ARTERIO-VENOUS ANASTOMOSES IN THE HUMAN EXTERNAL EAR

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Having found that arterio-venous anastomoses were present in the external ears (auricles) of a number of species of animal (Daniel & Prichard, 1956), we examined some human ears and found that large numbers of these highly specialized blood vessels, which form direct communications between arteries and veins, were present also in man. In this paper we describe the distribution and morphology of these arterio-venous anastomoses in the human ear.

METHODS

The ears were obtained at necropsy from eight subjects of both sexes ranging in age from 17 to 90, and from one newborn infant. They were excised with about 1·0 cm. of skin surrounding their line of attachment. Two specimens were injected with neoprene latex, and five with Berlin blue (2 % in distilled water). The remaining ears were not injected and were used solely for histological study. These ears were cut into horizontal blocks which were fixed either in 10 % formol-saline or in formol-mercuric-chloride (9 parts of saturated solution of mercuric chloride, 1 part of 40 % formaldehyde). After fixation the blocks were embedded in paraffin and cut at 7 μ, several serial runs being made of each block. In some preparations the serial block method was used (Beesley & Daniel, 1956). Most of the sections were stained alternately with Ehrlich's haematoxylin and eosin, Weigert's elastic method, and iron haematoxylin and van Gieson; some were stained by Masson's trichrome method.

The injection masses were introduced by syringe into one or more of the main intrinsic arteries of the ear (after insertion of cannulae under the binocular dissecting microscope), the cut ends of other arteries having been located and tied off. It was found to be important to warm the ears very thoroughly in hot water before introducing the mass, so that the fat was soft and did not impede the injection.

The casts of the vessels of the ears injected with neoprene were not examined until the tissue had been completely macerated. For maceration the ear was first placed in concentrated hydrochloric acid at 37° C. for 24 hr., after which it was thoroughly washed and transferred to 10 % potassium hydroxide at 37° C. for about 48 hr. to remove the remaining fat. After the cast had been again thoroughly washed it was examined in water under the dissecting microscope.

The ears which had been injected with Berlin blue were not fixed until after the skin and much of the subcutaneous fatty tissue had been stripped (under the dissecting microscope) from both surfaces with a sharp scalpel, leaving the vascular

layer overlying the perichondrium more or less exposed on each surface of the cartilage. The specimens were then placed in 10% formol-saline. After fixation any remaining areolar tissue still covering the layer of blood vessels on the perichondrium was carefully dissected away (again under the dissecting microscope). Small areas of perichondrium, together with the overlying vascular layer, were then stripped from the cartilage, dehydrated, cleared and mounted on a slide in Canada balsam. These preparations gave a good general view of the vessels but were too thick to be satisfactory when stained. Other preparations were therefore made in which small strips of the vascular layer were gently detached from the perichondrium. These were stained with Ehrlich's haematoxylin before being cleared and mounted on a slide. We found that the stripping of the thin layers of fixed tissue was more easily done after the specimen had been immersed in absolute alcohol for a little while. These 'strip' preparations were supplemented by sections cut on the freezing microtome at various thicknesses, but the latter did not in general prove to be so useful, although they had the advantage of showing the arterio-venous anastomoses in the corium as well as those in the perichondrium.

RESULTS

Arterio-venous anastomoses were found to be present in large numbers in the human external ear. They were seen in the specimens taken from the young adults, the aged, and the newborn infant. Many of the arterio-venous anastomoses lie on or in the perichondrium on both aspects of the cartilage. Others are present in the skin, where they lie in the deeper layer of the corium, and others again are present in the subcutaneous fatty tissue. In the lobule of the ear they are also found lying deep within the fatty tissue.

The injected preparations showed that the arterio-venous anastomoses spring from vessels of varying calibre, ranging from extremely small arteries to arteries with an internal diameter of about 80–100 μ. The size of the potential lumen of different arterio-venous anastomoses is very variable, and is related to the size of the artery from which the vessel springs—the larger this artery the larger the potential lumen of the anastomosis. In some of the injected preparations nearly all the arterio-venous anastomoses showed a very small lumen, due to the constricted state of these vessels. In other similar preparations, however, the arterio-venous anastomoses appeared to be widely open, and in these the internal diameter of the majority was about 20 μ, though in many it was in the region of 40 μ, and in some of the larger arterio-venous anastomoses the internal diameter was as much as 50–60 μ. The largest arterio-venous anastomoses lie on the perichondrium, but small ones are also present at this depth.

Individual arterio-venous anastomoses also differ somewhat from one another in shape. A few are rather tortuous, but in the majority marked convolutions are absent. Some make a more or less complete S-bend (Pl. 1, fig. 3); others make only a single bend, sometimes with a wide arc (Pl. 1, fig. 1), but more often with a sharp hairpin turn (Pl. 1, figs. 2, 4, 5). Frequently, however, there is no conspicuous bend in the course of the vessel, which appears as a relatively straight connexion between an artery and a vein (Pl. 1, fig. 4).

The distinction between arteries and veins in thin strips of injected tissue, cleared but unstained (Pl. 1, fig. 1), presented no real difficulty, thanks to differences in the characteristic form and pattern of the arterial and venous systems. No valves were found in the veins of the human ear, although they are present in the veins of the ears of some animals (Daniel & Prichard, 1956). Many of such strips of injected tissue were, however, stained with haematoxylin to demonstrate the cellular features of the walls of the various vessels. The specimens illustrated in Pl. 1, figs. 2–5, were necessarily photographed at relatively low magnifications, but with the higher powers of the microscope a surprising amount of cellular detail can be seen in such material, despite the thickness of the tissue.

The walls of the arterio-venous anastomoses are not nearly so thick as are those of arterio-venous anastomoses which we have seen in some other sites, e.g. the fingers and toes of man, and they contain many fewer layers of cells (cf. Pl. 2, figs. 6, 7). The thickest wall seen in an arterio-venous anastomosis in a human ear measured about 25 μ, but this was unusual, and many of the arterio-venous anastomoses had walls no thicker than about 10 μ. The structure of the vessel wall, however, is essentially the same as that of arterio-venous anastomoses in other regions. The media is composed of variable numbers of epithelioid and smooth muscle cells (Pl. 2, figs. 8–13). The latter may be of the type normally seen in the wall of a small artery, or may be of modified form, having shorter and thicker nuclei. The epithelioid cells have large nuclei, which are round, oval or polyhedral in shape and often pale in colour when stained with basic dyes (Pl. 2, fig. 9). The nuclear membrane is prominent, but there is no well-defined nucleolus. The cytoplasm of these epithelioid cells stains poorly with acid dyes and the cell outline is usually ill defined. The two types of cells are disposed somewhat irregularly in the vessel wall, though the epithelioid cells tend to lie nearer or even adjacent to the lumen and the smooth muscle cells to be situated more peripherally (Pl. 2, fig. 10). Near the arterial end of an arterio-venous anastomosis smooth muscle cells predominate, but further along the vessel epithelioid cells are the more numerous. In the mid-part of the anastomosis the epithelioid cells tend to be packed closely together (Pl. 2, figs. 6, 12), sometimes in irregular groups, but towards the venous end they are often seen forming a single layer lying adjacent to the lumen (Pl. 2, fig. 13). In paraffin sections many of the arterio-venous anastomoses are rather inconspicuous structures because of their relatively small size and particularly their not very thick walls (Pl. 2, figs. 11, 13).

Except for traces of elastic tissue which may be seen near the arterial end of an arterio-venous anastomosis an internal elastic lamina is absent. Towards the venous end a layer of elastic tissue may often be seen around the outer surface of the vessel wall. Endothelial cells are present along the lumen of an arterio-venous anastomosis, but it is doubtful whether they form a continuous lining to the vessel since epithelioid cells are often seen projecting into the lumen. In paraffin sections the lumen itself is frequently extremely small and irregular in shape (Pl. 2, figs. 6 and 8–10).

DISCUSSION

So far as we are aware, the presence of arterio-venous anastomoses in the external ear of man has not hitherto been definitely established. Sucquet (1862) postulated that they were present in this site, but his opinion was based on inference. Neither Berlinerblau (1875) nor Hoyer (1877) could find arterio-venous anastomoses in the human ear. Vastarini-Cresi (1903) was uncertain as to whether they existed or not. Grant & Bland (1931) were unable to find them in the one specimen they examined. The studies reported in this paper have shown that arterio-venous anastomoses are in fact present in the human ear in large numbers, but that in general they are far less conspicuous structures than they are in the ears of some species of animal (Daniel & Prichard, 1956) and in human fingers and toes. No actual counts were made, but there did not appear to be any obvious difference between the young adults and the aged, either in the number of arterio-venous anastomoses present or in their cellular features. Masson (1937), on the other hand, found that the arterio-venous anastomoses in the fingers and toes became less numerous in old age, and Popoff (1934) believed that those present in the digits of old people were atrophic.

The arterio-venous anastomoses in the human ear can be identified in paraffin sections, stained by routine methods, by the epithelioid cells and the modified smooth muscle cells which are present in variable numbers in the vessel wall. An internal elastic lamina is absent in the segment where the epithelioid cells are most numerous. In these respects the arterio-venous anastomoses in the human ear show the same structural features as are seen in arterio-venous anastomoses found in other sites, e.g. the fingers and toes of man (Popoff, 1934; Masson, 1937, and earlier papers), the external ear of the rabbit (Grant, 1930) and of many other animals (Goodall, 1955; Daniel & Prichard, 1956), the tongue of the dog, the sheep and the goat (Prichard & Daniel, 1953, 1954), and the nasal mucosa and tip of the nose of various animals (Dawes & Prichard, 1953). On the other hand, in the arterio-venous anastomoses situated in the human ear, the walls of the vessels are as a rule far less thick than are the walls of arterio-venous anastomoses present in some other sites. For this reason, in histological preparations the arterio-venous anastomoses in the human ear do not so readily attract attention as do those, for example, in the human finger or toe (Pl. 2, fig. 7), in the ears of the sheep, goat and pig (Daniel & Prichard, 1956, figs. 18–22) and in the tongue of the sheep and goat (Prichard & Daniel, 1954, figs. 8–15). It is presumably the relatively inconspicuous nature of the arterio-venous anastomoses in the human ear which has caused them to be overlooked by those who have searched for this type of vessel in this site.

It should be emphasized that many of the arterio-venous anastomoses in the human external ear are difficult to recognize in paraffin sections, partly because their walls are not very thick, but also because the cells in the walls are often less densely packed than they are in arterio-venous anastomoses in other sites. The examination of serial sections, variously stained, was of great assistance in the present work, both for tracing the course of the vessels and for determining the absence of an internal elastic lamina—a distinguishing feature of all arterio-venous anastomoses. In addition, the injected preparations proved a valuable complement to the straightforward histological material, in demonstrating the arterio-venous

anastomoses in a wholly different manner. In this particular investigation the use of the neoprene technique did not yield as useful results as it had done in previous studies (Daniel, Dawes & Prichard 1953; Prichard & Daniel, 1954; Xuereb, Prichard & Daniel, 1954*a*, *b*), and we learnt more about the arterio-venous anastomoses from the preparations injected with Berlin blue, particularly in regard to their relations to other vessels and their morphology.

Some of the arterio-venous anastomoses in the human ear have the tortuous course characteristic of this type of vessel in many other sites, but the majority are not greatly convoluted and some are almost straight. According to Clara (1927), Stolzenburg (1937), and Schumacher (1938), some of the arterio-venous anastomoses in the rabbit's ear are short and relatively straight, and have no epithelioid cells in their walls, but only typical or scarcely modified smooth muscle cells. As far as we have been able to determine, however, all the arterio-venous anastomoses in the human ear, whether relatively straight or convoluted, have at least a few cells in their walls which are of epithelioid type. This was also true of the arterio-venous anastomoses in the ears of many species of animal (Daniel & Prichard, 1956).

Various workers have considered the possible origin of the epithelioid cells in the walls of arterio-venous anastomoses, usually suggesting that they represent either metaplasia of fully developed smooth muscle cells or a special developmental form of primitive smooth muscle cells (for a discussion of this subject see Clara, 1939). More recently Murray & Stout (1942), as a result of tissue culture work with glomus tumours (tumours of arterio-venous anastomoses), have reported that the epithelioid cell can be identified as the pericyte of Zimmermann (1923). We have not been able to find any report of a glomus tumour in the ear, but it is probable that with increasing recognition of these tumours they will be diagnosed in this site as they have been in the fingers and elsewhere.

Whatever may be the origin of the epithelioid cells in arterio-venous anastomoses it seems clear that they must be concerned with the opening and closing of the vessel. How they accomplish this is not yet known, though Schumacher (1938) suggested that they change the size of the lumen by swelling or shrinking. He further suggested that during their shrinkage these cells might liberate a substance resembling acetylcholine. Grant (1930), who by direct visual observation studied the behaviour of arterio-venous anastomoses in the ear of the living rabbit, found that these vessels closed in response to adrenaline and to faradic stimulation of the sympathetic nerves, and that they opened in response to acetylcholine and histamine. Tischendorf (1938) came to a similar conclusion as to the effects of adrenaline and histamine, although he studied the problem by an entirely different method. The abundant nerve supply of arterio-venous anastomoses has been shown by studies of these vessels in the rabbit's ear (Grant, 1930), the digits of man (Masson, 1937), the tongue of the dog (Brown, 1937), the tongue of the sheep (Prichard & Daniel, 1954), and the sheep's ear (Daniel & Prichard, 1956). A histochemical study is being planned to determine whether the nerve endings on arterio-venous anastomoses are adrenergic or cholinergic in character.

It seems probable that an important, if not the most important, function of the arterio-venous anastomoses in the human external ear is to keep the ear warm in a cold environment, by opening up and permitting a greatly increased flow of blood

through the organ. The external ear is an extremity which is particularly liable to be affected by a cold environment because of its shape and structure. Not only is it a thin sheet of tissue but, except in the lobule, about a quarter of its thickness is taken up by cartilage, and there is thus between the two surfaces a layer which, being devoid of blood vessels, provides no warmth whatever. It is perhaps because of this relatively thick avascular layer that many arterio-venous anastomoses are present at a fairly deep level, namely on or in the perichondrium, as well as in the skin.

It is generally believed that the arterio-venous anastomoses which are present in the fingers and toes are a device for maintaining the temperature of these extremities when they are subjected to cold. That these extremities show a peculiar reaction to cold was first reported by Lewis (1930) and Grant & Bland (1931). These workers found that some minutes after the finger, for example, had been placed in crushed ice, and while it was still being cooled, the temperature of this finger ceased to fall and showed a sudden conspicuous rise. The more recent work on cold vasodilatation has lately been reviewed by Burton & Edholm (1955). Grant & Bland (1931) believed that the sudden rise of temperature during cooling was due to an increased blood flow through the part, which was brought about mainly by the opening up of the arterio-venous anastomoses in these extremities. Both Lewis (1930) and Grant & Bland (1931) noted a similar reaction to cooling in the human external ear but, failing to find arterio-venous anastomoses in this site, Grant & Bland attributed the response here to another mechanism. The present work makes it clear that the reaction to cooling shown by the ear can in fact also be attributed to the opening up of arterio-venous anastomoses. The theory that arterio-venous anastomoses play a major part in the phenomenon of cold vasodilatation gains support from Grant's (1930) visual observations on the arterio-venous anastomoses in the ear of the living rabbit, for he actually saw these vessels opening up when the ear had been cooled for a while. This work was confirmed and extended by Grant, Bland & Camp (1932). However, the arterio-venous anastomoses in the rabbit's ear also open up when the body of the animal is heated (Grant, 1930; Clark & Clark, 1934), which suggests that in this animal the arterio-venous anastomoses in the ear also serve as a means of lowering body temperature. For various reasons, including the small size of the ear in proportion to the size of the body, is seems unlikely that the arterio-venous anastomoses in the human ear play more than a minor part in lowering the body temperature.

In the rabbit's ear the arterio-venous anastomoses were seen by Grant (1930) and Clark & Clark (1934) to show spontaneous rhythmic changes in calibre. It seems likely that it is some such phasic activity of the arterio-venous anastomoses which gives rise to the 'hunting' phenomenon seen in cold vasodilatation, a phenomenon which was observed by Lewis (1930) in man in various peripheral sites, including the ear, and which has been studied more recently by many workers (see Burton & Edholm, 1955).

All the available evidence, direct or indirect, points to the fact that arterio-venous anastomoses are highly active blood vessels, which are capable both of complete closure and of wide dilatation, and that according to circumstances they fluctuate in calibre between these two extremes. Although the purposes which they serve and the mechanism by which they are controlled are not yet fully known there can be

no doubt that arterio-venous anastomoses are vessels of profound significance. In sites such as the human ears and digits their numbers are so great and their potential calibres so large that the amount of blood flowing through the part must be enormously different according to whether the arterio-venous anastomoses are open or closed. Consequently, when observations are made on the circulation through territories where arterio-venous anastomoses are known to be present it is essential to consider the contribution made by the activity of these vessels.

SUMMARY

Arterio-venous anastomoses are present in large numbers in the human external ear, both in and beneath the skin and on and in the perichondrium. They are not so tortuous as those present in the fingers and toes of man, and their walls are not so thick, containing fewer layers of cells. The actual cells, however, in the vessel wall are of a similar kind, consisting of epithelioid cells and smooth muscle cells, mainly of modified type, which are present in variable numbers and are irregularly disposed. An internal elastic lamina is absent.

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EXPLANATION OF PLATES

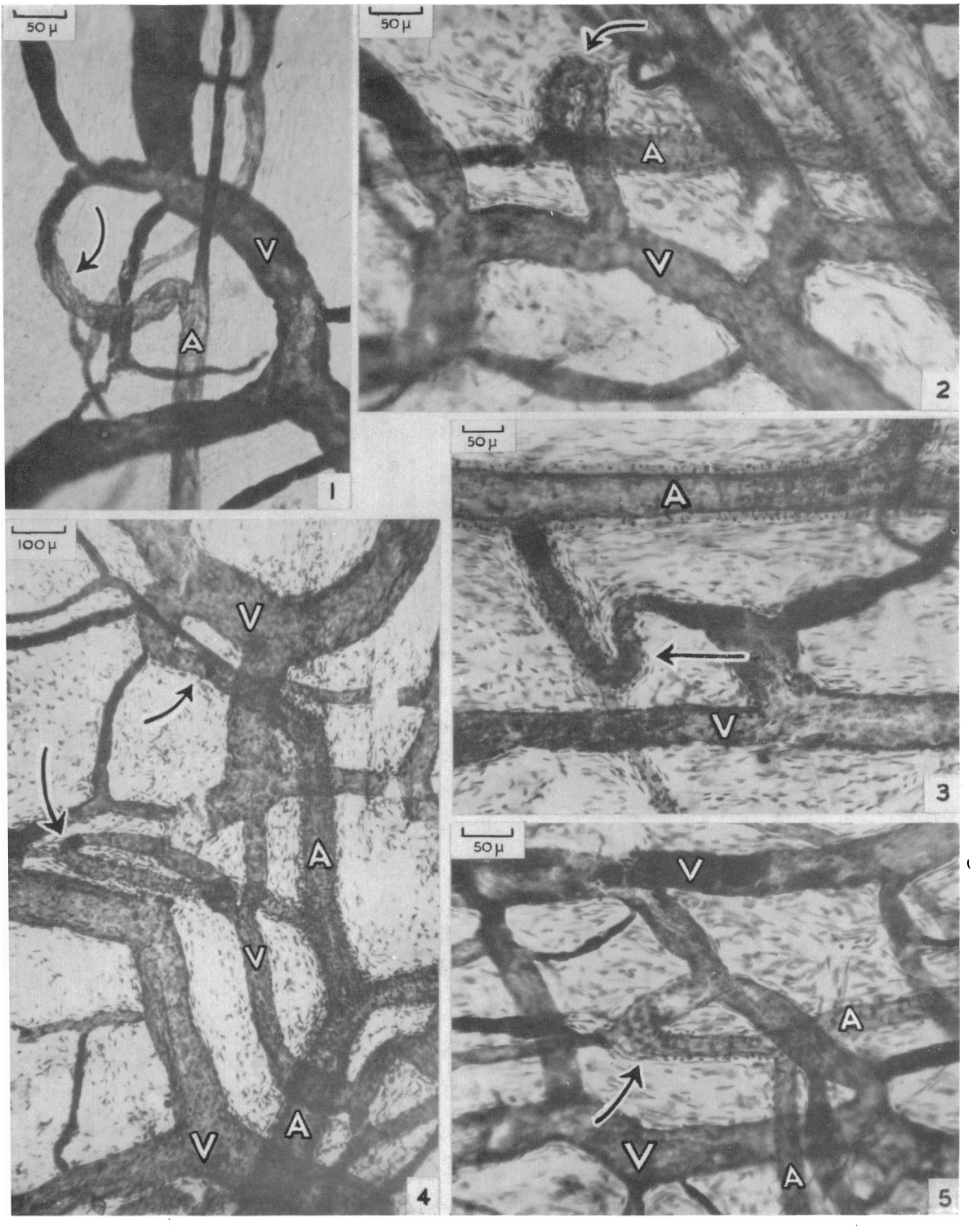
PLATE 1

Figs. 1–5. Preparations of the human external ear injected with Berlin blue. These thin pieces of tissue, stripped from the cartilage, show arterio-venous anastomoses (at arrows) in the vascular layer overlying the perichondrium. Note the large size of these channels which form direct connexions between arteries (*A*) and veins (*V*), and their relatively simple form. The examples illustrated show the typical shapes of the majority of the arterio-venous anastomoses found in the human ear, although more convoluted ones are also present. Note in figs. 1, 2 and 5 a single bend, in fig. 3 an S-bend, and in fig. 4 a long hair-pin bend (lower left arrow) and an almost straight vessel (upper arrow). The walls of these arterio-venous anastomoses are much less thick and cellular than are those of the arterio-venous anastomoses present in some other sites, e.g. in the human digits (see Pl. 2), and in the ears of some animals (Daniel & Prichard, 1956). Fig. 1 shows an injected preparation cleared and mounted unstained. Figs. 2–5 show similar preparations stained with haematoxylin before being cleared.

PLATE 2

Fig. 6. Arterio-venous anastomosis in the corium of a human external ear, cut transversely (left), with its afferent artery approaching it from the right. Note the thick wall of the arterio-venous anastomosis, containing many epithelioid cells packed closely together, and the very small, irregular lumen. This is an example of one of the more conspicuous arterio-venous anastomoses found in the human ear, but even so it is not so prominent a structure as are the arterio-venous anastomoses present in the fingers and toes of man (see fig. 7). Iron haematoxylin and Van Gieson.

Fig. 7. Arterio-venous anastomosis in the pad of a human toe, cut transversely. Compare this figure with fig. 6 and the other illustrations on this Plate, and note the much thicker wall of this arterio-venous anastomosis and its more numerous layers of cells. The actual cells, however, which consist of many epithelioid and some modified smooth muscle cells, are similar to those which form the walls of arterio-venous anastomoses in the human ear. Haematoxylin and eosin.



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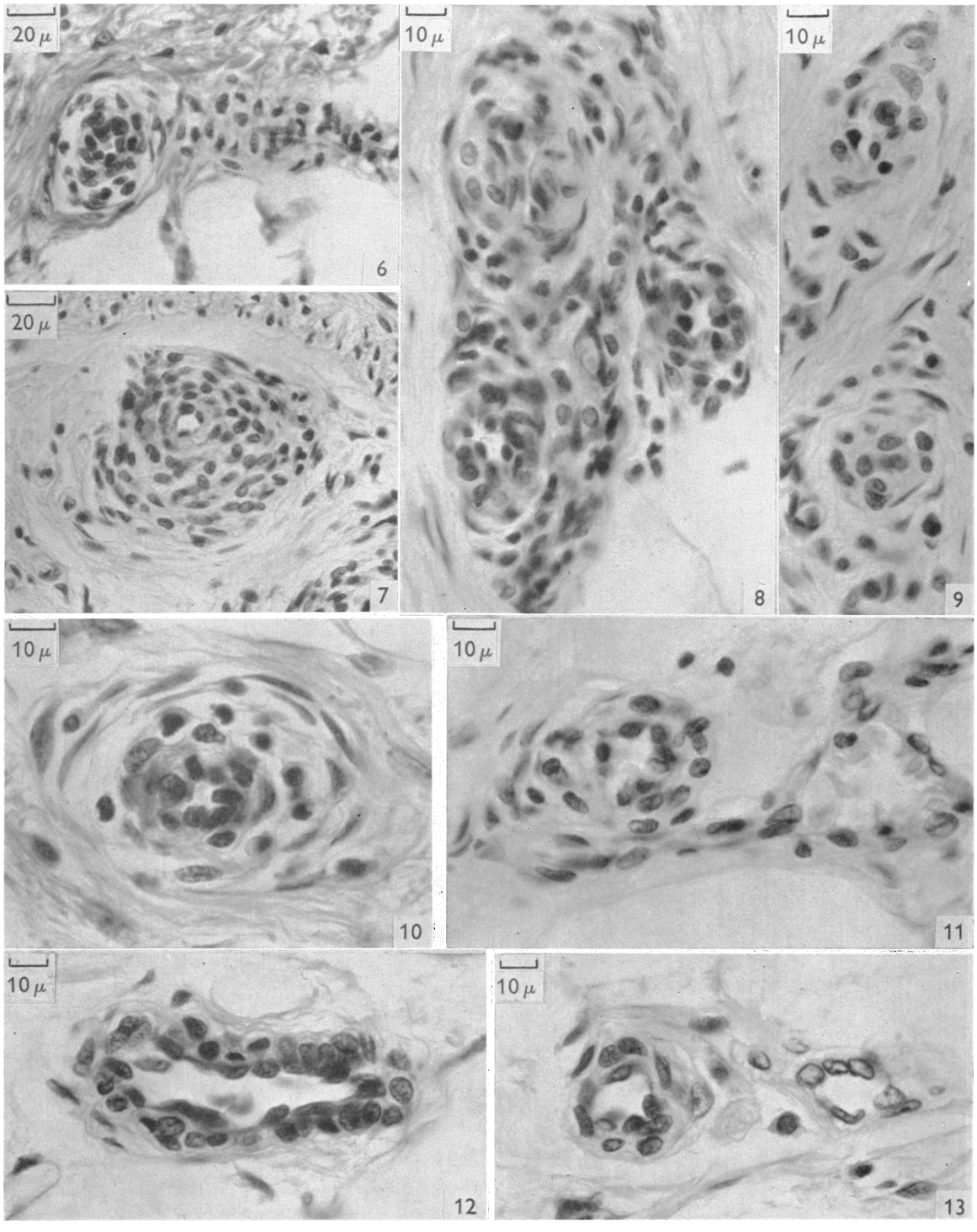

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Fig. 8. Three segments of a tortuous arterio-venous anastomosis lying within the perichondrium of the human external ear. Note the great cellularity of the wall of this relatively complex vessel. Haematoxylin and eosin.

Fig. 9. Two arterio-venous anastomoses in the corium of the human external ear. In the lower of these two vessels the pallid nuclei of several epithelioid cells are seen; its lumen is closed. Haematoxylin and eosin.

Fig. 10. Arterio-venous anastomosis in the corium of a human external ear cut transversely. Epithelioid and modified smooth muscle cells are present in the wall of this vessel. The lumen is small and cells project into it. Haematoxylin and eosin.

Fig. 11. Arterio-venous anastomosis (left) and small vein (right) lying on the perichondrium of a human external ear. This example is typical of many arterio-venous anastomoses in the human ear, which have only a relatively small number of epithelioid cells in their walls and are consequently not very conspicuous. Haematoxylin and eosin.

Fig. 12. Arterio-venous anastomosis, situated in the centre of the fatty tissue of the lobule of a human external ear, cut mainly longitudinally. Note the many cells, mainly epithelioid, packed closely together in the not very thick vessel wall. Haematoxylin and eosin.

Fig. 13. Two segments of an arterio-venous anastomosis lying just beneath the corium of a human external ear. The segment on the left is typical of the mid-portion of a small arterio-venous anastomosis in the human ear, and shows epithelioid and some modified smooth muscle cells irregularly disposed in the vessel wall. On the right the anastomosis is approaching its venous end and shows epithelioid cells, with very pale nuclei and prominent nuclear membranes, bordering the lumen. Haematoxylin and eosin.