Chapter 530

Non-Inferiority Tests for the Difference of Two Means in a Higher-Order Cross-Over Design

Introduction

This procedure calculates power and sample size for non-inferiority tests which use the difference in the means of a higher-order cross-over design. Measurements are made on individuals that have been randomly assigned to one of several treatment sequences. Only a brief introduction to the subject will be given here. For a comprehensive discussion on the subject, refer to Chen et al. (1997) and Chow et al. (2003).

Cross-Over Designs

Senn (2002) defines a *cross-over* design as one in which each subject receives all treatments at least once and the object is to study differences among the treatments. The name *cross-over* comes from the most common case in which there are only two treatments. In this case, each subject *crosses over* from one treatment to another. It is assumed that there is a *washout* period between treatments during which the response returns to its baseline value. If this does not occur, there is said to be a *carryover* effect.

A 2x2 cross-over design refers to two treatments (periods) and two *sequences* (treatment orderings). One sequence of treatments is treatment A followed by treatment B. The other sequence is B and then A. The design includes a washout period between responses to make certain that the effects of the first drug do no carryover to the second. Thus, the groups of subjects in this design are defined by the sequence in which the two treatments are administered, not by the treatments they receive.

Higher-Order Cross-Over Designs

Chen et al. (1997) present the results for four cross-over designs that are more complicated than the 2x2 design. Assume that the two treatments are labeled A and B. The available designs are defined by the order and number of times the two treatments are administered.

Balaam's Design

Balaam's design has four sequences with two treatments each. It is popular because it allows the intrasubject variabilities to be estimated. The design is

Sequence	**Period 1**	**Period 2**
1	A	A
2	B	B
3	A	B
4	B	A

Two-Sequence Dual Design

This design has two sequences with three periods each. It is popular because it allows the intrasubject variabilities to be estimated. The design is

Sequence	Period 1	Period 2	Period 3
1	A	B	B
2	B	A	A

Four-Period Design with Two Sequences

This design has two sequences of four periods each. The design is

Sequence	Period 1	Period 2	Period 3	Period 4
1	A	B	B	A
2	B	A	A	B

Four-Period Design with Four Sequences

This design has four sequences of four periods each. The design is

Sequence	Period 1	Period 2	Period 3	Period 4
1	A	A	B	B
2	B	B	A	A
1	A	B	B	A
2	B	A	A	B

Advantages of Cross-Over Designs

A comparison of treatments on the same subject is expected to be more precise. The increased precision often translates into a smaller sample size. Also, patient enrollment may be easier to obtain because each patient will receive both treatments.

Disadvantages of Cross-Over Designs

The statistical analysis of a cross-over experiment is more complex than a parallel-group experiment and requires additional assumptions. In a cross-over experiment, it may be difficult to separate the treatment effect from the time effect and the carry-over effect of the previous treatment.

These cross-over designs cannot be used when the treatment (or the measurement of the response) alters the subject permanently. Hence, it cannot be used to compare treatments that are intended to provide a cure.

Because subjects must be measured at least twice, it may be more difficult to keep patients enrolled in the study. This is particularly true when the measurement process is painful, uncomfortable, embarrassing, or time consuming.

The Statistical Hypotheses

Both non-inferiority and superiority tests are examples of directional (one-sided) tests. Remember that in the usual t-test setting, the null (H0) and alternative (H1) hypotheses for one-sided tests are defined as

$$H_0: \delta \leq A \quad \text{versus} \quad H_1: \delta > A$$

Rejecting H0 implies that the mean is larger than the value *A*. This test is called an *upper-tailed test* because H0 is rejected only in samples in which the difference in sample means is larger than *A.*

Following is an example of a *lower-tailed test*.

$$H_0: \delta \geq A \quad \text{versus} \quad H_1: \delta < A$$

Non-inferiority and *superiority* tests are special cases of the above directional tests. It will be convenient to adopt the following specialize notation for the discussion of these tests.

Parameter	**PASS Input/Output**	**Interpretation**
μ_T	Not used	*Treatment mean*. This is the treatment mean.
μ_R	Not used	*Reference mean*. This is the mean of a reference population.
M_{NI}	NIM	*Margin of non-inferiority.* This is a tolerance value that defines the magnitude of the amount that is not of practical importance. This may be thought of as the largest change from the baseline that is considered to be trivial. The absolute value is shown to emphasize that this is a magnitude. The sign of the value will be determined by the specific design that is being used.
δ	D	*True difference*. This is the value of $\mu_T - \mu_R$, the difference between the treatment and reference means. This is the value at which the power is calculated.

Note that the actual values of μ_T and μ_R are not needed. Only their difference is needed for power and sample size calculations.

Non-Inferiority Tests

A *non-inferiority test* tests that the treatment mean is not worse than the reference mean by more than the equivalence margin. The actual direction of the hypothesis depends on the response variable being studied.

Case 1: High Values Good, Non-Inferiority Test

In this case, higher values are better. The hypotheses are arranged so that rejecting the null hypothesis implies that the treatment mean is no less than a small amount below the reference mean. The value of δ is often set to zero. The following are equivalent sets of hypotheses.

$H_0: \mu_1 \leq \mu_2 - |M_{NI}|$ versus $H_1: \mu_1 > \mu_2 - |M_{NI}|$

$H_0: \mu_1 - \mu_2 \leq -|M_{NI}|$ versus $H_1: \mu_1 - \mu_2 > -|M_{NI}|$

$H_0: \delta \leq -|M_{NI}|$ versus $H_1: \delta > -|M_{NI}|$

Case 2: High Values Bad, Non-Inferiority Test

In this case, lower values are better. The hypotheses are arranged so that rejecting the null hypothesis implies that the treatment mean is no more than a small amount above the reference mean. The value of δ is often set to zero. The following are equivalent sets of hypotheses.

$H_0: \mu_1 \geq \mu_2 + |M_{NI}|$ versus $H_1: \mu_1 < \mu_2 + |M_{NI}|$

$H_0: \mu_1 - \mu_2 \geq |M_{NI}|$ versus $H_1: \mu_1 - \mu_2 < |M_{NI}|$

$H_0: \delta \geq |M_{NI}|$ versus $H_1: \delta < |M_{NI}|$

Test Statistics

The analysis for assessing equivalence (and thus non-inferiority) using higher-order cross-over designs is discussed in detail in Chapter 9 of Chow and Liu (2000). Unfortunately, their presentation is too lengthy to give here. Their method involves the computation of an analysis of variance to estimate the error variance. It also describes the construction of confidence limits for appropriate contrasts. One-sided confidence limits can be used for non-inferiority tests. Details of this approach are given in Chapter 3 of Chow et al. (2003). We refer you to these books for details.

Power Calculation

The power of the non-inferiority and superiority tests for the case in which higher values are better is given by

$$Power = T_V\left(\left(\frac{\delta - \varepsilon}{\sigma_W\sqrt{b/n}}\right) - t_{V,1-\alpha}\right)$$

where *T* represents the cumulative *t* distribution, *V* and *b* depend on the design, σ_W is the square root of the within mean square error from the ANOVA table used to analyze the cross-over design, and *n* is the average number of subjects per sequence. Note that the constants *V* and *b* depend on the design as follows.

The power of the non-inferiority and superiority tests for the case in which higher values are worse is given by

$$Power = 1 - T_V\left(t_{V,1-\alpha} - \left(\frac{\varepsilon - \delta}{\sigma_W\sqrt{b/n}}\right)\right)$$

The constants *V* and *b* depend on the design as follows:

Design Type	**Parameters (*V*,*b*)**
Balaam's Design	*V* = 4n – 3, *b* = 2.
Two-Sequence Dual Design	*V* = 4n – 4, *b* = 3/4.
Four-Period Design with Two Sequences	*V* = 6n – 5, *b* = 11/20.
Four-Period Design with Four Sequences	*V* = 12n – 5, *b* = 1/4.

Example 1 – Finding Power

Researchers want to calculate the power of a non-inferiority test using data from a two-sequence, dual cross-over design. The margin of equivalence is either 5 or 10 at several sample sizes between 6 and 66. The true difference between the means under is assumed to be 0. Similar experiments have had a standard deviation (σw) of 10. The significance level is 0.025.

Setup

If the procedure window is not already open, use the PASS Home window to open it. The parameters for this example are listed below and are stored in the **Example 1** settings file. To load these settings to the procedure window, click **Open Example Settings File** in the Help Center or File menu.

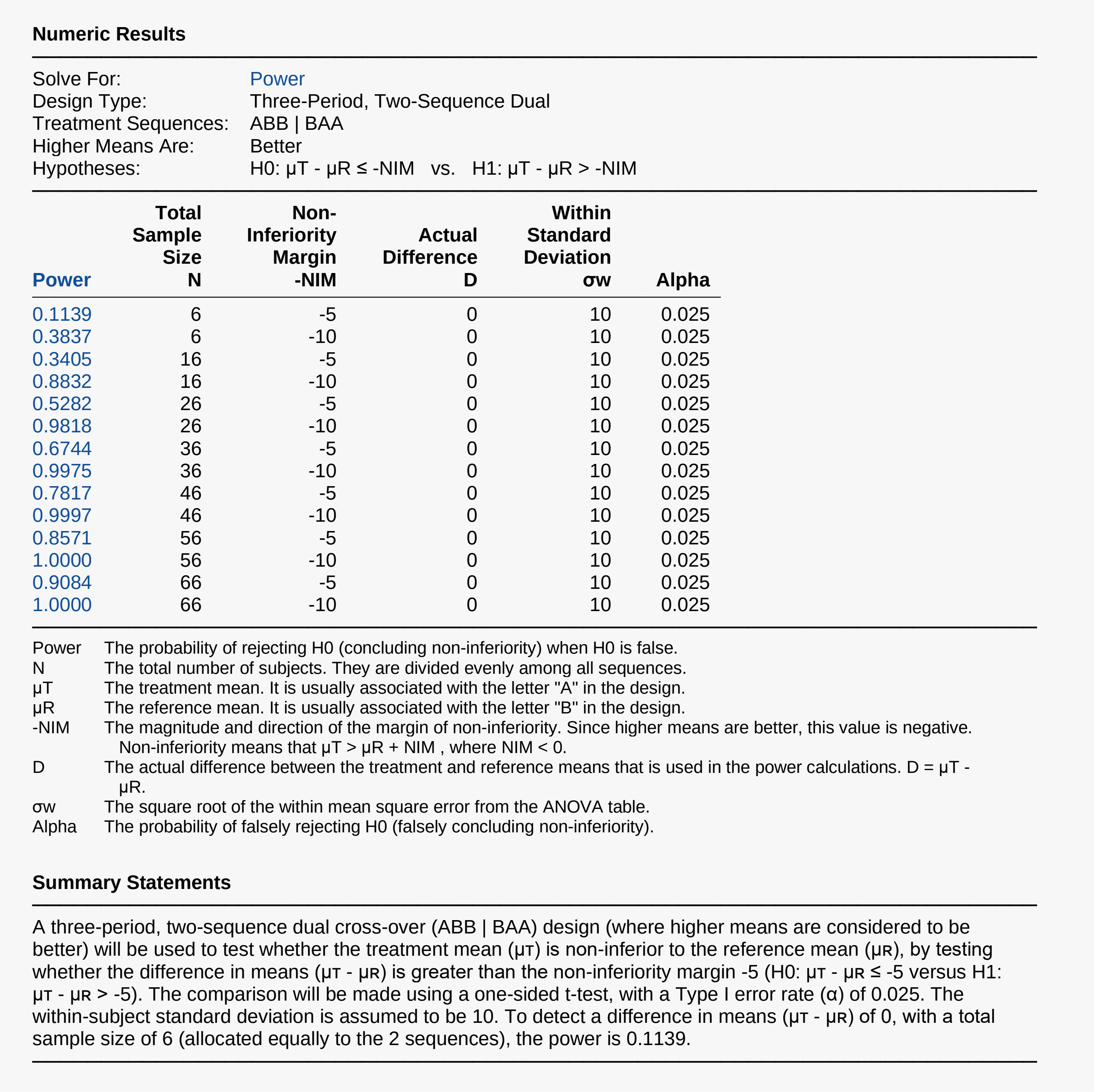
Design Tab

Solve For	**Power**
Design Type	**3x2 (Three-Period, Two-Sequence Dual: ABB|BAA)**
Higher Means Are	**Better**
Alpha	**0.025**
N (Total Sample Size)	**6 to 66 by 10**
NIM (Non-Inferiority Margin)	**5 10**
D (Difference)	**0**
Specify σ as σw or σb and ρ	**σw (Within Std Error)**
σw (Within Std Dev)	**10**

Output

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

Numeric Reports



Numeric Results

```
Solve For:              Power
Design Type:            Three-Period, Two-Sequence Dual
Treatment Sequences:    ABB | BAA
Higher Means Are:       Better
Hypotheses:             H0: μT - μR ≤ -NIM   vs.   H1: μT - μR > -NIM

                Total          Non-                      Within
               Sample   Inferiority      Actual        Standard
                 Size        Margin  Difference       Deviation
Power               N          -NIM           D              σw    Alpha

0.1139              6            -5           0              10    0.025
0.3837              6           -10           0              10    0.025
0.3405             16            -5           0              10    0.025
0.8832             16           -10           0              10    0.025
0.5282             26            -5           0              10    0.025
0.9818             26           -10           0              10    0.025
0.6744             36            -5           0              10    0.025
0.9975             36           -10           0              10    0.025
0.7817             46            -5           0              10    0.025
0.9997             46           -10           0              10    0.025
0.8571             56            -5           0              10    0.025
1.0000             56           -10           0              10    0.025
0.9084             66            -5           0              10    0.025
1.0000             66           -10           0              10    0.025

Power   The probability of rejecting H0 (concluding non-inferiority) when H0 is false.
N       The total number of subjects. They are divided evenly among all sequences.
μT      The treatment mean. It is usually associated with the letter "A" in the design.
μR      The reference mean. It is usually associated with the letter "B" in the design.
-NIM    The magnitude and direction of the margin of non-inferiority. Since higher means are better, this value is negative.
          Non-inferiority means that μT > μR + NIM , where NIM < 0.
D       The actual difference between the treatment and reference means that is used in the power calculations. D = μT -
          μR.
σw      The square root of the within mean square error from the ANOVA table.
Alpha   The probability of falsely rejecting H0 (falsely concluding non-inferiority).
```

Summary Statements

A three-period, two-sequence dual cross-over (ABB | BAA) design (where higher means are considered to be better) will be used to test whether the treatment mean (μT) is non-inferior to the reference mean (μR), by testing whether the difference in means (μT - μR) is greater than the non-inferiority margin -5 (H0: μT - μR ≤ -5 versus H1: μT - μR > -5). The comparison will be made using a one-sided t-test, with a Type I error rate (α) of 0.025. The within-subject standard deviation is assumed to be 10. To detect a difference in means (μT - μR) of 0, with a total sample size of 6 (allocated equally to the 2 sequences), the power is 0.1139.

Dropout-Inflated Sample Size

Dropout Rate	Sample Size N	Dropout- Inflated Enrollment Sample Size N'	Expected Number of Dropouts D
20%	6	8	2
20%	16	20	4
20%	26	33	7
20%	36	45	9
20%	46	58	12
20%	56	70	14
20%	66	83	17

Dropout Rate The percentage of subjects (or items) that are expected to be lost at random during the course of the study and for whom no response data will be collected (i.e., will be treated as "missing"). Abbreviated as DR.

N The evaluable sample size at which power is computed (as entered by the user). If N subjects are evaluated out of the N' subjects that are enrolled in the study, the design will achieve the stated power.

N' The total number of subjects that should be enrolled in the study in order to obtain N evaluable subjects, based on the assumed dropout rate. N' is calculated by inflating N using the formula N' = N / (1 - DR), with N' always rounded up. (See Julious, S.A. (2010) pages 52-53, or Chow, S.C., Shao, J., Wang, H., and Lokhnygina, Y. (2018) pages 32-33.)

D The expected number of dropouts. D = N' - N.

Dropout Summary Statements

Anticipating a 20% dropout rate, 8 subjects should be enrolled to obtain a final sample size of 6 subjects.

References

Chow, S.C. and Liu, J.P. 1999. Design and Analysis of Bioavailability and Bioequivalence Studies. Marcel Dekker. New York

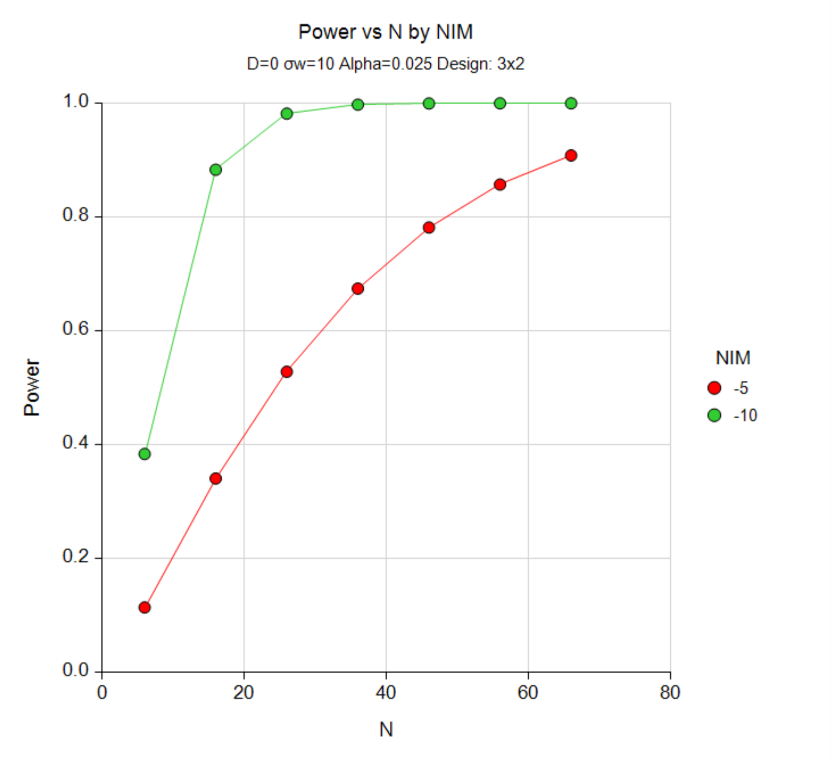
Chow, S.C., Shao, J., and Wang, H. 2003. Sample Size Calculations in Clinical Research. Marcel Dekker. New York.

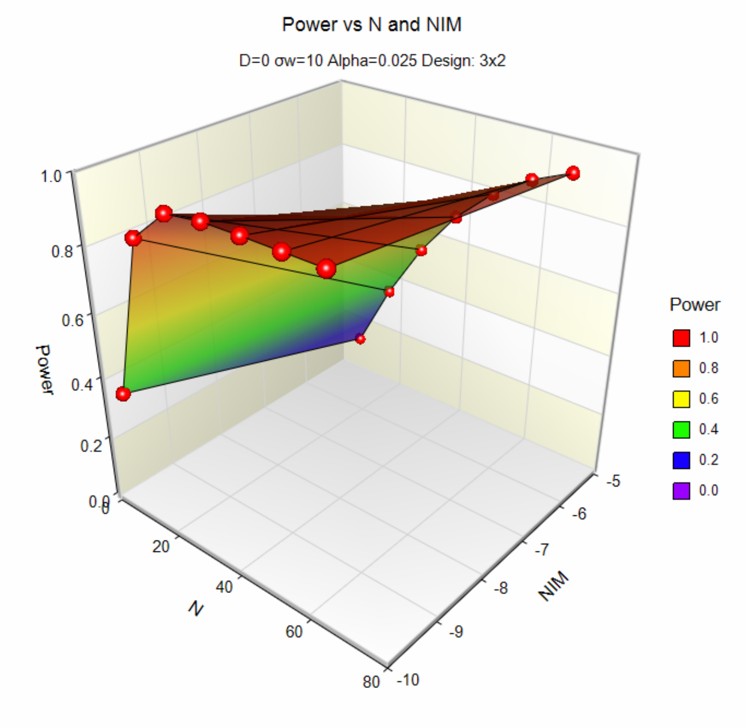
Chen, K.W., Chow, S.C., and Li, G. 1997. 'A Note on Sample Size Determination for Bioequivalence Studies with Higher-Order Crossover Designs.' Journal of Pharmacokinetics and Biopharmaceutics, Volume 25, No. 6, pages 753-765.

This report shows the power for the indicated scenarios.

Plots Section

Plots





These plots show the power versus the sample size.

Example 2 – Finding Sample Size

Continuing with Example 1, the researchers want to find the sample size needed to achieve both 80% power and 90% power when the number of subjects is equal for each sequence.

Setup

If the procedure window is not already open, use the PASS Home window to open it. The parameters for this example are listed below and are stored in the **Example 2** settings file. To load these settings to the procedure window, click **Open Example Settings File** in the Help Center or File menu.

```
Design Tab
Solve For ......................................................Sample Size (Equal Per Sequence)
Design Type..................................................3x2 (Three-Period, Two-Sequence Dual: ABB|BAA)
Higher Means Are .........................................Better
Power...........................................................0.80 0.90
Alpha............................................................0.025
NIM (Non-Inferiority Margin) ..........................5 10
D (Difference) ...............................................0
Specify σ as σw or σb and ρ.........................σw (Within Std Error)
σw (Within Std Dev)......................................10
```

Output

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

```
Numeric Results

Solve For:              Sample Size (Equal Per Sequence)
Design Type:            Three-Period, Two-Sequence Dual
Treatment Sequences:    ABB | BAA
Higher Means Are:       Better
Hypotheses:             H0: μT - μR ≤ -NIM   vs.   H1: μT - μR > -NIM

               Total          Non-
              Sample   Inferiority        Actual      Within
                Size        Margin    Difference    Standard
Power              N          -NIM             D   Deviation
                                                          σw    Alpha

0.8153            50            -5             0          10    0.025
0.8343            14           -10             0          10    0.025
0.9084            66            -5             0          10    0.025
0.9184            18           -10             0          10    0.025
```

When the non-inferiority margin is -5, 66 subjects are needed to achieve 90% power and 50 subjects are needed to achieve at least 80% power.

Example 3 – Validation

We could not find a validation example for this procedure in the statistical literature, so we will have to generate a validated example from within **PASS**. To do this, we use the Equivalence Tests for the Difference of Two Means in a Higher-Order Cross-Over Design procedure which was validated. By setting the upper equivalence limit to a large value (we used 22), we obtain results for a non-inferiority test.

A Balaam design is being planned. Suppose the square root of the within mean square error is 0.10, the equivalence limit is 0.20, the difference between the means is 0.05, the power is 90%, and the significance level is 0.05 (see the Example4 template). **PASS** calculates a sample size of 16.

Setup

If the procedure window is not already open, use the PASS Home window to open it. The parameters for this example are listed below and are stored in the **Example 3** settings file. To load these settings to the procedure window, click **Open Example Settings File** in the Help Center or File menu.

```
Design Tab
Solve For ...................................................Sample Size (Equal Per Sequence)
Design Type..................................................2x4 (Balaam: AA|BB|AB|BA)
Higher Means Are.............................................Better
Power........................................................0.90
Alpha........................................................0.05
NIM (Non-Inferiority Margin) ................................0.2
D (Difference) ..............................................0.05
Specify σ as σw or σb and ρ..................................σw (Within Std Error)
σw (Within Std Dev)..........................................0.10
```

Output

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

```
Numeric Results

Solve For:              Sample Size (Equal Per Sequence)
Design Type:            Two-Period, Four-Sequence (Balaam)
Treatment Sequences:    AA | BB | AB | BA
Higher Means Are:       Better
Hypotheses:             H0: μT - μR ≤ -NIM   vs.   H1: μT - μR > -NIM

                Total          Non-                       Within
               Sample   Inferiority        Actual      Standard
                 Size        Margin    Difference     Deviation
Power               N          -NIM             D            σw      Alpha
0.9495             16          -0.2          0.05           0.1       0.05
```

PASS has also obtained a sample size of 16 using the non-inferiority procedure.