Ultracentrifugal Analysis of Serum Lipoproteins in Nephrotic Syndrome of Rats.* (21227)

LENA A. LEWIS AND WALTER HEYMANN.

From the Research Division of Cleveland Clinic Foundation, and Department of Pediatrics, Western Reserve University School of Medicine, Cleveland, O.

The intravenous injection of nephrotoxic serum (N.T.S.) obtained from rabbits produces in rats a chronic renal disease in which hyperlipemia is always present. It simulates the nephrotic syndrome as it is seen in infants and children(1). The lipoprotein pattern has been studied in the human disease(2,3) and it seemed of value to determine whether similar changes occur in the rat.

Methods. Rats of the Long-Evans and Sprague-Dawley strains were fed dog chow† and water. Renal disease was induced according to the technic previously described(1) when they were 4-5 weeks of age. Animals were chosen that had a well-established disease for various lengths of time. Blood for serum protein, cholesterol and total lipid determinations was obtained by cardiac puncture 4-5 days before exsanguination when lipoprotein determinations were made. Serum lipoproteins were determined at a density of 1.21 by the method of Gofman(4) as modified by Lewis, Green and Page(5).

Results. Ultracentrifugation of lipoprotein concentrates of normal rat serum demonstrates 4 components. Dependent on decreasing density, their flotation rates are –S 1-10, 10-25, 25-40 and >70. Ninety to 120 mg of higher density –S 1-10 and 15-40 mg per 100 ml of –S 10-25 component are present, while there is only 15-30 mg of –S 25-40 component. The concentration of low density –S >70 material varies greatly (0 to 100 mg per 100 ml).

The lipoprotein concentration in serum of rats showing the nephrotic syndrome was much greater than that of normals (Table I). While the concentration of all fractions was sometimes increased the greatest increment was usually in the lower density fractions. The –S (40-70) component which is not usually present in measurable amount in normal rats was significantly increased. The –S 10-25 fraction was also often very large, and appeared heterogeneous as indicated by resolution of numerous small peaks.

There seemed to be no correlation between duration of the disease and the type or extent of lipoprotein changes. Rat no. 7 which had had the syndrome for 10 months showed the same type of change that rat no. 13 showed within 17 days. The lipoprotein changes of rats no. 3 and 4 with disease of longest duration and of lesser severity were less than in the other nephrotic animals.

In rats studied 17 days after injection of nephrotoxic serum, there was a total lipoprotein of 700 to 900 mg, or 15 to 35% of the total serum protein, which was 3 to 5 g per 100 ml. Rats which had had the disease 11 to 16 months had smaller concentration of lipoprotein 185 to 600 mg, or 3 to 12% of the total protein which was 4.9 to 6.0 g per 100 ml.

Discussion. While the absolute concentration of the lower density –S >70, 40-70 and 25-40 lipoproteins in the sera of nephrotic rats is not as great as in the nephrotic syndrome in the human being(3) the kind of change is similar. The normal concentration of –S 25-40 and 40-70 in human beings is much greater than in the rat so that the percentage increase in the two species may be comparable. Both species also show an increased concentration of –S 10-25 lipoprotein component.

The –S 25-40 fraction corresponds to the Sf 3-10 fraction determined at a density of 1.063, and the –S 40-70 to Sf 10-20. Gofman *et al.*(6) have observed an increased concentration of the Sf 10-20 class of lipoproteins in nephrotic sera and have emphasized the importance of this group of macromolecules in atherogenesis.

The change in the serum lipoproteins of rats injected with N.T.S. is very much greater than that observed in rats with acute renal

* Aided by grants from the Public Health Service, Dept. of Health, Education and Welfare.

† Friskies, Albers Milling Co., Kansas City, Mo.

TABLE I. Serum Lipoproteins, Determined at Density 1.21, Cholesterol, Total Lipids and Total Proteins of 13 Rats with the Nephrotic Syndrome.

Duration of disease	Severity of disease	Total protein, g/100 ml	Cholesterol, mg/100 ml	Total lipid, mg/100 ml	-S>70	40–70	25–40	10–25	1–10
					mg/100 ml				
Control	—	5.2	60	360	99	—	26	20	114
"	—	5.2	75	440	39	—	20	38	91
16 mo	2+	5.96	75	1600	33	—	21	—	129*
13	3+	5.83	105	1460	42	21	33	106	152
12	4+	4.88	140	1680	164	19	71	141	223
11	4+	5.29	208	2360	77	24	75	141	182
10	4+	5.17	242	1880	282	38	59	106	152
7	4+	5.63	270	1900	398	—	60	86	315*
7	4+	3.42	550	3310	—§	282	282	—	101*
4 wk	4+	4.63	155	4720	170	—	34	57	199
17 days	4+	2.96	485	5880	552‡	40	152	—	199*
17	4+	3.29	400	5240	>400	—	129†	71	168
17	4+	3.96	244	2650	282‡	16	47	164	164

* Spreading peak -S 1-15. † No clear resolution -S 25-70. ‡ -S 70-400; -S>400++++.
§ -S>400++++.

disease resulting from desoxycorticosterone acetate priming followed subsequently by renin injection(7). This may be due to differences in extensiveness of the lesions or duration of the disease.

Summary. Ultracentrifugal analyses at a density of 1.21 of serum lipoproteins in the nephrotic syndrome of rats induced by the intravenous injection of N.T.S. obtained from rabbits, showed greatly increased concentration of all lipoprotein components but chiefly in the lower density fractions. The amount of change was not directly correlated with the duration of the disease, but was generally comparable to its severity. The changes observed in the rat were similar in type to those in the nephrotic syndrome of children.

1. Heymann, W., and Lund, H. Z., *Pediatrics,* 1951, v7, 691.

2. Longworth, L. G., and MacInnes, D. A., *J. Exp. Med.,* 1940, v71, 77.

3. Lewis, L. A., and Page, I. H., *Circulation,* 1953, v7, 707.

4. Gofman, J. W., Lindgren, F., Elliott, H., Mantz, W., Hewitt, J., Strisower, B., and Herring, V., *Science,* 1950, v111, 166.

5. Lewis, L. A., Green, A. A., and Page, I. H., *Am. J. Physiol.,* 1952, v171, 391.

6. Gofman, J. W., Lindgren, F., Jones, H. B., Lyon, T. P., and Strisower, B., *J. Gerontol.,* 1951, v6, 105.

7. Lewis, L. A., Masson, G. M. C., and Page, I. H., *Fed. Proc.,* 1954, v13.

Received July 19, 1954. P.S.E.B.M., 1954, v86.

Participation of Brown Fat in Pathogenesis of Experimental Poliomyelitis of Monkeys.* (21228)

GREGORY SHWARTZMAN AND STANLEY M. ARONSON.

From the Department of Microbiology, Mount Sinai Hospital, New York City.

Progression studies have served to indicate some of the pathways by which peripherally administered poliomyelitis virus (Type 2) reaches the CNS in cortisone-treated hamsters (1-3). Somatic lesions in two types of tissues were noted during the anteneural phase of the experimental disease. A myositis was demonstrable(4) as well as lesions in brown (hibernating) fat(5). High concentrations of virus were present in these foci. The following studies deal with the possible role of brown

* Aided by a grant from the National Foundation for Infantile Paralysis.