female cross respectively suggests that levels of serum LDH in normal mice are influenced by genetic factors[9].

Résumé. Nous avons étudié, chez la souris normale, l'effet du sexe, de l'âge et de la constitution génétique sur l'activité du sérum LDH. Le seul résultat positif constaté est un rapport entre l'âge et la race. Nous avons établi la portée normale de l'activité du LDH dans cinq races de souris. Son augmentation n'est pas provoquée par une infection due au virus polyome, mais peut être mise en relation avec la constitution génétique de l'animal.

[9] *Acknowledgments.* The author is indebted to Dr. TH. R. CARSKI and Dr. D. M. FORSYTH for their contributions to this work. – Supported by grant No. C-5742 from the National Cancer Institute, U.S. Public Health Service.

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Immunelectrophoretic Analysis of the Lymph

The lymphatics transport from the tissue spaces not only plasma proteins filtered from the capillaries but also proteins (enzymes, hormones etc.) produced by the tissue cells. The usual procedures of protein fractionation, including the method of paper electrophoresis which is most commonly applied for the separation of protein components of the lymph, do not disclose any significant difference in the composition of blood plasma and lymph[1]. The method is apparently not sensitive enough to reveal the presence of minimal quantities of specific proteins formed.

It was supposed that proteins produced in the tissues could be detected in the lymph flowing from the respective organ by immunelectrophoretic analysis.

Mongrel dogs of both sexes, under chloralose general anaesthesia, were used. The cervical and intestinal lymphatic trunes and the main lymphatic channel of the liver were exposed and polyethylene tubes were inserted. In some cases, the thoracic duct was cannulated in the cervical region and lymph was collected also from the capsular and hilar lymphatics of the kidney. Blood samples were drawn from the carotid artery.

Immunelectrophoretic analysis of blood plasma and lymph was performed according to the micromethod of

[1] I. RUSZNYÁK, M. FÖLDI, and GY. SZABÓ, *Lymphatics and Lymph Circulation* (Pergamon Press, Oxford-London-New York-Paris 1960).

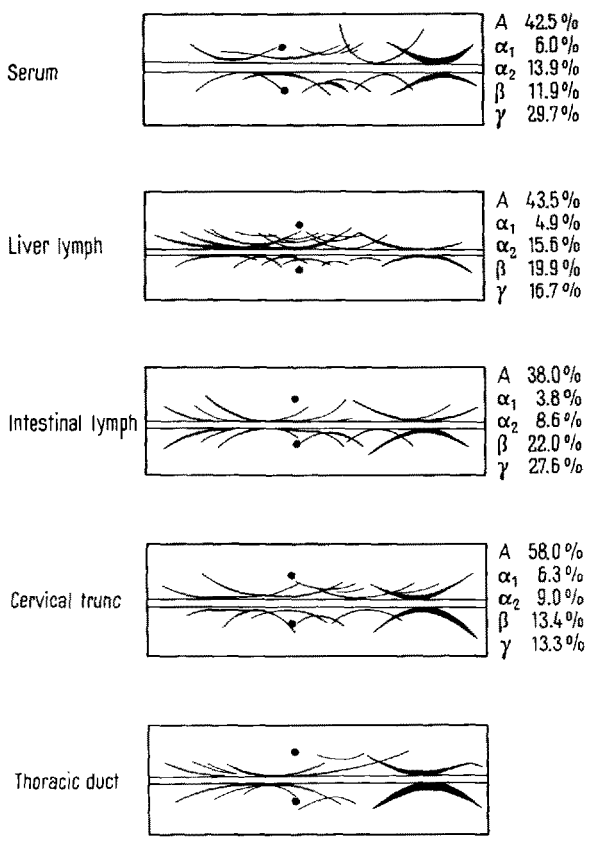

Fig. 1. Immunelectrophoretic and paper electrophoretic analysis of dog serum and lymph.

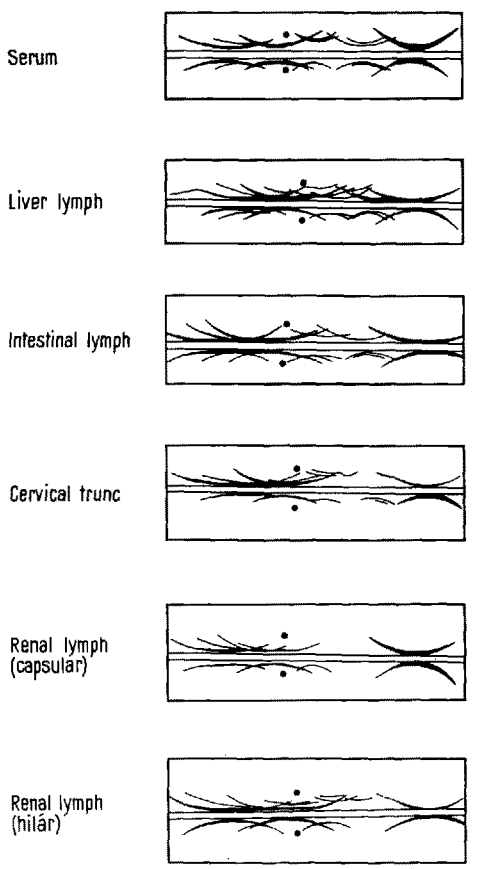

Fig. 2. Immunelectrophoretic pattern of blood serum and lymph samples of different origin of the same animal.

SCHEIDEGGER[2] on microscopic slides covered with 2 ml of gel in barbiturate buffer (pH 8.6, 0.05 μ). Antiserum against dog plasma was prepared by injection into rabbits. The samples were also fractionated by paper electrophoresis.

Nine experiments were performed. Eight samples of hepatic, intestinal and cervical, four samples of renal and two of thoracic duct lymph were analysed. No particular differences were disclosed by paper electrophoresis in the composition of lymph samples of different origin and of blood plasma. The same protein fractions were present in lymph and blood plasma and only minor quantitative differences could be detected. By immunelectrophoretic analysis of the liver lymph, however, several precipitation lines, corresponding to α- and β-globulin fractions, could be observed, which were present neither in the lymph samples of other origin nor in the plasma of the same animal.

As is well known, the liver plays an important part in the production of plasma proteins. It can be assumed, therefore, that proteins formed by the liver are at least partly transported to the general circulation by the lymphatics. Protein components formed in minimal quantities are strongly diluted in the blood plasma and cannot be detected there even by the highly sensitive immunelectrophoretic method.

Our investigations present evidence to the hepatic production of such protein components, but do not permit their exact identification.

Zusammenfassung. Vergleichende immunelektrophoretische Untersuchungen des Blutplasmas, der Nieren-, Leber-, Darm- und Tr.-cervicalis-Lymphe zeigten mit Ausnahme der Leberlymphe in den Lymphproben die gleichen Proteinfraktionen wie im Blutplasma. Im Immunelektrophorogramm der Leberlymphe waren an den Stellen der α- und β-Globulinfraktionen mehrere Präzipitinstreifen sichtbar, die im Plasma bzw. in den Lymphproben anderen Ursprungs nicht vorhanden waren.

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National Institute of Traumatology, First and Third Departments of Medicine, University Medical School, Budapest (Hungary), October 4, 1962.

[2] J. J. SCHEIDEGGER, Int. Arch. All. 7, 103 (1955).

Changes in Vascular Permeability and Dermal Mucopolysaccharides in the Female Rat after Parathyroidectomy

In the parathyroidectomized female rat the ovary can compensate for the parathyroid function in the homeostasis of plasma calcium[1–4], probably by stimulating the calcium absorption. Nevertheless, the survival period of parathyroidectomized female rats is generally shorter than that of the control animals, and death ensues from banal infections to which the operated rats become particularly susceptible. Infection seems to be facilitated either by decrease of immunological reactions or by easier penetration and spreading of the infectious agents. It was also observed that the inflammatory reaction—and subsequent necrosis of the skin—produced experimentally by subcutaneous injections of croton oil in the parathyroidectomized female rats is wider spread in these animals than in the controls. This has been explained by the hypothesis that in the operated rats a higher tissue permeability was induced by changes in the calcium metabolism[5]. Previous works have shown an influence of the parathyroids on connective tissue: bone decalcification provoked by hyperparathyroidism is associated with modifications of the organic bone matrix and this is believed to be the primary action of the parathyroid hormone[6–8]. A modification of the mucopolysaccharide components of serum has also been described[9].

In the present paper, experiments on the effects of the parathyroidectomy on the vascular permeability and on the connective tissue are described.

Female Sprague-Dawley rats were parathyroidectomized when weighing 150–200 g. In the first experiment vascular permeability changes were investigated by the method of WILHELM et al.[10]. The experiment was carried out on 9 parathyroidectomized rats, 15, 30 and 60 days after the operation, and on 10 controls (some of them subjected to sham-operation). The abdomen of the rats was accurately shaved 48 h before the test; 0.2 ml of 5% solution of Pontamine Sky-Blue 6 BX (Gurr's) were injected intravenously into the tail or the jugular veins. Half an hour later, 0.1 ml of rat serum was injected intradermally into the shaved area, in order to provoke an inflammatory reaction, induced by the plasma factor (P.F./native) of WILHELM et al.[10]. This mild inflammatory agent was chosen in order to follow the evolution of the inflammatory reaction more easily. The changes in permeability of the capillaries were evaluated according to the time taken for the skin to stain and the extension and intensity of the stain around the site of injection of the rat serum. The spreading of the colour around the injection site begins earlier in the parathyroidectomized than in the control rats: 1 h after the injection the spreading of the colour and its intensity is definitely greater in the operated rats (Figure).

A difference was already noticed in the animals operated 15 days before; it was more marked in the groups operated 30 and 60 days before.

The animals were subsequently bled in order to determine the calcium content in serum, after which they were dissected; a final evaluation of the skin reaction was made

[1] P. ROWINSKI and G. MANUNTA, Boll. Soc. Ital. Biol. sper. *27*, 37 (1951).
[2] P. ROWINSKI and G. MANUNTA, Rend. Acc. Naz. Lincei *10*, 495 (1951).
[3] G. MANUNTA, Rend. Acc. Naz. Lincei *12*, 104 (1952).
[4] A. MILLO and A. BEGLIOMINI, Riv. di Biol. *50*, 405 (1958).
[5] G. MANUNTA, Riv. di Biol. *50*, 279 (1958).
[6] I. GERSH and H. R. CATCHPOLE, Amer. J. Anat. *85*, 457 (1949).
[7] MINNIE HELLER-STEINBERG, Amer. J. Anat. *89*, 347 (1951).
[8] M. B. ENGEL and H. R. CATCHPOLE, Proc. Soc. exp. Biol. Med. *84*, 336 (1953).
[9] M. R. SHETLAR, R. H. BRADFORT, W. JOEL, and R. P. HOWARD, *The Parathyroids.* Proceedings of a Symposium on Advances in Parathyroid Research (Charles C. Thomas Publ., Springfield 1961), p. 144.
[10] D. L. WILHELM, P. Y. MILL, ELIZABETH M. SPARROW, MARGARET E. MACKAY, and A. A. MILES, Brit. J. exp. Path. *39*, 228 (1958).