Brien et al (2011) have discussed 12 general principles in designing two-phase experiments.

The first principle is always having a skeleton ANOVA table to evaluate the design.

I have generalised the method in constructing the ANOVA table for any two-phase experiment.

Always start with a simple composed randomisation method, which is all factors from one tier are randomised to second tier and all factor in the second tier is randomised to the third tier.

The treatment should always confounded with the factor the contribute the smallest variation.

The replication is required when there is uncontrolled variation in the Phase 2 experiment. This is typical the case in the biological experiments. This is because the variation of between using the different machines can always be very different. Especially if the given machine is just been introduced, then the replication of the sample s from the Phase 1experiment is essential. Thus, the Principle 11 is not required.

The treatment from the Phase 2 experiments is generally not the main interests for the biologists. For the case of MudPIT experiment, it is the tag effects. For the cases of microarray experiments, it is dye effects. For the next gene sequencing, it can be the effects using different barcoding. The biologists only interest in the different between the treatments of interest from the Phase 1 experiment. Thus, the Principle 12 is not required.

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