THE EFFECT OF ADENINE NUCLEOTIDES ON THE PERMEABILITY OF THE SKIN VESSELS IN RATS

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Of great importance in the pathogenesis of inflammation are the so-called "endogenous factors" of inflammation, which are physiologically active products of metabolism and tissue destruction. They include histamine, acetylcholine, sympathin, serotonin, and a group of polypeptides (leukotaxin, bradykinin), active proteins, and products of nuclein metabolism.

An increase in the permeability of the vessels, along with the emigration of leukocytes, is one of the earliest signs of inflammation. Numerous observations [3, 5, 6, 7, 9, 10] have shown that during the action of an inflammatory agent—trypan blue, when injected intravenously, rapidly penetrates into the focus of inflammation. The role of several different factors (histamine, leukotoxin, hyaluronidase) in the increasing of the vascular permeability in inflammation has also been investigated.

We have found [2, 4] that, together with other physiologically active substances, the adenine nucleotides play a definite role in the pathogenesis of the principal phenomena of inflammation — in the disturbance of the vascular permeability, in the emigration of the leukocytes, and in their phagocytic function. In the early stage of development of inflammation, there is an increase in the ATP content of the blood flowing from the focus of inflammation, and also in the inflamed tissue itself.

In investigations on rabbits we demonstrated the importance of the adenine nucleotides in the disturbances of vascular permeability. We found that ATP causes a marked increase in the permeability of the skin vessels in normal animals. For instance, after the intradermal injection of 1000-200 γ of ATP (sometimes after 20 γ)—if, at the same time, a 1% solution of trypan blue (5 ml/kg body weight) is injected intravenously—the dye is rapidly deposited in the area of the skin into which the ATP was injected. The intensity of deposition of the dye in these areas is largely determined by its concentration in the blood. With a decrease in the amount of dye injected (2.5-2.0 ml/kg body weight), the reaction develops later and is weaker. A dose of 1 ml/kg does not produce the concentration in the blood necessary for the rapid detection of its passage into the tissue, and is therefore unsuitable for this purpose.

In order to continue the study of the problem of the increase in the vascular permeability under the influence of compounds of the adenylic system, we conducted a series of observations on the permeability of the skin vessels in white rats.

EXPERIMENTAL METHOD

Experiments were carried out on 40 white rats weighing 140-170 g, in which the hair of the skin of the abdomen had been clipped 24 hours beforehand. An intraperitoneal injection of 0.8-1.0 ml of a 1% solution of trypan blue was given. After an interval of 3-4 minutes had elapsed, ATP or adenylic acid in a dose of 1000-250 γ (occasionally 25 γ ATP) in 0.1 ml of physiological saline was injected into the skin of the abdomen (at a distance of 1 cm from the linea alba). The solutions were prepared at the moment of injection. ATP preparations from the Ivanovo Meat Combine, and also imported adenylic acid and MAP in ampules, were used. In control experiments, 0.1 ml of physiological saline was injected. In order to achieve greater clarity, in some experiments the solutions of ATP or adenylic acid were injected into the skin on one side of the abdomen, and physiological saline was injected into the symmetrically opposite skin. Increased permeability of the vessels was judged by the intensity of staining of the area of skin into which the test substances were injected. Observations were maintained for a period of 2 hours.

EXPERIMENTAL RESULTS

From 10 to 15 minutes after the injection of ATP (in some cases after 30 minutes) the dye appeared in the skin around the site of injection. The intensity of staining gradually increased, and the dye spread diffusely over the whole area, to reach maximum saturation after 1.5-2 hours (Fig. 1). The diameters of the stained areas in the course of a marked reaction to injection of 250, 500, and 1000 γ ATP were 11-14 mm. In response to

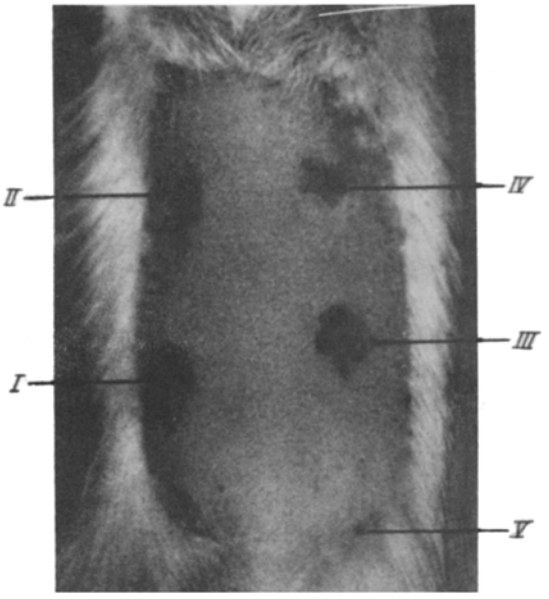

Fig. 1. Deposition of trypan blue, after injection of 1 ml of a 1% solution in areas of the skin of the abdomen in a rat following injection of ATP (in 1 ml of physiological saline) in these areas. I) 1000 γ; II) 500 γ; III) 250 γ; IV) 25 γ; V) physiological saline (absence of reaction).

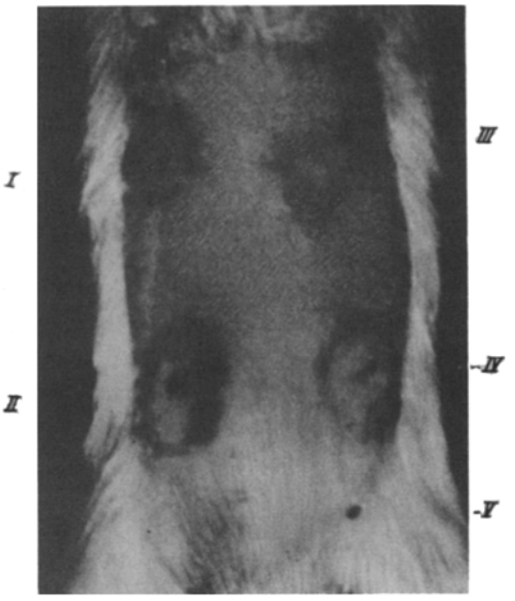

Fig. 2. Deposition of trypan blue (1 ml of a 1% solution injected intraperitoneally) in areas of the skin of the abdomen after injection of ATP and MAP into these areas. I) 250 γ ATP; II) 500 γ ATP; III) 250 γ MAP; IV) 500 γ MAP; V) site of injection of physiological saline. The deposition of the dye was more intensive at the sites of injection of ATP.

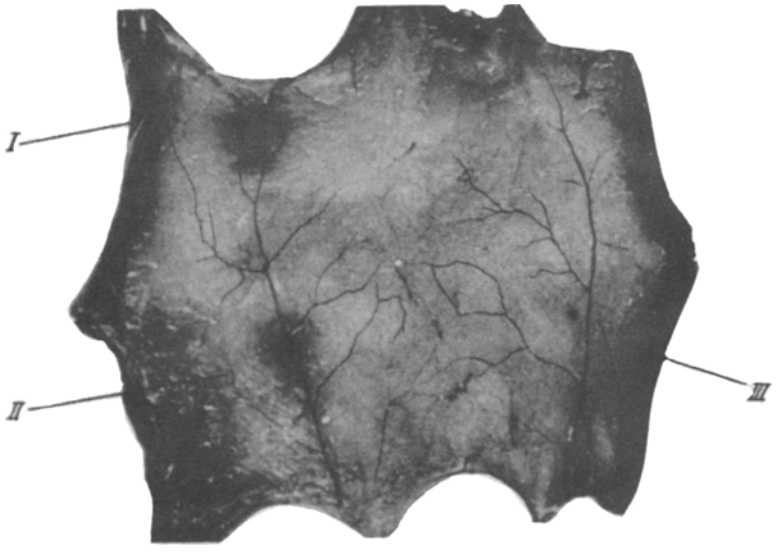

Fig. 3. Deposition of trypan blue (1 ml of a 1% solution injected intraperitoneally) in areas of the skin after injection therein of I) 500 γ ATP; II) 250 γ ATP in 0.1 ml of physiological saline. The point on the right – the site of injection of 0.1 ml of physiological saline (marked).

the action of the same dose of adenylic acid, staining appeared later and was less intensive (Fig. 2). In the control areas of skin, into which physiological saline was injected, sometimes no deposition of dye was found, and only in the center, at the site of puncture, did a blue spot develop. In some experiments the skin was also examined from the side of the dermis, where the reaction was particularly clearly expressed (Fig. 3).

It was not always possible to establish any definite relationship between the intensity of staining and the dose of the substance injected. At the same time, a clear relationship was found between the intensity of staining and the quantity of trypan blue injected. It thus became obvious that it was necessary to take account of the minimum quantity of dye required for demonstration of the effect of staining the tissue.

The investigations showed that ATP has a stronger effect on the permeability of the vessels than adenylic acid, which plays an important part in the stimulation of phagocytosis. The importance of adenine nucleotides and of decomposition products of nucleic acids in increasing the vascular permeability has also been demonstrated by other researchers [8, 12, 13].

SUMMARY

The effect of adenine nucleotides in causing a vascular permeability is discussed. As established in experiments on white rats, ATP causes a clear increase of vascular permeability in the skin of rats. This is detectable with the aid of vital trypan blue staining following intraperitoneal injection of the stain. The action of adenylic acid on the vascular permeability is less pronounced than that of ATP.

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