MATTERS ARISING

V-test is not a statistical test of 'homeward' direction

THE V-test[1] is widely used by behaviourists to evaluate circular data from studies of animal orientation. However, the statistic has been almost universally misinterpreted.

Batschelet[1] very clearly defined the scope of the test when he wrote: "the null hypothesis that we are going to test is randomness, which means that the angles of the sample are independent observations from a uniform circular distribution". However, he further stated that "the V-test leads to significance only when there is sufficient clustering around the predicted direction". It has been this statement, apparently, that has led to some misunderstanding. We can reject the null hypothesis when we have sufficient clustering around the predicted direction. However, the mean direction may still be significantly different from the predicted direction.

Nevertheless, it has been generally assumed that the test will determine whether a sample is oriented in a predicted direction. For example, in four recent papers[2], the V-test was used, in each case as if rejection of the null hypothesis supported the conclusion that animals were orienting in the appropriate direction. Samples with significant V probabilities were commonly called 'homeward directed'. There has even developed a myth that the V-test complements the Rayleigh test, the latter determining whether a sample is nonrandom, and the former whether the nonrandom sample is properly directed. Thus, Hartwick *et al.*[3] said that their data were "evaluated for nonrandomness by the Rayleigh test" and "for homeward directedness by the V-test".

This, unfortunately, is not valid. The null hypothesis of the V-test is the same as for the Rayleigh test, as we have pointed out. The two tests use different methods, but they have the same function. It is true that the calculation of the V statistic uses a predicted direction, but only so that the sample's component on that axis can be compared with the distribution of cosines from "samples from a uniform circular population" (ref. 4). In rejecting the null hypothesis, the researcher can claim that his data are not from a uniform circular population but he cannot claim that the data are from a population of samples oriented in the predicted direction.

A test that addresses the latter question is described elsewhere[5]. The null hypothesis states that the sample mean is drawn from the population of means around the predicted direction. After the nonrandomness of the sample has been demonstrated by either the Rayleigh test or the V-test, the Watson test can be used to determine whether a confidence interval around the sample mean includes the predicted direction. If the theoretical mean is not within the 95% confidence interval, then the null hypothesis is rejected, and the alternative hypothesis accepted, that the sample mean is not in the predicted direction.

As both tests use the sample mean's component in the predicted direction, sometimes a misinterpreted V-test may seem to give the same results as the Watson test. However, this is not always so; a case in point is the paper by Mather and Baker[6]. Here, the mice transported in the reversed magnetic field had a second-order mean direction of 131°, which is significant by the V-test, using as the predicted direction 180°, the reverse of that expected for the controls. The authors stated that the orientation of the experimental animals was "significantly clustered around 180°". However, use of the more appropriate Watson test shows that the 95% confidence interval around the experimental sample does not include 180°. Consequently, there is no evidence for precisely reverse orientation. From the V-test, we know that the sample mean has a nonrandom component in the predicted direction, but we cannot conclude that the mean itself is in the predicted direction. Thus we cannot conclude anything about the appropriateness of the orientation to the experimental manipulation.

Clearly, Mather and Baker are not alone in their misinterpretation; many others in the field have also used the V-test in the same way. Now seems like a suitable time for reform.

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MATHER AND BAKER REPLY—Aneshansley and Larkin have raised an important point concerning the interpretation of the V-test, a statistical test that is widely applied in the study of animal orientation. It is now apparent that for several years many behaviourists, following Batschelet[1], have misinterpreted the V-test by assuming that because there is a nonrandom vector in a predicted direction, the sample mean is not significantly different from that predicted.

Aneshansley and Larkin have used our paper[2] on magnetic orientation by mice to illustrate their point. After commenting on our application of the V-test, the authors state that no conclusions can be made about the appropriateness of the orientation to the experimental manipulation. While we accept the comments regarding the V-test, we stress that our main conclusions concerning rodent magnetic orientation are still valid: reversal of the direction of the ambient magnetic field during displacement subsequently influences the animals' ability to determine the direction of home. Most important is the fact that experimental manipulation of the magnetic field results in a significant difference in the directional preferences of control and experimental mice (Watson's $U^2_{n,m}$ nonparametric two-sample test). Moreover, both for controls and experimental animals, the mean orientation has a nonrandom component in the predicted direction (that is, 0° and 180° respectively). Also, the mean for the controls is actually in the predicted direction (Watson's F-test). We can only not say (nor did we in our paper) that the mean for the experimental animals was also in the predicted direction.

We agree with Aneshansley and Larkin, therefore, that we have not presented evidence for precisely reverse orientation but that was never our intention. The main point of our paper was to demonstrate a magnetic sense of direction in a rodent and its involvement in route-based navigation. This still stands.

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Molecular packing in collagen

IN RECENT papers, data from chemical cross-linking[1] and electron microscopy[2] were quoted to challenge aspects of the quasi-hexagonal model[3] for the molecular packing in collagen. Any models must, of course, account for X-ray, electron

0028–0836/81/380239—02$01.00