:
       rho
   0.1316563

   b. **All multiple models between S_{TG}-S_{AC} and temperature + longevity:**

   - lm(formula = AC_TGSkew ~ log2(Temperature + 2) + log2(Tm), data = SynNuc)
   (Intercept)   0.140140 0.043625 3.212 0.00167 **
   log2(Temperature + 2) 0.056133 0.009553 5.876 3.42e-08 ***
   log2(Tm)   0.024618 0.006086 4.045 8.99e-05 ***
   Residual standard error: 0.08634 on 128 degrees of freedom
     (4973 observations deleted due to missingness)
   Multiple R-squared: 0.24,   Adjusted R-squared: 0.2282
   F-statistic: 20.22 on 2 and 128 DF, p-value: 2.346e-08

   - lm(formula = AC_TGSkew ~ scale(Temperature + 2) + scale(Tm),
     data = SynNuc)
   (Intercept)   0.421980 0.007940 53.148 < 2e-16 ***
   scale(Temperature + 2) 0.045806 0.007493 6.113 1.10e-08 ***
   scale(Tm)   0.026502 0.006590 4.022 9.82e-05 ***
   Residual standard error: 0.08541 on 128 degrees of freedom
     (4973 observations deleted due to missingness)
   Multiple R-squared: 0.2563, Adjusted R-squared: 0.2447
   F-statistic: 22.06 on 2 and 128 DF, p-value: 5.876e-09

   c. **PICs**

   # lambda [ML] : 0.990
   # Coefficients:
   # Estimate Std. Error t value Pr(>|t|)
   # (Intercept) 0.3185648 0.1248595 2.5514 0.01191 *
   # log2(Temperature + 2) 0.0186083 0.0086042 2.1627 0.03242 *
   # log2(Tm) 0.0099184 0.0048863 2.0298 0.04445 *

   d. **All multiple models between fraction of A and temperature + longevity:**

   - lm(formula = FrA ~ scale(Temperature) + scale(Tm), data = SynNuc)
    (Intercept)   0.224467 0.004409 50.908 < 2e-16 ***
   scale(Temperature) -0.015662 0.004161 -3.764 0.000254 ***
   scale(Tm)   -0.006686 0.003660 -1.827 0.070052 .
    Residual standard error: 0.04743 on 128 degrees of freedom
    (4973 observations deleted due to missingness)
    Multiple R-squared: 0.1051, Adjusted R-squared: 0.09109
    F-statistic: 7.514 on 2 and 128 DF, p-value: 0.0008213
   - lm(formula = FrA ~ log2(Temperature + 2) + log2(Tm), data = SynNuc)
    (Intercept)   0.309754 0.024281 12.757 < 2e-16 ***
   log2(Temperature + 2) -0.017552 0.005317 -3.301 0.00125 **
   log2(Tm)   -0.005453 0.003387 -1.610 0.10989
    Residual standard error: 0.04806 on 128 degrees of freedom
     (4973 observations deleted due to missingness)
    Multiple R-squared: 0.08145, Adjusted R-squared: 0.0671
   F-statistic: 5.675 on 2 and 128 DF, p-value: 0.004351

- lm(formula = FrA ~ log2(Temperature + 2) * log2(Tm), data = SynNuc)
(Intercept)   0.422770 0.050387 8.390 8.02e-14 ***
log2(Temperature + 2)   -0.043535 0.011464 -3.798 0.000225 ***
log2(Tm)   -0.055592 0.019986 -2.782 0.006234 **
log2(Temperature + 2):log2(Tm) 0.011796 0.004637 2.544 0.012156 *
Residual standard error: 0.04706 on 127 degrees of freedom
(4973 observations deleted due to missingness)
Multiple R-squared: 0.126,   Adjusted R-squared: 0.1053
F-statistic: 6.102 on 3 and 127 DF, p-value: 0.0006525

e. All multiple models between other fractions and temperature + longevity:

- lm(formula = FrC ~ log2(Temperature + 2) + log2(Tm), data = SynNuc)
(Intercept)   0.120175 0.013771 8.727 1.2e-14 ***
log2(Temperature + 2) -0.010514 0.003016 -3.487 0.000671 ***
log2(Tm)   -0.006856 0.001921 -3.568 0.000506 ***
Residual standard error: 0.02726 on 128 degrees of freedom
  (4973 observations deleted due to missingness)
Multiple R-squared: 0.1307, Adjusted R-squared: 0.1172
F-statistic: 9.626 on 2 and 128 DF, p-value: 0.0001275
- lm(formula = FrT ~ log2(Temperature + 2) + log2(Tm), data = SynNuc)
(Intercept)   0.249276 0.026709 9.333 4.08e-16 ***
log2(Temperature + 2) 0.023023 0.005849 3.936 0.000135 ***
log2(Tm)   0.014498 0.003726 3.891 0.000160 ***
Residual standard error: 0.05286 on 128 degrees of freedom
  (4973 observations deleted due to missingness)
Multiple R-squared: 0.1562, Adjusted R-squared: 0.143
F-statistic: 11.85 on 2 and 128 DF, p-value: 1.904e-05

f. All multiple models between other nucleotide skews and temperature + longevity:

- lm(formula = CtoTSkew ~ log2(Temperature + 2) + log2(Tm), data = SynNuc)
(Intercept)   -0.39217 0.06129 -6.399 2.70e-09 ***
log2(Temperature + 2) -0.05852 0.01342 -4.361 2.64e-05 ***
log2(Tm)   -0.03714 0.00855 -4.344 2.82e-05 ***
Residual standard error: 0.1213 on 128 degrees of freedom
  (4973 observations deleted due to missingness)
Multiple R-squared: 0.1863, Adjusted R-squared: 0.1735
F-statistic: 14.65 on 2 and 128 DF, p-value: 1.866e-06
- lm(formula = GtoASkew ~ log2(Temperature + 2) + log2(Tm), data = SynNuc)
 (Intercept)   -0.006336 0.082967 -0.076 0.9392
log2(Temperature + 2) 0.043809 0.018168 2.411 0.0173 *
log2(Tm)   0.011212 0.011575 0.969 0.3345
Residual standard error: 0.1642 on 128 degrees of freedom
  (4973 observations deleted due to missingness)
Multiple R-squared: 0.04399, Adjusted R-squared: 0.02905
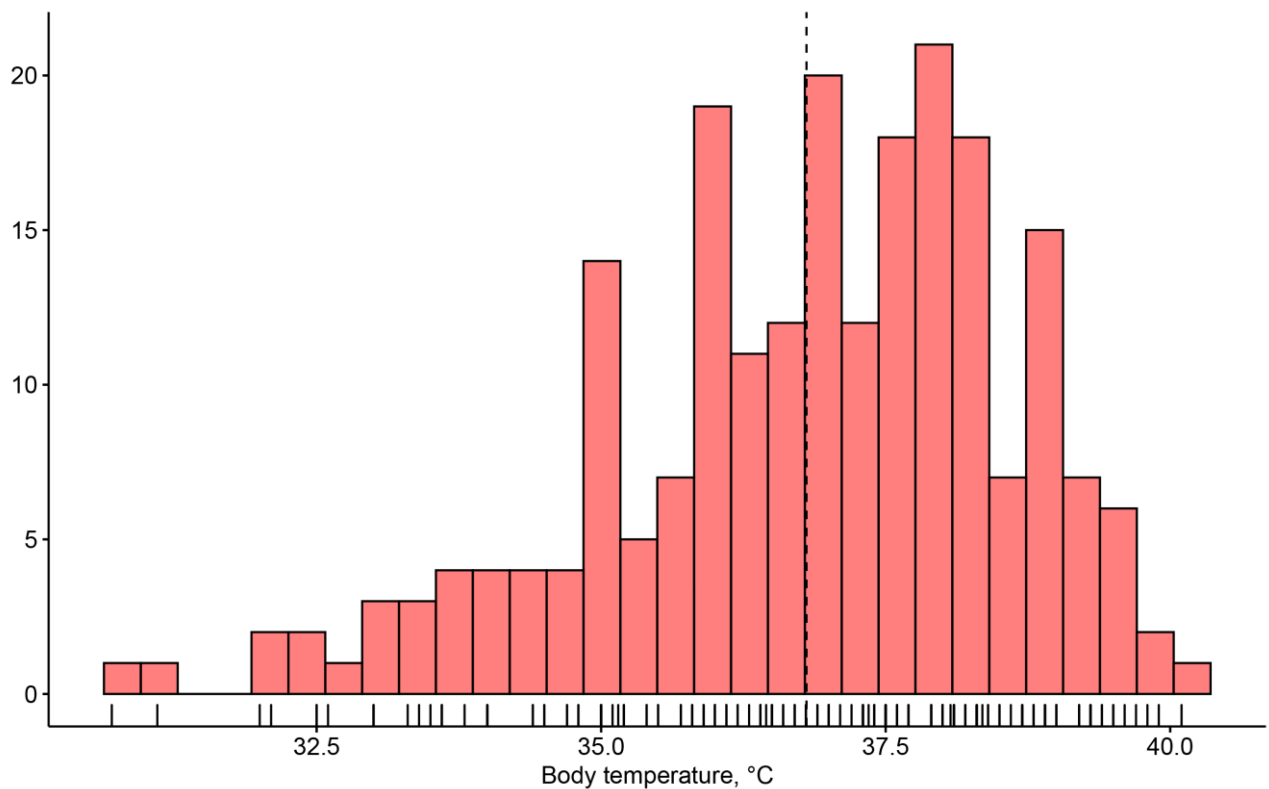F-statistic: 2.945 on 2 and 128 DF, p-value: 0.05618

3. Whole genomes of Mammals versus temperature and longevity (Generation length):
    a. **Average body temperature in Vertebrate classes (AnAge DB):**
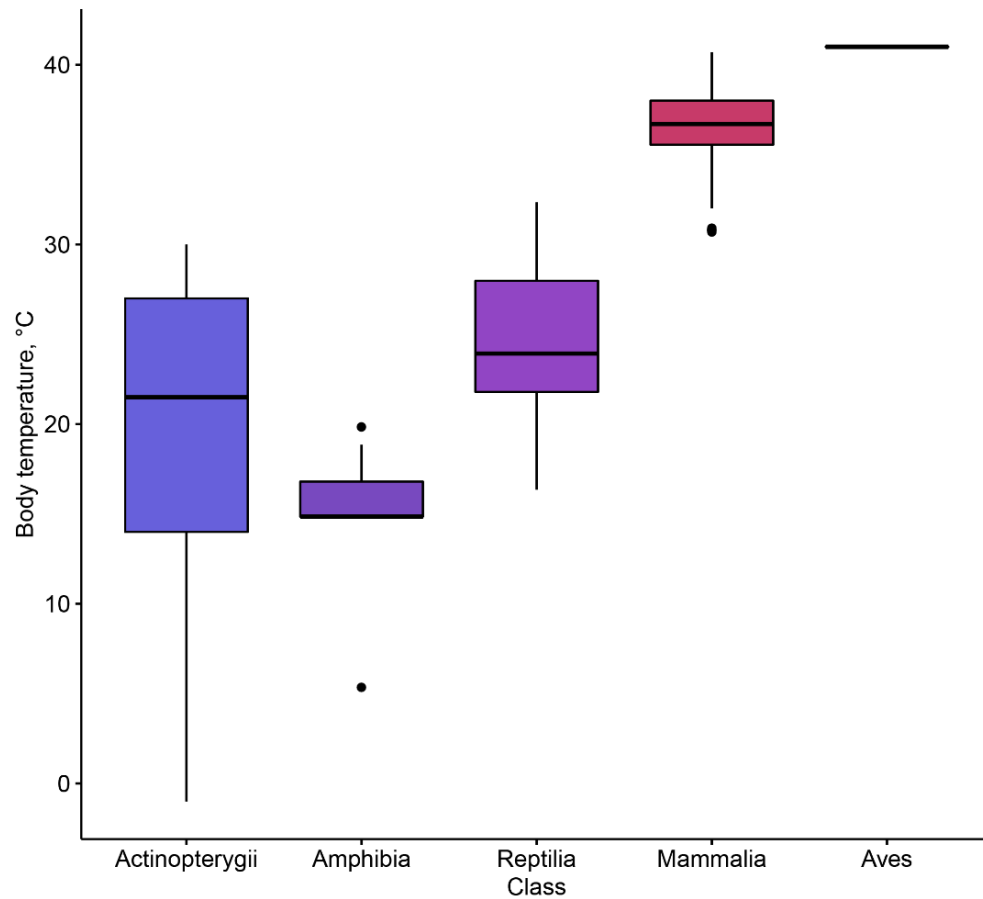
    b. **Average $A_H > G_H / T_H > C_H$ in Vertebrate classes:**
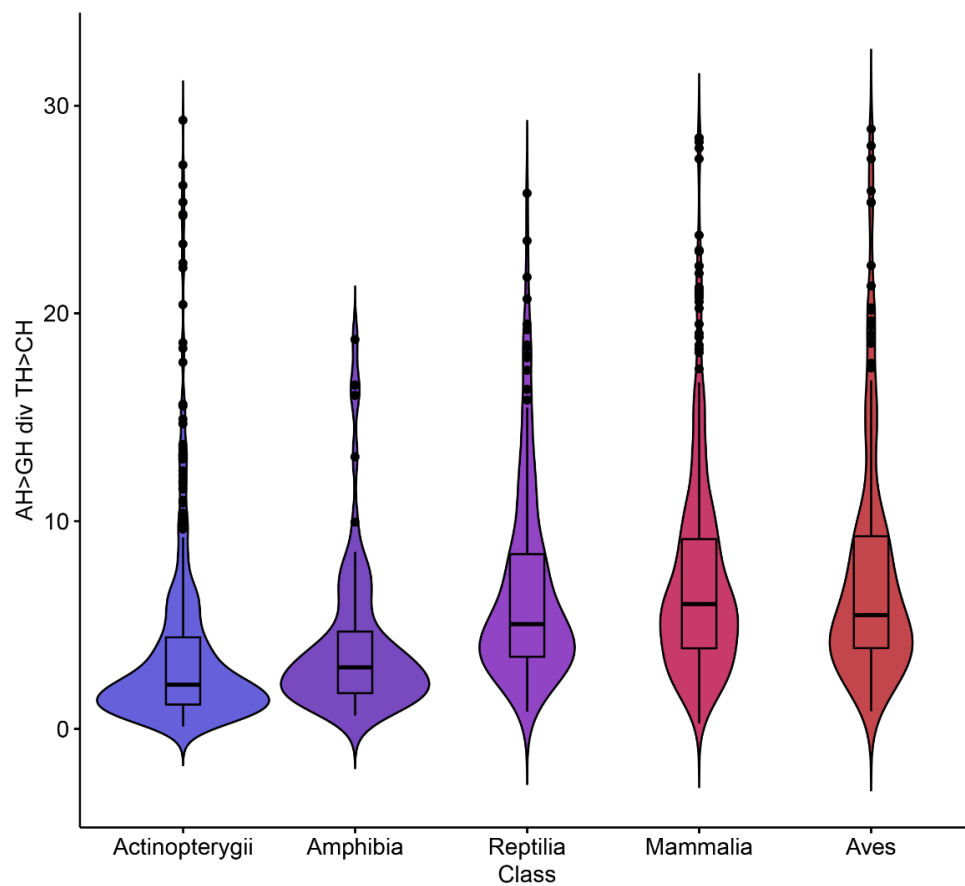
c. A range of known mammalian body temperature:

d. Multiple model between S_{TG}-S_{AC} skew and temperature + longevity:

$$S_{TG}\text{-}S_{AC}$$

- lm(formula = AC_TGSkew ~ scale(Temper) + scale(GenerationLength_d),
  data = allparameters)

```
(Intercept)             0.505937  0.006107  82.839  < 2e-16 ***
scale(Temper)           0.011729  0.006047   1.940  0.053679 .
scale(GenerationLength_d) 0.028182  0.007514   3.751  0.000225 ***
Residual standard error: 0.09029 on 221 degrees of freedom
 (425 observations deleted due to missingness)
Multiple R-squared:  0.07463, Adjusted R-squared:  0.06626
F-statistic: 8.912 on 2 and 221 DF,  p-value: 0.0001896
```

e. Multiple model between S_{TG}-S_{AC} skew and longevity + all cold groups (incl. cold species t° mean <36.8):

- lm(formula = TG_ACSkew ~ log2(GenerationLength_d) + scale(allcolddummy), data = allparameters)

```
 (Intercept)             0.314532  0.030680 -10.252  < 2e-16 ***
log2(GenerationLength_d) 0.018255  0.002761  -6.611 8.00e-11 ***
scale(allcolddummy)      0.017912  0.003106   5.768 1.25e-08 ***
Residual standard error: 0.07848 on 646 degrees of freedom
Multiple R-squared:  0.1192,  Adjusted R-squared:  0.1164
F-statistic: 43.69 on 2 and 646 DF,  p-value: < 2.2e-16
```

f. Multiple model between AG skew and all cold groups (incl. cold species t° mean <36.8):

- lm(formula = AtoGSkew ~ scale(Temper) + scale(GenerationLength_d), data = allparameters)

```
(Intercept)            -0.19540   0.01201 -16.264  < 2e-16 ***
scale(Temper)          -0.02751   0.01189  -2.313  0.0216 *
scale(GenerationLength_d) -0.09565   0.01478  -6.471 6.18e-10 ***
Residual standard error: 0.1776 on 221 degrees of freedom
 (425 observations deleted due to missingness)
Multiple R-squared:  0.176,    Adjusted R-squared:  0.1686
F-statistic: 23.61 on 2 and 221 DF,  p-value: 5.103e-10
```

g. Multiple model between fraction of A and all cold groups (incl. cold species t° mean <36.8):

- lm(formula = FrA ~ log2(GenerationLength_d) + scale(allcolddummy), data = allparameters)

(Intercept)   0.326964   0.016474 19.848   < 2e-16 ***
log2(GenerationLength_d) -0.012105   0.001483 -8.164 1.70e-15 ***
scale(allcolddummy)   0.010043   0.001668   6.023 2.88e-09 ***
Residual standard error: 0.04214 on 646 degrees of freedom
Multiple R-squared: 0.1527,   Adjusted R-squared: 0.1501
F-statistic: 58.21 on 2 and 646 DF, p-value: < 2.2e-16

- lm(formula = FrA ~ log2(GenerationLength_d), data = allparameters)

(Intercept)   0.338815   0.016796 20.173   <2e-16 ***
log2(GenerationLength_d) -0.013177   0.001512 -8.718   <2e-16 ***
Residual standard error: 0.04328 on 647 degrees of freedom
Multiple R-squared: 0.1051,   Adjusted R-squared: 0.1037
F-statistic: 76 on 1 and 647 DF, p-value: < 2.2e-16

- lm(formula = FrA ~ scale(GenerationLength_d) + scale(allcolddummy), data = allparameters)

(Intercept)   0.193148   0.001694 114.050   < 2e-16 ***
scale(GenerationLength_d) -0.009939   0.001706 -5.825 9.03e-09 ***
scale(allcolddummy)   0.010528   0.001706   6.170 1.20e-09 ***
Residual standard error: 0.04314 on 646 degrees of freedom
Multiple R-squared: 0.1119,   Adjusted R-squared: 0.1092
F-statistic: 40.7 on 2 and 646 DF, p-value: < 2.2e-16

h. Multiple model between fraction of A and temperature + longevity:

- lm(formula = FrA ~ scale(GenerationLength_d) + scale(Temper), data = allparameters)

(Intercept)   0.196165   0.003304 59.376 < 2e-16 ***
scale(GenerationLength_d) -0.021853   0.004065 -5.376 1.93e-07 ***
scale(Temper)   -0.006774   0.003271 -2.071   0.0395 *
Residual standard error: 0.04884 on 221 degrees of freedom
  (425 observations deleted due to missingness)
Multiple R-squared: 0.1306, Adjusted R-squared: 0.1227
F-statistic: 16.59 on 2 and 221 DF, p-value: 1.933e-07

i. Multiple model between fraction of A and cold species (t° mean <36.8):

- lm(formula = FrA ~ scale(GenerationLength_d) + scale(colddummy), data = allparameters)

(Intercept)   0.193148   0.001726 111.933 < 2e-16 ***
scale(GenerationLength_d) -0.010730   0.001731 -6.199 1.02e-09 ***
scale(colddummy)   0.006220   0.001731   3.594 0.000351 ***
Residual standard error: 0.04396 on 646 degrees of freedom
Multiple R-squared: 0.078,   Adjusted R-squared: 0.07514
F-statistic: 27.32 on 2 and 646 DF, p-value: 4.058e-12

j. Fraction of A versus longevity in mammals: