8. Papermaster, B. W., Bradley, S. G., Watson, D. W., Good, R. A., *J. Exp. Med.*, 1962, v115, 1191.

9. Smith, R. J., *Ciba Foundation Symposium on the Cellular Basis of Immunity*, Edited by G. Wolstenholme and M. O'Connor, 1959.

10. Archer, O. A., Pierce, J. C., Papermaster, B. W., Good, R. A., *Nature*, 1962, v195, 191.

11. Dalmasso, A., Martinez, C., Good, R. A., PROC. SOC. EXP. BIOL. AND MED., 1962, v110, 205.

12. Good, R. A., Martinez, C., Archer, O. A., Papermaster, B. W., *J. Clin. Invest.*, 1962, v41, 1361.

Received May 14, 1962. P.S.E.B.M., 1962, v111.

Polybasic Electrolytes and Colloid Osmotic Pressure of Plasma, Serum and Bovine Serum Albumin Solution.* (27700)

BOYD GOETZMAN† (Introduced by M. B. Visscher)
Department of Physiology, University of Minnesota, Minneapolis

Several polybasic macromolecular substances and basic dyes produce pulmonary edema in the perfused lung(1,2). There are three plausible mechanisms for this phenomenon. These substances 1) may produce arteriolar occlusion by agglutination of red cells, 2) may cause polymerization of plasma protein, thus reducing the colloid osmotic pressure, or 3) there may be chemical action on the capillary endothelium which increases permeability to protein. The first possibility has been shown to occur. Sela and Katchalski(3) have reviewed the literature relating to this phenomenon. The contributions of the other 2 mechanisms have not been established.

The purpose of this study was to investigate the effect of the basic dyes, Neutral red and Methylene blue, and the polybasic macromolecular substances, protamine (spermine · 4HCl) and Polybrene (hexadimethrine bromide) on the colloid osmotic pressure of plasma, serum, and a 6% albumin solution *in vitro*. Of the 4 substances tested, all except Neutral red were found to produce pulmonary edema by Visscher *et al.*(1).

Methods and materials. The colloid osmotic pressure of the test solutions was measured with an adaptation of a Statham strain gauge head$_2$, a 0-75 cm Hg strain gauge, and a Sanborn recorder(4). The membrane used was dialysis membrane supplied by Visking Corp. The closed lower chamber was filled with the reference buffer solution and the colloid containing solution to be tested (0.5 cc) was placed in the upper chamber of the head which was open to atmospheric pressure. The reference buffer solution was Krebs-Ringer buffer solution (0.81% NaCl, 0.041% KCl, 0.0049% $CaCl_2$, 0.019% KH_2PO_4, 0.034% $MgSO_4$, 0.15% Na_2HPO_4) pH = 7.4. The measurements were made at laboratory room temperature, approximately 22°C.

Pressure equilibrium was obtained in about 35-45 minutes with the plasma and in 20-30 minutes with the albumin solution.

The albumin solution was prepared by dissolving 0.6 g of powdered bovine serum albumin, MW = 69,000, in 10 cc of the Krebs-Ringer buffer. All of the substances to be tested were prepared in buffer solutions and in such concentrations that the plasma and albumin solutions were not diluted by more than 4% upon their addition. The final concentrations of the test substances are indicated in Table I.

Results. The effects of the various substances on the measured colloid osmotic pressures are summarized in Table I. In the concentrations employed none of the materials tested had significant effect on the C.O.P. The polybasic macromolecular substances changed the final pH of the test solutions by only 0.1 pH unit. The volume of diluent employed had a dilution effect of less than 1 mm Hg.

Conclusion. It has been shown that several materials causing pulmonary edema in isolated dog lungs under constant perfusion conditions have no significant effect on the plas-

* Supported by USPHS grant.

† Trainee under USPHS grant.

TABLE I.

Colloidal solution	Substance tested	C. O. P. ± S.E. mm Hg
Dog serum	—	24.8 ± .8
"	80 mg% Toluidine blue	22.9 ± 1.1
Dog plasma*	—	23.7 ± 1.6
"	5 mg% Protamine	25.5 ± 1.0
"	20 mg% Neutral red	22.7 ± 1.2
6% serum albumin	—	24.5 ± .5
"	10 mm% Polybrene	24.3 ± .7
"	10 mg% Protamine	24.1 ± .9
"	80 mg% Neutral red	25.5†

* Prepared by adding 20 ml of buffered 2% ethylene diaminetetraacetate to 180 ml whole blood and then centrifuging.

† Average of only 2 measurements, all other values are averages of 4 or more measurements.

ma colloid osmotic pressure when added in concentrations slightly above those expected in clinical use. Therefore, an effect on the colloid osmotic pressure seems highly unlikely as part of the mechanism in production of pulmonary edema by basic dyes and polybasic macromolecular substances.

1. Visscher, M. B., Absolon, K., Ballin, H., *Univ. of Minn. Med. Bull.*, 1962, v33, 181.

2. Visscher, M. B., *J. Lancet*, 1962, v82, 43.

3. Sela, M., Katchalski, E., *Advances in Protein Chem.*, 1959, v14, 391.

4. Meehan, J. P., Hyman, C., Sasnow, M., *Measurement of Colloid Osmotic Pressure with an Unbounded Strain Gage Transducer*, Statham Laboratories, Instrument Notes #15, Univ. of Southern Calif. School of Med., May 1950.

Received May 14, 1962. P.S.E.B.M., 1962, v111.

A Dietary Factor Influencing Lymphoid Tumor of the Chicken.* (27701)

C. OLSON, T. N. FREDRICKSON,† S. TEKELI AND H. R. BIRD

Departments of Veterinary Science and Poultry Husbandry, University of Wisconsin, Madison

It is now recognized that a virus of a lymphoid tumor(1) when given in low doses to day-old chickens, will cause a significant number of them to develop lymphoid tumors (lymphocytoma or lymphomatosis) after a long latent period(2). If a large dose of virus is given to day-old chickens, they develop erythroblastosis after a short incubation period(3). The resistance of the chicken is another factor subject to genetic influence(4) and immunity of the host stimulated by prior exposure to viable tumor cells(5). Probably many birds harbor the virus but do not develop the disease. Such birds may shed the virus and infect very young chickens(6). The effect of certain mineral and vitamin levels as nutritional factors was found without significant effect on fowl paralysis(7). Vit. E deficiency was implicated by the reports of some workers(8); other workers(9,10) could not support this contention. A decrease in incidence of neoplastic disease was noted by Waters(11) when the diet of non-inoculated chickens in genetic experiments was changed from one containing cod-liver oil to one containing a synthetic form of vit. D. Three experiments have been done which indicate that one particular lot of cod-liver oil in the diet of chickens increased the incidence of lymphoid tumors in experimentally exposed birds. Another lot of cod-liver oil did not have this property. Experimentally induced erythroblastosis was not affected by cod-liver oil in the diet. These results are briefly presented here.

Materials and methods. The basal diet had the following percentage composition: ground yellow corn 52.4285, wheat standard middlings 5, wheat bran 5, soybean oil meal (solvent) 25, alfalfa meal 5, meat scrap 5, steemed bone meal 1, calcium carbonate 1, iodized salt 0.5, manganese sulphate (Mn SO 4 • H_2O) 0.025, riboflavin supplement (4 g riboflavin per lb.) 0.01, vitamin B_{12} supple-

* Published with the approval of the Director of Wisconsin Agri. Exp. Station and supported in part by a Research Contract with U. S. Dept. of Agriculture.

† Now at U.S.D.A. Regional Poultry Disease Laboratory, East Lansing, Mich.