Chapter 300

Tests for Two Between-Subject Variances in a 2×2M Replicated Cross-Over Design

Introduction

This procedure calculates power and sample size of tests of between-subject variabilities from a 2×2M replicated cross-over design for the case when the ratio assumed by the null hypothesis is equal to one. This routine deals with the case in which the statistical hypotheses are expressed in terms of the ratio of the between-subject variances.

This design is used to compare two treatments which are administered to subjects in different orders. The design has two treatment sequences. Here, *M* is the number of times a particular treatment is received by a subject.

For example, if *M* = 2, the design is a 2×4 replicated cross-over. The two sequences might be

sequence 1: C T C T

sequence 2: T C T C

It is assumed that either there is no carry-over from one measurement to the next, or there is an ample washout period between measurements.

Technical Details

This procedure uses the formulation given in Chow, Shao, Wang, and Lokhnygina (2018), pages 213 - 216.

Suppose x_{ijkl} is the response in the *i*th sequence (*i* = 1, 2), *j*th subject (*j* = 1, …, *Ni*), *k*th treatment (*k* = T, C), and *l*th replicate (l = 1, …, M). The mixed effect model analyzed in this procedure is

$$x_{ijkl} = \mu_k + \gamma_{ikl} + S_{ijk} + e_{ijkl}$$

where μ_k is the *k*th treatment effect, γ_{ikl} is the fixed effect of the *l*th replicate on treatment *k* in the *i*th sequence, S_{ij1} and S_{ij2} are random effects of the *ij*th subject, and e_{ijkl} is the within-subject error term which is normally distributed with mean 0 and variance $V_k = \sigma_{Wk}^2$.

Unbiased estimators of these variances are found after applying an orthogonal transformation matrix *P* to the x's as follows

$$z_{ijk} = P' x_{ijk}$$

where *P* is an *m* × *m* matrix such that $P'P$ is diagonal and $\mathrm{var}\left(z_{ijkl}\right) = \sigma_{Wk}^2$.

Let $N_s = N_1 + N_2 - 2$. In a 2×4 cross-over design the z's become

$$z_{ijk1} = \frac{x_{ijk1} + x_{ijk2}}{2} = \bar{x}_{ijk\cdot}$$

and

$$z_{ijk2} = \frac{x_{ijk1} + x_{ijk2}}{\sqrt{2}} = \bar{x}_{ijk\cdot}$$

In this case, the within-subject variances are estimated as

$$s_{WT}^2 = \frac{1}{N_S(M-1)} \sum_{i=1}^{2} \sum_{j=1}^{N_i} \sum_{l=1}^{M} \left(z_{ijTl} - \bar{z}_{i.Tl}\right)^2$$

and

$$s_{WC}^2 = \frac{1}{N_S(M-1)} \sum_{i=1}^{2} \sum_{j=1}^{N_i} \sum_{l=1}^{M} \left(z_{ijCl} - \bar{z}_{i.Cl}\right)^2$$

Similarly, the between-subject variances are estimated as

$$s_{BT}^2 = \frac{1}{N_S} \sum_{i=1}^{2} \sum_{j=1}^{N_i} \left(\bar{x}_{ijT.} - \bar{x}_{i.T.}\right)^2$$

and

$$s_{BC}^2 = \frac{1}{N_S} \sum_{i=1}^{2} \sum_{j=1}^{N_i} \left(\bar{x}_{ijC.} - \bar{x}_{i.C.}\right)^2$$

where

$$\bar{x}_{i.k.} = \frac{1}{N_i} \sum_{j=1}^{N_i} \bar{x}_{ijk.}$$

Now, since $E(s_{BK}^2) = \sigma_{BK}^2 + \sigma_{WK}^2/M$, estimators for the between-subject variance are given by

$$\hat{\sigma}_{BK}^2 = s_{BK}^2 - \hat{\sigma}_{WK}^2/M$$

The sample between-subject covariance is calculated using

$$s_{BTC}^2 = \frac{1}{N_S} \sum_{i=1}^{2} \sum_{j=1}^{N_i} \left(\bar{x}_{ijT.} - \bar{x}_{i.T.}\right)\left(\bar{x}_{ijC.} - \bar{x}_{i.C.}\right)$$

Using this value, the sample between-subject correlation is easily calculated.

Testing Variance Inequality

The following three sets of statistical hypotheses are used to test for between-subject variance inequality.

$$H_0: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} \geq 1 \quad \text{versus} \quad H_1: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} < 1,$$

$$H_0: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} \leq 1 \quad \text{versus} \quad H_1: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} > 1,$$

$$H_0: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} = 1 \quad \text{versus} \quad H_1: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} \neq 1,$$

Let $\eta = \sigma_{BT}^2 - \sigma_{BC}^2$ be the parameter of interest. The test statistic is $\hat{\eta} = \hat{\sigma}_{BT}^2 - \hat{\sigma}_{BC}^2$.

Two-Sided Test

For the two-sided test, compute two limits, $\hat{\eta}_L$ and $\hat{\eta}_U$, using

$$\hat{\eta}_L = \hat{\eta} - \sqrt{\Delta_L}$$

$$\hat{\eta}_U = \hat{\eta} + \sqrt{\Delta_U}$$

Reject the null hypothesis if $\hat{\eta}_L > 0$ is or $\hat{\eta}_U < 0$.

The Δs are given by

$$\Delta_L = \lambda_1^2 h\left(\frac{\alpha}{2}, N_s - 1\right) + \lambda_2^2 h\left(1 - \frac{\alpha}{2}, N_s - 1\right) + h\left(\frac{\alpha}{2}, N_s(M-1)\right)\frac{\hat{\sigma}_{WT}^4}{M^2} + h\left(1 - \frac{\alpha}{2}, N_s(M-1)\right)\frac{\hat{\sigma}_{WC}^4}{M^2}$$

$$\Delta_U = \lambda_1^2 h\left(1 - \frac{\alpha}{2}, N_s - 1\right) + \lambda_2^2 h\left(\frac{\alpha}{2}, N_s - 1\right) + h\left(1 - \frac{\alpha}{2}, N_s(M-1)\right)\frac{\hat{\sigma}_{WT}^4}{M^2} + h\left(\frac{\alpha}{2}, N_s(M-1)\right)\frac{\hat{\sigma}_{WC}^4}{M^2}$$

where

$$h(A,B) = \left(1 - \frac{B}{\chi_{A,B}^2}\right)^2$$

$$\lambda_i^2 = \left(\frac{s_{BT}^2 - s_{BC}^2 \pm \sqrt{(s_{BT}^2 + s_{BC}^2)^2 - 4(R0)s_{BTC}^4}}{2}\right) \text{ for } i = 1,2$$

and $\chi_{A,B}^2$ is the upper quantile of the Chi-square distribution with B degrees of freedom.

One-Sided Test

For the lower, one-sided test, compute the limit, $\hat{\eta}_U$, using

$$\hat{\eta}_U = \hat{\eta} + \sqrt{\Delta_U}$$

Reject the null hypothesis if $\hat{\eta}_U < 0$.

The Δ_U is given by

$$\Delta_U = h(1-\alpha, N_s - 1)\lambda_1^2 + h(\alpha, N_s - 1)\lambda_2^2 + h\big(1-\alpha, N_s(M-1)\big)\frac{\hat{\sigma}_{WT}^4}{M^2} + h\big(\alpha, N_s(M-1)\big)\frac{\hat{\sigma}_{WC}^4}{M^2}$$

Power

Two-Sided Test

The power of the two-sided test is given by

$$\text{Power} = 1 - \Phi\left(z_{1-\frac{\alpha}{2}} - \frac{(R_1 - 1)\sigma_{BC}^2}{\sqrt{\sigma^{*2}/N_s}}\right) + \Phi\left(z_{\alpha/2} - \frac{(R_1 - 1)\sigma_{BC}^2}{\sqrt{\sigma^{*2}/N_s}}\right)$$

where

$$R_1 = \frac{\sigma_{BT}^2}{\sigma_{BC}^2}$$

$$\sigma_{BT}^2 = R_1\sigma_{BC}^2$$

$$\sigma^{*2} = 2\left[\left(\sigma_{BT}^2 + \frac{\sigma_{WT}^2}{M}\right)^2 + \left(\sigma_{BC}^2 + \frac{\sigma_{WC}^2}{M}\right)^2 + \frac{\sigma_{WT}^4}{M^2(M-1)} + \frac{\sigma_{WC}^4}{M^2(M-1)} - 2R_1\sigma_{BC}^4\rho^2\right]$$

where *R1* is the value of the variance ratio stated by the alternative hypothesis and $\Phi(x)$ is the standard normal CDF.

A simple binary search algorithm can be applied to the power function to obtain an estimate of the necessary sample size.

One-Sided Test

The power of the lower, one-sided test, $H_0: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} \geq 1 \text{ versus } H_1: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} < 1,$ is given by

$$\text{Power} = \Phi\left(z_\alpha - \frac{(R_1 - 1)\sigma_{BC}^2}{\sqrt{\sigma^{*2}/N_s}}\right)$$

The power of the upper, one-sided test, $H_0: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} \leq 1 \text{ versus } H_1: \frac{\sigma_{BT}^2}{\sigma_{BC}^2} > 1,$ is given by

$$\text{Power} = 1 - \Phi\left(z_{1-\alpha} - \frac{(R_1 - 1)\sigma_{BC}^2}{\sqrt{\sigma^{*2}/N_s}}\right)$$

Example 1 – Finding Sample Size

A company has developed a generic drug for treating rheumatism and wants to compare it to the standard drug in terms of the between-subject variability. A 2 x 4 cross-over design will be used to test the inequality using a two-sided test.

Company researchers set the variance ratio under the null hypothesis to 1, the significance level to 0.05, the power to 0.90, M to 2, and the actual variance ratio values between 0.6 and 1.4. They also set σ^2_{BC} = 0.4, σ^2_{WT} = 0.2, σ^2_{WC} = 0.3, and ρ =0.75. They want to investigate the range of required sample size values assuming that the two sequence sample sizes are equal.

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	**Sample Size**
Alternative Hypothesis	**Two-Sided (H1: $\sigma^2_{BT}/\sigma^2_{BC} \neq 1$)**
Power	**0.90**
Alpha	**0.05**
Sequence Allocation	**Equal (N1 = N2)**
M (Number of Replicates)	**2**
R1 (Actual Variance Ratio)	**0.6 0.7 0.9 1.1 1.3 1.4**
σ^2_{BC} (Control Variance)	**0.4**
σ^2_{WT} (Treatment Variance)	**0.2**
σ^2_{WC} (Control Variance)	**0.3**
ρ (Treatment, Control Correlation)	**0.75**

Output

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

Numeric Results

Solve For: Sample Size
Hypotheses: H0: $\sigma^2_{BT}/\sigma^2_{BC} = 1$ vs. H1: $\sigma^2_{BT}/\sigma^2_{BC} \neq 1$

Power		Sequence Sample Size			Number of Replicates	Between-Subject Variance		Within-Subject Variance		Between-Subject (Treatment, Control) Correlation	
Target	Actual	N1	N2	N	M	Ratio R1	Control σ^2_{BC}	Treatment σ^2_{WT}	Control σ^2_{WC}	ρ	Alpha
0.9	0.9008	142	142	284	2	0.6	0.4	0.2	0.3	0.75	0.05
0.9	0.9001	259	259	518	2	0.7	0.4	0.2	0.3	0.75	0.05
0.9	0.9000	2527	2527	5054	2	0.9	0.4	0.2	0.3	0.75	0.05
0.9	0.9000	2816	2816	5632	2	1.1	0.4	0.2	0.3	0.75	0.05
0.9	0.9007	356	356	712	2	1.3	0.4	0.2	0.3	0.75	0.05
0.9	0.9002	214	214	428	2	1.4	0.4	0.2	0.3	0.75	0.05

Target Power	The desired power value entered in the procedure. Power is the probability of rejecting a false null hypothesis.
Actual Power	The actual power achieved. Because N1 and N2 are discrete, this value is usually slightly larger than the target power.
N1	The number of subjects in sequence 1.
N2	The number of subjects in sequence 2.
N	The total number of subjects. N = N1 + N2.
M	The number of replicates. That is, it is the number of times a treatment measurement is repeated on a subject.
R1	The value of the between-subject variance ratio at which the power is calculated.
σ^2_{BC}	The between-subject variance of measurements in the control group.
σ^2_{WT}	The within-subject variance of measurements in the treatment group.
σ^2_{WC}	The within-subject variance of measurements in the control group.
ρ	The between-subject correlation of the average subject treatment-group measurements versus the average subject control-group measurements.
Alpha	The probability of rejecting a true null hypothesis.

Summary Statements

A 2×2M replicated cross-over design will be used to test whether the between-subject variance of the treatment (σ^2_{BT}) is different from the between-subject variance of the control (σ^2_{BC}) by testing whether the between-subject variance ratio (σ^2_{BT} / σ^2_{BC}) is different from 1 (H0: σ^2_{BT} / σ^2_{BC} = 1 versus H1: σ^2_{BT} / σ^2_{BC} ≠ 1). Each subject will alternate treatments (T and C), with an assumed wash-out period between measurements to avoid carry-over. With 2 replicate pairs, each subject will be measured 4 times. For those in the Sequence 1 group, the first treatment will be C, and the sequence is [C T C T]. For those in the Sequence 2 group, the first treatment will be T, and the sequence is [T C T C]. The comparison will be made using a two-sided, variance-difference test (treatment minus control) as described in Chow, Shao, Wang, and Lokhnygina (2018), with a Type I error rate (α) of 0.05. For the control group, the between-subject variance (σ^2_{BC}) is assumed to be 0.4, and the within-subject variance is assumed to be 0.3. The within-subject variance of the treatment group is assumed to be 0.2. The between-subject correlation between the average treatment measurement per subject and the average control measurement per subject is assumed to be 0.75. To detect a between-subject variance ratio (σ^2_{BT} / σ^2_{BC}) of 0.6 with 90% power, the number of subjects needed will be 142 in Group/Sequence 1, and 142 in Group/Sequence 2.

Dropout-Inflated Sample Size

	Sample Size			Dropout-Inflated Enrollment Sample Size			Expected Number of Dropouts		
Dropout Rate	**N1**	**N2**	**N**	**N1'**	**N2'**	**N'**	**D1**	**D2**	**D**
20%	142	142	284	178	178	356	36	36	72
20%	259	259	518	324	324	648	65	65	130
20%	2527	2527	5054	3159	3159	6318	632	632	1264
20%	2816	2816	5632	3520	3520	7040	704	704	1408
20%	356	356	712	445	445	890	89	89	178
20%	214	214	428	268	268	536	54	54	108

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.
N1, N2, and N The evaluable sample sizes at which power is computed. If N1 and N2 subjects are evaluated out of the N1' and N2' subjects that are enrolled in the study, the design will achieve the stated power.
N1', N2', and N' The number of subjects that should be enrolled in the study in order to obtain N1, N2, and N evaluable subjects, based on the assumed dropout rate. After solving for N1 and N2, N1' and N2' are calculated by inflating N1 and N2 using the formulas N1' = N1 / (1 - DR) and N2' = N2 / (1 - DR), with N1' and N2' 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.)
D1, D2, and D The expected number of dropouts. D1 = N1' - N1, D2 = N2' - N2, and D = D1 + D2.

Dropout Summary Statements

Anticipating a 20% dropout rate, 178 subjects should be enrolled in Group 1, and 178 in Group 2, to obtain final group sample sizes of 142 and 142, respectively.

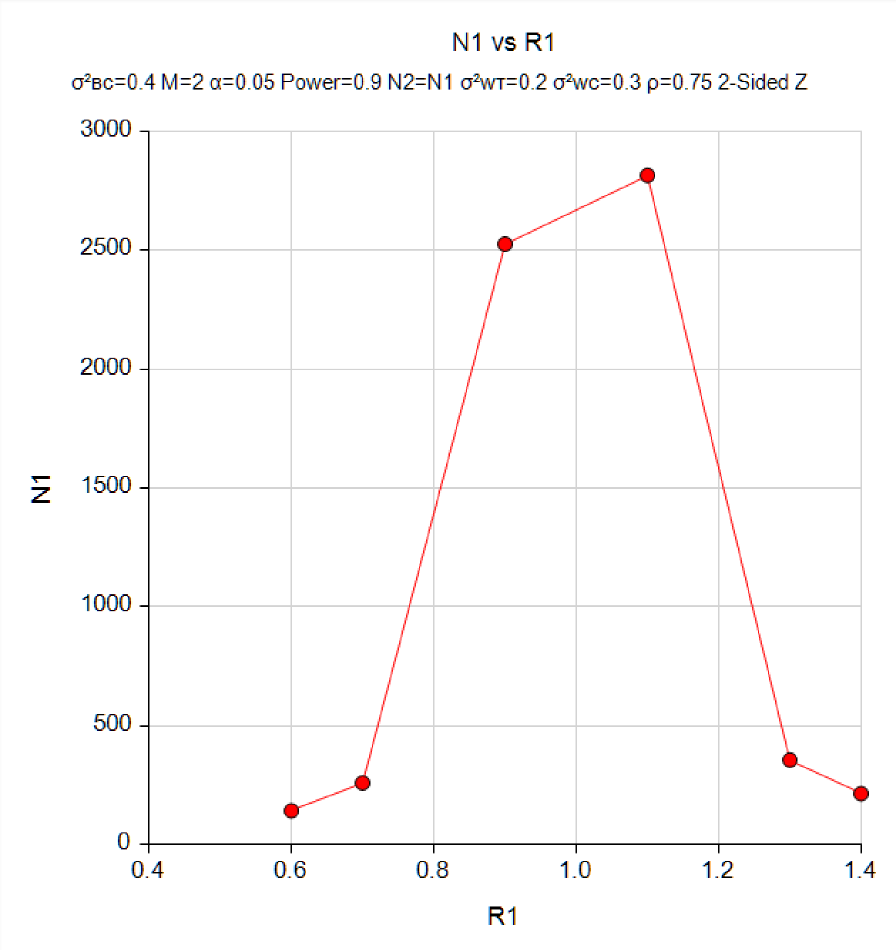
References

Chow, S.C., Shao, J., Wang, H., and Lokhnygina, Y. 2018. Sample Size Calculations in Clinical Research, Third Edition. Taylor & Francis/CRC. Boca Raton, Florida.
Chow, S.C., and Liu, J.P. 2014. Design and Analysis of Clinical Trials, Third Edition. John Wiley & Sons. Hoboken, New Jersey.

This report gives the sample sizes for the indicated scenarios.

Plots Section

Plots



This plot shows the relationship between sample size and R1.

Example 2 – Validation using Chow and Liu (2014)

We will use an example from Chow and Liu (2014) page 517 to validate this procedure.

In this example, significance level = 0.05, power = 0.80, M = 2, σ_{BT} = 0.3, σ_{BC} = 0.4, σ_{WT} = 0.2, σ_{WC} = 0.3, and ρ = 0.75. From these values, we find that R1 = 0.5625. The resulting sample size is found to be 66 per 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**
Alternative Hypothesis	**Two-Sided (H1: $\sigma^2_{BT}/\sigma^2_{BC} \neq 1$)**
Power	**0.80**
Alpha	**0.05**
Sequence Allocation	**Equal (N1 = N2)**
M (Number of Replicates)	**2**
R1 (Actual Variance Ratio)	**0.5625**
σ^2_{BC} (Control Variance)	**0.16**
σ^2_{WT} (Treatment Variance)	**0.04**
σ^2_{WC} (Control Variance)	**0.09**
ρ (Treatment, Control Correlation)	**0.75**

Output

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

Numeric Results

Solve For: Sample Size
Hypotheses: H0: $\sigma^2_{BT}/\sigma^2_{BC} = 1$ vs. H1: $\sigma^2_{BT}/\sigma^2_{BC} \neq 1$

Power		Sequence Sample Size			Number of Replicates	Between-Subject Variance		Within-Subject Variance		Between-Subject (Treatment, Control) Correlation	
Target	Actual	N1	N2	N	M	Ratio R1	Control σ^2_{BC}	Treatment σ^2_{WT}	Control σ^2_{WC}	ρ	Alpha
0.8	0.8022	66	66	132	2	0.563	0.16	0.04	0.09	0.75	0.05

The sample sizes match Chow and Liu (2014).