The results suggest that cholera toxin acts indirectly on many components in the enzyme chain of PG synthesis and metabolism, shifting their equilibrium in one direction or the other from the normal level.

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EXPERIMENTAL STUDY OF THE POSSIBILITY OF ISOLATED PERFUSION OF THE LYMPHATIC SYSTEM

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Among the many functions of the human lymphatic system, its role in transport has been the least adequately studied. It has become recognized that movement of lymph takes place only in the central direction on account of the valve system of the lymphatic vessels, rhythmic contractions of the diaphragm, pulsation of arteries, and active and passive movements. As regards relations between the lymphatic and blood systems, two diametrically opposite opinions are held. The supporters of lymphovenous junctions [1, 7, 9, 11, 13, 14] claim that anastomoses between lymphatic and venous vessels have now been proved reliably, but this is evidently true only of pathologically formed connections arising in the presence of chronic lymphatic stasis of varied origin. Other workers [2-4, 8, 10, 12], on the other hand, consider that the absence of any additional sites of junction of the blood and lymphatic vessels than the places of entry of the main lymphatic trunks into the venous system in the neck is an undisputed fact. Yet it is important to know whether lymphovenous anastomoses do or do not exist. Since under ordinary conditions no additional sites of drainage of lymphatic vessels into veins have been found, it would seem perfectly possible to create isolated perfusion of the lymphatic system by means of endolymphatic infusion with simultaneous drainage of the thoracic duct. The investigation described below was carried out to study this problem.

EXPERIMENTAL METHOD

The investigation was conducted in three directions: the volume flow rate of Evans' blue T-1824, injected endolymphatically was studied (by this method, passage of the dye into the blood system is not observed); in addition, the functional capacity of the lymphatic system to retain various drugs, low-molecular-weight substances, and microorganisms was tested, and the possibility of creation of an extracorporeal lymphatic circulation was investigated.

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Experiments of series I were carried out on 22 fresh cadavers. The thoracic duct was drained, the iliac and right subclavian veins were catheterized, and 5 ml of 0.5% Evans' blue in 80-100 ml isotonic solution was then injected intranodularly. The indicator was injected beneath the capsule into the left (seven cadavers), right (seven cadavers), and left and right (eight cadavers) inguinal lymph nodes simultaneously through a horseshoe-shaped self-fixing needle by means of an automatic perfusion pump (Infusion Pump, Sweden) at a rate of 0.1 to 0.5 ml/min. Passage of the dye from the lymphatic system into the blood system was detected spectrophotometrically (SF-4 instrument) by measuring the increase in optical density of T-1824 in blood plasma taken from the iliac vein compared with that in blood plasma from the right subclavian vein (control).

Experiments of series II were carried out on 50 mongrel dogs by the following general technique. Under intravenous thiopental anesthesia a catheter was inserted into the inferior vena cava through the greater saphenous vein. A lymphatic vessel on the outer surface of the leg was then catheterized and, finally, the thoracic duct was drained. Its proximal portion was ligated before its point of entry into the vein. In this series five experiments were carried out with perfusion with Evans' blue, staphylococci, kanamycin, thiopental sodium, and sodium chloride. In all cases perfusion was carried out by the automatic pump with a volume flow rate of 0.3 ml/min. The appearance of Evans' blue in the biological media was verified photometrically, kanamycin by the agar diffusion method [5], staphylococci by the appearance of growth on yolk-salt agar, the passage of a lethal dose of barbiturates through the lymphatic system by a biological test on mice, and the passage of low-molecular-weight substances (1-3% KCl solutions) through the lymphatic system by determination of potassium in the lymph, blood plasma, and urine by flame photometry.

In series III (15 dogs) lymph from the thoracic duct was returned to the peripheral lymphatic vessels in the lower limbs, i.e., an extracorporeal lymphatic circulation was set up. In this series of experiments the lymph drainage and toxicity of the lymph [6] from the thoracic duct were determined before and during perfusion.

EXPERIMENTAL RESULTS

Analysis of the results of the experiments on cadavers showed that in 15 (of 22) cases Evans' blue did not penetrate into the venous system. In five of the seven cases in which the rate of injection exceeded 0.3 ml/min, passage of the indicator into the ipsilateral iliac vein was observed. In two cases, it was considered that the dye leaked into the venous system because of injury to the sinuses and blood vessels of the lymph node by the injection needle. Considering these errors, later during experiments on dogs the preparations were injected, not into the lymph nodes, but into lymphatic vessels, and the rate of injection did not exceed 0.3 ml/min into each peripheral vessel. To avoid artificial passage of the dye into the blood system, the rate of perfusion of the peripheral lymphatic vessels, in our opinion, ought not to exceed 0.3 ml/min, for a rise of pressure in the lymphatic capillaries leads to partial leaking of the injected preparations into the venous component of the blood vascular system at the lymph node level.

Five groups of animals (10 dogs in each group) were used in the experiments of series II. In group 1, by means of the automatic perfusion pump 2 ml of a 0.5% solution of Evans' blue was injected endolymphatically, followed by 60 ml of isotonic solution. After the beginning of perfusion blood and lymph samples were taken every 10 min for 2 h. Blood and lymph obtained before injection of the T-1824 indicator served as the control. In no case was the passage of Evans' blue from the lymphatic into the blood system discovered.

The ability of the lymph nodes to retain microbial cells was tested in the animals of group 2. After endolymphatic infusion of staphylococci into the dogs and subsequent seeding, single colonies were obtained from the lymph in the case of perfusion of 10^9 microbial cells/kg body weight for 20 min. After perfusion of $2 \cdot 10^9$ microbial cells/kg body weight for 10 min continuous growth of staphylococci was obtained from lymph of the thoracic duct. In no case were staphylococci grown from the blood.

The animals of group 3 were perfused with kanamycin disulfate (50 ml/kg), which was detected in the lymph in a concentration of 40 μg/ml after 10 min, whereas only traces of this antibiotic (5 μg/ml) appeared in the blood of animals with effect from 20 min after the beginning of perfusion of the lymphatic system with the antibiotic.

In group 4 the dogs were perfused with a dose of thiopental sodium 10 times greater than the highest sessional dose (100 mg/kg). Absence of passage of the substance into the blood system was judged from the weak anesthetic effect obtained in the animals. Meanwhile intraperitoneal injection of 1 ml of lymph taken from the thoracic duct of dogs 10 min after the beginning of perfusion, into albino mice, caused not only anesthetic sleep, but also death of some of the mice.

In group 5 the animals were perfused with low-molecular-weight substances (1% and 3% KCl solutions). Only 10 min after the beginning of perfusion considerable quantities of potassium (up to 40 mmoles/liter) were discovered in lymph flowing from the thoracic duct; its concentration in the blood and urine increased only by 0.5-1 mmole/liter. It should be pointed out that parallel investigations of protein in lymph from the thoracic duct of the animals of this group showed considerable (three-fourfold) increases in its concentration after the beginning of perfusion. In our opinion this is a manifestation of Donnan equilibrium — an essential condition for binding and retaining in the lymphatic system concentrations of potassium, the ions of which (30 nm) are many times smaller than the capillary pores (over 1000 nm), that would be dangerous for the body.

Data obtained on cadavers and in experiments on animals confirm autonomy of the lymphatic system from the blood system. This enabled us in the experiments of series III (15 dogs) to set up a closed perfusion system, or extracorporeal lymphatic circulation. New properties of the transport function of the lymphatic system were discovered in these experiments: 5-7 min after the beginning of closed perfusion the lymph outflow through the thoracic duct increased from 0.22 ± 0.01 to 1.8 ± 0.07 ml/min ($P < 0.001$), but the toxicity of the lymph rose by 1.5-2 times compared with the toxicity of lymph obtained from these dogs by ordinary drainage of the thoracic duct.

The experiments thus showed that during drainage of the thoracic duct and intralymphatic infusions with a volume flow rate of not more than 0.3 ml/min into each peripheral vessel conditions are created for isolated perfusion of the lymphatic system with various substances, sometimes in increased concentrations. Isolated perfusion of the lymphatic system increases lymph production and the toxicity of lymph flowing from the thoracic duct, and this may be utilized for the purposes of detoxication and intensive treatment.

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