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Lack of Genetic Control of the Lymphatic Pathway. Studies on Rats

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Summary

The lymphatic route from the testis to the cisterna chyli is studied in two inbred and one random bred strain of rats. The pathway varied similarly in all three groups with no indication of genetic control.

Introduction

It has previously been reported that the lymphatics from the right testis of rats sometimes bypass all lymph nodes and drain directly to the cisterna chyli (4). There are some old observations which indicate that this direct pathway to the blood stream can be found in endocrine glands in animals (2) and it has been observed in the thyroid gland in man (5).

In experiments on rats, Engeset (4) observed that lymphatics from the testis went directly to the cisterna chyli in about 1/3 of the animals. One of the rat strains primarily studied (Hooded strain A, reference 4) has since been inbred. In the present study testis lymphography has been performed in this strain in order to see if this direct lymphatic communication is genetically controlled.

Material

This study has been performed on 39 animals in 3 groups.

Group I: 24 rats from the inbred Hooded strain A. This strain has been bred as a closed popu-

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Table 1 The route of lymphatic drainage from rat testes studied by injection of metallic mercury. A dash indicates where no node is visualized on the main pathway to the cisterna chyli.

Parents	♀	Rat No.	Group I			
			Left side	Right side		
600	595	606	Renal node	Renal node		
		607	-	Renal node		
		608	-	-		
		609	-	-		
		610	-	-		
606	605	617	-	-		
		618	-	-		
606	603	623	Lumbar + renal node	Lumbar + renal node		
		624	-	-		
		625	-	-		
		626	-	-		
		627	Renal node	-		
		628	-	-		
		629	Lumbar node	-		
		630	-	-		
		606	602	325-1	Renal node	-
				325-2	Renal node	-
606	604	326-1	-	-		
		326-2	Lumbar node	-		
		326-3	-	-		
		326-4	-	-		
		326-5	-	-		
		326-6	-	Lumbar node		
		326-7	-	-		
			Group II			
7642	7639	7781	Renal node	-		
		7782	-	-		
		7783	-	-		
		7784	Renal node	Lumbar node		
7642	7637	7736	Lumbar node	-		
		7738	-	-		
7642	7638	7741	-	-		
7642	7636	7753	-	-		
7642	7638	7790	-	-		
			Group III			
Random		1-1	-	-		
		1-2	-	-		
		1-3	-	-		
		1-4	Renal node	-		
		1-5	Renal node	-		
		1-6	Renal node	Renal node		

llation for many years, thereafter inbred for 12 generations. Skin grafts are not rejected among these animals. From this strain five males from one litter were examined (Fig. 1 and Table 1). One of them sired four litters with four different siblings. The male offspring of these litters and the five males of the parent generation were the animals evaluated for the study. The lymphatic pathway of the father was obviously not known until after the offspring had been sired.

Group II: 9 rats from an inbred strain bearing renal tumors. The animal tested did not have tumors and are all offspring of the same male by four different sibs.

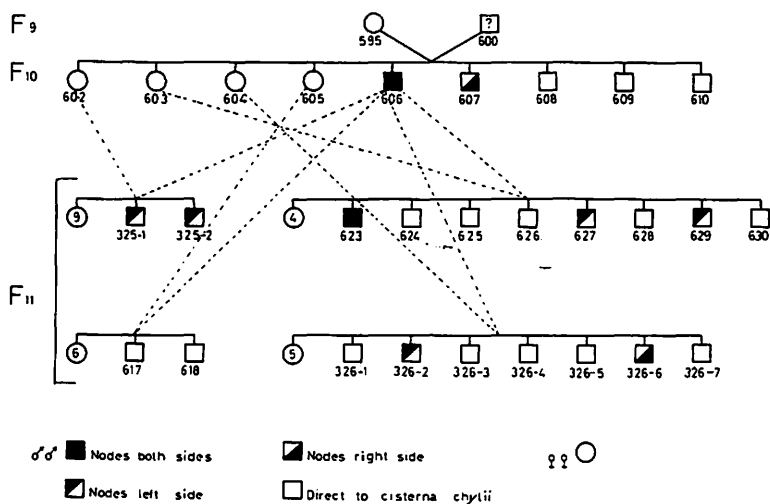


Fig. 1 Pedigree of hooded rats in Group I.

Group III: 6 rats from a commercial randomly bred Wistar strain (Wistar/AF/Han/Mol/(Han 67)).

Methods

The animals were killed with ether. The testes were then exposed by opening the scrotum. About 0.02 ml of 11% Patent Blue Violet was deposited in each testis for visualization of the lymph vessels. Two lymphatics are always easy to detect. One of them is seen at the proximal pole of the testis. Small branches emerging from the capsule of the testis converge into one vessel which traverses the surface of the upper pole of the testis under the caput epididymis. The other lymphatic suitable for injection lies on the dorso-medial aspect of the testis, at the point where the serous membrane is reflected from the epididymis into the testis. It runs from the distal to the proximal pole of the testis, parallel to the blood vessels. Like the other vessel, it is not covered by fat or connective tissue. These two lymphatics converge into one vessel either at the upper pole of the testis or in the funiculus.

Lymphatic punctures were made under the dissecting microscope. Metallic mercury – as a contrast medium – was injected into the lymphatic at the upper pole of the testis through a metal cannula with an external diameter of 0.45 mm. The injection was performed with a 1 ml syringe that was driven mechanically by a synchronous motor. The injection rate was 0.01 ml/min. 0.04–0.08 ml of mercury was injected in each side. After injection of both testes the animals were X-rayed. If the lymphatics were not sufficiently filled with contrast medium, the animals were reinjected with 0.04–0.06 ml of mercury and rephotographed.

The animals were then dissected under a dissecting microscope. The abdomen was opened and lymph vessels and nodes were exposed from testes to the cisterna chyli.

Results

The findings by lymphography are shown in Table 1, and typical examples are shown in Fig. 2–5.

In group I – 15 of 24 rats had lymph vessels passing from the testes directly to the cisterna chyli on both sides without passing through any lymph node. The direct route on only one side

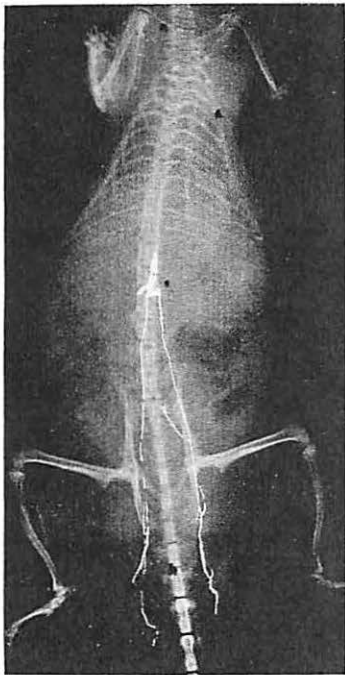


Fig. 2 Rat 624. Direct bilateral lymphatic pathway from testes to cisterna chyli. On left side a branch to lumbar node.



Fig. 3 Rat 326-5. The two lymphatics anastomose on left side and proceed as one trunk directly to cisterna chyli. Branch to left lumbar node.

was observed in 7 rats: 2 of them on the left, and 5 on the right side. In one rat the lymphatics passed through renal nodes on both sides; in another one through lumbar and renal nodes on both sides.

In group II — 6 of 9 rats had lymph vessels passing directly to the cisterna chyli on both sides. In 2 rats there was a direct route on the right side. On the left, in one of them, lymph vessels passed through the renal node and in the other through the lumbar node. In one rat the lymphatics passed through the renal node on the left and through the lumbar node on the right side.

In group III — 3 of 6 rats had lymph vessels passing directly to the cisterna chyli on both sides: in two rats through renal nodes on left side and in another one through renal nodes on both sides.

In all rats from all groups the lymph nodes were normal and normally situated on dissection. On the lymphograms of rats with a direct route from the testes to the cisterna chyli, injected lymphatics bypassing normal nodes were seen.

Discussion

Retroperitoneal lymph nodes in the rat are the caudal node, solitary, situated on the midline, just distal to the bifurcation of the aorta, the iliac nodes close to the external iliac artery, the lumbar nodes on each side of the aorta, just above the bifurcation, and the 1-4 renal nodes usually 1 on each side of the aorta in the region of the renal artery. In our study the animals were dissected after lymphography and we found that in all rats the lymph nodes were normal and normally situated.

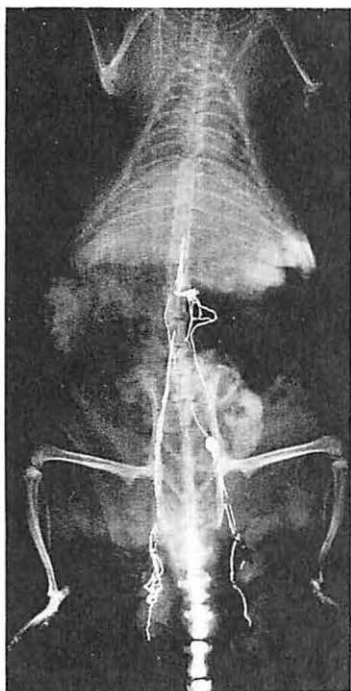


Fig. 4 Rat 606, sire. The lymphatic pathway to renal nodes on both sides, on left side visualized on X-ray, on right side found on dissection. Branch to left lumbar node with extravasation of mercury.

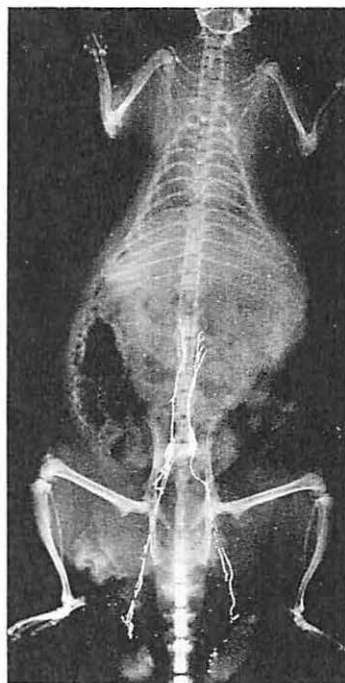


Fig. 5 Rat 623. The lymphatic pathway through lumbar nodes on both sides, visualized, and thence to the renal nodes found on dissection.

Metallic mercury has been employed in the present investigation because, in addition to excellent roentgen contrast, it also makes the lymphatics clearly visible for dissection. During dissection it can be pushed forward in the vessels and their branches can easily be observed. Mercury enters very fine branches of the lymph vessels but it has difficulty passing through nodes and bypasses the lymph nodes if there are vessels going that way. Mercury is therefore suitable for studies of bypassing vessels such as in the presented experiments. The fact that Engeset (3) found spreading of injected Walker carcinoma to the lungs only in those rats with a direct route, supports the validity of the conception that injection with mercury is a reliable indication of lymphatic topography.

The findings by lymphography are shown in Table 1 and typical examples are shown in Figs. 2-5. The study confirms the previous observation that more than half of the rats have lymph vessels passing from the testes directly to the cisterna chyli, without passing through any lymph node, on both sides. In the present study 24 out of 39 animals had this pathway. The direct pathway was seen more frequently on the right side than on the left. The direct route was observed in all three strains examined without any marked difference between the strains. The variation in lymphatic topography within each strain, together with the small differences between the three strains indicate that the pathway is not genetically controlled. The pedigree of the inbred hooded rats, Fig. 1, shows that ♂ 606, the sire of all individuals in F_{11} by his 4 sisters, had a lymphatic pathway through the nodes on both sides. Among those offspring, variations in the pathway were observed (Fig. 1). An accurate genetic analysis of this trait, if it is one, is complicated by the impossibility of similar studies in females.

Unfortunately, the sire of the males in Group II was not studied, but here again, the offspring of one male of an inbred strain by four of his sibs revealed the same diversity of the lymphatic pathway from the testis. Only six of the commercially obtained and randomly bred Wistar rats were studied and here there were three which bypassed all nodes while three went to the renal node.

This study was made on rats available at the time, and was not planned far enough in advance to have included as many generations as would have been desirable, but we believe that the evidence is sufficient to indicate that the topography of the lymphatic pathway from the testis to the thoracic duct is not genetically controlled, as there is just as much diversity in the inbred as in the randomly bred animals.

The fact that the testis lymphatics usually bypass the lumbar and iliac nodes may be a result of the embryonic topography. The testis develops in the renal area close to the cisterna chyli. This occurs in all mammals, but in man no direct pathway has been observed. One explanation could be that there are many more lymph nodes in man than in rats. The chance that the lymph vessels should bypass all these nodes which are believed to develop from the lymph vessels must be markedly reduced.

However, a direct pathway has been observed in dogs (4) which have more numerous lymph nodes than rats. This last observation has also been made using mercury as contrast medium. It may be that this pathway is detectable especially with mercury as contrast medium and that similar studies should be performed on human cadavers to see if it also exists in man.

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