when the agar medium of Tatum and Beadle[5] was used and the rate of growth was determined by measuring the diameter of the colonies. It should be stated that this fungus is variable, the amount of growth fluctuating over a wide range under apparently identical conditions. Nevertheless, the fundamental principle of its behavior remains the same, namely, that the effectiveness of *p*-aminobenzoic acid as a growth factor decreases with the increase in pH.

An opposite pH effect was observed by Stokes, Foster and Woodward[6] with a pyridoxin-requiring mutant of *Neurospora sitophila*. These investigators found that under certain conditions of nitrogen nutrition the fungus could synthesize pyridoxine at a rate necessary for normal growth if the pH remained above 6.2. However, in a medium containing no *p*-aminobenzoic acid the pH exerted no controlling effect on the *Neurospora crassa* mutant used in our work. In the presence of the vitamin the fungus attains maximum growth within a few days, whereas in its absence no growth will occur during that time. This failure to grow may continue for two or three weeks, but eventually, and then within only a few days, a rich growth will ensue regardless of the pH value. From the weight of the mycelium produced in such cultures, as well as from microbiological assay of the culture filtrate, it is evident that through some adaptive process the organism develops a latent ability to synthesize *p*-aminobenzoic acid during the prolonged incubation period. The fact that this synthesis and the growth resulting from it are not fundamentally influenced by the pH of the culture medium indicates that the pH effects observed in the early growth must be ascribed to changes in the effectiveness of the *p*-aminobenzoic acid.

Since *p*-aminobenzoic acid has a dissociation constant of about 2×10^{-5}, at pH 4.8 it exists in solution as equal amounts of molecules and ions. At pH 5.8 the molecular form decreases from 50 to 10 per cent. and above that value the portion present as the molecule drops almost tenfold with each unit rise in pH. Therefore, the efficiency of the vitamin in the nutrition of this organism appears to be a function of the molecular form rather than of the ion.

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RAPID AND STERILIZING EFFECT OF PENICILLIN SODIUM IN EXPERIMENTAL RELAPSING FEVER INFECTIONS AND ITS INEFFECTIVENESS IN THE TREATMENT OF TRYPANOSOMIASIS (TRYPANOSOMA LEWISI) AND TOXOPLASMOSIS[1, 2]

THE following preliminary report deals with the results obtained with penicillin sodium therapy in the following experimental infections: (1) trypanosomiasis, *T. lewisi,* in laboratory rats of a Wistar strain believed to be free from *Haemobartonella muris;* (2) toxoplasmosis, and (3) relapsing fever in Swiss mice. The penicillin sodium used in these experiments was kindly furnished by Dr. Chester Keefer.

(1) *Trypanosomiasis.* Six rats weighing about 70 grams each were used in testing the therapeutic value of penicillin sodium in trypanosomiasis. Treatment was started on 4 of the rats 6 days after their inoculation with a dilute suspension of blood containing adult trypanosomes. The infections in the 2 untreated rats served as controls. The routine therapy covered a period of 48 hours and consisted of the subcutaneous injection of 2,000 Oxford units of penicillin sodium dissolved in 1 cc distilled water every 3 hours, night and day, for 2 rats, and intraperitoneal injections of the drug in a similar manner for the other 2 rats. The total dose received by each of the 4 rats was 32,000 units, or 429,000 units per kilogram. Parasite counts were made 24 hours after the initial dose and daily thereafter for 5 days. No significant difference was noted between the counts for the treated and untreated animals. All infections ran a typical course. The trypanosomes in the blood of the treated animals appeared active, unharmed, and infected other rats, producing again typical infections.

(2) *Toxoplasmosis.* In the toxoplasma experiments the mice were infected by the intraperitoneal inoculation of large doses of a strain of *Toxoplasma* highly pathogenic for mice. Sixteen mice were infected.

Treatment of lot I, consisting of 4 mice, was started the 5th day after infection; each received 9,000 units, 500 units intraperitoneally in 0.5 cc saline every 3 hours. The treatment of lot II, also consisting of 4 mice, was started on the 9th day after infection. Each mouse received 500 units intraperitoneally in 0.5 cc saline to make a total dosage ranging from 6,500 to

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[2] A preliminary report.

9,000 units. The treated mice died in the same interval as the infected, untreated controls.

(3) Relapsing fever (*"S. novyi"* strain). Eleven mice were inoculated intraperitoneally with 0.25 cc heparinated pooled blood from five mice showing heavy infections with *S. novyi.* Twenty-four hours later the infections were moderately heavy and treatment of six of the mice was started with penicillin sodium. The remaining 5 mice served as controls. The treated mice received intraperitoneally 1,000 units in 1 cc saline for a first dose. Every 3 hours thereafter, for 48 hours, each animal received an additional 500 units of the drug. The total dose for each treated mouse was 9,000 units. The first effects of the drug were observed 6 hours after the first dose. At this time the infections had decreased in intensity to about one fortieth in the treated mice, whereas they increased about 50 per cent. in the untreated animals. At the end of 27 hours no spirochaetes were microscopically visible in the treated mice, whereas in the untreated animals they averaged 140 in a single oil immersion field. Sixty hours after treatment was started, 2 of the apparently cured mice were sacrificed. The citrated heart blood of each mouse was inoculated intraperitoneally into two new mice. No infections resulted.

In a second experiment a relapsing fever mouse, sacrificed after receiving 4,000 units in 19 hours, was found to be a carrier, although no spirochaetes were found in a thick drop of its blood.

From the results of these preliminary experiments, it is evident that penicillin sodium, in the very large doses employed, was inactive against *Trypanosoma lewisi* and *Toxoplasma,* but was spectacularly effective in the treatment of relapsing fever.

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HEPATIC DYSFUNCTION IN MALARIA

EVERY student of the subject is aware that enlargement and tenderness of the liver and even jaundice may occur in the various forms of malaria. Nevertheless, relatively little attention has been given to the possibility that hepatic dysfunction and its associated derangements in metabolism can exist in this disease. Slatineau and Sibi,[1] and Phocas[2] did note that some degree of transitory hepatic insufficiency exists in nearly all cases, but their data has not received general acceptance. More recently Kopp and Solomon[3] studied the influence of induced tertian malaria on the liver function of nine patients under treatment for general paresis. They noted a transient disturbance in liver function which usually cleared up within three to six weeks after the termination of the malaria.

With the increasing incidence of malaria consequent to the war, it became important to establish further the probability that an associated liver dysfunction is more frequent than is generally acknowledged. Towards this end, the status of the function of the liver was studied in a series of malaria patients by means of various tests and constituents of the blood. The present preliminary report deals with two cases without a previous history of malaria before the present attack, six with recurrent malaria and a history dating back as long as six months to two years, and two patients with a definite history but no evidence of malaria at the time of study.

The preliminary pertinent data obtained at various intervals during the patients' hospitalization are summarized in Table 1 and reveal that every patient with malaria had an abnormal cephalin cholesterol flocculation test. The majority of the patients also demonstrated an increased sedimentation rate, an anemia, a high serum globulin and, in fewer instances, a slightly increased icteric index. For purposes of comparison another flocculation test, the Kahn test for syphilis, was performed at the same time as the cephalin cholesterol flocculation test. In every instance a negative result was obtained.

Many studies with the cephalin cholesterol test have established that this procedure is an excellent, sensitive measure of hepatic damage. The data summarized previously, therefore, may be interpreted as indicating that in nearly every case of malaria some degree of liver damage may exist. Since the initial test was performed in several instances before any therapy was instituted, it is probable that neither atabrine nor quinine are factors in the production of the hepatic damage.

The presence of liver damage in malaria necessitates revision of modern treatment, since not only is it necessary to administer the specific drugs aimed at the elimination of the plasmodia, but it is necessary also to institute measures which may result in a restitution of the liver to normal. Such measures are the administration of high carbohydrate, high protein, high vitamin diets and not the administration of "only fluids during the course of the fever," as is advocated by some of the leading students of malaria.

The efficacy of a diet aimed at improving the status of the liver is suggested by the fact that patients thus treated may show a rapid disappearance of the positive cephalin cholesterol flocculation test, while patients not treated in this manner show a positive floc-

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[2] E. Phocas, *Rev. Med. et Hygiene Tropicales,* 29: 246, 1937.

[3] I. Kopp and H. C. Solomon, *Am. Jour. Med. Sci.,* 205: 90, 1943.