the 17th day. Implantation proceeded normally, as far as could be determined macroscopically, in all the remaining experiments, and at least some of the rats in each experiment carried living young to the 21st day. The percentage of completely maintained rats, as well as the ratio of living young to total number of implantations, increased with increase in the amount of progesterone administered.

The placentomata which formed in the sterile horn of the uteri of all the rats of Exp. III-VI were larger in every case than the implantation sites in the pregnant horn. This would indicate that the formation of decidual tissue in the rat is dependent, not only upon the size of the progesterone dose,[11] but possibly upon the strength of the traumatic stimulus as well, since it is most likely that the trauma of the uterine epithelium produced by the implanting egg is not of the same order of magnitude as that used in the artificial production of placentomata.

The possibility that contaminants in the progesterone preparation might have influenced the results must be admitted, but we do not believe that they played an important part. In other experiments,[11, 12] using the same type of preparations, we found no quantitative or qualitative differences between the non-crystalline and crystalline progesterones.

Summary. Rats castrated on the 4th day of pregnancy were maintained in pregnancy until the 21st day with daily doses of progesterone of 1 or 2 Rb.U.

11470 P

Precipitation Pattern of Serum Proteins in Phenylpyruvic Oligophrenia.

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Jervis, *et al.*,[1] have shown that the serum of phenylpyruvic oligophrenic individuals contains an abnormal amount of phenylalanine. In view of the recognized effects of small amounts of amino acids on the molecular dispersion of the proteins[2] it seemed possible that

12 Rothchild and Meyer, *Anat. Rec.*, 1939, **75**, supl. 1, 71.

1 Jervis, G. A., Block, R. J., Bolling, D., and Kanze, E., in press.

2 Tiselius, A., *Ann. Rev. Biochem.*, 1939, **8**, 155.

abnormalities in the state of the serum proteins might occur in this disease. Perlzweig, Kondritzer and Bruch[3] found that in pathological conditions fractional precipitation of the serum proteins with gradually increasing quantities of potassium phosphate at pH 6.5-6.8, according to Butler and Montgomery,[4] frequently reveals significant changes which are not brought out by the conventional methods for the determination of albumins and globulins by precipitation with an arbitrarily fixed concentration of a neutral salt.

In the present investigation the fractional precipitation procedure was applied to the serum proteins of 8 physically healthy patients in whom a diagnosis of phenylpyruvic oligophrenia had been made, and 8 healthy persons as controls. Nineteen portions of each sample of serum were precipitated with an equimolar KH_2PO_4-K_2HPO_4 buffer which ranged in molality from 1.2 to 3.0 mols in 0.1 mol increments. In 5 of the patients and 2 of the normal subjects 0.5 cc portions of freshly centrifuged serum were added to 15 cc portions of buffer (serum dilution 1:31). In the remaining experiments in which less serum was available the serum was diluted with an equal volume of physiological saline; then 0.5 cc portions were added to 10 cc portions of the buffer solutions (serum dilution 1:42). After standing overnight at room temperature the precipitate was filtered off and the total nitrogen in solution was determined (microkjeldahl) on a suitable aliquot. The value was corrected for the N.P.N., determined on a trichloracetic acid filtrate of the original serum, and the percentage of the total protein remaining in solution at each molality of phosphate was calculated. The data obtained in experiments in which the dilution of the serum was 1:42 were calculated to a dilution of 1:31 and combined with the data at the latter dilution. The average values obtained in the 8 normal and the 8 phenylpyruvic sera were plotted against the concentrations of the phosphate solutions (Fig. 1).

A small but consistent difference between the two solubility-precipitation curves in the middle range of phosphate concentration is apparent; more protein appears to have been precipitated from the sera of the phenylpyruvics than from those of the healthy controls. As the differences were small the data were subjected to rigorous statistical analysis with the kind assistance of Dr. Joseph Zubin. The methods which Fisher[5] developed for analyzing small series of

[3] Perlzweig, W. A., Kondritzer, A. A., and Bruch, E., *Proc. Am. Soc. Biol. Chem.*, 1938, **32**, xcii.

[4] Butler, A. M., and Montgomery, H., *J. Biol. Chem.*, 1932, **99**, 173.

[5] Fisher, R. A., *Statistical Methods for Research Workers*, 1934, Edinburgh and London.

data were applied in the modification of Snedecor.[6] The F value obtained gives a measure of the probability that the difference between 2 means is significant. For averages obtained on 2 series of 8 determinations each the critical values of F those which could arise by chance not more than 1 to 5 times in 100, lie between 9.07 and 4.67 respectively. It will be seen from the chart that F values in this range were obtained for 3 phosphate concentrations between 2.0 and 2.3 mols per liter.* The chance that fortuitous differences of this degree of significance would occur at adjacent points is exceedingly small.

Each value for the percentage of the protein remaining in solu-

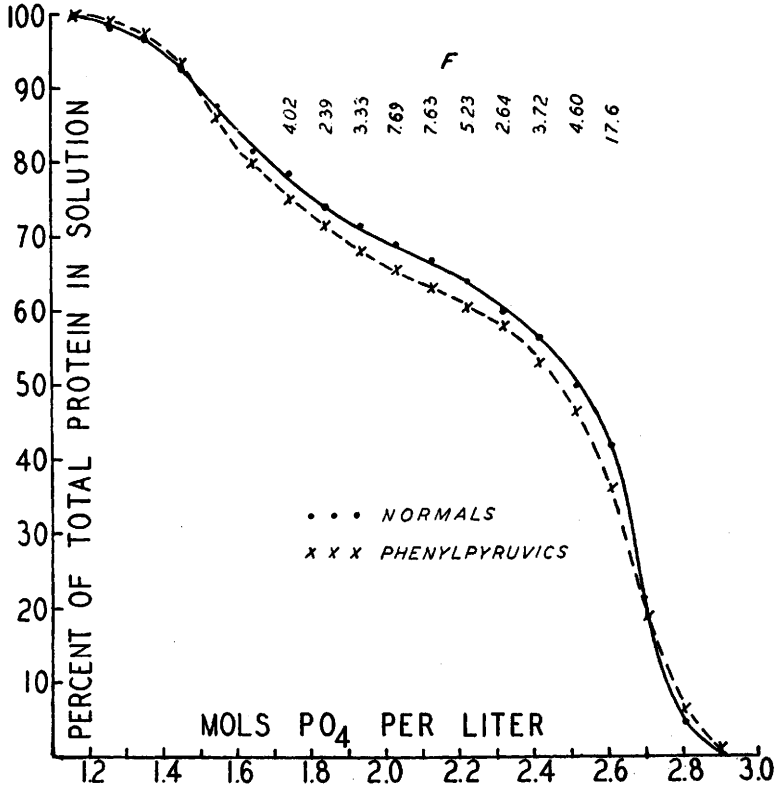

FIG. 1.

[6] Snedecor, G. W., *Analysis of Variance*, 1394, Collegiate Press, Inc., Ames, Iowa.

* For this range of phosphate concentrations the effect of the difference in dilution (1:31 and 1:42) on the solubility of the serum proteins was shown during the statistical analysis to be of no significance.

tion was calculated from three nitrogen estimations (protein and N.P.N. in the original serum, and the total nitrogen of the filtrate). The chances for a cumulative error are, therefore, rather high. However, all determinations were carried out in the same apparatus under identical conditions, as nearly as they could be controlled, and hence the statistical treatment evaluated automatically the effect of errors arising in the analysis. Furthermore, errors in the determination of the protein and N.P.N. of the original serum would have a uniform effect throughout for each sample of serum and could not account for significant deviations in a portion of the curve.

It is concluded that there was a small but significant increase above normal in the globulin fraction of the serum proteins of the individuals with phenylpyruvic oligophrenia studied in this investigation. It is possible that the increase may have resulted from factors other than the particular pathology involved in this disease.†

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Response of Various Breeds of Rabbits to Hamilton and Schwartz Test for Parathyroid Secretion.

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In our earlier work[1] with the Hamilton and Schwartz[2] test for parathyroid hormone, we used 2 breeds of rabbits, raised by ourselves; a black and white Dutch strain and a gray Belgian strain, which included some albinos. All these rabbits gave a positive test when injected with 10 Hansen units of parathormone per kg. In a few instances injection with as little as 4 units per kg resulted in a positive reaction. Similar responses were given by hybrids of these 2 strains. (The H. and S. test depends upon the fact that successive feedings of $CaCl_2$ result in smaller and smaller rises in serum Ca, so that after the 3rd or 4th administration of 100 mg of Ca as $CaCl_2$ normal rabbits will show a rise of serum Ca of less than 1.2 mg per 100 cc, whereas if more parathyroid hormone than that normally circulating is present, a greater rise of serum Ca results, roughly proportional to the quantity of hormone administered.)

† The phenylpyruvic blood samples were obtained from inmates of Letchworth Village through the courtesy of Dr. Harry C. Storrs, Superintendent.

1 Baumann, E. J., and Sprinson, D. B., *Am. J. Physiol.*, 1939, **125**, 741.

2 Hamilton, B., and Schwartz, C., *J. Pharm. and Exp. Therap.*, 1932, **46**, 285.