Model Based Statistics in Biology.
Part V. The General Linear Model.
Chapter 17.6 Model Revision

ReCap. Part I (Chapters 1,2,3,4), Part II (Ch 5, 6, 7)
ReCap Part III (Ch 9, 10, 11), Part IV (Ch13, 14)
17 Poisson Response Variables
17.1 Poisson Regression
17.2 Single Categorical Explanatory Variable (Log-linear Model)
17.3 Single Categorical Explanatory Variable (Sensitivity Analysis)
17.4 Two or More Categorical Explanatory Variables
17.5 Poisson ANCOVA
17.6 Model Revision

Ch17.xls

on chalk board

ReCap Part I (Chapters 1,2,3,4) Quantitative reasoning
ReCap Part II (Chapters 5,6,7) Hypothesis testing and estimation
ReCap (Ch 9, 10,11) The General Linear Model with a single explanatory variable.
ReCap (Ch 12,13,14,15) GLM with more than one explanatory variable
ReCap (Ch 16,17)

Today: Model revision for Poisson regression

Wrap-up.

Example: biological assay.

Does a suspected toxin reduce the fecundity (brood size) of the assay organism *Ceriodaphnia dubia* ?

Assay data by Bailer and Oris taken from:

Nicholas Lange, Louise Ryan and Lynne Billard, David Brillinger, Loveday Conquest, Joel Greenhouse. 1994 *Case Studies in Biometry* John Wiley & Sons, Inc.
Preliminary computations: proportions and variances.

We begin by computing the mean and variance in fecundity (brood size) at each dose.

Brood 3 statistics

Conc	0	80	160	235	310
Mean	13.90	14.80	11.50	6.70	0.00
Var	4.77	3.07	0.94	8.68	0.00
Var/Mean	0.34	0.21	0.08	1.30	

It is evident that the variance is not a fixed value.
Instead of assuming homogeneous variances (as with GLM), we are going to assume that the residuals arise from a Poisson distribution (GzLM with Poisson response).

1. Model

Verbal model.
Count is the number of offspring in the third brood of each *C. dubia*.
Conc is the concentration of the some substance (micrograms/L)

Graphical model

Plot of fecundity versus toxin concentration shows a curvilinear relation.

Response variable: Count
Explanatory variable: experimentally fixed concentration of substance Conc
Explanatory variable has multiplicative effect fecundity.

Write formal model as a multiplicative effect $Count = e^{(\mu)} + : Poisson\ error$

$$\mu = \beta_o + \beta_{Conc} \cdot Conc$$

The link between the Count and the structural model μ is: $\ln(Count) = \mu$

This avoids negative predicted values of fecundity

2. Execute analysis.

Place data in model format:

Column labelled Count, with response variable # of animals

Column labelled Conc, with explanatory variable concentration

```
data A;
  input Conc Count;
cards;
0        10
0        15
0        17
0        15
0        15
0        15
0        15
0        12
0        11
0        14
80       16
80       16    etc
;
```

SAS command file

In a package with spreadsheet format, there will be two columns (variables) and 50 rows for this data set.

Code the GzLM model statement in statistical package

$Count = e^{(\mu)} + : Poisson\ error$

$\mu = \beta_o + \beta_{Conc} \cdot Conc$

```
Proc Genmod;
  Model Count = Conc/
  Link=log dist=poisson type1 type3;
```

SAS command file

```
MTB >
```

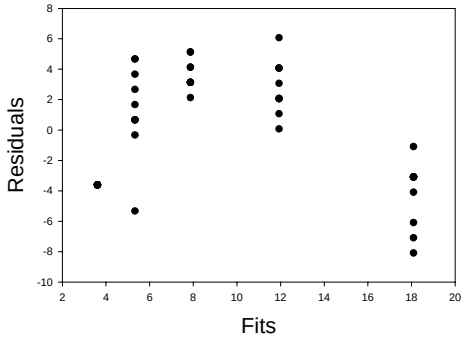
Minitab command lines

Click Stat

Minitab sequence to produce line commands

3. Evaluate model

Conclusion: Revise model.
Use concentration as a categorical variable.
Return to step 2.



2. Execute analysis.

```
Proc Genmod;
 Class Conc;
  Model Count = Conc/
  Link=log dist=poisson type1 type3;
```

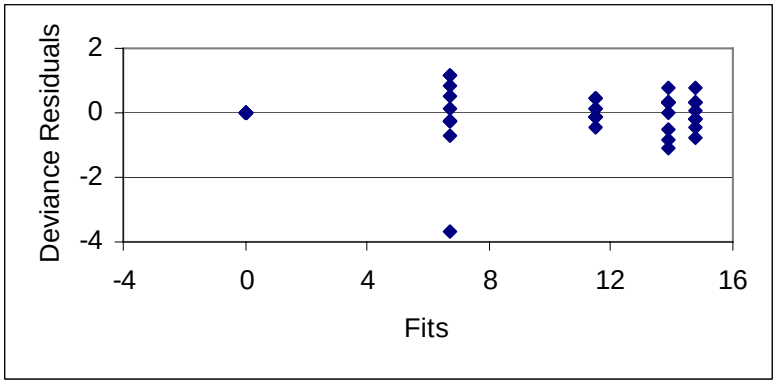
SAS command file

3. Evaluate model

a. No need to evaluate straight line assumption because no straight line was fit.

b. Residuals homogeneous ?
Yes, except for outlier.

Later, we will evaluate the sensitivity of the results to this outlier.



4. State population and whether the sample is representative.

Population.

All possible outcomes if the experiment were repeated on *C. dubia* with the same toxin and experimental protocol.

5. Decide on mode of inference. Is hypothesis testing appropriate?
Yes, as we wish to declare overall yes/no decision about effect of aflatoxin on brood size.

6. State Ho/HA pair (some analyses may require several pairs).
State test statistic, its distribution (t or F), and tolerance of Type I error.

HA: $\beta_{Conc} \neq 0$ hence: $Count = e^{\left(\beta_o + \beta_{Conc} \cdot Conc\right)} \neq constant$

H0: $\beta_{Conc} = 0$ hence: $Count = e^{\left(\beta_o\right)} = constant$

Tolerance for Type I error. $\alpha = 5\%$

7. Calculate change in fit (ΔG) due to explanatory variables.
For the Generalized Linear Model, step 6 is modified: we calculate the change in deviance ΔG rather than the SS for each term in the model.

Here is the output from the SPlus package.

```
     Df Deviance Resid. Df Resid. Dev
NULL                    49   269.7944
Conc  4  246.108         45    23.6864
```

SPlus output

The improvement in fit is ΔG = 269.79 – 246.11 = 23.69 on 4 df.
Calculate p-value from Chisquare distribution.

Is the change in fit ΔG better than by chance ?
The p-value reported for ΔG = 23.69 is p = 0.000092
The p-value is small, hence ΔG is too large to be due to chance.
For generalized linear models, we compute a p-value on ΔG, not on the deviance itself G.

8. Evaluate results if assumptions not met. Recompute estimates and p-values if necessary.
The p-value is far from the criterion of significance so the presence of the outlier is unlikely to lead to an incorrect decision. We check this judgement by running the analysis without the outlier.

```
     Df Deviance Resid. Df Resid. Dev
NULL                    48   250.8443
Conc  4 241.2769         44     9.5674    0.048
```

SPlus output

The decision was unchanged but there was a substantial change in the p-value. Our experience with p-values from F-ratios, which rarely change by a factor of 5, did not apply here.

9. Declare decision. p = 0.048 hence reject H_o and accept H_A

The substance is toxic, causing a reduction of fecundity as concentration increases.

10. Analysis of parameters.

Here is the SAS output.

Parameter		DF	Estimate	Standard Error	Wald 95% Confidence Limits		Chi-Square	Pr > ChiSq
Intercept		1	-22.9644	0.1222	-23.2039	-22.7250	35333.4	<.0001
conc	0	1	25.5963	0.1487	25.3048	25.8878	29619.4	<.0001
conc	80	1	25.6590	0.1472	25.3704	25.9476	30365.4	<.0001
conc	160	1	25.4068	0.1537	25.1055	25.7080	27327.5	<.0001
conc	235	0	24.9719	0.0000	24.9719	24.9719	.	.
conc	310	0	0.0000	0.0000	0.0000	0.0000	.	.
Scale		0	1.0000	0.0000	1.0000	1.0000		

SAS output

Te estimates and confidence limits are converted back to predicted values via the link function. For example:

$$\exp(-22.9644 + 25.5963) = 13.9$$

This is the predicted value of brood size at zero concentration of the toxin.
Here are the expected or predicted values for each concentration.

Parameter		DF	Estimate	Standard Error	Wald 95% Confidence	Limits
Intercept		1	-22.96	0.1222	-23.20	-22.73
conc	0	1	13.90	0.1487	8.17	23.64
conc	80	1	14.80	0.1472	8.73	25.09
conc	160	1	11.50	0.1537	6.70	19.75
conc	235	0	7.44	0	5.86	9.46
conc	310	0	0	0	0	0

SAS output

The estimates are statistically indistinguishable at 0, 80, and 160 micrograms/L.
Brood size drops at 235 micrograms/L.
Brood size at 310 micrograms/L differs substantially from brood size at low concentrations of the toxin.