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THE DESIGN OF FORESTRY FIELD TRIALS

The appropriate experimental design to use for seedling out planting trials will vary due to numerous factors such as number of treatments, site heterogeneity, desired inference of results, and of course practicality. There are however a number of principles (e.g. randomization, replication) which such be followed in any design.

The first task is to define the problem. What is the specific question(s) to be asked? This question is developed into a null hypothesis which must be statistically rejected before the alternate hypothesis (there are differences between the treatments) can be accepted.

Treatments must be applied to more than one experimental unit (group of seedlings) to be replicated. The experimental unit is the basic unit of material to which one level of treatment or combination of treatments has been applied. This could be one seedling in a Styroblock™, one row of seedlings in a Styroblock™, one Styroblock™, one pallet, or one greenhouse, etc.; depending to how many seedlings comprised one treatment application). This is essential, as without replication, the validity of your conclusions can not be measured. The most common pitfall regarding replication is the use of pseudo-replication, which occurs when the sampling units (seedlings comprising one experimental unit) are regarded as replicates in the analysis.

The sample size must be large enough so that enough seedlings will be available for all phases of the study. Various factors must be taken into account, such as survival, type of measurements (non-invasive vs. destructive), duration of experiment, and the desired precision of the measurement. For example, to measure mean survival to the nearest 1%, 100 seedlings or more (e.g. 4 replications of 25 seedling plots) would be needed per treatment; compared to precision of 6 to 7% obtained from a design comprising 3 replications of 5 seedlings per plot. A minimum of 3 replications is recommended, but this depends on: the desired detectable level of difference (%) between 2 treatments, the coefficient of variation (variation among plots receiving the same treatment as a percentage of the treatment mean), and the type 1 error rate (significance level) and degrees of freedom in the design. Basically, if the number of treatments is small, then the replications should be increased to increase the sensitivity of the experiment to detecting differences between treatments. Single tree plots (one seedling per treatment) are not recommend due to possible losses of replicates because of mortality; but have the advantage of increasing precision by removing site variation on very non-uniform sites (see blocking below). Also, the use of a row of buffer trees (not included in measurements) around the experimental trees is recommended.

If we have a trial with only 2 levels of one treatment (e.g. fertilizer and control), the results may be analyzed using a t-test or one-way ANOVA. In most cases, the chose of which experimental design to use will depend mainly on site uniformity and number of treatments. If we have a small number of treatments, and a very homogenous trial site, than the completely randomized design (CRD) may be used. This is the most efficient design, in that the smaller number of experimental units required decrease the cost and work. However, it is not used that often by researchers as they feel uncomfortable assuming that their experimental area is truly homogenous. As the

number of replications should be increased when using smaller numbers of treatments, it may be just as easy (at the out planting end) to increase the levels of an existing treatment (e.g. fertilizer) or include other treatments (e.g. species) instead of increasing the number of replications; which would increase the applicability of results gleaned from the experiment.

Example: Completely Randomized Design with 1 Treatment at 2 levels applied to one species/SL/stock type combination at the nursery:

Treatment: Slow Release Fertilizer Levels: 1- 20 kg/m3
2- 0 (control)

LAYOUT: site (S) - 1
species (Sp) -1
fert rates (Rf) - 2
reps (R) - 8 rows of seedlings per fert rate
sampling units - 20 seedlings per row

Total 1S x 1Sp x 2Rf x 8R = 16 experimental units x 20 = 320 seedlings + 70 buffer seedlings = 390 seedlings on site. Also requires 160 seedlings for pre-out planting measurements, thus 550 seedlings total.

This is a very simple design that would yield a minimum of information (i.e. whether there is a difference in measured attributes between fertilized and control seedlings). To be effective, it would have to be laid out in an area of the site that had uniform characteristics such as soil, slope, drainage, etc. The shape could be rectangle or square - whatever encompasses the most uniform area. The rows must be arranged randomly with the trial area. Row spacing and tree spacing within row would depend on species, site conditions, and the duration of the experiment. Long term results (e.g. > 10 years) require operational spacing (approximately 3x3 m), although if planned, trees could be removed throughout the course of the experiment to eventually arrive at the proper spacing. Minimum suggested spacing is 1 m between rows and 0.5 cm between plants within rows. The amount of replications could be decreased if the trial was repeated on the same site in various locations, or repeated at different sites. This experiment could also be laid out as a randomized complete block, if the site conditions are very heterogeneous (see below).

An alternative is to use blocking in the experimental design. The Randomized Complete Block (RCB) experimental design is very common in forestry. The idea is to make the difference between blocks as large as possible, but the differences within blocks as small as possible. All treatments occur randomly within each block, with the blocks in effective supplying the replication. The optimum plot size will vary, but has been recommended as 500 m^2. The blocks are not assigned randomly. Blocks should always be set up across a gradient (i.e. up and down a slope rather than across a slope).

Example RCB Mycorrhiza Trial with 3 treatments each having 2 levels:

Treatments: Site (S) - 1
Species (Sp) -1 Dfc
Seedlots (Sl) - 2
Application dates (D) - 2

Rates (R) - 2 (manufacturers' suggested rate and control)
Blocks (B) - 4 (2Sl x 2D x 2R x 20 seedlings per row = 160 seedlings/block)

Total: 1Sp x 2 Sl x 2D x 2R x 4B = 32 experimental units x 20 = 640 seedlings + 102 buffer seedlings = 742 seedlings per site. Also requires 320 seedlings for pre-outplanting measurements, thus 1062 seedlings total.

For this design to be more effective than a completely randomized design, it is best if laid out in such a fashion that site conditions within blocks are uniform, while site conditions between blocks are not. The blocks could be physically side by side, but this does not have to be the case.

All treatment combinations must occur randomly within each block. Each treatment combination (experimental unit) is made up of 1 row of 20 seedlings, as it is felt this would give adequate measurement precision, and allow for seedling mortality, damage and sampling throughout the experiment. If seedlings are to be destructively sampled (e.g. tissue analysis) midway through the experiment, all 20 seedlings need not be measured each sampling date. For example, 10 randomly selected (non-deformed) seedlings per row, could be measured each sampling date. Row spacing and tree spacing within row would depend on species, site conditions, and the duration of the experiment. Long term results (i.e. > 10 years) require operational spacing (e.g. approximately 3x3 m), although if planned, trees could be removed throughout the course of the experiment to eventually arrive at the proper spacing. Minimum suggested spacing is 1 m between rows and 0.5 cm between plants within rows.

As you can see, for each trial, the first step is deciding what you would like to learn from each particular experiment. Probably the next logical step would be to determine what outplanting sites are available. After a brief consultation to learn all the particulars, a specific experimental trial working plan can be written up.

PRT Altogether a better approach
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