Dose-Time Response Modeling of Rat Brain AChE Activity: Propoxur Inhalation Exposure

June 23, 2005

1 Preamble

Here is some code to set up the analysis: loading required libraries and datasets, and defining some functions.

First, `CarbamateData` loads the full dataset for this risk assessment, and causes the library `DRUtils` to be loaded.

```
> library(CarbamateData)

Loading required package: DRUtils
Loading required package: nlme
Loading required package: tcltk
```

Set up lattice to use B&W instead of color:

```
> library(lattice)
> ltheme <- canonical.theme(color = FALSE)
> ltheme$strip.background$col <- "transparent"
> lattice.options(default.theme = ltheme)
```

Use package `Hmisc` for some formatting support.

```
> library(Hmisc)

Hmisc library by Frank E Harrell Jr

Type library(help='Hmisc'), ?Overview, or ?Hmisc.Overview')
to see overall documentation.

NOTE:Hmisc no longer redefines [.factor to drop unused levels when
subsetting.  To get the old behavior of Hmisc type dropUnusedLevels().

Attaching package 'Hmisc':


        The following object(s) are masked from package:stats :

         ecdf
```

The rat gavage data for this analysis are in `PadillaData`. The following code prints out documentation for the datasets in use:

```
> printDataDoc(PadillaData)
```

```
----------------------------------------
Data set: PadillaData

Dataset creation date: Wed Jun 22 12:07:28 2005
Script name: /home/setzer/tasks/CarbamateCRA/EmpiricalDoseResponsesFinal/Data/getdata.R
Script last modified: 2005-06-22 12:05:47
sysname: Linux
release: 2.4.24-openmosix2
version: #1 Wed Jul 28 14:33:52 CEST 2004
nodename: node24.cluster2.org
machine: i686
login: unknown
user: setzer
----------------------------------------
```

The following function turns out to be quite useful on subsetted dataframes. It just eliminates unused levels of all factors in the data frame:

```
> CleanUp <- function(x) {
+     for (nm in names(x)) {
+         if (is.factor(x[, nm]))
+             x[, nm] <- factor(x[, nm])
+     }
+     x
+ }
```

To get starting values, we often have to extract values from a previously fit model. The following function simplifies that. The argument `what` is a regular expression:

```
> getParms <- function(what, Par) {
+     Par[grep(what, names(Par))]
+ }
```

This script is for modeling the dose-time response for rat brain. via gavage dosing. It includes an acute study that includes a time course at one dose and a dose-response at around the time of peak effect.

All the data used for this DR model are in `AggData`.

Set up the analysis dataset (the data were inadvertantly duplicated in `AggData`. The second copy has NA for `tmpstds`, which serves as a convenient flag for deleting the second copy. `tmonstdy` differs between males and females by a week. To simplify the modeling, combine the pairs into a single level.

```
> dta <- CleanUp(subset(AggData, chemical %in% "PROPOXUR" & dsmtd ==
+     "INHALATION" & !is.na(tmpstds) & sctn == "WHOLEBRAIN" & !is.na(cheact),
+     select = c("dose", "cheact", "sd", "n", "sex", "tmonstdy",
+         "species", "mrid")))
> dta$ftmonstdy <- factor(dta$tmonstdy)
> lvls <- levels(dta$ftmonstdy)
> lvls[c(1, 3, 5)] <- lvls[c(2, 4, 6)]
> dta$ftm2 <- dta$ftmonstdy
> levels(dta$ftm2) <- lvls
> idta <- with(dta, PhonyDF(dose, n, cheact, sd, "dose", "cheact",
+     dta[, c("sex", "ftm2")]))
```

Summary of this dataset:

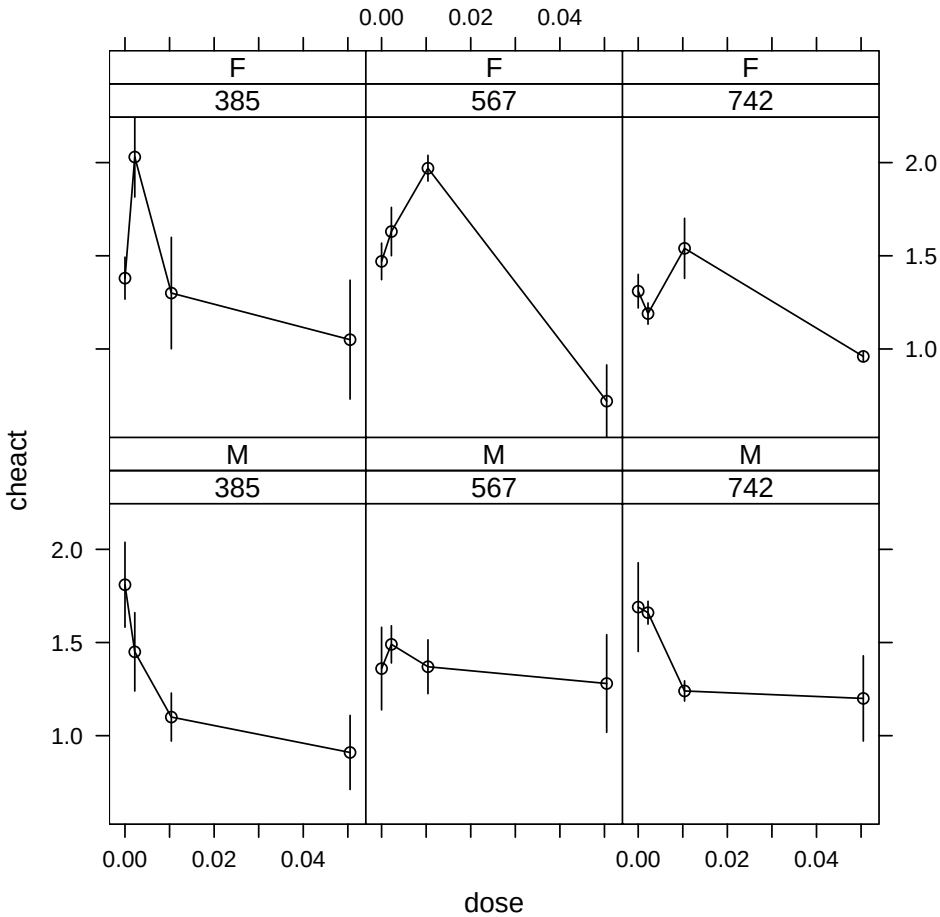
```
> summary(dta)
```

```
      dose             cheact            sd               n         sex
 Min.   :0.00000   Min.   :0.720   Min.   :0.0300   Min.   :4.000   F:12
 1st Qu.:0.00165   1st Qu.:1.198   1st Qu.:0.1098   1st Qu.:5.000   M:12
 Median :0.00630   Median :1.365   Median :0.1745   Median :5.000
 Mean   :0.01578   Mean   :1.380   Mean   :0.1798   Mean   :4.875
 3rd Qu.:0.02042   3rd Qu.:1.562   3rd Qu.:0.2547   3rd Qu.:5.000
 Max.   :0.05050   Max.   :2.030   Max.   :0.3640   Max.   :5.000
    tmonstdy     species        mrid    ftmonstdy  ftm2
 Min.   :378.0   RAT:24   42648001:24   378:4     385:8
 1st Qu.:385.0                          385:4     567:8
 Median :563.5                          560:4     742:8
 Mean   :561.2                          567:4
 3rd Qu.:735.0                          735:4
 Max.   :742.0                          742:4
```

```
> with(dta, table(dose, tmonstdy))
```

```
        tmonstdy
dose     378 385 560 567 735 742
  0      1   1   1   1   1   1
  0.0022 1   1   1   1   1   1
  0.0104 1   1   1   1   1   1
  0.0505 1   1   1   1   1   1
```

Plots of dose-response for each sex and time point, with 95% confidence intervals.



2 Dose-Response Modeling

From the figure above, it looks as if there is a lot of variability around whatever dose-response there is, and little consistency in dose-response shape. Fit a linear model to log cheact in dose, and allow the slope to vary among times.

```
> idta$lncheact <- log(idta$cheact)
> drmod1 <- lme(lncheact ~ sex:ftm2 - 1 + dose + sex:dose, data = idta,
+     random = ~dose - 1 | ftm2)
> summary(drmod1)

Linear mixed-effects model fit by REML
 Data: idta
       AIC      BIC   logLik
  10.16805 37.08152 4.915977

Random effects:
 Formula: ~dose - 1 | ftm2
            dose  Residual
StdDev: 1.534347 0.2193163
```

```
Fixed effects: lncheact ~ sex:ftm2 - 1 + dose + sex:dose
                  Value Std.Error  DF   t-value p-value
dose         -11.060385 1.6582000 107 -6.670115   0.000
sexF:ftm2385   0.507345 0.0611741 107  8.293462   0.000
sexM:ftm2385   0.344153 0.0556829 107  6.180590   0.000
sexF:ftm2567   0.476361 0.0557206 107  8.549112   0.000
sexM:ftm2567   0.417491 0.0556686 107  7.499576   0.000
sexF:ftm2742   0.362234 0.0557206 107  6.500910   0.000
sexM:ftm2742   0.435199 0.0556686 107  7.817678   0.000
sexM:dose      4.450249 1.9716771 107  2.257088   0.026
 Correlation:
             dose   sF:238 sM:238 sF:256 sM:256 sF:274 sM:274
sexF:ftm2385 -0.348
sexM:ftm2385 -0.003  0.074
sexF:ftm2567 -0.334  0.127 -0.033
sexM:ftm2567  0.002 -0.037  0.119  0.069
sexF:ftm2742 -0.334  0.127 -0.033  0.122 -0.035
sexM:ftm2742  0.002 -0.037  0.119 -0.035  0.120  0.069
sexM:dose    -0.601  0.293 -0.274  0.281 -0.278  0.281 -0.278

Standardized Within-Group Residuals:
        Min          Q1         Med          Q3         Max
-3.07397309 -0.62762065  0.02125666  0.60609957  2.45860275

Number of Observations: 117
Number of Groups: 3
```

There is clearly a dose-response here, and possibly difference in slopes between the sexes. To facilitate calculating BMDs, etc. refit with a different parameterization (a separate slope for each sex):

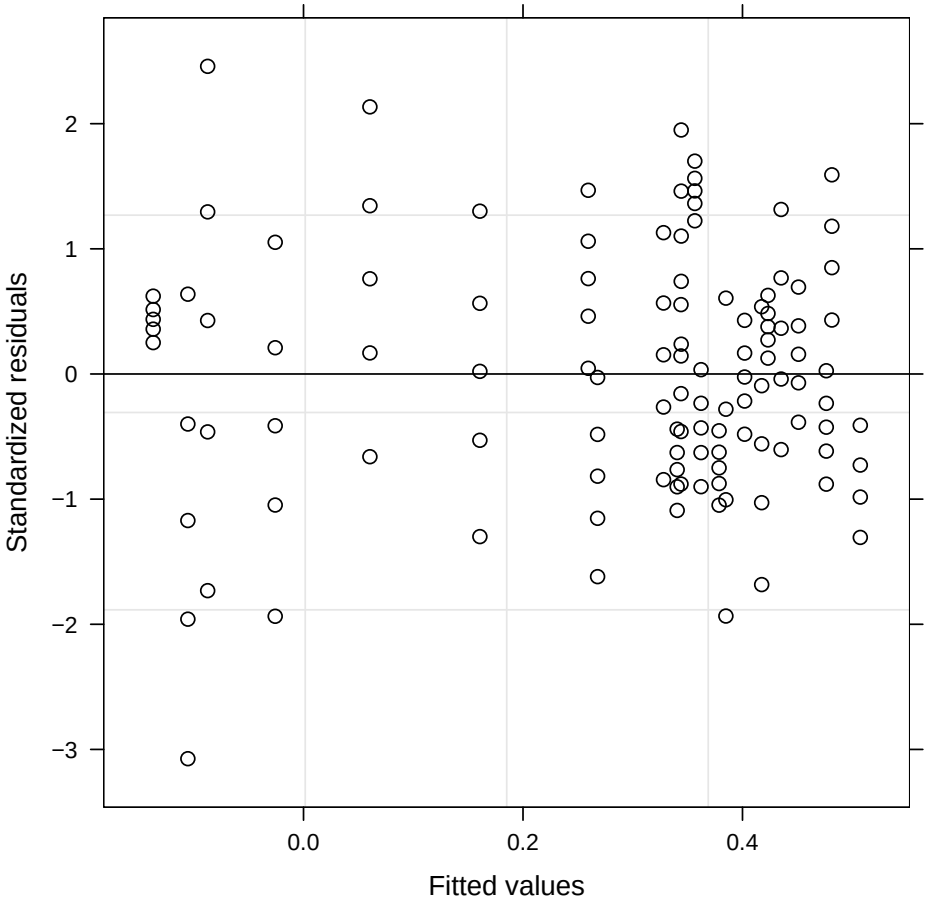
```
> drmod2 <- lme(lncheact ~ sex:ftm2 - 1 + sex:dose, data = idta,
+     random = ~dose - 1 | ftm2)
> summary(drmod2)

Linear mixed-effects model fit by REML
 Data: idta
       AIC      BIC   logLik
  10.16805 37.08152 4.915977

Random effects:
 Formula: ~dose - 1 | ftm2
            dose  Residual
StdDev: 1.534347 0.2193163

Fixed effects: lncheact ~ sex:ftm2 - 1 + sex:dose
                  Value Std.Error  DF   t-value p-value
sexF:ftm2385   0.507345 0.0611741 107  8.293462   0e+00
sexM:ftm2385   0.344153 0.0556829 107  6.180590   0e+00
sexF:ftm2567   0.476361 0.0557206 107  8.549112   0e+00
sexM:ftm2567   0.417491 0.0556686 107  7.499576   0e+00
sexF:ftm2742   0.362234 0.0557206 107  6.500910   0e+00
sexM:ftm2742   0.435199 0.0556686 107  7.817678   0e+00
sexF:dose    -11.060385 1.6582000 107 -6.670115   0e+00
sexM:dose     -6.610136 1.6454071 107 -4.017325   1e-04
 Correlation:
```

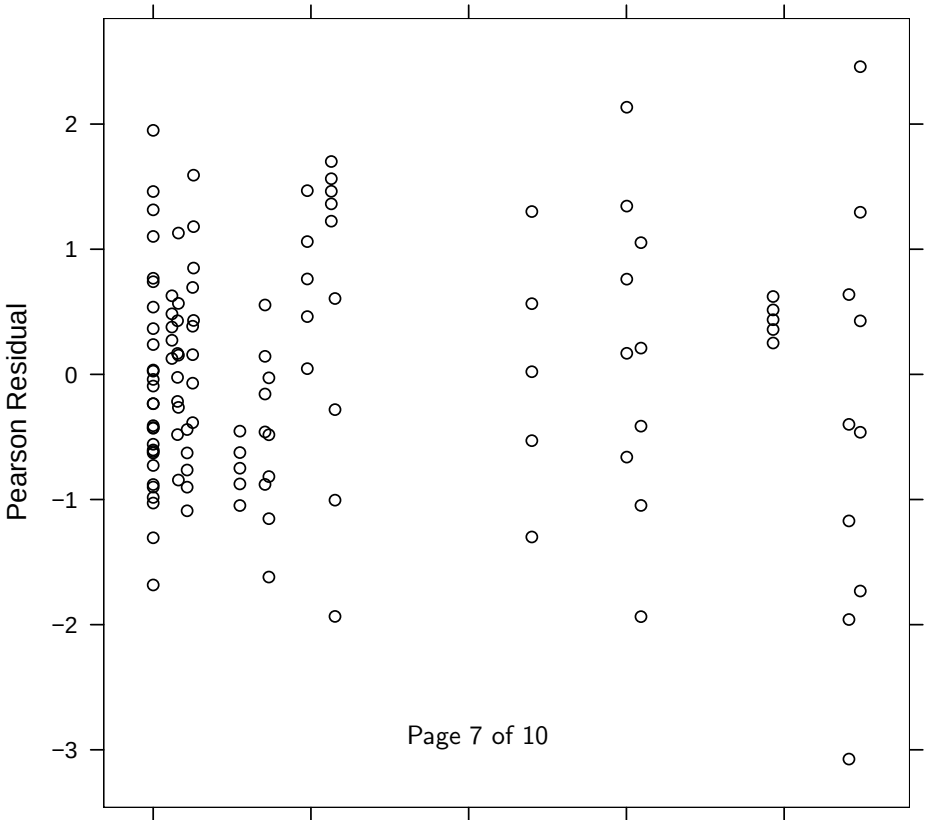
```
             sF:238 sM:238 sF:256 sM:256 sF:274 sM:274 sxF:ds
sexM:ftm2385  0.074
sexF:ftm2567  0.127 -0.033
sexM:ftm2567 -0.037  0.119  0.069
sexF:ftm2742  0.127 -0.033  0.122 -0.035
sexM:ftm2742 -0.037  0.119 -0.035  0.120  0.069
sexF:dose    -0.348 -0.003 -0.334  0.002 -0.334  0.002
sexM:dose     0.000 -0.331  0.000 -0.331  0.000 -0.331  0.288

Standardized Within-Group Residuals:
        Min          Q1         Med          Q3         Max
-3.07397309 -0.62762065  0.02125666  0.60609957  2.45860275

Number of Observations: 117
Number of Groups: 3
```

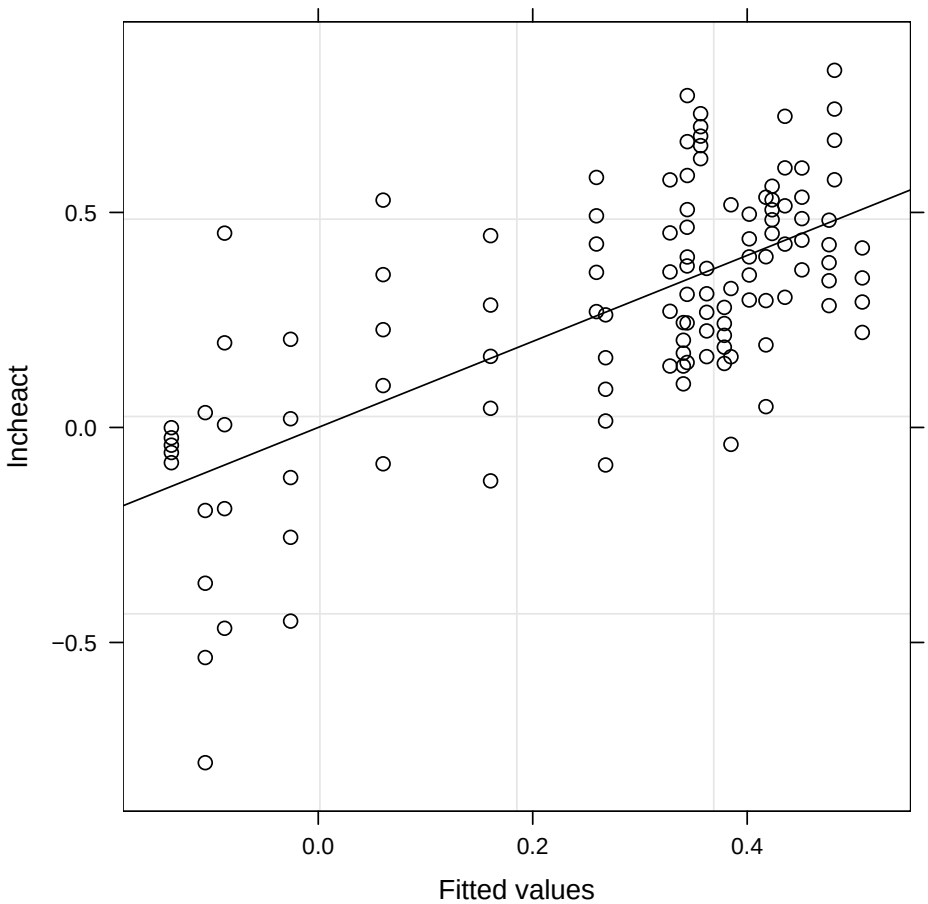
We will use the results in `drmod2` to estimate BMDs.

3 Model Summary

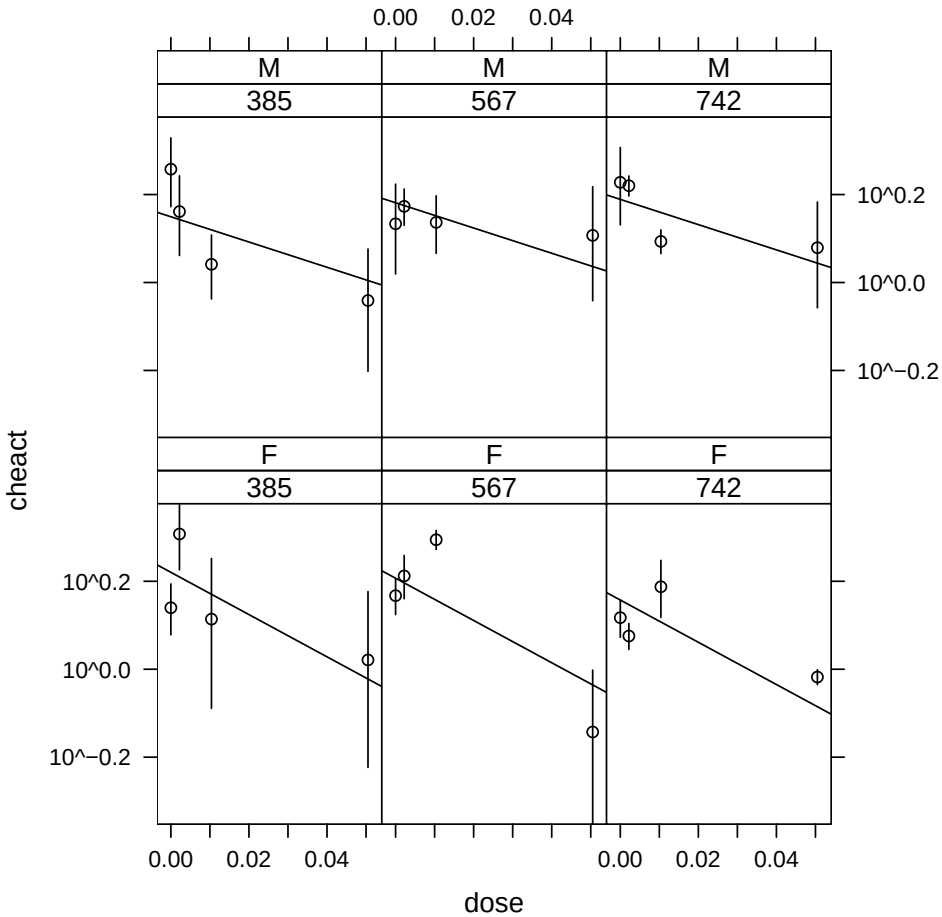
Diagnostic plots for this model:


Standardized residuals
Fitted values
2
1
0
−1
−2
−3
0.0
0.2
0.4


Pearson Residual
2
1
0
−1
−2
−3



The following figure shows the mean AChE values and their 95% confidence intervals, and the fitted line from `drmod2`.



4 Calculating BMDs

We have essentially fit the model

$$Y = Ae^{m \times dose}$$

with a constant coefficient of variation model for the error variance. In this model, for a BMD that results in BMR$\times$100% inhibition, the BMD is calculated as:

$$\frac{log(1 - BMR)}{m}$$

For consistency with the other endpoints and models, and to be able to calculate RPFs with confidence intervals, we need log BMD and its standard error. log BMD is just the log of the above quantity, and its standard error is, based on the delta method:

$$\hat{\sigma}_{lD} = \frac{\hat{\sigma}_m}{m}$$

The following code calculates the BMDs, etc.

```
> tTab <- summary(drmod2)$tTable
> Ints <- intervals(drmod2, which = "fixed")$fixed
> Ints90 <- intervals(drmod2, which = "fixed", level = 0.9)$fixed
> MBMD <- log(1 - 0.1)/Ints["sexM:dose", "est."]
> MBMDL <- log(1 - 0.1)/Ints90["sexM:dose", "lower"]
```

```
> MBMD.CI <- log(1 - 0.1)/Ints["sexM:dose", c("lower", "upper")]
> MlD <- log(MBMD)
> MlD.se <- tTab["sexM:dose", "Std.Error"]/tTab["sexM:dose", "Value"]
> FBMD <- log(1 - 0.1)/Ints["sexF:dose", "est."]
> FBMDL <- log(1 - 0.1)/Ints90["sexF:dose", "lower"]
> FBMD.CI <- log(1 - 0.1)/Ints["sexF:dose", c("lower", "upper")]
> FlD <- log(FBMD)
> FlD.se <- tTab["sexF:dose", "Std.Error"]/tTab["sexF:dose", "Value"]
```

5 Summary

The critical estimates from this analysis are listed below. They are printed with greater than usual precision, in case they are to be used in further computation. For reporting, round to two or three significant digits. BMD has units mg/m^3.

species RAT

mrid `[1] "42648001"`

Female `lD` **(standard error)** -4.6537371420051 (-0.149922448391222)

Male `lD` **(standard error)** -4.13897150632694 (-0.248921839773616)

Female BMD (95% CI) 0.0095259355530021 (0.00734343808141065, 0.0135543349923242)

Male BMD (95% CI) 0.0159392364810191 (0.0106727011926277, 0.0314667938820087)

Female BMDL, the one-sided lower 95% CL 0.00762835103001739

Male BMDL, the one-sided lower 95% CL 0.0112802950155485

Save everything:

```
> save.image(file = "InhalationRatBrainDR.RData")
```

Save the results for incorporating into a database:

```
> propoxur.inhalation.brain <- list(mrid = levels(dta$mrid), species = levels(dta$species),
+     BMDs = list(female = list(lD = FlD, lD.se = FlD.se, BMD = FBMD,
+         BMD.CI = FBMD.CI, BMDL = FBMDL), male = list(lD = MlD,
+         lD.se = MlD.se, BMD = MBMD, BMD.CI = MBMD.CI, BMDL = MBMDL)))
> save(propoxur.inhalation.brain, file = file.path("..", "..",
+     "01Summaries", "propoxur.inhalation.brain.RData"))
```