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Microvascularisation of the pineal gland in the rat

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The authors investigated the pineal blood supply in the rat, including microvascularisation, using different methods of vascular corrosion cast technique and two methods of inspection: scanning electron microscopy (SEM) and light microscope (LM). The animals were divided into three groups and injected with two types of casting media. It was found that the pineal gland is highly vascularised. Arterial supply is from the branches of the medial posterior choroidal artery which originates from the posterior cerebral artery. All veins drain into the great cerebral vein directly without forming the pineal vein. However, we noticed a short venous trunk between the great cerebral vein and the confluence of the sinuses. This venous trunk had not been described in literature till now and does not have a name in anatomical nomenclature. The authors showed the relationship of the pineal gland to adjacent venous vessels.

key words: vascular corrosion cast, scanning electron microscopy, pineal gland, pineal arteries and veins, great cerebral vein, confluence of sinuses

INTRODUCTION

The detailed blood supply and its relations to surrounding brain areas have been studied in different species using different methods [2–4, 10]. Casting techniques and casting media for the demonstration of blood vessels have been extensively developed. Good casting media have sufficiently low viscosity to penetrate the capillary system and endure the overheating caused by electron beam. Some of the important casting media mentioned in literature were neoprene latex, nylon, aniline blue-gelatin mixture, polyester plastic and resin [3, 5]. In our investigations the microvascular system of the rat pineal organ was studied using various injection techniques, however the best effect was obtained using Mercox resin.

MATERIAL AND METHODS

Thirty adult male Wistar rats, weighing from 250––350 g, were used. The animals were divided into three groups. The first group was anaesthetised, the thoracic cavity was opened and after exposing the heart and cutting off apex, perfused through the left ventricle with 0.9% solution of NaCl and injected via ascending aorta with plastogen. Skulls were opened and area of pineal gland was inspected using LM. Next the specimens were corroded with 40% solution of KOH for studying the vasculature in LM. The second group was anaesthetised and using the same preparations injected with 20 ml of Mercox-Cl-2b. Five skulls were opened and inspected in LM, all ten specimens were corroded in 40% KOH solution. After 48 hours they were rinsed with detergent and studied in LM or after special preparation under the scanning electron microscope (SEM). The third group was anaesthetised and heparinised by an intraperitoneal injection of 5000 I.U. of heparin diluted in 2 ml of 0.9% NaCl solution. After perfusing with Mercox-Cl-2b the rats were placed in water bath for 24 hours. The rats' heads were corroded in 40% KOH solution for 24–48 hours, rinsed in distilled water

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with detergents for 24 hours, distilled water for 1–2 hours, dried, dissected and mounted using LM, sputtercoated with gold and inspected in SEM (Cambridge Stereoscan 600 at 15 kV).

RESULTS

The rat's pineal gland is a cone-shaped, elongated organ. The greatest part of the pineal gland is situated superficially, in front of the cerebellum, between superior colliculluses of the tectal plate, the rest is reduced. The pineal gland is inferior to the great cerebral vein, posterior and inferior to the internal cerebral veins and posterior to the great vein of prosencephali (Fig. 1). The pineal gland is highly vascularised (Fig. 3, 8). The arteries tend to branch off into the capillary network as in most parts of the brain (Fig. 8). The gland is supplied by 2–6 branches from the medial posterior choroidal artery, which originates from the posterior cerebral arteries [1, 4, 10]. These vessels had a very characteristic shape and course after dividing into precapillary network. Branching off was correlated with reduction of diameter and precapillaries tend to branch off at a right angle (Fig. 8). All the vessels can be identified by endothelial cell imprint pattern. We observed vessels with oval to longish endothelial nuclei imprint patterns orientated in parallel to the long axis of the vessel (Fig. 6, 7). According to the literature [6, 7] these vessels can be identified as arteries. We also noticed vessels with roundish imprints that were randomly orientated (Fig. 4, 6, 7). This description is characteristic of veins [6, 7]. Venous drainage consists of superficial collecting venules which are 40–60 μm in diameter (Fig. 3). There are about 10–16 peripherial venules and 1 or 2 central veins all draining the capillary network of the pineal gland to the great cerebral vein (Fig. 5). The great cerebral vein drains usually directly to the confluence of the sinuses (Fig. 2) or, as we noticed, via the short venous trunk between the great cerebral vein and the confluence of sinuses. The venous trunk joins the great cerebral vein with internal cerebral veins into a U-shaped vessel. This vessel drains directly into the confluence of sinuses (Fig. 9).

We did not find any route in pineal drainage into adjacent brain areas. There were no connections even on the precapillary level. However our study did not concern vessels situated in the proximal and intermediate parts of the pineal gland.

DISSCUSION

The arterial branches to the pineal gland of Wistar rat observed in this study are from the medial posterior choroidal artery, which originates from the posterior cerebral arteries. This seems to be in correlation with data reported in man [8, 12], rat [1, 4], mouse [1], rabbit [10]. Venous drainage consists of 10–16 peripheral veins and 1–2 central veins. These data are similar to those given in literature except for the additional central vein. All veins drain into the great cerebral vein directly without forming the pineal vein. However, we noticed a short venous trunk between great cerebral vein and confluence of sinuses. This seems to correspond to straight sinus in man. We did not find any route in pineal drainage into adjacent brain areas, confirming the hypothesis of Quay suggesting the retrograde perfusion [9]. The

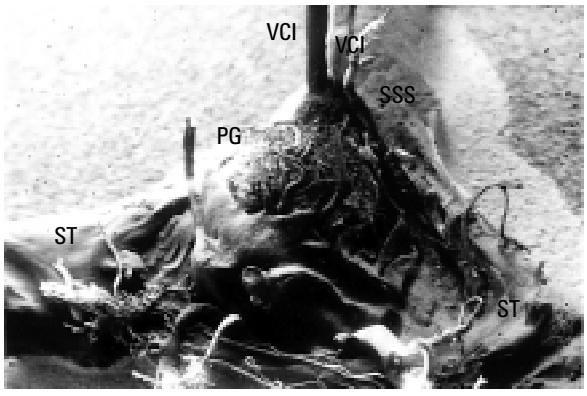

Figure 1. SEM of the pineal gland, relationship to surrounding vessels. The pineal gland is inferior to the great cerebral vein and the confluence of sinuses. Cast mounted upside down, top side is rostral. Magnification bar = 1 mm; PG — pineal gland; ST — transverse sinus; SSS — superior sagittal sinus; VCI — internal cerebral vein.

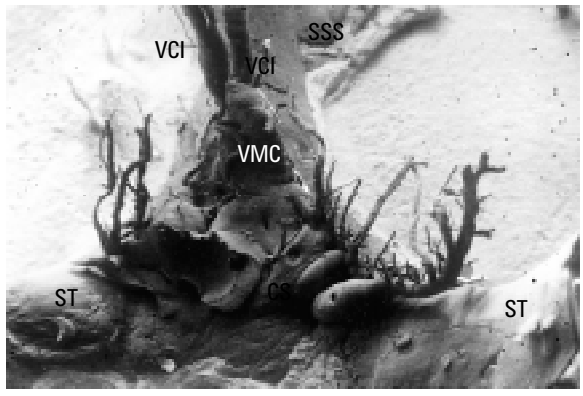

Figure 2. SEM of sinuses adjacent to the pineal gland. This figure shows the great cerebral vein drains into the confluence of sinuses. Pineal gland cut off, cast mounted upside down, top side is rostral. Magnification bar = 1 mm; VMC — great cerebral vein; ST — transverse sinus; SSS — superior sagittal sinus; VCI — internal cerebral vein.

Figure 3. SEM of collecting venules with imprinted endothelial cell pattern, very dense vessels network drains into collecting venules. Cast mounted upside down, top side is rostral. Magnification bar = 200 μm; V — collecting vein.

Figure 4. SEM of collecting venules with imprinted endothelial cell pattern (>), vessels with roundish imprints that were randomly orientated. Cast mounted upside down, top side is rostral. Magnification bar = 200 μm.

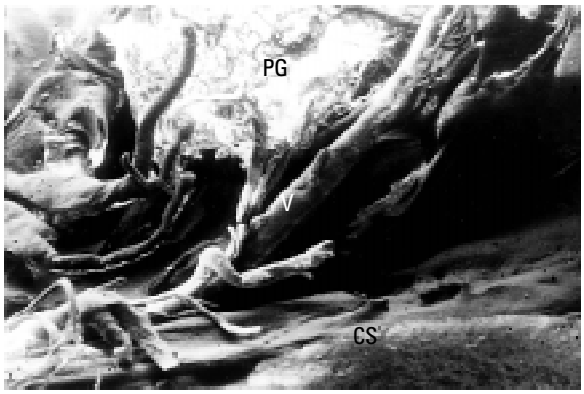

Figure 5. SEM of collecting venules and central vein all draining to the great cerebral vein. Cast mounted upside down, top side is rostral. Magnification bar = 400 μm; PG — pineal gland cast; V — central vein; CS — confluence of sinuses.

Figure 6. Precapillary and capillary network of the pineal gland, artery-like precapillary and vein-like precapillary, identification by endothelial cell imprint pattern. Cast mounted upside down, top side is rostral. Magnification bar = 100 μm; A — artery-like precapillary; V — vein-like precapillary.

Figure 7. SEM. Artery-like precapillary (A) and vein-like precapillary (V), all the vessels can be identified by endothelial cell imprint pattern. Arteries have oval to longish endothelial nuclei imprint patterns orientated in parallel to the long axis of the vessel, veins have roundish imprints that are randomly orientated. Cast mounted upside down, top side is rostral. Magnification bar = 100 μm.

Figure 8. This figure shows SEM of very dense capillary network. Branching off is correlated with reduction of diameter and precapillaries tend to branch off at a right angle. Cast mounted upside down, top side is rostral. Magnification bar = 40 μm.

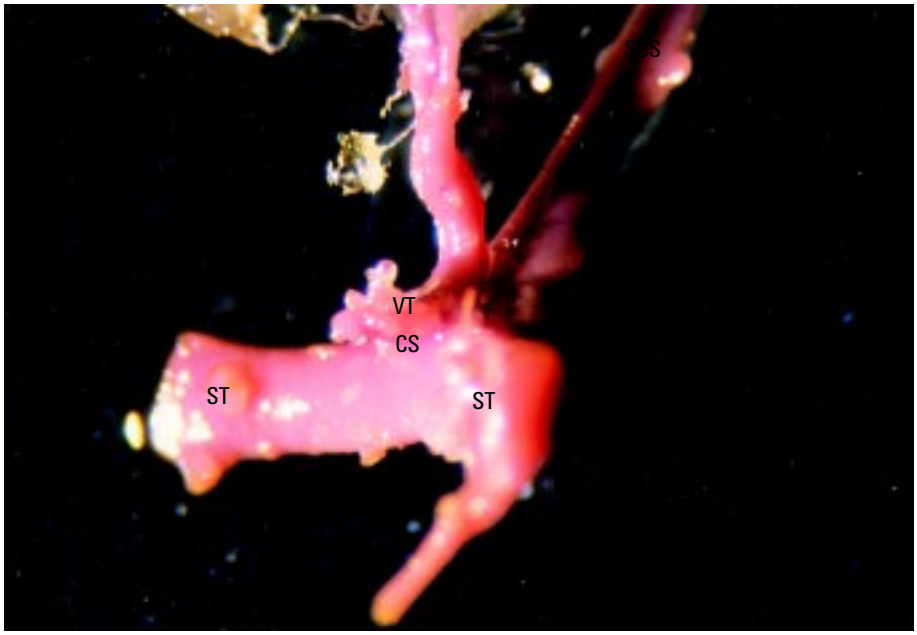

Figure 9. LM shows the venous trunk connecting the great cerebral vein and the confluence of sinuses, the alternative way of draining had not been mentioned in the literature yet. Plastogen cast, top side is rostral. ST — transverse sinus; SSS — superior sagittal sinus; CS — confluence of sinuses; VT — venous trunk.

results of the present observations confirmed that pineal gland is highly vascularised, and its capillary network is at least as dense as in other parts of rat's brain. The rat's pineal gland is classified as $\alpha\beta$C type [11] due to the fact that the bulk of the pineal gland has superficial position, the proximal and intermediate parts being greatly reduced in size. Therefore our study concerns only vessels adjacent to the bulk of pineal gland. The intermediate and proximal parts of the pineal gland need further investigation of microvasculature.

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