PROTEIN CONTENT OF OEDEMA FLUID IN LYMPHOEDEMA

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Lymphangiography (Kinmonth *et al.*, 1955, 1957) has demonstrated the structural defects of the lymphatic system in primary lymphoedema. This report is concerned with some of the physiological disturbances caused by lymphatic malfunction in patients with primary lymphoedema.

Method.—Oedema fluid was obtained from the subcutaneous tissue of patients suffering from chronic primary lymphoedema of the legs. A small-bore Southey tube was used to collect the fluid, and a sufficient quantity was obtained to enable the total protein concentration to be estimated by a biuret method in 38 patients. In addition, the albumin/globulin ratio and the paper electrophoretic pattern of this protein was determined in 20 patients. The majority of patients were ambulant up to the time of collection of the sample of oedema fluid.

Results

Protein Content of Oedema Fluid.—Fig. 1 shows the protein level of the oedema fluids from 38 patients with primary lymphoedema. The patients are grouped into three

Fig. 1.—Protein content of oedema fluids from 38 patients with primary lymphoedema. Protein level for each patient is charted individually to nearest 0.5 g. Mean values for moderate and severe groups are indicated at right of chart.

categories according to the clinical degree of their oedema. The mean protein content of the oedema fluid for the whole group was 2.8 g. per 100 ml. The protein levels ranged from 1 to 5.5 g. per 100 ml., and the series as a whole would fit into the high protein category of oedema fluid as proposed by Crockett (1956). Fluids with the highest levels of protein tended to be found in patients with severe clinical disease, although the mean values of the moderate and severe groups of patients did not differ significantly.

Albumin/Globulin Ratios.—The A./G. ratios in the sera and oedema fluids of 20 patients were determined, and are shown plotted against one another in Fig. 2. For most patients the A./G. ratio was higher for the oedema fluid than for the serum, the mean difference being 1.2 ± 0.44 (S.E.), which is significantly greater than zero at the 5%

level ($t=2.72$, $n=19$, $0.01 < P < 0.05$). The theory of restricted diffusion (Pappenheimer, 1953) implies that the ability of protein molecules to pass through the normal capillary wall varies inversely with their size. The small albumin molecules have least difficulty in negotiating the molecular sieve of the normal capillary wall, and their relative concentration rises in the oedema fluid. Albumin

Fig. 2.—A./G. ratios of serum and oedema fluid plotted against one another in 20 patients and lymphoedema. Each patient is charted individually, and in those falling to the right of the oblique line the A./G. ratio was increased in oedema fluid.

molecules are osmotically more active than the other components of the plasma proteins, and their increased preponderance in the oedema fluid will increase the magnitude of the physiological disturbance.

Electrophoresis.—The electrophoretic pattern of the oedema fluids differed consistently from that of their parent sera in that the α_2 -globulin fraction was either absent or greatly diminished (Fig. 3). This relative absence is

Fig. 3.—Paper electrophoresis patterns of serum and oedema fluid from a patient with lymphoedema, and of fluid from an inflamed bursa. The α_2 -globulin band is greatly diminished in oedema fluid.

probably due to the large molecular size of the α_2 -globulin, which would encounter considerable physical restriction in diffusing through the normal blood capillary wall. The significance of this observation and that of the increase in

albumin concentration in the oedema fluid lies in their indication that blood capillary permeability remains unaltered in uncomplicated lymphoedema. Pathological processes, such as inflammation, which render the capillary wall abnormally permeable produce extravascular fluids that have an electrophoretic pattern resembling closely their parent serum. Fig. 3 includes the electrophoretic pattern of fluid obtained from a slightly inflamed bursa, and it can be seen that all components of the plasma protein are represented. It was also observed that in the electrophoretic pattern of oedema fluids containing less than 2.5 g. of protein per 100 ml. the β -globulin band was a prominent feature and second only to albumin in intensity. It is probable that this band is due to the iron-binding globulin (siderophyllin or transferrin). This substance has a molecular size only slightly larger than that of albumin, and has been found in nephrotic urine (Neale, 1955).

Discussion

An important function of the lymphatic system is to remove protein molecules from the tissue spaces. Lymphatic insufficiency interferes with this function, and we have shown (Taylor *et al.*, 1957) that plasma proteins are removed abnormally slowly from the subcutaneous tissues in lymphoedema. On these grounds it can be expected that oedema fluid in lymphoedema will contain an abnormally high quantity of protein. The investigation presented here confirms this, and the results are in general agreement with the observations of Drinker *et al.* (1934) on four patients, Watson (1953) on five patients, and Crockett (1956) on 10 patients with lymphoedema.

The Starling hypothesis (Starling, 1896) of tissue-fluid formation depends on the high protein content of the plasma to provide an osmotic force sufficient to promote absorption of tissue fluid into the vascular bed at the venous end of the capillary loop. In lymphatic insufficiency the small quantity of plasma protein that normally leaks through the capillary wall will accumulate in the tissue spaces. This extravascular protein then exerts an osmotic pressure in opposition to that of the plasma protein, and in consequence absorption of water from the tissue spaces is impaired. If the disturbance is great enough, water will be retained in the tissues and clinical oedema will be produced. A measurement of the colloid osmotic pressure made on the oedema fluid from one of our patients gives an indication of the magnitude of the abnormal extravascular osmotic force. This fluid contained 2.5 g. of protein per 100 ml. and had a colloid osmotic pressure of 14 cm. of water as measured by the method of Rowe (1954). The plasma colloid osmotic pressure is normally 36 cm. of water (Landis, 1934), and it can be seen that the osmotic force of the extravascular protein is of such a magnitude as to influence the hydrodynamic balance considerably. In these circumstances water will be retained in the tissue spaces until dilution and rising tissue tension restore equilibrium.

Summary and Conclusions

Oedema fluid from the legs of 38 patients with chronic primary lymphoedema was examined for protein content. Values ranging from 1 to 5.5 g. per 100 ml. were found, the mean level being 2.8 g. per 100 ml. The A./G. ratio was found to be increased in the oedema fluids, and electrophoresis showed that the large-molecule protein fractions such as α_2 -globulin were retained inside the blood capillary wall. It is concluded that lymphatic failure results in an accumulation of protein molecules in the tissue spaces and that water retention and oedema are produced by this abnormal extravascular osmotic force. Capillary permeability is unaltered in uncomplicated lymphoedema.

We thank Mr. M. P. Curwen for statistical advice, Dr. D. S. Rowe for the measurements of osmotic pressure, Dr. E. Smith

for assistance with the electrophoresis, and Miss D. Aitken for technical assistance. The investigation was done during the tenure of a B.M.A. scholarship held by one of us (G. W. T.).

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TREATMENT OF FAT EMBOLISM WITH HEPARIN

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Aird (1949), writing of fat embolism, states that "most patients who suffer from fat embolism probably recover, but those in whom the diagnosis is made during life usually die." Faced with a severe case of this condition in 1956, one of us (R. H. S.) consulted the Department of Pharmacology of the University of Birmingham and was advised to try the use of heparin. This patient recovered, and heparin has since been used successfully in two further cases.

The pharmacological basis of the treatment is the effect *in vivo* of heparin "clearing" lipaemic plasma—that is, rendering translucent plasma which has become opalescent after a fatty meal (Hahn, 1943). French, Robinson, and Florey (1953) suggested, therefore, that the fat globules present in the blood of patients suffering from fat embolism might be similarly lysed, reducing the risk of infarction.

As a result of experiments with the heparin treatment of artificially induced fat embolism in rats, Gardner and Harrison (1957) came to the conclusion that a clinical trial of heparin in human fat embolism would not be justified. They found that, in their animals, heparin both accelerated death and increased the number of deaths after fat embolism. In view of these findings we present below three human cases in which heparin was successfully used. Despite the view of de Takáts (1945) that there may be regression of symptoms to complete recovery, no matter how serious the initial symptoms, these three were of such severity that we would otherwise have expected them all to be fatal.

Case 1

A soldier aged 23, a pillion passenger on a motor-cycle, received a closed fracture of the mid-shaft of the right femur on May 15, 1955. This was treated by extension, with a Steinmann pin through the tibial tuberosity, on a Thomas splint. No blood transfusion was given at this stage. On the evening of the 16th his temperature was 101.6° F. (38.7° C.), but the pulse and respirations were normal. On May 17 his temperature was 102.8° F. (39.3° C.), pulse 86,