Comments and Communications

Mitochondria or Microorganisms?

A PAPER by U. N. Lanham, entitled "Observations on the Supposed Intracellular Symbiotic Microorganisms of Aphids," contains some interesting observations of a preliminary nature (*1*). However, the facts reported do not warrant the statement that "the aphid particles lack DNA, and . . , they are not microorganisms." Failure to obtain certain staining reactions generally characteristic of DNA does not prove that a particular object of microscopic dimensions is not a microorganism. Certain stages of the erythrocytic cycle of malaria parasites do not show any structures that give the Feulgen reaction (*2*), although the reaction can be brought out if the malarial pigment is first removed (*3*). Even under the latter conditions, the chromatin stains very faintly in large trophozoites and in gametocytes. A similar negative or barely visible Feulgen reaction has been noted in certain insect oocysts (*4*), cells which nevertheless almost certainly contain DNA. It is possible that the symbiotes of aphids exhibit different staining reactions at different stages of their developmental cycle.

Especially important in the demonstration that certain insect symbiotes are undoubtedly symbiotic microorganisms have been the researches of A. Koch (*5*), M. Blewett and G. Fraenkel (*6*), and N. C. Pant and G. Fraenkel (*7*). These workers have shown that several species of beetles which contain yeastlike symbiotes are able to grow on diets deficient in certain vitamins, whereas the same species, if experimentally deprived of their symbiotes, require these vitamins in their diet. Moreover, two of the species of symbiotes have been grown in culture, and the cultures have been used to infect symbiote-free insects.

Certainly much remains to be learned as to the nature and function of the intracellular symbiotes of aphids and related insects. Equally certainly, more than microscopic observations on one species of aphid will be needed before definitive conclusions can be drawn.

WILLIAM TRAGER

The Rockefeller Institute for Medical Research
New York

References

1. LANHAM, U. N. *Science*, **115**, 459 (1952).
2. SCHÄFFER, K. *Acta Tropica*, **2**. 17 (1945).
3. DEANE, H. W. *J. Cell. Comp. Physiol.*, **26**, 139 (1945).
4. BAUER, H. *Z. Zellforsch. u. mikroskop. Anat.*, **18**, 254 (1933).
5. KOCH. A. *Naturwissenschaften*, **21**, 543 (1933).
6. BLEWETT, M., and FRAENKEL, G. *Proc. Roy. Soc.* (*London*), B, **132**, 212 (1944).
7. PANT, N. C., and FRAENKEL, G. *Science*, **112**, 498 (1950).

A NEGATIVE result, such as failure to demonstrate the presence of a given substance, is, of course, usually unsatisfactory, and although such a result might lead one or another investigator to the conclusion that the substance in question is really absent, it cannot be regarded as affording absolute proof. The comments by Dr. Trager on the difficulties of staining *Plasmodium* by the Feulgen technique are not strictly relevant, since the conclusions in question were based on the use of acid-Giemsa; but it is quite possible that Giemsa would also fail in this instance. Certain bacteria also are reported to have nuclei resistant to acid-Giemsa, although with some of these (including *Azotobacter*) I have had little difficulty in obtaining densely staining internal structures of some kind, following fixation of dry films in absolute methyl alcohol.

The example of the microorganisms of beetles (*Stegobium* and *Lasioderma*) studied by Koch and others may or may not be relevant to the aphid problem. These microorganisms are said to be transmitted from one generation to another on the eggshells, which have to be eaten by the newly hatched larva. Nothing like this occurs in aphids, in which a cytoplasmic mode of transmittal has been postulated. Also, the beetle microorganisms are found free in the digestive tract; or, when intracellular, the host cells are associated with the digestive tract. There is no conclusive evidence of extracellular occurrence of the aphid particles, and the cells containing them ("mycetocytes") have nothing to do with the digestive tract. Studies of intracellular "microorganisms" in insects have suffered from a certain amount of uncritical jumbling together of unrelated evidence from diverse groups of insects, or even from protozoans and leguminous plants, when this seemed expedient.

In aphids, it seems well established that every individual, of every species, has "mycetocytes" that are packed with the particles in question. From the biological viewpoint, the hypothesis that they are symbiotic microorganisms seems to be a more unlikely, difficult, and complex one than the hypothesis that they are intracellular particulates of the nature of mitochondria. The reasons for assuming the more complex hypothesis to be true from rather early in the history of the subject, and for placing the burden of proof on those holding the simpler hypothesis, can be discussed adequately only by a detailed analysis of the literature of microbial symbiosis in both animals and plants.

The conclusion that the intracellular particles of aphid "mycetocytes" lack DNA and are therefore not microorganisms, reached on the basis of the previously mentioned negative evidence, should also be considered in the light of the following estimate of the major points of the controversy: (1) The aphid particles look like microbes when stained with the older standard stains. It should be noted, however, that when techniques are available which bear critically on the problem, such as the acid-Giemsa technique, the evidence from staining is, at least so far, negative. (2) Proponents of the microbial theory can point out that the aphid particles do not resemble classical mitochondria because of their stability and their size. However, the studies of Watanabe and Williams (*J. Gen. Physiol.,* 34, 675 [1951]) on the sarcosomes of insect flight muscles disclose another type which approaches the stability and size of the aphid particles. (3) The aphid particles are supposed to be added intact to the developing egg from the "mycetocytes" of the parent, so that the egg can be regarded as being infected by exogenous microorganisms. But if one examines the critical review of, and careful original work on, the subject of the ontogeny of the "mycetocytes" by Uichanco (*Philippine J. Sci.,* 24, 143 [1924]), one will find no direct or conclusive evidence for the continuity of the particles of the parental mycetocytes with the particles that suddenly appear in the developing egg or embryo in the oviduct. (4) The aphid particles are said to have been grown *in vitro.* All such claims need verification. Some reportedly successful experiments involve very simple techniques and can easily be repeated. My own attempts to cultivate them, including the use of hanging drop techniques where individual particles could be observed, were not successful.

If the aphid particles are cell particulates, mitochondrial in nature, the question is raised as to the function of cells apparently containing little more than mitochondria. These cells are intimately associated with the fat body in aphids, and what are possibly similar cells in some other insects are also imbedded in the fat body. Since mitochondria are centers of Krebs cycle activity, it might be tentatively postulated that there is a division of labor among the cells of the fat body, certain cells ("mycetocytes") being primarily responsible for providing the energy for the synthesis of fats, proteins, and glycogen, with the typical fat cells being responsible for the storage of these materials. Present work on the particles of the "mycetocytes" of aphids and other insects is exploring this possibility. The presence of Krebs cycle enzymes in the particles would not, of course, directly disprove the microbial hypothesis. The presence or absence of DNA appears to be the most critical question in this respect, and I feel that until someone can show conclusively that the particles contain DNA granules or nuclei, they should be regarded as cell particulates rather than as microorganisms.

U. N. Lanham

Department of Zoology, University of Michigan

Book Reviews

Botany of Sugarcane. C. van Dillewijn. Waltham, Mass.: Chronica Botanica; New York: Stechert-Hafner, 1952. 371 pp. $6.00.

A comprehensive new book of value to sugar technologists and cane growers alike has been produced by Dr. van Dillewijn, formerly director of the Sugar Experiment Station, Cheribon, Java. The title page appears to suggest that the book is part of a series in preparation, or proposed, entitled "Handbook of Sugarcane." Volume I, *Botany of Sugarcane,* is devoted to the structure, growth, and physiology of the sugar-cane plant, with emphasis on the application of scientific studies to crop production. Chapters 1–6, inclusive, comprise a richly illustrated description of the outer and inner morphology of sugar cane, with separate discussions of the stem, bud, leaf, flower, and root and an evaluation of the characteristics useful in identification of sugar-cane varieties. Sources used by Dr. van Dillewijn in preparing this portion of the book consist of the published records of specialists from late in the past century to the most recent years.

Section II, under the general heading "Physiology," contains chapters on germination, tillering, growth, vegetative composition, chemical composition, nutrition, water relations, photosynthesis, and respiration.

For cane growers without access to the large volume of technical literature on sugar cane this book will provide in compact form an excellent, up-to-date, and full account of research on the biology of sugar cane. Of special interest are the careful descriptions of root development and tillering and the progressive development of ratoon crops after the plant cane crop is harvested. For the first time in a sugar-cane handbook space is given to the important growth-regulating substances. The whole treatment of growth processes contains much that is useful in practical manipulation of the crop. The chapters on nutrition and on water relations similarly present fundamental information on fertilizer and water requirements and the interrelation of these factors as they influence growth and sugar production. There are 617 references to technical literature.

E. W. Brandes

Palm Beach Research Farm
Canal Point, Florida

Short-Wave Radiation Phenomena, Vols. I and II. August Hund. New York–London: McGraw-Hill, 1952. 1382 pp. $20.00 the set.

It is not always easy to formulate a concrete opinion about a book of such tremendous proportions as this 1382-page opus. In the present case, this is even more difficult because it is hard to identify the reader to whom the work might have been addressed. It is cer-