

This Week's Citation Classic

Lund J W G, Kipling C & Le Cren E D. The inverted microscope method of estimating algal numbers and the statistical basis of estimations by counting. *Hydrobiologia* 11:143-70, 1958.
[Freshwater Biological Association, Ferry House, Ambleside, Westmorland, England]

Certain methods for estimating the abundance of the cells of algae and other microscopic aquatic organisms are described. One method is considered in detail, including the preceding sampling, sub-sampling, and preparing the material for counting on an inverted microscope, with special reference to the statistical errors involved. [The SCF[®] indicates that this paper has been cited over 180 times since 1961.]

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"Soon after I joined the staff of the Freshwater Biological Association, then a group of ten scientists in a mock Victorian castle by the side of Windermere, and began estimating the abundance of algae by counting cells, I had to consider the human and statistical errors involved in any method. Some were related to sampling systems and techniques, others to the manipulation of the material in preparing it for counting, using a normal or inverted microscope or a specially designed chamber (e.g., a haemocytometer). It was also necessary to find out to what extent two or more workers obtained the same answer when counting the same preparation, for example, a single sedimentation. Then there was the statistical variation if a series of sub-samples were taken from a mixed sample. It was necessary to determine the statistical errors inherent in counting randomised particles, which could be single cells or groups of cells (colonies or filaments). The individuals would follow a Poisson distribution, but if they contained more than one cell, there would be an additional and independent statistical error related to the

mean number of cells per individual and the distribution of the varying numbers in the sample.

"Arising from discussions of these problems we came to the conclusion that there was a need for a guide for beginners or others, written in English, and that, for our purpose, the main emphasis should be on the inverted microscope method of Utermöhl,¹ whose papers were in German.

"It is interesting to note that, unlike most of the papers considered in this series, there was nothing novel in our work. Even the method of sampling by a flexible tube had been described previously.² Twenty-five years ago, when this work was done, there were relatively few statistical texts in English for biologists and, excellent though some were, they involved a mathematical approach which may well have seemed difficult or forbidding because, also unlike today, a course on statistics was not an essential part of the training of biologists.

"We tried to describe methods suitable for a wide range of circumstances, to use a commonsense approach with a minimum of statistical theory and jargon, and to make the paper easily understandable. Its frequent citation suggests that the simple, practical, and easily understandable methodology based on actual tests by several workers filled a need. In relation to this, it is noteworthy that even recently an introductory text on the statistics of estimating aquatic organisms, with special reference to benthic invertebrates, has run into a second edition and sold more copies than any other "Scientific Publication" of the Freshwater Biological Association.³

"I was fortunate in having as collaborators a statistician (Kipling), who came to biology after war work on torpedoes and rockets, and a biologist (Le Cren), experienced in the use of statistics in fishery investigations in Britain and America. It would have been more appropriate if the authors had been listed alphabetically, in which case my name would have come last; but my coauthors, in their modesty, considered that I, the algal ecologist, should be the senior author."

1. Utermöhl H. Über das umgekehrte Mikroskop. *Arch. Hydrobiol. Plankt.* 22:643-5, 1931.
2. Land J W G. Studies on *Asterionella*. 1. The origin and nature of the cells producing seasonal maxima. *J. Ecology* 37:389-419, 1949.
3. Elliott J M. *Some methods for the statistical analysis of samples of benthic invertebrates*. Ambleside, England: Freshwater Biological Association, 1977. Scientific Publication no. 25. 160 p.