

5. H. U. Choi and R. Carubelli, *Biochemistry* 7, 4423 (1968).
6. L. Warren, *J. Biol. Chem.* 234, 1971 (1959).
7. R. Carubelli, R. E. Trucco, R. Caputto, *Biochim. Biophys. Acta* 60, 196 (1962); B. H. Taha and R. Carubelli, *Arch. Biochem. Biophys.* 119, 55 (1967); D. R. P. Tulsiani and R. Carubelli, *Fed. Proc.* 29, 3034 (1970).
8. O. H. Lowry, N. J. Rosebrough, A. L. Farr, R. J. Randall, *J. Biol. Chem.* 193, 265 (1951).
9. P. Talalay, W. H. Fishman, C. Huggins, *ibid.* 166, 757 (1946).
10. W. C. Schneider and G. H. Hogeboom, *ibid.* 198, 155 (1952).
11. H. Suzuki, M. Ohsawa, H. Endo, *Endocrinol. Jap.* 16, 121 (1969).
12. D. E. Comings and T. Kakefuda, *J. Mol. Biol.* 33, 225 (1968).
13. N. Keller, thesis, University of Oklahoma (1970); M. J. Griffin and N. Keller, in preparation.
14. H. B. Bosmann and R. A. Winston, *J. Cell Biol.* 45, 23 (1970).
15. Supported in part by NIH grants AI-04954, NS-09176, and CA-10614. R.C. is recipient of a PHS research career development award (5-K3-AM-38649).

1 June 1970; revised 1 October 1970

Analysis of Data in a Study of Nest Parasitism, Productivity, and Clutch Size in Purple Martins

Moss and Camin (1) used the paired-comparisons *t*-test on mean daily weights of treated and untreated nestling purple martins for 29 days, and concluded that the treated nestlings were heavier ($P < .001$). The test is inappropriate because the 29 "paired comparisons" are not independent, the same birds having been weighed day after day, not to mention that the error variance is probably also changing from day to day.

Similarly, Moss and Camin indicate significant differences in mean maximum weight, resting their tests on standard deviations in each group, without regard to whether there are significant differences between nests. Although perhaps in these data nest means do not differ significantly by the standard of variability of nestlings in the same nest, it cannot be argued that the nest component of variance is zero. Hence, their tests should have used numbers of degrees of freedom derived from numbers of nests, not nestlings, and are mostly nonsignificant. For example, the 1967 comparison of treated to untreated has only two degrees of freedom for nests within treatment. Hence, where Moss and Camin indicate $P < .001$, actually the .05 level is barely reached on the assumption that there is little or no variance between nests. In fact, a comprehensive analysis is needed. Their reference 12, mentioning that a higher mean weight for a brood of five than for (two broods of) four "may be attributable to unusual food-gathering proficiency of a single pair of parents," is on the right track, but apparently they did not perceive how to perform a correct test of significance.

As a final point, in examining a 2×3 table of frequencies of brood sizes according to treatment, Moss and Camin appear to have tested broods of size three against those of size five,

omitting broods of four because they were equally frequent for treated and untreated conditions. Presumably their interest lay in progressive change, to which broods of size four are as relevant as other brood sizes. An appropriate test finds that there is progressive change ($P < .01$).

H. W. NORTON

Department of Animal Science,
University of Illinois, Urbana 61801

Reference

1. W. W. Moss and J. H. Camin, *Science* 168, 1000 (1970).
- 4 June 1970

Norton (1) is correct in objecting to the use of the paired-comparisons *t*-test as a test for seasonal difference between martin nestling weights (2). However, how crucial is this objection? Statistical tests are often robust, even when an occasional rule of application is unintentionally violated. Would another test give different or comparable results? To examine the validity of his objections, we elected to test our data again following a randomization approach (3). With 500 iterations, probability values for equivalence of mean daily weight for each day over the course of the 1966 nesting season were obtained. These were then combined (4) to obtain a combined probability value $P_c = .005$ for equivalence of mean weight of treated and untreated nestlings over the course of the season. Thus, our conclusion that parasitized nestlings are lighter than those unparasitized remains unchanged (2).

Norton's second criticism relative to our testing of mean maximum nestling weight (MNW) seems somewhat more valid. Reanalysis of our data by using a randomization approach yielded probability values that were in fact higher than those published initially in our table 2 (2). In the majority of cases these lay somewhat above the .05 level

generally taken as an indicator of statistical significance. However, comparisons within a single year, although still strongly suggestive, are of less interest biologically than conclusions drawn on the basis of several years' data. To test whether unparasitized and parasitized nestlings have the same mean MNW one can combine the seven relevant, recomputed probabilities from modified table 2 to obtain $P_c = .007$, which allows us to reject firmly a hypothesis of equal weight for parasitized and unparasitized young of equal brood size. The case for decrease in mean MNW with increase in brood size is somewhat less strong over the five seasons of data, but is still quite suggestive; a combined probability $P_c = .06$ for equivalence of mean MNW indicates an overall trend toward decrease in weight with increase in brood size. Considering separately brood sizes of three versus four, and four versus five, and combining recomputed probabilities, we obtain for the former case $P_c = .49$, for the latter $P_c = .098$. Thus, a decrease in mean MNW is much more marked as we go from brood sizes of four to five, than from three to four, as suggested earlier in our reference 12 (2). In contrast, combining recomputed probabilities in a test of mean MNW reached by unparasitized broods of n versus that reached by parasitized broods of $n-1$ yields a value of $P_c = .96$, which confirms our earlier statement that there is no apparent difference in this case. Finally, if we must include data on broods of four when comparing broods of three and five, then we are glad to know that the probability value determined with this test is somewhat lower than that which we previously reported.

In summary, Norton's conclusions may be of interest from a statistical viewpoint, but their force is considerably blunted by the fact that they do not affect the conclusions drawn in our report.

W. WAYNE MOSS

Academy of Natural Sciences,
19th and Parkway,
Philadelphia, Pennsylvania 19103

JOSEPH H. CAMIN

Department of Entomology,
University of Kansas, Lawrence 66044

References

1. H. W. Norton, *Science*, this issue.
2. W. W. Moss and J. H. Camin, *ibid.* 168, 1000 (1970).
3. R. R. Sokal and F. J. Rohlf, *Biometry. The Principles and Practice of Statistics in Biological Research* (Freeman, San Francisco, 1969), p. 629.
4. ———, *ibid.*, p. 621.
- 25 September 1970