PATHOLOGICAL PHYSIOLOGY AND GENERAL PATHOLOGY

THE MECHANISM OF EXCRETION OF DYSENTERY ANTIGEN BY THE KIDNEYS

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(Received May 17, 1958. Presented by Active Member of the AMN SSSR L. A. Zil'ber)

Many investigations have been devoted to the excretion of viruses and bacterial antigens by the kidneys. These have supplied information about viruses and antigens excreted, but not about the mechanisms concerned in their excretion. Only in recent years has research been undertaken, under the direction of L. A. Zil'ber and A. D. Ado, into the mechanisms of excretion of viruses and antigens by the kidneys [1-5].

The objects of our present investigation were: to study the influence of proteinuria due to hyperproteinemia on the process of excretion of antigen of the Flexner dysentery bacillus; to ascertain the laws governing the excretion of antigen during maximum secretion of diodrast; and to discover any changes in the excretion of antigen during selective poisoning of the cells of the tubules by 2,4-dinitrophenol.

EXPERIMENTAL METHOD

Hyperproteinemia was induced by the repeated intraperitoneal injection of homologous serum in doses of 50, 70, 90 and 100 ml per 1 kg body weight of the animal, with intervals of 2-3 days between injections. The plasma proteins were determined with a refractometer. The quantity of protein in the urine was studied by the Roberts-Stol'nikov method. The concentration of diodrast in the blood was kept between 15-30 mg% by infusion of a solution or by repeated injection of a 10% solution in small doses (6-10 ml) in each clearance period of 20 minutes. Diodrast was determined by the method described by N. A. Ratner [6]. The maximum secretion of diodrast was calculated from the formula

$$K_{MS} = K_M D - 0.73\, K_P F_K,$$

where K_{MS} – maximum secretion; K_M – concentration of diodrast in the urine; D – the minute diuresis; K_P – concentration of diodrast in the plasma; F_K – filtration as determined by creatinine; 0.73 – readily soluble fraction of diodrast excreted by filtration (coefficient of filtration of diodrast).

Poisoning with 2,4-dinitrophenol was carried out with 10 mg of the drug per 1 kg body weight of the animal. In the experiments we used healthy dogs whose kidneys had been shown to be free from any abnormality of function by repeated investigations.

Complete Flexner dysentery antigen, obtained from the I. I. Mechnikov Moscow Institute of Vaccines and Sera, was injected intravenously or intramuscularly in a dose of 0.1 - 0.4 mg per 1 kg body weight of the animal. The quantity of antigen in the serum and urine was estimated by the cold complement fixation reaction. Precipitated polyvalent dysentery serum, with a titer according to the complement fixation reaction of 1: 2,000,000, obtained from the Moscow Institute of Vaccines and Sera, was used in the performance of the reactions. By the use of this serum it was possible to detect the presence of 0.0000005 g or 0.0005 mg of antigen in 1 ml of fluid tested.

TABLE 1

The Effect of Proteinuria on the Excretion of Dysentery Antigen by the Kidneys

Name of dog	Minute diuresis ml/min	Creatinine clearence index ml/min	Antigen				Plasma proteins				
			concentration in serum, mg%	concentration in urine, mg%	concentration index	clearance index, ml/min	concentration in blood	concentration in urine, %	filtration, mg/min	excretion in urine mg/min	reabsorption mg/min
Roza	0.8	68	0.4	0.2	0.5	0.4	—	—	—	—	—
Knopka	0.5	66.4	0.4	0.2	0.5	0.25	—	—	—	—	—
Volna	0.7	91.8	0.2	0.1	0.5	0.35	—	—	—	—	—
Norma	0.5	98.7	0.4	0.2	0.5	0.25	—	—	—	—	—
Roza	0.7	62	0.4	0.8	2	1.4	10.28	0.65	27.34	4.55	23.79
Knopka	0.4	66	0.4	0.8	2	0.8	9.78	0.33	27.1	1.32	25.8
Volna	0.41	71.7	0.2	0.4	2	0.82	10.80	0.264	33.18	1.08	32.1
Norma	0.5	80.5	0.4	0.4	1	0.5	9.81	0.429	33.8	2.14	31.7
Lapusha	1.0	96	0.4	0.8	2	2.0	—	—	—	—	—
Laika	1.0	108	0.4	0.8	2	2.0	—	—	—	—	—
Laska	0.6	74	0.4	0.8	2	1.2	—	—	—	—	—
Roza	0.5	68	0.4	0.8	2	1.0	—	—	—	—	—
Lapusha	1.0	98	0.4	3.2	8	8.0	10.41	0.165	43.85	1.65	42.2
Laika	0.7	105	0.8	6.4	8	5.6	10.69	0.099	48.12	0.92	47.2
Laska	0.4	66	0.4	3.2	8	3.2	10.85	0.099	28.7	0.39	28.3
Roza	0.5	68	0.4	3.2	8	4.0	10.17	0.132	29.64	0.66	28.9

TABLE 2

The Excretion of Dysentery Antigen by the Kidneys of Dogs during Maximum Secretion of Diodrast

Name of Dog	Minute diuresis ml/min	Creatinine clearance index ml/min	Antigen				Diodrast				
			concentration in serum, mg%	concentration in urine mg%	concentration index	clearance index ml/min	concentration in plasma mg%	concentration in urine, mg%	filtration, mg/min	maximum secretion, mg/min	excretion in urine mg/min
Laika	2.0	113.0	0.4	0.8	2	4	—	—	—	—	—
Malyshka	1.9	91.0	0.4	0.8	2	3.8	—	—	—	—	—
Lapusha	1.5	92	0.4	0.8	2	3.0	—	—	—	—	—
Laika	2.1	113.6	0.4	1.6	4	8.4	19.7	4 000	16.33	67.6	84.0
Malyshka	2.0	100	0.4	1.6	4	8.0	16.1	4 770	11.75	83.6	95.4
Lapusha	1.0	102.3	0.4	1.6	4	4.0	30.5	7 890	22.7	56.1	78.9

EXPERIMENTAL RESULTS

In the first series of investigations the influence of proteinuria on the excretion of dysentery antigen was studied in 8 dogs.

As seen from Table 1, proteinuria increases the excretion of dysentery antigen by the kidneys of both immunized and unimmunized dogs, but this effect is much more marked in the immunized animals.

In unimmunized dogs the antigen is excreted slowly the concentration index is 0.5, the clearance index varies between 0.25 and 0.4 ml/min. In proteinuria due to hyperproteinemia the antigen is excreted more intensively; the concentrating power of the kidneys in respect of the antigen is increased to give an index of 2; with diuresis close to that of the control experiment, clearance increases and varies between 0.5 and 1.4 ml/min. After active immunization the influence of proteinuria on clearance of antigen is still more pronounced. Under identical experimental conditions, with equal diuresis and the same concentration of antigen in the plasma, proteinuria brings about a marked increase in the antigen clearance index, which reaches 4-8 ml/min compared with 0.5-1.4 ml/min in unimmunized animals.

The increased excretion of dysentery antigen during proteinuria, due to hyperproteinemia, is evidently explained by the greater reabsorption of serum protein than of antigen. An explanation of the mechanism of this relationship between reabsorption of antigen and of serum protein requires a special investigation.

Some workers [7] have shown that secretion of para-aminohippuric acid by the tubule cells leads to depression of reabsorption of ascorbic acid. In view of this finding, in the next series we attempted to explain the effect of maximum secretion of diodrast on the process of excretion of dysentery antigen in the final urine. The results of experiments performed on 3 dogs are shown in Table 2. As seen from this table, maximum secretion of diodrast is accompanied by slight stimulation of the excretion of dysentery antigen by the kidneys of dogs. In experiments without diodrast and with the same concentration of antigen in the serum in immunized animals, the concentration index was 2 and the clearance 4 ml/min. With maximum secretion of diodrast and the same concentration of antigen and minute clearance, the concentration index rose to 4 and the clearance to 8.4 ml/min.

The results of this series of experiments show that maximum secretion of diodrast is accompanied by a considerable increase in the excretion of dysentery antigen by the kidneys of immunized dogs. How can this

TABLE 3

The Excretion of Dysentery Antigen by the Kidneys of Dogs during Poisoning with 2,4-Dinitrophenol

Name of dog	Dose of 2,4-dinitrophenol, mg/kg	minute diuresis, ml/min	Creatinine clearance index, ml/min	Antigen			
				concentration in serum, mg%	concentration in urine, mg%	concentration index	clearance index ml/min
Lapusha	—	1,8	94	0,4	0,8	2	3,6
Mal'va	—	2,2	110	0,4	0,8	2	4,4
Malyshka	—	1,5	114	0,4	0,8	2	3,0
Laika	—	1,6	102	0,4	0,8	2	3,2
Lapusha	10	1,9	96	0,4	1,6	4	7,6
Mal'va	10	2,0	110	0,8	3,2	4	8,0
Malyshka	10	1,5	112	0,4	1,6	4	6,0
Laika	10	1,5	—	0,4	1,6	4	6,0

mechanism of increased antigen clearance during maximum secretion of diodrast be explained? According to the same workers [7] maximum secretion of one substance inhibits the reabsorption of another.

If this view[7] is accepted, it can be assumed that in our experiments the increased excretion of antigen is connected with some degree of inhibition of its reabsorption from the lumen of the tubules.

On these grounds it may be postulated that some particles of dysentery antigen pass through the glomerular membrane and part of these are reabsorbed during their passage along the tubules. Reabsorption of antigen by the cells of the tubules may be inhibited by maximum secretion of diodrast or stimulated by reabsorption of protein; as a result of this much more antigen is excreted in the final urine.

In our next series of investigations we studied the effect of selective poisoning of the tubule cells during excretion of dysentery antigen by the kidneys of dogs. The animals were poisoned with 2,4-dinitrophenol, which in a dose of 10 mg per 1 kg body weight causes selective damage to the cells of the tubules. As we showed, in spite of the early development of a febrile reaction to the 2,4-dinitrophenol, signs of tubular damage were seen from 2-3 hours after the beginning of its administration. Accordingly we gave dinitrophenol to the animals from 2-3 hours before injecting the antigen. Experiments on 4 immunized dogs enabled us to discover certain distinctive features of the excretion of antigen during poisoning of the tubule cells. In Table 3 are shown the experimental results obtained after poisoning the dogs with 2,4-dinitrophenol.

In immunized animals poisoned with 2,4-dinitrophenol, the concentration index was twice as high as in the previous experiments (before poisoning).

The blood antigen clearance value is also correspondingly raised to 6-8 ml/min. The increased excretion of dysentery antigen in the poisoned animals is evidently connected with a fall in reabsorption of antigen, since in the doses in which we administered it, 2,4-dinitrophenol causes selective damage to the cells of the tubules without affecting the permeability of the glomerular membrane.

From an analysis of all the results it may be concluded that many particles of dysentery antigen pass through the glomerular membrane without damaging it. During its passage along the tubules part of the antigen is resorbed by the cells of the proximal division of the tubules by the same mechanism as that responsible for the reabsorption of protein present in the lumen of the tubules. The intensity of reabsorption of antigen depends on the immunological state of the animal and also on the processes of secretion and reabsorption of other substances (components) passing through the kidney at the same time as the antigen.

SUMMARY

Proteinurea caused by hyperproteinemia considerably intensifies the excretion of the dysentery antigen by the dog's kidneys. The mechanism of increased urinary excretion of the antigen in hyperproteinemia is connected

with its reduced resorbtion. The maximal secretion of cardiotrast as a result of development of competitive relations with the antigen reabsorption unhibitis the absorption of the latter and increases its excretion into the final portion of the urine. 2,4-Dinitrophenol poisoning causes a selective affection of the tubular cells and limits the resorbtion of the antigen.

LITERATURE CITED

[1] A. D. Ado, Antigens as Extraordinary Stimuli of the Nervous System, Moscow, (1952).*

[2] A. D. Ado, A. A. Pol'ner and M. M. Khakberdyev, Uspekhi Sovremennoi Biol. 1, 70-82 (1957).

[3] L. A. Zil'ber, Voprosy Virusologii 1, 45-54 (1956).

[4] L. A. Zil'ber, A. K. Shubladze and A. D. Sheboldaeva, Zhur. Mikrobiol. Épidemiol. i. Immunobiol. 4, 531-545 (1937).

[5] V. A. Parnes, N. D. Petrova, E. V. Volina and Z. A. Avenirova, Zhur. Mikrobiol. Épidemoil. i Immunobiol. 5, 40-44 [1950].

[6] N. A. Ratner, Changes in the Function of the Kidneys in Hypertension, Moscow (1953).*

[7] E. E. Selkurt, Am. J. Physiol. 142, 182-190 (1944).

* In Russian.