



# BIOMETRICS INFORMATION

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PAMPHLET NO. # 5

DATE: April 12, 1988

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SUBJECT: Understanding Replication and Pseudo-replication

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The layout of an experiment is not understood unless the manner of treatment replication is understood. Recognizing when treatments are not actually replicated (pseudo-replication) requires determining the actual experimental units for each treatment. This determination is often more difficult than one might expect.

A treatment is replicated when it is applied to two or more experimental units (e.u.'s) in one of two main ways:

- 1) in a completely randomized design treatments are randomly assigned to e.u.'s.
- 2) in a randomized block design treatments are randomly assigned to one e.u. per block.

Since blocks are often called "reps" the term replication is restricted by some to mean blocks. Unfortunately, this can cause confusion so that a REP term is occasionally included in the ANOVA table of a completely randomized design, when it should be indicated by the error term (e.u.'s nested within treatments).

When thinking about the replication of an experiment it is important to recognize what the e.u.'s are (it is possible for different treatments in an experiment to have different e.u.'s, as occurs with the split-plot). An experimental unit is a basic unit of experimental material to which one level of a treatment or one combination of levels of treatments is applied. An e.u. may be comprised of many sampling units (s.u.) on which actual measurements are made. A common example is the use of a row of 50 trees as an e.u. The actual measurements are made on the individual trees in the row but it is the row mean which represents the e.u. in an ANOVA. Thus an experiment may have 30 rows of trees as e.u.'s but 30x50 measurements will have to be made to get the 30 row means. If the 30x50 numbers obtained are seen as replicates then there appears to be a lot more data than there really is. These numbers are considered PSEUDO-REPLICATES.

Replication is used in experiments to help answer the following question: "Are the observed treatment effects due to the treatment or due to inherent differences between the e.u.'s that would have been observed anyway?" By applying each treatment level to several e.u.'s an estimate of the "natural" variability of the e.u.'s can be separated from and compared with treatment differences (see pamphlet #2). A major problem of experiments with no replication (which may be obscured by pseudoreplication) is that the above question cannot be answered statistically. Of course, the experimenter may argue on biological grounds for treatment differences but others are free to disagree. So questions the experiment was to answer may remain unanswered.

The use of the pseudo-replicate variation to test for treatment differences in an ANOVA situation is incorrect although it is very tempting to do so. The F-tests are more likely to show a treatment effect when, in fact, there is none since within e.u. variation should be smaller than between e.u. variation.

There are legitimate reasons for designing experiments where at least one of the treatments is not replicated. Most commonly there is a lack of resources such as when experimental units are greenhouses or plots for different mechanical site preparations. Although these experiments may provide useful information they suffer from a serious design problem which must be recognized.

**Reference:**

Hurlbert, Stuart H., 1984, *Pseudo-replication and the design of ecological field experiments*, Ecological Monographs, 54:187-211.

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