eastern Asiatics whose growth is still Malthusian in character must be added most of the population of the Near East, of Africa and of a considerable proportion of Central and South America. Merely as an illustration of what is happening it may be noted that the Indians increased by about 83 million in the 20 years following 1921—from 306 million to 389 million.

Dr. Mather seems to think that these "backward" peoples will not long continue to grow as fast as their "subsistence" increases and therefore, will not feel any increased pressure on their food supply or on their economy as a whole. The reviewer can not agree with this view. He believes they are feeling increased pressure already in many places and will feel much more as their conditions begin to improve. He believes that the political and economic life of the world is not organized to take care of an increase of 50 to 75 million in India in each decade between now and the end of the century. Only increased pressure on subsistence which will raise the current high death rate to about the level of the birth rate will prevent this, for there is no one fairly familiar with India's population who believes there will be any significant decline in her birth rate within five to six decades. Furthermore, even after the birth rate begins to decline the death rate will decline even faster for two to three decades thus raising the rate of growth. If the entire population of south and east Asia (excepting Japan) were to increase as rapidly as that of India (1931–1941) it would grow by at least 140 million in 1940–50 (this is more than the entire population of the United States) and would increase even more rapidly thereafter. Is there any reasonable hope that China and India can care for 80 to 85 per cent. of such an increase within their own borders for the next half or three quarters of a century? The reviewer believes the answer must be, no! Then we must ask if there is any hope of political reorganization which will give these countries access to the land and minerals of which Dr. Mather says there are "enough and to spare." The reader will give his own answer.

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SCRIPPS FOUNDATION FOR
RESEARCH IN POPULATION PROBLEMS

SPECIAL ARTICLES

INTERACTION BETWEEN CRYSTALLINE PENICILLIN AND HUMAN PLASMA PROTEINS[1]

PLASMA proteins which were formerly conceived to be only inert colloids, having to do with fluid equilibrium, are now known to be able to bind with small molecules of physiological importance. Schonholzer[2] concluded from his electrophoretic experiments that there is a union of azo-dyes of sulfanilamide with serum albumin. Davis[3,4] showed binding of sulfonamides with serum albumin by equilibrating the drugs between a plasma protein solution and a buffer solution. On the other hand, Kimmig and Weselmann[4] showed, by the technique of cataphoresis, that sulfonamides are "adsorbed" to serum albumin and that dissociation can be produced by animal charcoal. The ability of plasma protein to combine with phenol red[5,6,7] has also been demonstrated by the spectrophotometric absorption method.

Since penicillin is now widely used clinically, it is of extreme interest and importance to ascertain whether it will combine with human plasma protein. For that purpose, we have used as test proteins electrophoretically homogeneous γ-globulin and albumin, as well as a mixture of α- and β-globulins, at the concentration present in normal human plasma. Thus 7 ml of a protein solution, pH 7.8, containing 125 micrograms of crystalline sodium salt of penicillin per ml were put into a Cellophane bag and equilibrated with 14 ml of a phosphate buffer solution of penicillin of the same concentration and the same pH. After 18 to 24 hours of continuous rocking at 1° C. the concentrations of penicillin, in plasma and in buffer, were determined. It was found that, if albumin is used, the antibiotic activity of the protein solution after equilibration is greater than that of the dialysate (see table 1). Moreover, the greater part of

TABLE 1

THE ANTIBIOTIC ACTIVITY OF EXTERNAL AND INTERNAL FLUIDS, BEFORE AND AFTER DIALYSIS, IN OXFORD UNITS PER ML*

		External fluid (Penicillin in buffer)	Internal fluid (Penicillin and albumin in buffer)
Experiment I	Before dialysis	187	187
	After dialysis	151	284
Experiment II	Before dialysis	178	187
	After dialysis	150	246

* 1.0 mg of the crystalline sodium salt of penicillin = 1,650 Oxford Units.

[1] From the Laboratory of Physical Chemistry and the Division of Microbiology, The Squibb Institute for Medical Research, New Brunswick, N. J.

[2] G. Schonholzer, *Klin. Wchnschr.*, 19: 790, 1940.

[3] B. D. Davis, SCIENCE, 95: 78, 1942.

[4] B. D. Davis, *Jour. Clin. Investigation*, 22: 753, 1943.

[5] J. Kimmig and H. Weselmann, *Dermat. u. Syph.*, 182: 436, 1941.

[6] A. Grollman, *Jour. Biol. Chem.*, 64: 141, 1925.

[7] H. W. Robinson and C. G. Hogden, *Jour. Biol. Chem.*, 137: 239, 1941.

the original antibiotic activity can be accounted for. Within the limits of testing procedures, there is no demonstrable union between penicillin and any of the globulins.

To demonstrate further that a union between penicillin and albumin took place, we have attempted to isolate the penicillin-albumin complex by precipitating it in aqueous 50 per cent. alcohol solution at 5° C. The unbound penicillin is soluble in such a concentration of alcohol. The precipitated protein-penicillin complex was re-dissolved with water and re-precipitated with 50 per cent. alcohol without appreciable loss of penicillin activity. To obtain a dried powder of the new compound, the alcohol precipitate was dissolved with water and the solution was lyophylized.

Unlike the sulfonamide-albumin complex, which Davis and others believe is devoid of bacteriostatic property, the penicillin-albumin complex does possess antibiotic activity. If chemical union and not physical adsorption takes place between the antibiotic agent and the albumin, the complex is expected to be more slowly excreted than penicillin itself because of the increase of molecular size. A few preliminary biological studies of this protein have been made. It was found that mice receiving the complex intramuscularly did not excrete the antibiotic agent into the urine as rapidly as mice receiving the same dose of free penicillin by a similar route. The importance of discovering a means to retard the rate of excretion of penicillin has been generally recognized. Partially successful attempts to obtain delayed action of penicillin by use of diodrast[8] or para-amino-hippuric acid[9] have been reported. Likewise, the use of a suspension of penicillin in oil or in oil containing 0.75 to 6.0 per cent. beeswax[10] has been reported to give delayed excretion. However, we are not aware of any report in the literature on a chemical compound of penicillin possessing a delayed excretion rate. It must be stated here that at present we have no evidence that a true chemical compound is formed when penicillin and albumin interact. Since the albumin used is of human origin, the resulting complex might be expected to possess little antigenicity in man. Its metabolism, its immunological properties as well as its stability toward acid, alkali and penicillinase will be reported elsewhere after detailed studies have been completed.

SUMMARY

Penicillin combines with human serum albumin, as demonstrated by dialysis experiments. Unlike the sulfonamide-albumin complex, the penicillin-albumin possesses antibiotic activity. Such a complex can be freed of unbound penicillin by repeated precipitation at 50 per cent. alcohol and obtained as a dried powder. Mouse excretion studies indicated that the penicillin-albumin complex was excreted more slowly than the sodium salt of penicillin.

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[9] K. H. Beyer, R. Woodward, L. Peters, W. F. Verwey and P. A. Mattis, SCIENCE, 100: 107, 1944.

[10] M. J. Romansky and G. E. Rittman, SCIENCE, 100: 196, 1944.

THE SEROLOGICAL DIAGNOSIS OF SYPHILIS[1]

IN an extensive investigation directed toward the development of a serodiagnostic method for the differentiation between syphilitic and biologic false positive sera, observations have been made which indicate that in the two groups of sera[2] the reactive antibodies differ from each other in certain significant respects. Since previous work has shown that even in strongly reactive syphilitic sera the antibodies constitute but a minute fraction of the total proteins,[3] and that isolation by specific flocculation with lipoidal antigen yields extremely low recovery of the purified product,[4, 5] a practical approach to the problem has been based on non-specific methods of characterization and fractionation of the sera, as described below:

(1) *Electrophoresis.* Electrophoretic analyses were made on 13 normal, 25 syphilitic and 45 biologic false positive sera. The sera from patients with syphilis[6] differed markedly from normal sera in exhibiting decreased albumin and increased *gamma* globulin con-

[1] The work described in this paper was done under contract, recommended by the Committee on Medical Research, between the Office of Scientific Research and Development and Duke University.

[2] A total of about 300 syphilitic and biologic false positive sera has been used for the investigation of the various phenomena described in this report. Sera accepted as syphilitic were those for which there was unequivocal clinical evidence for infection, whereas the group of allegedly false positive sera may have included some genuinely syphilitic specimens. However, among this group the occurrence of syphilis was fairly well excluded in those cases where the positive serological reaction could be shown to be of transitory nature, and accompanied by such etiologically unrelated diseases as malaria, virus pneumonia, mumps, smallpox vaccination, etc. Serum specimens have been obtained from various Army General Hospitals through Brigadier General Hugh Morgan, M.C., and Major Charles R. Rein, M.C.; from private hospitals through Drs. J. E. Moore and Paul Rosahn; from the Rapid Treatment Center, U. S. Public Health Service, Durham, N. C., through Surgeon Samuel Fisher, and from the clinics of Duke Hospital through Dr. J. L. Callaway.

[3] H. Eagle, "Laboratory Diagnosis of Syphilis," p. 168. St. Louis: C. V. Mosby Company, 1937.

[4] E. Witebsky, *Zent. f. Bakt.*, 122: 70, 1931; *Z. f. Immunf.*, 80: 323, 1933.

[5] O. Bier and E. Trapp, *Jour. Immunol.*, 40: 465, 1941.

[6] These particular sera were obtained from patients who had received therapy of one kind or another.