

## Model Based Statistics in Biology.

### Part V. The Generalized Linear Model.

#### Chapter 17.3 Single Categorical Explanatory Variable (Sensitivity Analysis)

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) GLM with more than one explanatory variable

**ReCap** (Ch 15) GLM review

**ReCap** (Ch 16)The generalized linear model.

**ReCap** (Ch 17) Regression and one-way ANOVA with Poisson errors.

Today: Power of a test. Poisson response variable with single categorical explanatory variable.

#### Wrap-up.

G-statistics measures goodness of fit of data to model, for any design.

We looked at two way classification (two nominal scale explanatory variables)

The power  $(1-\beta)=(1-\text{Type II error})$  is not worth calculation after analysis.

It is of interest to compute

the difference that could have been detected, given the error and n  
or, the sample size require to detect a specified difference.

Type II statistical error. Sensitivity and Power Analysis  
Standard practice in biology has been to report Type I error only, with no consideration of Type II error. One reason for this is that many applications are for nominal  $H_a$  for which Type II error (sensitivity) cannot be calculated easily, if at all. Generally one needs a specific  $H_a$  in order to calculate  $\beta$  and state sensitivity.

The tolerance for type I error  $\alpha$  is often set at 5% in biology.

Some rules of thumb about sensitivity.

Reducing the tolerance for type I error, aka  $\alpha$ , will increase

Type II error  $\beta$  if the number of observations  $n$  is fixed.

Tolerance of type I error  $\alpha$  often fixed at 5% not lower, for this reason.

An increase in number of measurements  $n$  decreases Type II error  $\beta$

This increase in number of measurements  $n$  increases the sensitivity of a test, allowing small contrasts can be detected.

Some tests have lower Type II error  $\beta$  than others:

For example, rescaling the quantity of interest to ranks typically reduces the sensitivity and raises type II error.

If the null hypothesis accepted we do not undertake power analysis (Hoenig)  
Instead we undertake a sensitivity analysis.

-Report detectable difference

-Calculate  $n$  required for specified difference

## Poisson frequencies: comparison of 4 proportions (Donax)

Analyses of poisson frequencies in biology are often made in a two-way classification of the response variable. In a two-way classification we

Donax data can be analyzed as poisson  
(Prey selection, as here)  
It could also be binomial  
(case-control estimate of selection intensity)  
Because each animal could either be eaten  
or not, with probability depending on colour.

are typically interested in the interaction term, rather than main effects. For example in the case of colour patterns in tiger beetles in four seasons (Sokal and Rohlf 1995 Box 17.8) we are not particularly interested in whether a different number of beetles were collected in each of the four seasons. We have no idea of whether equal effort was made to collect beetles in the four seasons, so it is of little interest to test whether the four frequencies differ from a 1:1:1:1 ratio. Similarly, we are not particularly interested in whether the proportion of Bright red and Not bright red differ from a 1:1 ratio in this particular case. Instead, we are interested in whether the proportions of bright red and not bright red beetles change with season. This is measured by an interaction term.

Another example. Do visual predators (birds) form search images and selectively remove common colour morphs from polymorphic prey populations. Predated and unpredated clams are collected from the beach, and sorted into colour categories.

The first explanatory variable is shell colour (dark, ray, tinge, white).  
The second explanatory variable is predation (yes or no).

This results in a 2 by 4 table.

The response variable is frequency of 488 Donax in these 8 cells.

We are interested in selective predation--in whether the relative frequency of shell colours taken by birds differs from that in the environment.

Draw picture



## Poisson frequencies (Donax)

The model is:

relative frequency of shell colours depends on whether or not we are looking at a predated or unpredated collection.

$$f = e^{\beta_{\text{prd}^* \text{clr}} N} + \text{residual}$$

$e^{\beta_{\text{prd}^* \text{clr}}}$  is the expected proportion of Donax in each of 4 colour categories and 2 states, predated by birds or not predated. This parameter will be estimated from the marginal totals. That is, we will use information about the number of Donax in the 4 categories, and about the total number predated or not predate, to estimate  $e^{\beta_{\text{prd}^* \text{clr}}}$  for each of the 8 observed frequencies that we have.

## Poisson frequencies (Donax continued)

If Two-way Poisson analysis covered (beetles) then skip ahead at this point to  
 $G = 4.16$      $p = 0.26$   
 accept  $H_0$

There are 8 observed frequencies

	Live	Predated	
Dark	24	4	28
Rays	118	35	153
Tinge	90	38	128
White	139	40	179
	371	117	488

The expected proportion of live donax, in the dark category, out of 488 Donax, is

$$e^{\beta_{\text{prd}^* \text{clr}}} = \frac{28}{488} \cdot \frac{371}{488} = 0.057 \cdot 0.76 = 0.044$$

In similar fashion, we estimate the other 7 values of the parameter  $e^{\beta_{\text{prd}^* \text{clr}}}$ . The 8 values of  $e^{\beta_{\text{prd}^* \text{clr}}}$  are

	Live	Predated
Dark	0.044	0.014
Rayed	0.238	0.075
Tinge	0.199	0.063
White	0.279	0.088

## Two-way G-test (continued)

We then use these to compute the fitted values  $e^{\beta}N$ , which appear here in 8 data equations.

$f$	$=$	$e^{\beta} \cdot N$	$+$	residual	$2 \ln L = 2 f \ln(f/e^{\beta}N)$
24	$=$	$0.044 \cdot 488$	$+$	2.71	5.76
118	$=$	$0.238 \cdot 488$	$+$	1.68	3.39
90	$=$	$0.199 \cdot 488$	$-$	7.73	-14.06
139	$=$	$0.279 \cdot 488$	$+$	.	5.89
4	$=$	$0.032 \cdot 488$		.	.
35	$=$	$0.026 \cdot 488$		.	.
38	$=$	$0.106 \cdot 488$		.	.
40	$=$	$0.084 \cdot 488$		.	.
$G = 2 \sum f \ln(f/e^{\beta}N)$					4.16

While these computations can be done in any statistical package, they are also easily completed in a spreadsheet.

The G-statistic is 4.16

The p-value for this G-statistic is computed from a Chi-square distribution with three degrees of freedom. The degrees of freedom are computed exactly the same as was the case with the interaction term in a two-way ANOVA. The degrees of freedom for the interaction term are calculated as the product of the degrees of freedom for the main effects:

$$df_{\text{prd} \cdot \text{clr}} = df_{\text{prd}} * df_{\text{clr}} = (2-1) * (4-1) = 3.$$

The Minitab command to compute the p-value is

More than 75.53% of the G-statistics obtained by chance will be less than our observed  $G = 4.16$ . And 24.47% will be greater. So we accept the null hypothesis that relative proportions of *Donax* colour morphs taken by birds matches that for the available prey. The symbol  $p_{\text{prd}|\text{clr}}$  is read "the portion predated, given the colour."

```
MTB > cdf 4.16;
SUBC> chisquare 3.

4.16    0.7553
```

$$H_0: p_{\text{prd}|\text{dark}} = p_{\text{prd}|\text{ray}} = p_{\text{prd}|\text{tinge}} = p_{\text{prd}|\text{white}}$$

We reject the alternative hypothesis

$$H_A: \text{ proportions are not equal.}$$

## Sensitivity Analysis

Skip ahead to here, if 2-way analysis (beetle example) covered in previous lecture.

Because we have accepted the null hypothesis, Type II error becomes of interest.

	$H_0$ True	$H_0$ False	Therefore
Accept $H_0$	ok	II	-->Reject $H_a$
Reject $H_0$	I	ok	-->Accept $H_a$

Zar (1996) gives many examples of computations of Type II error. However, these examples are usually for comparing two groups. I.e. comparing 2 means, or 2 proportions. Power (which is 1 - Type II error) is difficult to calculate if we have multiple groups.

The Donax analysis is an example. There are more than two groups, there are several coloration categories.

In these cases, it is of interest to look at the difference that could have been detected. It is also of interest to calculate the sample size needed to detect at specified difference.

In fact these questions are of more interest than calculating the Type II error rate.

The questions of detectable difference and adequate sample size arise when the null hypotheses (it is just due to chance) has been accepted.

To demonstrate this approach we will look at the the detectable difference for the Donax analysis, using an iterative method.

The difference between expected (no selective predation) and observed (weak effects if any) was 10 clams too many for Donax tinged with slight pink.

	Live	Predated	Expected	Deviation	Dev+	"Predated"
Dark	24	4	8	-4	-6	2
Rays	118	35	37	-2	-3	34
Tinge	90	38	28	+10	+10	43
White	139	40	44	-4	-6	38
	371	117	117	0	0	117

This number was successively increased until th G-statistic reached the criterion value where  $p < \alpha$ .

Dev0	Dev1	Dev2	Dev3	Dev4	Dev5
-4	-5	-5	-6	-6	-6
-2	-2	-2	-2	-2	-3
+10	+11	+12	+13	+14	+15
-4	-4	-5	-5	-6	-6

G-statistic rises as the deviation is increased.

When deviation reached +15,  
 $G = 10.37$   $df = 3$   $p = 0.0157$

It would have been possible to detect a deviation of 15 Donax "too many" in the tinged category.

The observed deviation was 10 out of 117 Donax or 9%

The detectable deviation was 15 out of 117 Donax of 13%

We can be sure that selection was less than 13%, given the sample size we were able to obtain.