BLOOD CULTURES IN EPILEPSY *

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In cultures from the blood of the following six cases of epilepsy we have failed to isolate the organism described by C. A. L. Reed¹ as the *Bacillus epilepticus*.

Dr. Reed gave us one of the cultures isolated by Dr. Hyatt from a case (S). As far as we could tell, this organism belonged to the *B. subtilis* group. Its morphology is well shown in the photomicrographs published by Dr. Reed in the article cited. It varied in its reaction to Gram's stain according to the medium on which it was cultivated. In size it resembled *B. subtilis* and formed terminal and subterminal spores which, when mature, were almost central. It grew rapidly at 37 C. or 24 C. with a smooth or wrinkled dirty white layer on any of the ordinary mediums. Blood serum was rapidly digested, as was also the casein clot in milk. Acid was produced from dextrose, levulose, galactose, saccharose and mannite. We decided to test its pathogenicity after we had proved to our satisfaction that it really could be isolated from the blood of epileptics.

Dr. Reed kindly offered to allow us to make cultures in his cases, which was done by us in four cited below in conjunction with his bacteriologist, Dr. Hyatt. Not only could we not isolate such an organism but also we failed to find any bacteria in smears stained by the Giemsa stain and by Abbott's spore stain and other methods. Dr. Hyatt showed us a blood smear containing the "spores" of *B. epilepticus*. We were not able to pass on the nature of these bodies, which were about the size of the spores of *B. subtilis*. We will only say that we have been unable to find similar bodies in smears made by us. Our reason for limiting ourselves to the study of a few cases was influenced not only by the pressure of other work; we also hoped that by the use of special mediums, which we had found suitable for the growth of many delicately growing parasites, we might be able to isolate something overlooked heretofore.

We submit the following condensed notes on the cases in which we made cultures:

CASE 1.—Mr. B., Cincinnati General Hospital, was referred to us by Dr. Reed as particularly favorable on account of the frequent attacks of grand mal. Three separate blood cultures were made:

(a) Feb. 24, 1916, 5 c.c. of blood were taken between the frequent attacks and distributed in two flasks of +1 broth and several deep tubes of +1 glucose agar. These all remained sterile and were discarded after fifteen days' incubation at 37 C.

(b) March 6, 10 c.c. of blood were drawn one hour after a typical attack of grand mal in which the patient fell down and injured himself. The blood was immediately distributed among four flasks, each containing 400 c.c. +1 broth; 2 deep tubes of +0.5 Martin's medium; two deep tubes of +2.0 Martin's medium, and six tubes of +0.5 Martin's medium which were solidified in the slanting position and two of these were made anaerobic. These all remained sterile during two weeks' incubation at 37 C.

Some of the sediment from the flasks was transplanted to +1 maltose and +1 human blood agar slants on the third day, but these subcultures too remained sterile.

(c) On the afternoon of March 6, the patient had a seizure, and while he was on the floor, Dr. Betzner drew 10 c.c. of blood and defibrinated it. This was distributed in flasks of +1 broth and +1 glucose broth and deep tubes of Martin's medium. After a month's incubation a few colonies of staphylococci were detected in some of the deep tubes of Martin's medium. The +1 broth flasks remained sterile, but a staphylococcus and a peculiar clostridium were isolated from the glucose-broth-blood culture. The clostridium multiplied very slowly in the blood sediment, and proved to be a partial tension organism. We have described its isolation in an article which is shortly to appear.² We subsequently encountered an aerobic strain of the same organism in a glucose-broth-blood culture in a case of phlebitis. Investigations showed that the clostridium had extremely resistant spores and probably came from the glucose.

After this we took care to autoclave all the mediums used in the following cultures ourselves.

CASE 2.—Mrs. H., Cincinnati General Hospital, was referred to us, February 28, by Dr. Roger Morris and Dr. T. H. Kelly. Blood was drawn from an arm vein two hours after a typical attack of grand mal. Five c.c. of blood were distributed in three aerobic agar plates, in four deep tubes of +0.5 Martin's medium, and in three flasks of +1 broth and one flask of +1 dextrose broth. These were incubated at 37 C. All these cultures remained sterile during twenty-two days' observation.

CASE 3.—From Miss G. R., aged 15, one of Dr. Reed's grand mal patients, from a private hospital, April 12, with Dr. Hyatt, we drew 10 c.c. of blood from an arm vein. About 5 c.c. of this were immediately distributed in one deep tube of +0.5 Martin's medium; two flasks containing 600 c.c. of +1 broth each and 1 flask containing 600 c.c. of +1 glucose broth, and into three +1 agar plates. The other 5 c.c. of blood were hemolyzed in sterile distilled water and taken back to the laboratory, where it was sedimented at high speed and the washed sediment and supernatant fluid planted in a variety of mediums. In all sixteen aerobic and anaerobic cultures were made in +1 glycerin agar, +0.5 Martin's medium, Martin's medium made up with monosodium phosphate, +0.5 Martin's pleuritic and on +1 human blood agar slants.

All of these cultures remained sterile during twenty-six days' observation, excepting that eight superficial colonies of *Staphylococcus albus* appeared on one of the +1 agar plates. This was discarded as an apparent contamination, as the room in the hospital in which the plates were made was dusty and drafty.

CASE 4.—Mr. L., aged about 45, one of Dr. Reed's cases, from a private hospital, had petit mal, but "has been growing worse." April 12, cultures were made from 10 c.c. of blood drawn at the same time and place as in Case 3. Cultures were distributed in the same way and in the same mediums. One plate showed a superficial colony of a large coccus. The rest of the cultures remained sterile as in Case 3.

CASE 5.—From Miss A. E., who had nocturnal attacks, referred by Dr. C. A. L. Reed, April 16, various types of mediums were inoculated with 10 c.c. of blood and incubated aerobically, at partial tension, and anaerobically with negative results during eight days' observation. About 2.5 c.c. of the blood was inoculated into ten tubes of +1 broth. Four of the tubes belonged to Dr. Hyatt. These we divided between us. Ours, including two of Dr. Hyatt's broth tubes, remained sterile. Dr. Reed later informed us that Dr. Hyatt had isolated the *Bacillus epilepticus* from one of the tubes taken by her.

CASE 6.—Mr. A., from the Jewish Hospital, one of Dr. Reed's patients, April 25, 1916, Dr. Reed notified us, was a

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1. Reed, C. A. L.: The *Bacillus Epilepticus*, THE JOURNAL A. M. A., May 20, 1916, p. 1607.

NOTE.—We have for publication next week a paper by Dr. W. B. Terhune, of the East Louisiana Hospital for the Insane, giving his results, which apparently confirm Dr. Reed's claims as to the causative agent of epilepsy; also a comment by Dr. Reed on this article and the one by Caro and Thom which follows.

2. Wherry, W. B., and Oliver, W. W.: Jour. Infect. Dis., to be published.

favorable case. We could not go ourselves to collect the blood, but Dr. Hyatt kindly collected about 5 c.c. in sterile sodium citrate solution (1.5 per cent. furnished by us). Some of the blood had been discharged into tubes of plain broth (made by Dr. Hyatt). Two of these inoculated broth tubes were turned over to us for observation. The citrated blood was centrifuged. The citrate plasma was removed and the sediment hemolyzed with distilled water. The hemolyzed sediment and plasma were then centrifuged at high speed for one hour and the sediments planted in deep tubes of +0.3 and +1.0 Martin's medium containing sterile pleuritic fluid. Two plates of combined sediments were made; one was kept aerobic and the other at partial tension at 37 C. All the cultures showed no growth during two weeks' observation.

A BACTERIOLOGIC STUDY OF THE BLOOD OF SEVENTY EPILEPTICS

WITH SPECIAL REFERENCE TO THE BACILLUS
EPILEPTICUS OF REED *

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The obscurity surrounding the fundamental causes of so complex a group of neurologic disorders as the epilepsies will, in itself, bring forth from time to time many new observations, both etiologic and therapeutic in nature. Recently, Dr. C. A. L. Reed of Cincinnati has written concerning the isolation of an organism believed by him to be the cause of the convulsion in so-called "idiopathic epilepsy." The value of such an observation can hardly be overestimated, particularly since it contains vast possibilities in the opening up of a new field of therapeutic endeavor. Accordingly, we felt it to be essential that this work should be corroborated, merely from the point of view of recovering the organism from the blood of epileptic patients.

A brief summary of Dr. Reed's work will aid in the presentation of the results of our investigations. In February, 1915, Dr. Reed¹ stated that "the rôle of constipation as a causative factor in epilepsy is due to the more than probable fact that the initial cause is to be found in a specific infection." So convincing was this conclusion that he felt the necessity of stating that it was probably a bacillus of the gas-forming group, which, together with its toxins, had the faculty of accumulating in the tissues and in the blood.

Following this hypothesis, Dr. Reed² reported the finding of a specific organism as the cause of "idiopathic epilepsy," which he believed to be the epileptococcus. This organism consisted "of granular bodies, sometimes arranged in diplococci, sometimes in chains, sometimes in clusters, while in certain cases all these forms were present in a single case." It was to be found in the blood either just before or just after a seizure, but in the interim it disappeared. Growth of the coccus occurred readily on ordinary mediums, and animal experimentation fulfilled the third of Koch's laws. In conclusion, the presence of the epileptococcus was stated definitely to be pathogenic for the disease.

In a later communication, Dr. Reed¹ stated that "epilepsy is an infection depending on a definite spore-bearing organism—the *Bacillus epilepticus*." The discarding of the epileptococcus was explained briefly (in a footnote) on the basis of the changing morphology of the bacillus, as shown at different stages of its development. This had given rise to some confusion, and the discredited researches of Bra are quoted in defense of this change in the causative agent. In this paper, it was stated that cultures from the blood of epileptics showed the organism present in a large number of cases, more abundant immediately preceding or during a convulsion, although frequently found at other times, and, less readily, in "long interval" cases. The duration of the disease had no influence, apparently, on the finding of the bacillus, for mention is made of a case of more than twelve years' duration in which cultures were found positive. The organism was said to grow slowly in bouillon or on agar-agar (taking from two to five days), and to stain best with carbolfuchsin, although other stains could be employed. Finally, injection of pure cultures of the organism into rabbits was stated to produce a syndrome analogous to epilepsy in man.

Brief mention of the work of Bra may not be inadvisable at this point. In 1902, she³ described the neurococcus as the essential cause of epilepsy. Besta, working in the laboratory of Ceni, failed to find the organism in a total of 375 cultures, of which 370 were sterile, the remaining five being contaminated. Ghiliarowsky of Moscow found the organism in insane patients (not epileptic), and concluded that it was a contamination. The following year, Tirelli and Brossa examined a total of fifty-eight cultures with negative results. Lannois and Lesieufs of Lyons also were unable to find the neurococcus.

With these facts in mind, we began an investigation along the lines taken by Reed and his assistants. In all, seventy cases of clinically idiopathic epilepsy, without any demonstrable organic lesion, have been studied. In this group were included forty-seven women and twenty-three men. In fifty-five of these patients no exciting cause could be found. In four-

TABLE 1.—AGE OF ONSET

Age	Number of Cases
Before end of first year.....	8
Between age of 1 and 5.....	14
Between age of 6 and 10.....	24
Between age of 11 and 15.....	23
Between age of 16 and 20.....	4
Between age of 21 and 30.....	6
Between age of 41 and 45.....	1
Total	70

teen cases the exciting cause was given as: pregnancy, 2; pneumonia, 1; scarlet fever, 1; dentition, 3; indigestion, 3; pertussis, 2; rachitis, 1, and measles, 1. In this group, one case (blood taken during status) was included in which the assigned cause was given as "head injury," but necropsy failed to reveal any lesion of the calvarium or brain structures. The age of onset of the disease varied from infancy to 41 years (Table 1). The age at time of first known convulsion is here taken as the age of onset.

The duration of the epilepsy was from three to forty-five years; periods of remission, no matter what their length, are included in the duration (Table 2).

In regard to the incidence of constipation, on which Reed laid considerable stress, it was found that all of the seventy patients were suffering in varying degrees from this complication.

3. Bra: Recherches microbiologiques sur l'épilepsie.

* From the laboratory and wards of the Monson State Hospital (for Epileptics), Palmer, Mass.

1. Reed, C. A. L.: The *Bacillus Epilepticus*: Third Report, THE JOURNAL A. M. A., May 20, 1916, p. 1607.

2. Reed, C. A. L.: Diagnostic Methods and Pathologic Constants in Idiopathic Epilepsy, THE JOURNAL A. M. A., Jan. 29, 1916, p. 336.