

Meta-Analysis for Longevity Summary Excluding HUM251

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1. Summary

This document reports the process taken in the model fitting stage of the meta-analysis in thermal longevity.

2. Setup

We first read in our data and select all of the effect sizes related to longevity. We do this using the following code.

```
### Read in effect size data
effectdata <- read.csv("Survival project all pairwise.es.csv")
longdata_warm <- subset(effectdata, Trait.category == "Longevity" &
  warm.cool == "Warm")
longdata_cool <- subset(effectdata, Trait.category == "Longevity" &
  warm.cool == "Cool")

alllong <- rbind(longdata_warm, longdata_cool)

### select data for analysis
rdata <- alllong

rdata <- subset(rdata, Paper.code != "HUM251")

rdata <- rdata %>%
  mutate(c_treattemp = treattemp - 25)
```

Next we create new columns in our dataframe which will serve as random factors in our multi-level meta analysis models. The following initialises four new columns, namely “obs”, “study_code”, “Species.phylo” and “species”. Lastly, we create a column name “precision” which is equal to the inverse standard error.

```
### Create random factors into data frame
rdata$obs <- factor(c(1:nrow(rdata))) # Unique observation code
rdata$study_code <- factor(rdata$Paper.code) # Model requires column names study_code
rdata$Species.phylo <- factor(rdata$Species.latin) # Species names for phylo matrix
rdata$species <- factor(rdata$Species.latin) # Another species column for random factor

precision <- sqrt(1/rdata$v) # inverse standard error
rdata[, "precision"] <- precision
```

The number of species and total number of studies present in the data are as follows.

```
nlevels(rdata$species) # Check number of species
```

```
## [1] 290
```

```
nlevels(rdata$study_code) # Check number of studies
```

```
## [1] 316
```

The final stage in the setup is to import a phylogenetic tree of the data. Below is the code used to produce the tree and a plot of the tree itself.

```
## import tree from map
tree1 <- read.nexus("all_longevity_excHUM251_tree.nex")
tree_grafen = compute.brlen(tree1, method = "Grafen", power = 1)
phylo_matrix <- vcv(tree_grafen, cor = TRUE, model = "Brownian") # Make phylogenetic matrix

## character(0)
```



3. Random effects models

In this section we determine which random effects to include in our model. For each model I have provided the code used to specify the structure of the model and a summary of the results. We begin with a model that includes all of the random factors we created earlier.

```
# Adding four random factors
meta2 <- rma.mv(es, v, random = list(~1 | Species.phylo, ~1 |
  species, ~1 | study_code, ~1 | obs), R = list(Species.phylo = phylo_matrix),
  test = "t", dfs = "contain", data = rdata, method = "REML")
```

```
summary(meta2)
```

```
##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3569.8004    7139.6008    7149.6008    7175.7500    7149.6444
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor      R
## sigma^2.1  0.0000  0.0001   290     no Species.phylo  yes
## sigma^2.2  0.0000  0.0003   290     no      species    no
## sigma^2.3  0.9558  0.9776   316     no  study_code    no
## sigma^2.4  7.6624  2.7681  1381     no        obs     no
##
## Test for Heterogeneity:
## Q(df = 1380) = 51322.8613, p-val < .0001
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub
## -0.1783  0.0979  -1.8205  0.0687  -0.3702  0.0137
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
i2_ml(meta2, method = c("ratio")) # Heterogeneity at each random factor level
```

```
##      I2_Total I2_Species.phylo      I2_species      I2_study_code      I2_obs
##      9.908166e+01      4.027427e-08      9.504325e-07      1.098812e+01      8.809354e+01
```

Accounting for non-independence of data points from the same experiment

The data has a nested structure. Each study (study_code) may have a number of experiments (effect.size.code) which share a common control temperature. Each effect size has its own unique code, obs. Effect sizes from the same experiment which share a control temperature are thought to be non-independent. The following code create a covariance matrix “VCV_shared” which assumes a correlation of 0.5 between effect sizes from the same experiment. We include this structure in our proceeding models.

```
rdata$shared_control <- factor(rdata$Effect.size.code)
vcv_shared <- impute_covariance_matrix(vi = rdata$v, cluster = rdata$shared_control,
  r = 0.5)
```

```
# Add new variance matrix into the mixed-effects
# meta-analysis model
```

```
meta3 <- rma.mv(es, vcv_shared, random = list(~1 | Species.phylo,
~1 | species, ~1 | study_code, ~1 | obs), test = "t", dfs = "contain",
R = list(Species.phylo = phylo_matrix), data = rdata, method = "REML")
```

```
summary(meta3)
```

```
##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3558.8224    7117.6449    7127.6449    7153.7941    7127.6886
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor      R
## sigma^2.1  0.0000  0.0000   290    no Species.phylo yes
## sigma^2.2  0.0000  0.0003   290    no      species    no
## sigma^2.3  0.6821  0.8259   316    no  study_code    no
## sigma^2.4  7.9265  2.8154  1381    no      obs      no
##
## Test for Heterogeneity:
## Q(df = 1380) = 69550.4868, p-val < .0001
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub      *
## -0.1901  0.0946  -2.0088  0.0446  -0.3756  -0.0046  *
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
i2_ml(meta3, method = c("ratio")) # Heterogeneity at each random factor level
```

```
##      I2_Total I2_Species.phylo      I2_species      I2_study_code      I2_obs
##      9.908064e+01      1.014580e-10      7.633043e-07      7.850134e+00      9.123051e+01
```

Model without phylogeny

The variance-covariance matrix for phylogenetic relatedness of included species has now been excluded as a random effect in the model (Chamberlain et al., 2012) as its inclusion did not improve model fit and the phylogenetic signal was very weak.

```
## without phylogeny
meta5 <- rma.mv(es, VCV_shared, random = list(~1 | species, ~1 |
  study_code, ~1 | obs), test = "t", dfs = "contain", data = rdata,
  method = "REML")

summary(meta5)

##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3558.8224    7117.6449    7125.6449    7146.5643    7125.6740
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor
## sigma^2.1  0.0000  0.0003   290     no      species
## sigma^2.2  0.6821  0.8259   316     no  study_code
## sigma^2.3  7.9265  2.8154  1381     no        obs
##
## Test for Heterogeneity:
## Q(df = 1380) = 69550.4868, p-val < .0001
##
## Model Results:
##
## estimate      se      tval   df    pval    ci.lb    ci.ub
## -0.1901  0.0946  -2.0088  289  0.0455  -0.3764  -0.0038 *
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

i2_ml(meta5, method = c("ratio")) # Heterogeneity at each random factor level

##      I2_Total      I2_species I2_study_code      I2_obs
## 9.908064e+01  9.571908e-07  7.850129e+00  9.123051e+01
```

Model without phylogeny or species

```
## without phylogeny or species
meta4 <- rma.mv(es, VCV_shared, random = list(~1 | study_code,
~1 | obs), data = rdata, test = "t", dfs = "contain", method = "REML")

summary(meta4)

##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3558.8224    7117.6449    7123.6449    7139.3344    7123.6623
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor
## sigma^2.1  0.6821  0.8259   316      no  study_code
## sigma^2.2  7.9265  2.8154  1381      no      obs
##
## Test for Heterogeneity:
## Q(df = 1380) = 69550.4868, p-val < .0001
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub
## -0.1901  0.0946  -2.0088  0.0446  -0.3756  -0.0046  *
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

i2_ml(meta4, method = c("ratio")) # Heterogeneity at each random factor level

##      I2_Total I2_study_code      I2_obs
##      99.080643      7.850129    91.230514
```

Model without phylogeny, species or study_code

```
## without phylogeny, species or study_code
meta7 <- rma.mv(es, VCV_shared, random = list(~1 | obs), data = rdata,
  test = "t", dfs = "contain", method = "REML")
```

```
summary(meta7)
```

```
##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3577.8245    7155.6489    7159.6489    7170.1086    7159.6576
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed  factor
## sigma^2    8.6967  2.9490   1381     no     obs
##
## Test for Heterogeneity:
## Q(df = 1380) = 69550.4868, p-val < .0001
##
## Model Results:
##
## estimate      se      zval      pval      ci.lb      ci.ub
## -0.2047  0.0817  -2.5071  0.0122  -0.3648  -0.0447  *
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

i2_ml(meta7, method = c("ratio")) # Heterogeneity at each random factor level

## I2_Total  I2_obs
## 99.08988 99.08988
```

We can see from the above that the best fitting model according to AIC is “meta4” which includes only the study code and the unique effect size code, obs. There is a AIC difference of 4 between the model meta4 and the next best model meta3 . We continue our analysis using meta4 as our base model.

4. Meta-regressions

Starting with the best fitting random-effect model from Section 3, “meta4” we now include single factors as a fixed effect. We initially explore the fixed factors

- **reftemp**: The experiment’s control (reference) temperature.
- **treattemp**: The treatment temperature, which we expect to have a non-linear relationship to longevity.
- **warm.cool**: A categorical variable indicating whether treatment is warmer or cooler than the reference temperature
- **diff**: The difference between the reference and treatment temperature.

Reference temperature

```
meta_trait_ref <- rma.mv(es, VCV_shared, mod = ~reftemp, random = list(~1 |  
  study_code, ~1 | obs), test = "t", dfs = "contain", data = rdata,  
  method = "REML")
```

```
summary(meta_trait_ref)
```

```
##  
## Multivariate Meta-Analysis Model (k = 1381; method: REML)  
##  
##      logLik      Deviance      AIC      BIC      AICc  
## -3555.9039    7111.8079    7119.8079    7140.7243    7119.8370  
##  
## Variance Components:  
##  
##      estim      sqrt  nlvls  fixed      factor  
## sigma^2.1  0.6742  0.8211   316     no  study_code  
## sigma^2.2  7.9293  2.8159  1381     no           obs  
##  
## Test for Residual Heterogeneity:  
## QE(df = 1379) = 69486.0682, p-val < .0001  
##  
## Test of Moderators (coefficient 2):  
## QM(df = 1) = 1.5364, p-val = 0.2152  
##  
## Model Results:  
##  
##      estimate      se      zval      pval      ci.lb      ci.ub  
## intrcpt    -1.3024  0.9023   -1.4434  0.1489   -3.0708  0.4661  
## reftemp     0.0447  0.0361    1.2395  0.2152   -0.0260  0.1155  
##  
## ---  
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Treatment temperature

```
meta_trait_treattemp <- rma.mv(es, VCV_shared, mod = ~c_treattemp,  
  random = list(~1 | study_code, ~1 | obs), test = "t", dfs = "contain",  
  data = rdata, method = "REML")
```

```
summary(meta_trait_treattemp)
```

```
##  
## Multivariate Meta-Analysis Model (k = 1381; method: REML)  
##  
##      logLik      Deviance      AIC      BIC      AICc  
## -3356.9436    6713.8872    6721.8872    6742.8036    6721.9163  
##  
## Variance Components:  
##  
##      estim      sqrt  nlvls  fixed      factor  
## sigma^2.1  1.1335  1.0647   316     no  study_code  
## sigma^2.2  5.3123  2.3048  1381     no           obs  
##  
## Test for Residual Heterogeneity:  
## QE(df = 1379) = 48854.1415, p-val < .0001  
##  
## Test of Moderators (coefficient 2):  
## QM(df = 1) = 489.6259, p-val < .0001  
##  
## Model Results:  
##  
##      estimate      se      zval      pval      ci.lb      ci.ub  
## intrcpt      -0.3127  0.0926   -3.3784  0.0007   -0.4941   -0.1313 ***  
## c_treattemp  -0.1907  0.0086  -22.1275 <.0001   -0.2076   -0.1738 ***  
##  
## ---  
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Warm vs Cool

```
meta_trait_warm <- rma.mv(es, VCV_shared, mod = ~warm.cool, random = list(~1 |  
  study_code, ~1 | obs), test = "t", dfs = "contain", data = rdata,  
  method = "REML")
```

```
summary(meta_trait_warm)
```

```
##  
## Multivariate Meta-Analysis Model (k = 1381; method: REML)  
##  
##      logLik      Deviance      AIC      BIC      AICc  
## -3326.1091    6652.2183    6660.2183    6681.1347    6660.2474  
##  
## Variance Components:  
##  
##      estim      sqrt  nlvls  fixed      factor  
## sigma^2.1  0.6254  0.7908   316     no  study_code  
## sigma^2.2  5.2758  2.2969  1381     no           obs  
##  
## Test for Residual Heterogeneity:  
## QE(df = 1379) = 46542.1152, p-val < .0001  
##  
## Test of Moderators (coefficient 2):  
## QM(df = 1) = 557.0028, p-val < .0001  
##  
## Model Results:  
##  
##      estimate      se      zval      pval      ci.lb      ci.ub  
## intrcpt      1.3461  0.1047   12.8548 <.0001    1.1409    1.5514 ***  
## warm.coolWarm -3.0965  0.1312  -23.6009 <.0001   -3.3537   -2.8394 ***  
##  
## ---  
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

We model warm versus cool without and intercept so we can visualise the estimates easier.

```
meta_trait_warm_nointer <- rma.mv(es, VCV_shared, mod = ~warm.cool -
  1, random = list(~1 | study_code, ~1 | obs), data = rdata,
  test = "t", dfs = "contain", method = "REML")

summary(meta_trait_warm_nointer)
```

```
##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3326.1091   6652.2183   6660.2183   6681.1347   6660.2474
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor
## sigma^2.1  0.6254  0.7908   316     no  study_code
## sigma^2.2  5.2758  2.2969  1381     no      obs
##
## Test for Residual Heterogeneity:
## QE(df = 1379) = 46542.1152, p-val < .0001
##
## Test of Moderators (coefficients 1:2):
## QM(df = 2) = 562.6656, p-val < .0001
##
## Model Results:
##
##      estimate      se      zval      pval      ci.lb      ci.ub
## warm.coolCool    1.3461  0.1047   12.8548 <.0001    1.1409    1.5514 ***
## warm.coolWarm   -1.7504  0.1051  -16.6536 <.0001   -1.9564   -1.5444 ***
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Difference

```
meta_trait_diff <- rma.mv(es, VCV_shared, mod = ~diff, random = list(~1 |
  study_code, ~1 | obs), data = rdata, test = "t", dfs = "contain",
  method = "REML")
```

```
summary(meta_trait_diff)
```

```
##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3343.4388    6686.8776    6694.8776    6715.7940    6694.9067
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor
## sigma^2.1  0.9889  0.9944   316      no  study_code
## sigma^2.2  5.2509  2.2915  1381      no      obs
##
## Test for Residual Heterogeneity:
## QE(df = 1379) = 46581.0991, p-val < .0001
##
## Test of Moderators (coefficient 2):
## QM(df = 1) = 520.9053, p-val < .0001
##
## Model Results:
##
##      estimate      se      zval      pval      ci.lb      ci.ub
## intrcpt  -0.2817  0.0894   -3.1495  0.0016   -0.4570   -0.1064  **
## diff      -0.1969  0.0086  -22.8234  <.0001   -0.2138   -0.1800  ***
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Treatment temperature as a cubic effect

We expect that treatment temperature will have a non-linear effect on longevity. We expect that the relationship will be cubic. More specifically, we expect that longevity will increase at cooler temperature and decrease at more extreme higher temperature. We also assume that this optimum temperature will be close to the control temperature. That said, we investigate **treattemp** as a cubic fixed effect.

```
meta_trait_treat3 <- rma.mv(es, VCV_shared, mod = ~poly(c_treattemp,
  degree = 3, raw = TRUE), random = list(~1 | study_code, ~1 |
  obs), test = "t", dfs = "contain", data = rdata, method = "REML")
```

```
summary(meta_trait_treat3)
```

```
##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3289.3464    6578.6928    6590.6928    6622.0588    6590.7541
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor
## sigma^2.1  0.6631  0.8143   316     no  study_code
## sigma^2.2  4.9962  2.2352  1381     no      obs
##
## Test for Residual Heterogeneity:
## QE(df = 1377) = 45364.6017, p-val < .0001
##
## Test of Moderators (coefficients 2:4):
## QM(df = 3) = 650.3631, p-val < .0001
##
## Model Results:
##
##              estimate      se      zval      pval      ci.lb      ci.ub
## intrcpt          -0.0693  0.0986   -0.7027  0.4822   -0.2626    0.1240
## poly(c_treattemp, degree = 3, raw = TRUE)1   -0.2894  0.0125  -23.2134 <.0001   -0.3139   -0.2650 **
## poly(c_treattemp, degree = 3, raw = TRUE)2   -0.0031  0.0008   -3.6610  0.0003   -0.0047   -0.0014 **
## poly(c_treattemp, degree = 3, raw = TRUE)3    0.0006  0.0001   10.6070 <.0001    0.0005    0.0008 **
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

For completeness we also investigate treatment temperature as a quadratic effect.

```
meta_trait_treat2 <- rma.mv(es, VCV_shared, mod = ~poly(c_treattemp,
  degree = 2, raw = TRUE), random = list(~1 | study_code, ~1 |
  obs), test = "t", dfs = "contain", data = rdata, method = "REML")
```

```
summary(meta_trait_treat2)
```

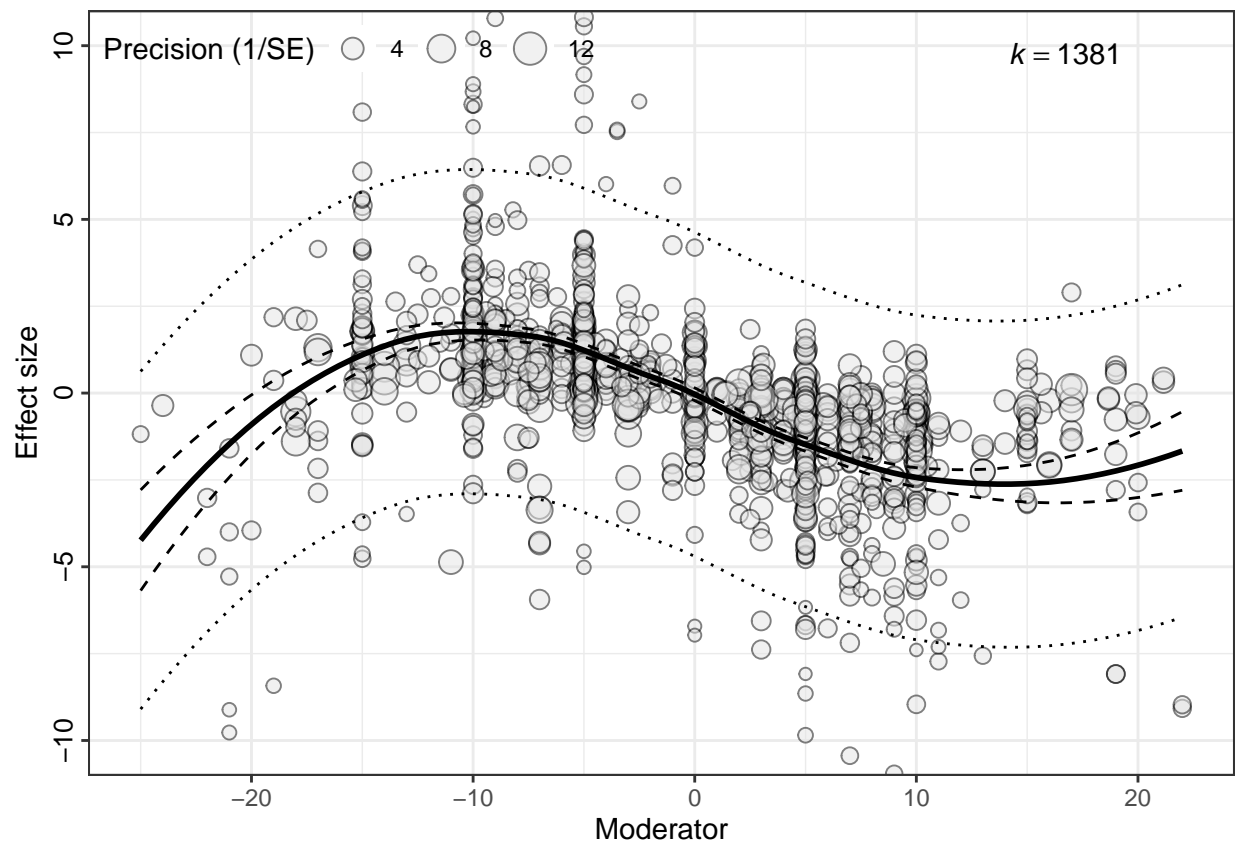
```
##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3342.2747    6684.5495    6694.5495    6720.6914    6694.5932
##
## Variance Components:
```

```

##
##          estim      sqrt  nlvls  fixed      factor
## sigma^2.1 1.1454  1.0702   316     no  study_code
## sigma^2.2 5.1999  2.2803  1381     no           obs
##
## Test for Residual Heterogeneity:
## QE(df = 1378) = 48666.3566, p-val < .0001
##
## Test of Moderators (coefficients 2:3):
## QM(df = 2) = 525.2602, p-val < .0001
##
## Model Results:
##
##          estimate      se      zval      pval      ci.lb      ci.ub
## intrcpt          -0.0166  0.1093   -0.1517  0.8794   -0.2309   0.1977
## poly(c_treattemp, degree = 2, raw = TRUE)1  -0.1974  0.0086  -22.8527 <.0001   -0.2143  -0.1805 **
## poly(c_treattemp, degree = 2, raw = TRUE)2  -0.0045  0.0009   -5.0642 <.0001   -0.0063  -0.0028 **
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

```

Below is a bubble plot of the fitted cubic model.



Modelling response with binned treatment temperatures

The last model we will try is one with categorised or ‘binned’ treatment temperatures. We categories our effect sizes into one seven bins depending on the treatment temperature. The bins and the number of effect sizes in each bin are given below

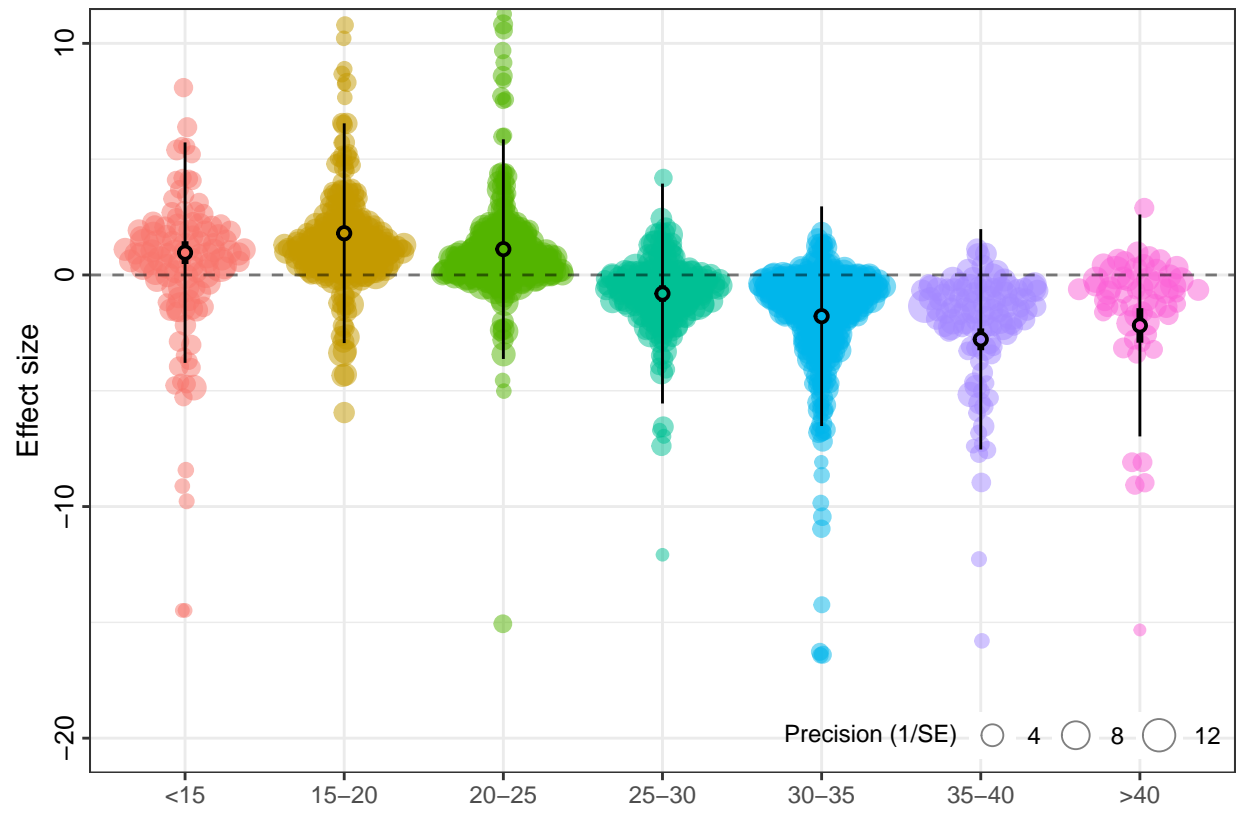
```
table(rdata$bin.temp)
```

```
##
##   <15   >40 15-20 20-25 25-30 30-35 35-40
##   110    55  249   316   189   354   108
```

```
meta_trait_bintemp <- rma.mv(es, VCV_shared, mod = ~bin.temp -
  1, random = list(~1 | study_code, ~1 | obs), data = rdata,
  method = "REML")
```

```
summary(meta_trait_bintemp)
```

```
##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3299.9928   6599.9856   6617.9856   6665.0150   6618.1176
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor
## sigma^2.1  0.7395  0.8599   316      no  study_code
## sigma^2.2  5.0928  2.2567  1381      no      obs
##
## Test for Residual Heterogeneity:
## QE(df = 1374) = 46257.8558, p-val < .0001
##
## Test of Moderators (coefficients 1:7):
## QM(df = 7) = 611.7116, p-val < .0001
##
## Model Results:
##
##      estimate      se      zval      pval      ci.lb      ci.ub
## bin.temp<15      0.9640  0.2503    3.8505  0.0001    0.4733    1.4546 ***
## bin.temp>40     -2.1798  0.3798   -5.7387 <.0001   -2.9243   -1.4353 ***
## bin.temp15-20     1.8019  0.1599   11.2684 <.0001    1.4885    2.1153 ***
## bin.temp20-25     1.1145  0.1435    7.7674 <.0001    0.8333    1.3957 ***
## bin.temp25-30    -0.8034  0.1823   -4.4072 <.0001   -1.1607   -0.4461 ***
## bin.temp30-35    -1.7814  0.1366  -13.0396 <.0001   -2.0492   -1.5137 ***
## bin.temp35-40    -2.7791  0.2403  -11.5643 <.0001   -3.2502   -2.3081 ***
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```



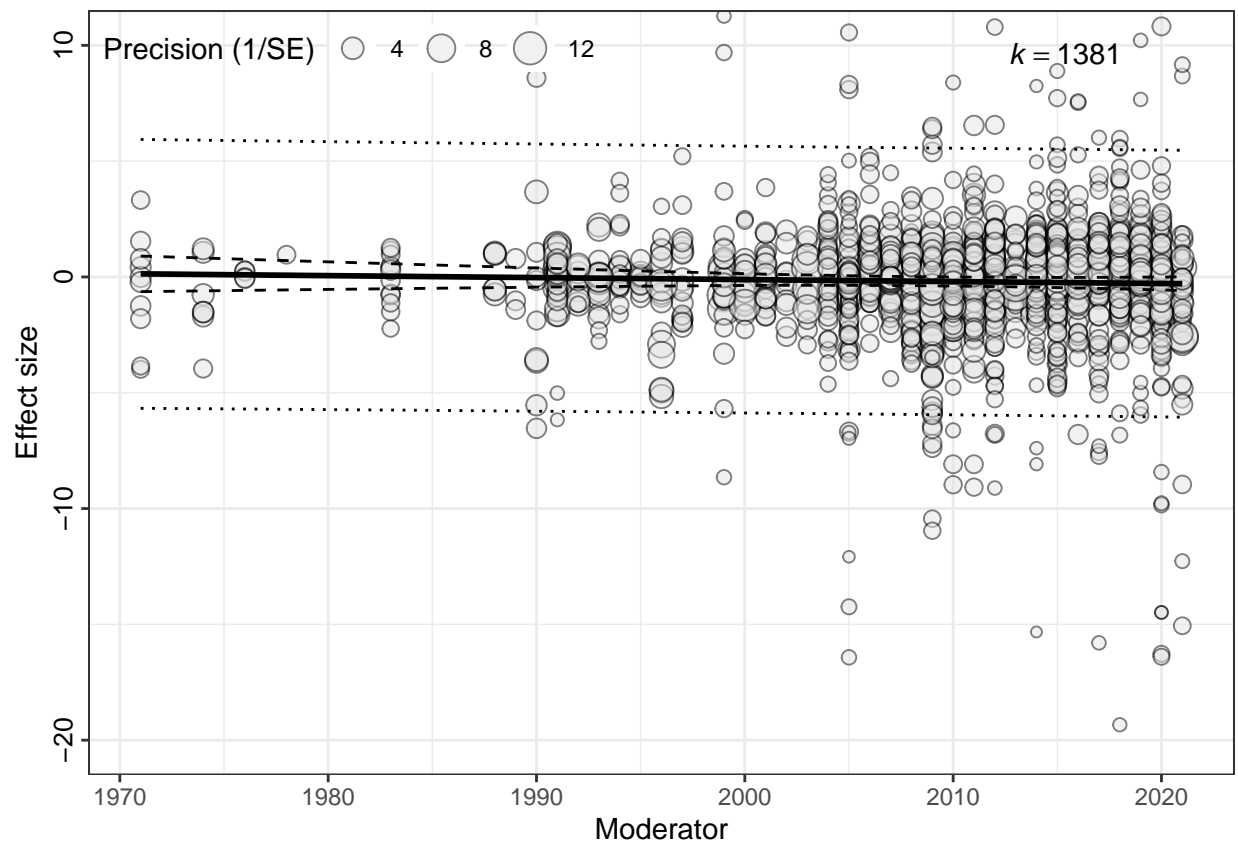
Publication Bias.

We fit meta-regression model with year as a moderator to see whether there is an publication bias. In other words, is effect size strongly correlated with publication year. We found now evidence to suggest that year has an effect on the report effect sizes. A summary of the model is given below.

```
meta_year <- rma.mv(es, VCV_shared, mod = ~Publication.year,
  random = list(~1 | study_code, ~1 | obs), data = rdata, test = "t",
  dfs = "contain", method = "REML")
```

```
summary(meta_year)
```

```
##
## Multivariate Meta-Analysis Model (k = 1381; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3556.2736    7112.5473    7120.5473    7141.4637    7120.5764
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor
## sigma^2.1  0.6932  0.8326   316     no  study_code
## sigma^2.2  7.9235  2.8149  1381     no      obs
##
## Test for Residual Heterogeneity:
## QE(df = 1379) = 69459.6629, p-val < .0001
##
## Test of Moderators (coefficient 2):
## QM(df = 1) = 0.7279, p-val = 0.3936
##
## Model Results:
##
##      estimate      se      zval      pval      ci.lb      ci.ub
## intrcpt      16.8856  20.0142   0.8437  0.3988  -22.3414   56.1127
## Publication.year -0.0085   0.0100  -0.8532  0.3936   -0.0280    0.0110
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```



Sensitivity Analysis

Here, we perform a sensitivity analysis by removing the smallest and largest 2.5% of effect sizes.

The 2.5th and 97.5th percentiles are given below.

```
print(minq)

##      2.5%
## -6.897064

print(maxq)

##      97.5%
##  6.432521
```

The sumamry of the effect sizes is now

```
summary(sdata$es)

##      Min. 1st Qu.  Median      Mean 3rd Qu.      Max.
## -6.8267 -1.2309 -0.0704 -0.1874  0.9824  6.3795
```

We run the cubic treatment temperature and the binned temperature models again with our new subsetting data i.e. only with data that is between the 2.5Th and 97.5Th percentile.

Treatment temperature as a cubic effect (sesnsitivity analysis)

We re-create the variance-covariance matrix with our new subsetting data, which we name sdata. Then we run the meta analysis model again with treatment temperature as quadratic fixed effect.

```
# recreate vcv_shared matrix
VCV_shared_sa <- impute_covariance_matrix(vi = sdata$v, cluster = sdata$shared_control,
  r = 0.5)
```

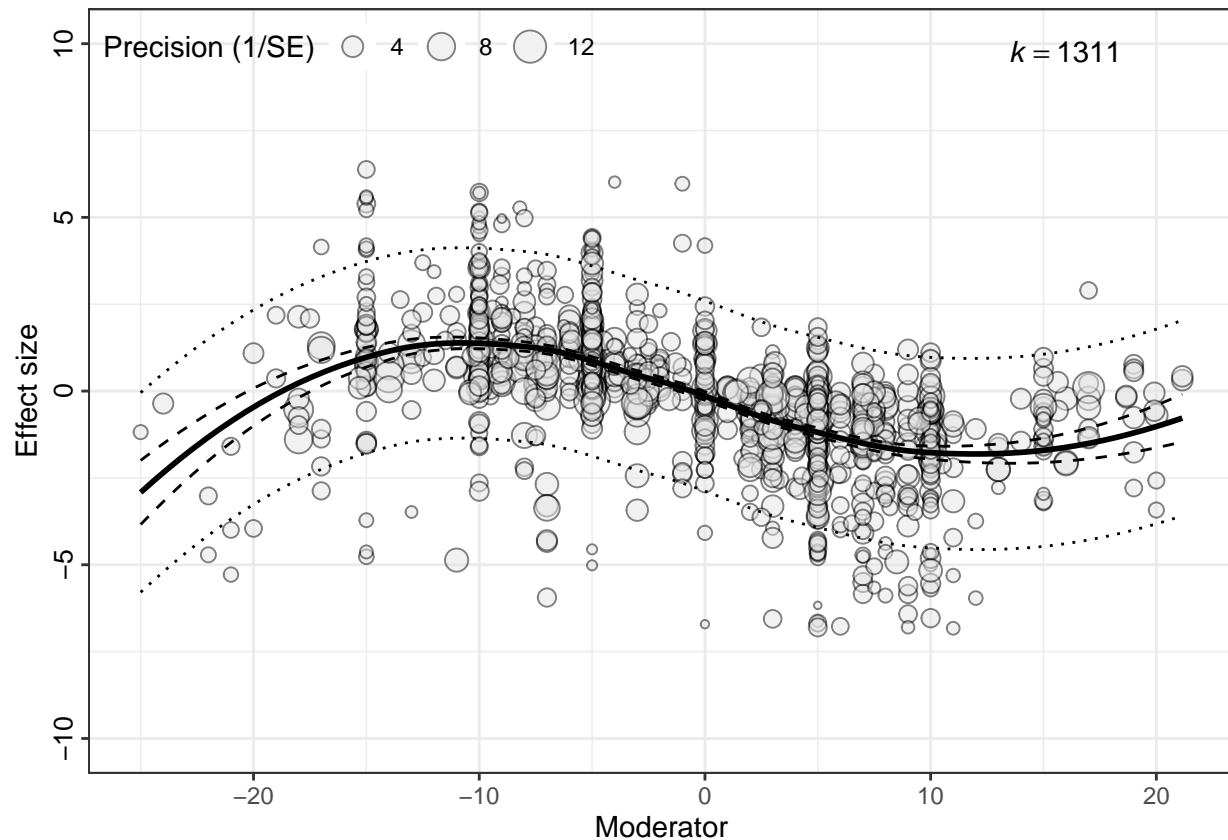
```
meta_sa_treat3 <- rma.mv(es, VCV_shared_sa, mod = ~poly(c_treattemp,
  degree = 3, raw = TRUE), random = list(~1 | study_code, ~1 |
  obs), test = "t", dfs = "contain", data = sdata, method = "REML")
```

```
summary(meta_sa_treat3)
```

```
##
## Multivariate Meta-Analysis Model (k = 1311; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -2303.7299   4607.4598   4619.4598   4650.5127   4619.5244
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor
## sigma^2.1  0.5165  0.7187   313     no  study_code
## sigma^2.2  1.4322  1.1967  1311     no           obs
##
## Test for Residual Heterogeneity:
## QE(df = 1307) = 28230.5429, p-val < .0001
##
## Test of Moderators (coefficients 2:4):
## QM(df = 3) = 1103.6042, p-val < .0001
##
```

```
## Model Results:
##
##
##               estimate      se      zval      pval      ci.lb      ci.ub
## intrcpt          -0.1607  0.0667   -2.4094  0.0160   -0.2914  -0.0300
## poly(c_treattemp, degree = 3, raw = TRUE)1  -0.2229  0.0073  -30.3953 <.0001   -0.2373  -0.2085 **
## poly(c_treattemp, degree = 3, raw = TRUE)2  -0.0009  0.0005   -1.7016  0.0888   -0.0020   0.0001
## poly(c_treattemp, degree = 3, raw = TRUE)3    0.0005  0.0000   13.3619 <.0001    0.0004   0.0006 **
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Below is a bubble plot of the fitted cubic model to the subsetted data.



Modelling response with binned treatment temperatures

Lastly, we rerun the model where our effect sizes are categorised into one seven bins depending on the treatment temperature. The bins and the number of effect sizes in each bin of the new subsetted data are given below

```
table(sdata$bin.temp)
```

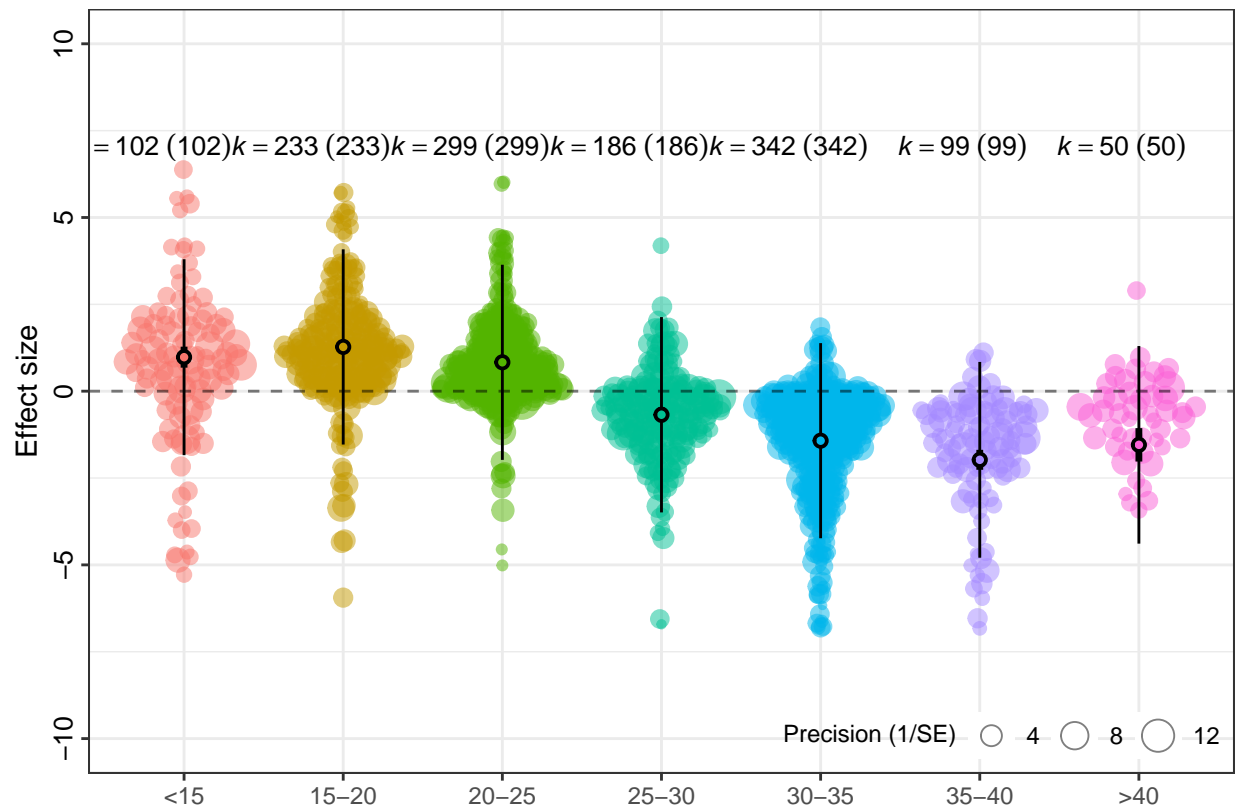
```
##
##  <15  >40 15-20 20-25 25-30 30-35 35-40
##  102   50  233  299  186  342   99
```

```
meta_sa_bintemp <- rma.mv(es, VCV_shared_sa, mod = ~bin.temp -
  1, random = list(~1 | study_code, ~1 | obs), test = "t",
  dfs = "contain", data = sdata, method = "REML")
```

```
summary(meta_sa_bintemp)
```

```
##
## Multivariate Meta-Analysis Model (k = 1311; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -2325.8125  4651.6249  4669.6249  4716.1837  4669.7640
##
## Variance Components:
##
```

```
##          estim      sqrt  nlvls  fixed      factor
## sigma^2.1 0.5448  0.7381   313     no  study_code
## sigma^2.2 1.5006  1.2250  1311     no           obs
##
## Test for Residual Heterogeneity:
## QE(df = 1304) = 29286.7914, p-val < .0001
##
## Test of Moderators (coefficients 1:7):
## QM(df = 7) = 1016.8182, p-val < .0001
##
## Model Results:
##
##          estimate      se      zval      pval      ci.lb      ci.ub
## bin.temp<15      0.9793  0.1540    6.3583 <.0001    0.6774    1.2812 ***
## bin.temp>40     -1.5440  0.2451   -6.2987 <.0001   -2.0245   -1.0636 ***
## bin.temp15-20     1.2743  0.0991   12.8569 <.0001    1.0800    1.4685 ***
## bin.temp20-25     0.8315  0.0895    9.2937 <.0001    0.6561    1.0068 ***
## bin.temp25-30    -0.6775  0.1094   -6.1911 <.0001   -0.8920   -0.4630 ***
## bin.temp30-35    -1.4249  0.0851  -16.7388 <.0001   -1.5918   -1.2581 ***
## bin.temp35-40    -1.9753  0.1467  -13.4636 <.0001   -2.2629   -1.6878 ***
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```



Other fixed effects

Habitat

```
meta_bintemp_habitat <- rma.mv(es, VCV_shared, mod = ~bin.temp *  
  Habitat, random = list(~1 | study_code, ~1 | obs), test = "t",  
  dfs = "contain", data = rdata, method = "REML")
```

```
summary(meta_bintemp_habitat)
```

```
##  
## Multivariate Meta-Analysis Model (k = 1381; method: REML)  
##  
##      logLik      Deviance      AIC      BIC      AICc  
## -3265.9266    6531.8532    6571.8532    6676.2020    6572.4791  
##  
## Variance Components:  
##  
##      estim      sqrt  nlvls  fixed      factor  
## sigma^2.1  0.5992  0.7741   316      no  study_code  
## sigma^2.2  5.0846  2.2549  1381      no      obs  
##  
## Test for Residual Heterogeneity:  
## QE(df = 1363) = 43855.7469, p-val < .0001  
##  
## Test of Moderators (coefficients 2:18):  
## QM(df = 17) = 638.3230, p-val < .0001  
##  
## Model Results:  
##  
##              estimate      se      zval      pval      ci.lb      ci.ub  
## intrcpt          -0.3772  1.4059   -0.2683  0.7885   -3.1327   2.3783  
## bin.temp>40        -3.1879  0.4479   -7.1176 <.0001   -4.0657  -2.3100 ***  
## bin.temp15-20       2.2452  1.5226    1.4745  0.1403   -0.7391   5.2295  
## bin.temp20-25       1.9369  1.6151    1.1993  0.2304   -1.2286   5.1025  
## bin.temp25-30      -1.6073  1.5002   -1.0714  0.2840   -4.5478   1.3331  
## bin.temp30-35      -0.3674  1.7231   -0.2132  0.8312   -3.7446   3.0099  
## bin.temp35-40      -3.8301  0.3435  -11.1515 <.0001   -4.5033  -3.1570 ***  
## HabitatBoth        -4.2719  2.2027   -1.9393  0.0525   -8.5891   0.0454 .  
## HabitatTerrestrial  1.4782  1.4285    1.0348  0.3008   -1.3216   4.2780  
## bin.temp15-20:HabitatBoth  2.2173  2.3714    0.9350  0.3498   -2.4306   6.8652  
## bin.temp20-25:HabitatBoth  2.8733  2.4362    1.1794  0.2382   -1.9015   7.6481  
## bin.temp25-30:HabitatBoth  3.7067  2.4508    1.5124  0.1304   -1.0968   8.5101  
## bin.temp30-35:HabitatBoth  1.5093  2.4692    0.6112  0.5410   -3.3303   6.3488  
## bin.temp35-40:HabitatBoth  3.6738  2.8372    1.2948  0.1954   -1.8871   9.2346  
## bin.temp15-20:HabitatTerrestrial -1.4870  1.5509   -0.9588  0.3377   -4.5267   1.5527  
## bin.temp20-25:HabitatTerrestrial -1.9102  1.6402   -1.1647  0.2442   -5.1249   1.3044  
## bin.temp25-30:HabitatTerrestrial -0.1171  1.5319   -0.0764  0.9391   -3.1195   2.8854  
## bin.temp30-35:HabitatTerrestrial -2.4634  1.7461   -1.4108  0.1583   -5.8858   0.9590  
##  
## ---  
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Geary's Test

Here we use Geary's test to determine which effect sizes to remove. According to Hedges et al 1999, effect sizes are deemed valid and accurate when the standardized mean of either the control or the treatment group is ≥ 3 . In other words, if

$$\frac{\bar{X}}{SD}\sqrt{N} \geq 3$$

As a sensitivity analysis we will determine which of our effect sizes do not meet this threshold i.e. are < 3 . We then rerun our best fitting models with these effect sizes removed.

```
# Read in data with calculated standardised mean
gearydata <- read.csv("Data/Gearys_test_data.csv")

# view the first fews rows of selected columns
gearydata %>%
  dplyr::select(Experiment.code, Trait.category, reftemp, treattemp,
    es, v, gtest) %>%
  head(., 30)

# How many effect sozes have a standardised mean < 3 ?
outliers_lon <- subset(gearydata, Trait.category == "Longevity" &
  gtest < 3) #82
outliers_sur <- subset(gearydata, Trait.category == "Survival" &
  gtest < 3) # 3
outliers_rep <- subset(gearydata, Trait.category == "Reproduction" &
  gtest < 3) # 171
```

There are 256 effect sizes in total that have a $gtest < 3$. 82 of these are longevity effect sizes, 3 are survival and 171 are reproduction.

We remove these effect sizes and rerun the best fitting model from earlier which is the model `meta_trait_treat2`, where temperature is modelled as a quadratic effect on reproduction.

Treatment temperature as a quadratic effect with Geary's outliers removed.

```
# Remove outliers
s.gearydata <- gearydata %>%
  subset(gtest > 3)

# Resubset reproduction dat
gdata_lon_warm <- subset(s.gearydata, Trait.category == "Longevity" &
  warm.cool == "Warm")
gdata_lon_cool <- subset(s.gearydata, Trait.category == "Longevity" &
  warm.cool == "Cool")

all_gdata <- rbind(gdata_lon_warm, gdata_lon_cool)

all_gdata <- all_gdata %>%
  mutate(c_treattemp = treattemp - 25)

### Create random factors into data frame
all_gdata$obs <- factor(c(1:nrow(all_gdata))) # Unique observation code
all_gdata$study_code <- factor(all_gdata$Paper.code) # Model requires column names study_code
```

```

all_gdata$Species.phylo <- factor(all_gdata$Species.latin) # Species names for phylo matrix
all_gdata$species <- factor(all_gdata$Species.latin) # Another species column for random factor

precision <- sqrt(1/all_gdata$v) # inverse standard error
all_gdata[, "precision"] <- precision

# recalculate v matrix
all_gdata$shared_control <- factor(all_gdata$Effect.size.code)
VCV_shared_ga <- impute_covariance_matrix(vi = all_gdata$v, cluster = all_gdata$shared_control,
r = 0.5)

meta_ga_treat3 <- rma.mv(es, VCV_shared_ga, mod = ~poly(c_treattemp,
degree = 3, raw = TRUE), random = list(~1 | study_code, ~1 |
obs), test = "t", dfs = "contain", data = all_gdata, method = "REML")

summary(meta_ga_treat3)

##
## Multivariate Meta-Analysis Model (k = 1310; method: REML)
##
##      logLik      Deviance      AIC      BIC      AICc
## -3195.6635    6391.3269    6403.3269    6434.3753    6403.3916
##
## Variance Components:
##
##      estim      sqrt  nlvls  fixed      factor
## sigma^2.1  0.6337  0.7960    311      no  study_code
## sigma^2.2  5.1381  2.2667   1310      no      obs
##
## Test for Residual Heterogeneity:
## QE(df = 1306) = 44067.8973, p-val < .0001
##
## Test of Moderators (coefficients 2:4):
## F(df1 = 3, df2 = 1306) = 215.6771, p-val < .0001
##
## Model Results:
##
##              estimate      se      tval      df      pval      ci.lb      ci.ub
## intrcpt          -0.0537  0.1011   -0.5316    307  0.5954   -0.2526    0.1441
## poly(c_treattemp, degree = 3, raw = TRUE)1  -0.3003  0.0130  -23.1438   1306 <.0001   -0.3258   -0.2748
## poly(c_treattemp, degree = 3, raw = TRUE)2  -0.0030  0.0009   -3.4836   1306  0.0005   -0.0047   -0.0013
## poly(c_treattemp, degree = 3, raw = TRUE)3    0.0007  0.0001   10.5347   1306 <.0001    0.0005    0.0009
##
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

```

Below is a bubble plot of the fitted quadratic model.

