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Estimation of the variation that can be attributed to different levels in a clinical trial of a vaccine against *Campylobacter* in broilers

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This presentation is focusing on the interaction between the experimental design of a vaccination trial and appropriate data analysis using a trial of a vaccine against *Campylobacter* in broilers as an example. This study was designed using four rotations with eight isolators per rotation (10 chickens per isolator). Treatment was administered at isolator level on day 14 (vaccine or placebo). The broilers were inoculated with *Campylobacter jejuni* at day 31 and slaughtered at day 42. The numbers of *Campylobacter* (cfu/g) were obtained in the laboratory using selective cultivation methods and log transformed to obtain a Gaussian distribution. Initially, the effect of the vaccine was analyzed using all data in a t-test. Subsequently, the t-test was stratified by rotation. Finally, mixed linear models were used, taking into account the physical hierarchical setup of the trial. Results from the t-test indicate an effect of the vaccine, whereas the result obtained from the complex model indicated high variability between birds and isolators but not significant vaccine effect. The apparent observed differences between vaccinated and placebo groups in the t-tests could be attributed to the variation between incubators. Broilers in the same isolator had more equal numbers of *C. jejuni* compared to chickens in other incubators. It is possible that chickens in the same isolator re-infect each other with *Campylobacter*. In this study, the design effect was considerable, reducing the effective sample size (67 animals instead of the 290 animals included). The clustered design used in this trial was trying to emulate the clustering effect found in broiler flocks and farms.