CORRESPONDENCE

This department is provided for the publication of informal communications which are of interest because they are informative or stimulating, and for the discussion of controversial matters.

LEPROLIN. BACILLI IN TISSUE

To the Editor:

One of the difficulties that we experience in our leprosy work here at the American Mission Hospital is inability to locate a source of material for skin-testing our cases. We deal only with outpatients, and though we ourselves can get some material that could be used for preparing leprolin, the outstanding lepromatous cases are irregular in attendance and often it is difficult to get enough of them at one time to prepare a reliable leprolin. Unless the product contains material from several cases I should question the results obtained with it. We would like to learn of any institution that may be in a position to supply us; and we imagine that there are many other places where that information would be useful.

Another difficulty that we have been having is in staining the leprosy bacillus by the carbol-fuchsin method in sections of tissues. Sometimes the bacilli are fairly well stained, but we cannot depend upon the findings with our present technique. We cannot be sure that bacilli are really absent when we fail to find them. It occurs to me that some of the readers of the Journal may have arrived at reliable methods of staining bacilli, and it would be helpful if they would record them.

American Mission Hospital
Assiut, Egypt.

Horace K. Giffen

Comment by Dr. H. W. Wade, Culion.—Referring to the second question in Dr. Giffen's communication, it has long been a matter of puzzlement why some technicians have a great deal of difficulty in staining leprosy bacilli in sections while others seem to have none. Because the bacillus is so easily and so positively stained in smears, it would seem that its satisfactory demonstration in sections should also be easy. The fact is that it is not a simple matter to meet the two-phase requirement of (*a*) demonstrating the bacilli, in anything like their real numbers, in (*b*) histological preparations that are good with respect to the condition and staining of

the tissue. It is easier to make the equivalent demonstration in sections of tuberculous lesions, and yet the fact that even there the situation is not entirely satisfactory is shown by the occasional appearance of new articles on that subject.

As far back as 1925 I made a rather extensive inquiry into the cause of the difficulty and found it to lie in the fact that, when bacteria of the acid-fast group are treated with oily substances, the physico-chemical complex which is responsible for the retention of the fuchsin stain tends to become so modified that it is readily extracted when alcohol is applied subsequently. This happens with both the tubercle and leprosy bacilli, but apparently more easily and completely with the latter as a rule, though there are very wide differences in the lability of either of these germs in the tissues from different patients. It happens even in the deparaffinization of sections by the usual method, the routine alcohol treatment after xylol being usually highly destructive.

A technique that in large degree overcomes this and certain other difficulties was developed at the time mentioned and communicated to several colleagues, and a modification of it was later published from the U. S. Army Medical Museum [Campbell, *Jour. Tech. Meth. & Bull. Internat. Assoc. Med. Mus.* No. 12 (1927) 129]. In the same publication (p. 130) Haythorn, referring to the tubercle bacillus, stated that the solutions used in fixing, imbedding, dehydrating and mounting paraffin sections remove substances necessary to the "proper maintenance of their acid-fast qualities," which is the first published statement of the kind that I have encountered. He had found fixation in formalin to be harmful. Lowe [*Indian Jour. Med. Res.* 22 (1934) 313] ascribed the difficulty with leprosy bacilli mainly to the effects of fixation, saying that all methods in common use markedly reduce acid-fastness and almost abolish alcohol-fastness. In sections of fixed tissue, he found, to follow xylol with alcohol has a particularly harmful effect, and he avoided the use of alcohol by drying the sections between xylol and water and for dehydrating after staining. I had attained the same end by the use of certain essential oils, thus avoiding both alcohol and desiccation. This technique, which has been in use in the Culion laboratories for nearly fifteen years, will be published in the near future.