Chapter 720

Probit Analysis

Introduction

Probit and logit analysis may be used for comparative LD_{50} studies for testing the efficacy of drugs designed to prevent lethality. This program module presents calculates power and sample size using the methodology outlined in Kodell, Lensing, Landes, Kumar, and Hauer-Jensen (2010).

Technical Details

Consider the following situation: suppose two, equal-sized groups of animals are exposed to the same death-causing agent such as radiation. The test group of animals is exposed to a countermeasure drug, while the control group is not. The study's objective is to test whether the radiation LD_{50} of the treatment group is significantly greater than that of the control group.

Probit and logit analysis are often used to study the relative potency of a test treatment over a control treatment. The probit and logit transformations are

$$Y = F^{-1}(P) = \beta_0 + \beta_1 \log_{10}(D)$$

where *F*(x) is the cumulative normal distribution for the probit analysis and the cumulative logistic distribution for the logit analysis, β_0 is the intercept, β_1 is the slope, and *D* is the dose of the agent (radiation). Define $LD_{50}(T)$ as the lethal dose for 50% of the treated population and $LD_{50}(C)$ as the lethal dose for 50% of the control population. Finney (1978) provides methodology for estimating the relative potency (efficacy) using parallel, log-dose regression lines.

Let

$$\rho = \frac{LD_{50}(T)}{LD_{50}(C)}$$

and

$$\log_{10}(\rho) = \log_{10}\{LD_{50}(T)\} - \log_{10}\{LD_{50}(C)\}.$$

The null and alternative hypotheses are

$$H_0 : \rho = 1$$
$$H_A : \rho > 1$$

or

$$H_0 : \log_{10}(\rho) = 0$$
$$H_A : \log_{10}(\rho) > 0$$

Probit Analysis

Let

$$\theta = \log_{10}(\rho) = \frac{\beta_{0C} - \beta_{0T}}{\beta_1}$$

This can be estimated by

$$\hat{\theta} = \log_{10}(\hat{\rho}) = \frac{\hat{\beta}_{0C} - \hat{\beta}_{0T}}{\hat{\beta}_1}$$

The variance of $\hat{\theta}$ is estimated using

$$\hat{V}(\hat{\theta}) = \frac{s^2}{\hat{\beta}_1^2}\left[\sum_{T,C}\frac{1}{\sum_{i=1}^{g} w_i n_i} + \left\{\frac{(\bar{y}_T - \bar{y}_C)^2}{\hat{\beta}_1^2}\right\}\left\{\frac{1}{\sum_{T,C}\sum_{i=1}^{g} w_i n_i (x_i - \bar{x})^2}\right\}\right]$$

where n_i is the number of animals in the ith dose group and

$$w_i = \frac{\phi\left(\Phi^{-1}(P_i)\right)^2}{P_i(1-P_i)} \text{ for the probit analysis}$$

and

$$w_i = P_i(1-P_i) \text{ for a logit analysis.}$$

Note that $\phi(x)$ is the normal density function and $\Phi^{-1}(x)$ is the normal c.d.f.

The test statistic for testing H_0 versus H_A is

$$T = \frac{\hat{\theta}}{\sqrt{\hat{V}(\hat{\theta})}}$$

which has a *t* distribution with $f = 2g - 3$ degrees of freedom.

Using several simplifications and approximations, Kodell, et al. (2010) show that the sample size per dose group is given by

$$n = \frac{2\left(t_{f,1-\alpha} + t_{f,1-\beta}\right)^2}{\{\beta_1 \log(\rho)\}^2 \sum_{i=1}^{g} w_i}$$

The power is given by

$$t_{f,1-\beta} = \sqrt{n\left(\frac{\{\beta_1 \log(\rho)\}^2 \sum_{i=1}^{g} w_i}{2}\right)} - t_{f,1-\alpha}$$

The relative potency is given by

$$\rho = 10^{A}$$

where

$$A = \sqrt{\frac{2\left(t_{f,1-\alpha} + t_{f,1-\beta}\right)^2}{n\beta_1^2 \sum_{i=1}^{g} w_i}}$$

Procedure Options

This section describes the options that are specific to this procedure. These are located on the Design tab. For more information about the options of other tabs, go to the Procedure Window chapter.

Design Tab

The Design tab contains most of the parameters and options that you will be concerned with.

Solve For

Solve For

This option specifies the parameter to be solved for from the other parameters. Under most situations, you will select either *Power* for a power analysis or *Sample Size* for sample size determination.

Select *Sample Size* when you want to calculate the sample size needed to achieve a given power and alpha level. Select *Power* when you want to calculate the power of an experiment.

Test

Model

Select whether a Probit or Logit analysis is planned.

This choice will result in about the same power and sample size, but it will change the slope. So, if you change this value and you have entered *Slopes* in the *Dose Input Option* option, you must change the slope values appropriately.

Power and Alpha

Power

This option specifies one or more values for power. Power is the probability of rejecting a false null hypothesis, and is equal to one minus Beta. Beta (consumer's risk) is the probability of a type-II error, which occurs when a false null hypothesis is not rejected. In this procedure, a type-II error occurs when you fail to reject the null hypothesis of equal thetas when in fact they are different.

Values must be between zero and one. Historically, the value of 0.80 (Beta = 0.20) was used for power. Now, 0.90 (Beta = 0.10) is also commonly used.

A single value may be entered here or a range of values such as *0.8 to 0.95 by 0.05* may be entered.

Alpha

This option specifies one or more values for the probability of a type-I error. A type-I error occurs when a true null hypothesis is rejected. In this procedure, a type-I error occurs when you reject the null hypothesis of equal thetas when in fact they are equal.

Values must be between zero and one. Historically, the value of 0.05 has been used for two-sided tests and 0.025 for one-sided tests.

You may enter a range of values such as *0.025 0.05 0.10* or *0.025 to 0.10 by 0.025*.

Sample Size

n (Sample Size per Dose-Group)

Enter a value (or range of values) for the sample size (number of subjects) in each dose-group. The total sample size, N, is this value times 2 (the number of groups) times the number of doses. These values are ignored when you are solving for *n*.

Effect Size

Target Response Proportions

Enter a set of response proportions, one per dose. Since these are proportions, they must be between 0 and 1.

It is assumed that the doses of the control group and the treated group will be chosen so that these target response proportions will be obtained.

The number of doses, *g*, is set implicitly as the number of values entered here. Popular designs have five-doses or seven-doses with proportions that are equally spaced, but you can enter as many as you like. Experimental design principles suggest having some values at each end (near 0 and near 1) and some in the middle.

Recommended Values

Kodell et al. recommend 0.05, 0.225, 0.5, 0.725, 0.95 for a five-dose design and 0.05, 0.2, 0.35, 0.5, 0.65, 0.8, 0.95 for a seven-dose design.

Input Dose Using

The power calculations are based on the value of the slope of the regression equation. This slope can be calculated from a set of doses and response proportions, or it can be entered as a single value.

Doses

A set of doses is entered and the slope is calculated from the doses and response proportions. The calculated slope is different depending on whether a probit analysis or logit analysis is selected. Only one slope may be generated. Note that the number of doses must match the number of target response proportions.

Slopes

One or more slopes may be entered. A separate calculation is made for each slope. These values depend on the model that is selected. If you change the model, you need to change these slopes appropriately.

Doses (Control Group)

Enter a set of doses for the control group, one for each Target Response Proportion. The program calculates the slope using the Target Response Proportions entered above. The number of doses must match the number of Target Response Proportions.

Input Notes

1. Each control dose should be chosen so that it will (approximately) result in the corresponding target response proportion (lethality). It is assumed that the treatment doses will be different from the control doses, but each will result in the corresponding target response proportion.

2. The calculated slopes will be different depending on whether you use a probit or a logit model.

Slopes

Enter a set of slope values. The analysis assumes that the regression lines are parallel, that the slopes of the control and treated groups are equal.

The slopes depend on the model selected. If you switch from a probit to logit model, you must change these slopes appropriately.

Rho (Relative Potency)

Enter one or more values for rho, the relative potency or dose reduction factor (DFR), the factor by which the treatment drug reduces the potency of the drug. This design assumes that the treatment decreases the dose potency, so LD50(Treatment) > LD50(Control).

Range

Values must be greater than 1. Typical values are 1.1, 1.2, etc.

You can enter a list of values such as 1.1 1.2 1.3 or 1.1 to 1.5 by 0.1

Example 1 – Power for Several Sample Sizes

This example will calculate power for several sample sizes of a probit analysis study designed to compare the efficacy of a new drug as a countermeasure to radiation-induced lethality. Experimenters want to size the study so that they can detect a relative potency of 1.1. They also want to study values of 1.05 and 1.15. They would like to study the power at a significance level of 0.025 of samples of 5 to 55 subjects.

They want to use a five-dose study with doses of 11, 12, 13, 14, and 15 chosen so that lethalities of about 0.05, 0.275, 0.5, 0.725, and 0.95 are obtained.

Setup

This section presents the values of each of the parameters needed to run this example. First, from the PASS Home window, load the **Probit Analysis** procedure window by selecting it from the **Survival** menu. You may then make the appropriate entries as listed below, or open **Example 1** by going to the **File** menu and choosing **Open Example Template**.

Option	Value
Design Tab	
Solve For	**Power**
Model	**Probit**
Alpha	**0.025**
n (Sample Size per Dose-Group)	**5 to 55 by 10**
Target Response Proportions	**0.05 0.275 0.5 0.725 0.95**
Input Dose Using	**Doses**
Doses (Control Group)	**11 12 13 14 15**
Rho (Relative Potency)	**1.05 1.1 1.15**
Plot Text Tab	
Decimal Places – Plot Probabilities	**3**

Annotated Output

Click the Calculate button to perform the calculations and generate the following output.

Numeric Results

Numeric Results for a Design with 5 Doses

Power	Group Sample Size (n)	Total Sample Size (N)	Relative Potency (Rho)	Slope (β1)	Alpha	Beta
0.13140	5	50	1.05	23.07	0.02500	0.86860
0.35831	15	150	1.05	23.07	0.02500	0.64169
0.57645	25	250	1.05	23.07	0.02500	0.42355
0.73782	35	350	1.05	23.07	0.02500	0.26218
0.84126	45	450	1.05	23.07	0.02500	0.15874
0.90340	55	550	1.05	23.07	0.02500	0.09660
0.45238	5	50	1.10	23.07	0.02500	0.54762
0.91337	15	150	1.10	23.07	0.02500	0.08663
0.98342	25	250	1.10	23.07	0.02500	0.01658
0.99541	35	350	1.10	23.07	0.02500	0.00459
0.99834	45	450	1.10	23.07	0.02500	0.00166
0.99927	55	550	1.10	23.07	0.02500	0.00073
0.80614	5	50	1.15	23.07	0.02500	0.19386
0.99367	15	150	1.15	23.07	0.02500	0.00633
0.99920	25	250	1.15	23.07	0.02500	0.00080
0.99980	35	350	1.15	23.07	0.02500	0.00020
0.99993	45	450	1.15	23.07	0.02500	0.00007
0.99997	55	550	1.15	23.07	0.02500	0.00003

Report Definitions
Power is the probability of rejecting a false null hypothesis. It should be close to one.
Group Sample Size (n) is the dose-group sample size.
Total Sample Size (N) is the total of all dose-group sample sizes.
Relative Potency (Rho) is the ratio: (50% Lethal Dose of Treatment) / (50% Lethal Dose of Control).
Slope (β1) is the slope of regressing the probits (or logits) on the log10 doses.
Alpha is the probability of rejecting a true null hypothesis. It should be small.
Beta is the probability of accepting a false null hypothesis. It should be small.

Summary Statements
In a study using probit analysis to compare the potency of a treatment and a control, 5 subjects are required from each of 10 dose groups. This results in a total sample of 50 subjects. This study achieves 13.140% power to detect an increase in relative potency to 1.05 using a one-sided t-test with a 0.02500 significance level. The common slope between the probits and the log doses was assumed to be 23.07.

This report shows the power for each of the scenarios. Note that the computed slope is about 23.

Lethality Report

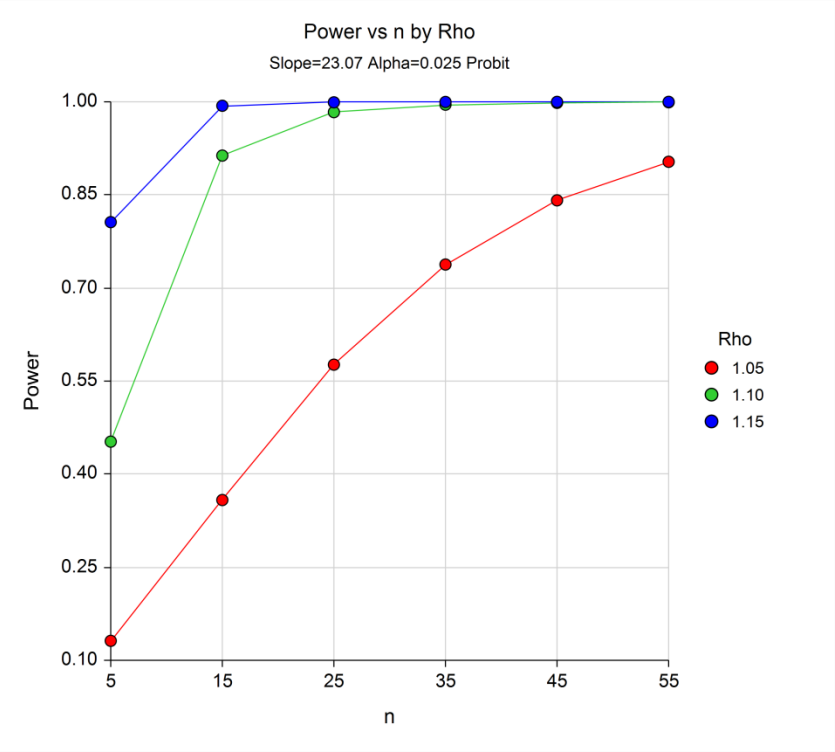
Lethality Report

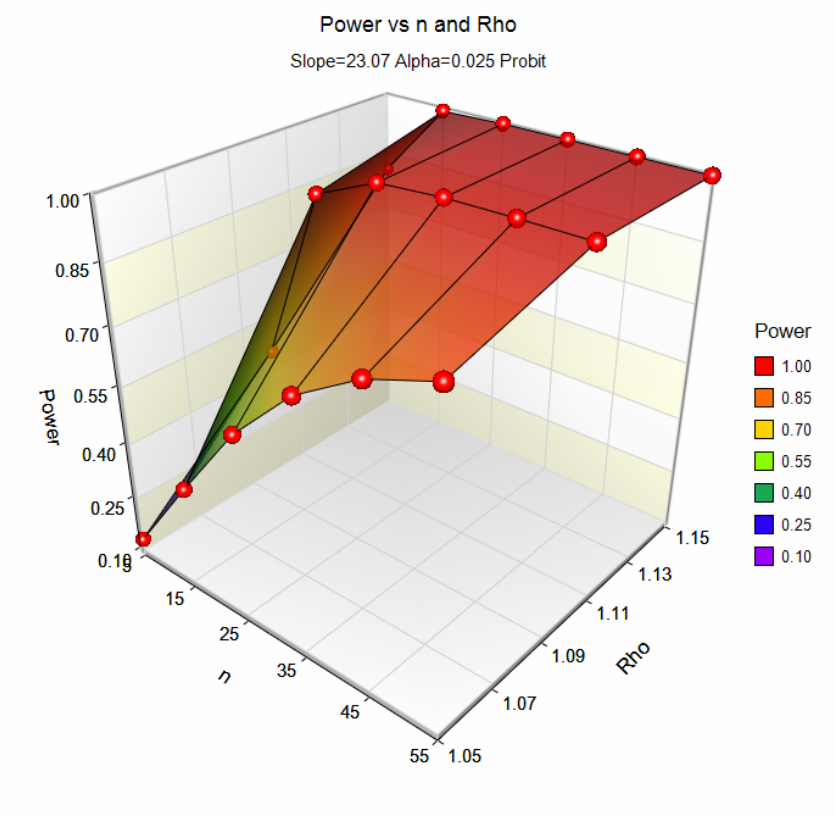
Group Number	Response Proportion (Lethality)	Weight (w)	Dose
1	0.05000	0.22394	11.00
2	0.27500	0.55843	12.00
3	0.50000	0.63662	13.00
4	0.72500	0.55843	14.00
5	0.95000	0.22394	15.00
Total		2.20135	

This report documents the individual values of the lethalities, weights, and doses.

Probit Analysis

Plots Section





Example 2 – Validation using Kodell, et al.

We will validate this procedure using the results of Kodell et al. (2010). On page 243, in Table 2 of their article, Kodell et al. give the following example. For a five-dose example with target lethalities of 0.05, 0.275, 0.5, 0.725, 0.95, a slope of 23.25, rho of 1.1, power of 0.9, and alpha of 0.05, they obtain an *n* of 11.

Setup

This section presents the values of each of the parameters needed to run this example. First, from the PASS Home window, load the **Probit Analysis** procedure window by selecting it from the **Survival** menu. You may then make the appropriate entries as listed below, or open **Example 1** by going to the **File** menu and choosing **Open Example Template**.

Option	Value
Design Tab	
Solve For	**Sample Size**
Model	**Probit**
Power	**0.90**
Alpha	**0.05**
Target Response Proportions	**0.05 0.275 0.5 0.725 0.95**
Input Dose Using	**Slopes**
Slopes	**23.25**
Rho (Relative Potency)	**1.1**

Output

Click the Calculate button to perform the calculations and generate the following output.

Numeric Results for a Design with 5 Doses

Power	Group Sample Size (n)	Total Sample Size (N)	Relative Potency (Rho)	Slope (β1)	Alpha	Beta
0.90538	11	110	1.10	23.25	0.05000	0.09462

Note that PASS has also calculated the required sample size at 11, or a total sample size of 110.