The Atrio-ventricular System of the Equine Heart.

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Although the chief contributions to the study and explanation of the atrio-ventricular system were made by Tawara in 1906 and by Keith and Flack in 1907, several portions of the system had been described many years earlier. Thus, Purkinje in the middle of the 19th century discovered the fibres which posterity has since named after him, while, practically simultaneously in 1893, His and Stanley Kent, quite independent of one another, drew attention to the structure which is still sometimes called the Bundle of His or, preferably, the A.V. Bundle.

Up to the publication of the works of His and Kent, it was believed that the musculature of the atria and ventricle was quite distinct and separate and that there was no sort of connection between the two. It is now known that the Bundle of His, generally called at the present time the atrio-ventricular bundle (or briefly the A.V. bundle) is a band of special kind of muscle which conducts the cardial impulse to contraction from the atrio-ventricular node to the ventricular musculature.

Keith has in a series of papers described the development of the mammalian heart and discussed the evolutionary significance of the various parts of the organ. His conclusions, now generally accepted, are that the A.V. system is derived from a structure which forms in lower vertebrates, where the heart is more primitive than in mammalia, a muscular continuity throughout the organ from the venous to the arterial end.

While the A.V. system exists in the heart of all mammals that have been examined it is not easy to demonstrate in most species. Thus owing to the naked eye prominence of the Purkinje fibres in the domestic ruminants, the entire system has been thoroughly examined in these animals. In most mammals, however, including horse and man, it requires histological examination to reveal some parts of the system, e.g. Purkinje fibres or the various nodes.

As is well known the A.V. system consists of (a) sino-auricular node or the node of Keith and Flack, (b) atrio-ventricular node or the node of Tawara (or 'Tawara-Aschoff), (c) the bundle of His or the A.V. bundle—the main stem—and its two chief septal divisions, which branch to the right and left ventricles respectively, and, finally, (d) the fibres of Purkinje which are terminal in distribution and connect the special fibres of the A.V. system with the ventricular musculature.

Kent has described other connections between the right atrium and the right ventricle, but as the significance of these structures is unknown and their functions even doubted under normal conditions they will not be further noticed in this paper. In connection with the presence of the A.V. system in the avian heart "there is a bundle of His or other specialised form of muscle tissue connecting the auricles with the ventricles," and "the sino-auricular node is also absent" (Ivy Mackenzie and Jane Robertson). Keith and Flack, however, had no doubt that the avian heart possessed an A.V. bundle, which others according to Morrison, failed to confirm. Recently this observation of Keith and Flack has received the support of Drennan, who dissected five ostrich hearts and in all was able to demonstrate a definite structure, which corresponds in origin with the homologous structure in the bovine heart. Apparently, however, he could not find any A.V. node. Although there is no branch which answers to the right septal division of the A.V. bundle of mammals yet the distribution of the left branch is very similar.

Drennan concludes from his observation "that there is a system of fibres in the ostrich heart somewhat similar to the A.V. bundle of the mammalian heart. The distribution also suggests a similar function, in short that they constitute a true avian auriculo-ventricular bundle."

THE SINO-AURICULAR NODE. THE NODE OF KEITH AND FLACK.

This was first described by Keith and Flack in 1907.

Where the cranial vena cava empties into the right atrium there is, on the external surface of the heart, a groove of varying length and depth—the sulcus terminalis—best seen and deepest at the cranial border of the organ. It corresponds to the crista terminalis on the inner aspect of the atrium on which the auricular columnae carneae end. These two structures indicate the junction of the primitive sinus of the embryo with the atrium. In the sulcus terminalis lies the sino-auricular node, which, in the horse, is not clearly defined.

Subepicardially, especially on the left face of the atrium and on the right face ventral to the sulcus terminalis there may be seen several nerves, derived from the cardiac nerves, which run into the sulcus where they become intimately fused with the sino-auricular node.

As already noticed this node is an ill defined structure in the equine heart. Removal of the epicardium and careful dissection in the sulcus will show a whitish body, irregular and vague in shape, measuring about 3 cm. long by .5 cm. broad. Its long axis is parallel with that of the sulcus, and it contains a considerable quantity of connective tissue.

In spite of long and repeated research, no special apparatus has ever been found in any mammal between the sino-auricular and the atrio-ventricular nodes. The bridge described as existing by Thorel and others has not been confirmed.

Lewis has definitely shown that the normal heart beat arises in this node and spreading in all directions through the ordinary atrial musculature eventually arrives at the node of Tawara.

A large well defined artery, derived from the Arteria Coronaria Dextra, occupies a constant position in the lower portion of the node. Its course may be traced for some distance and may be shown up by suitable injection. As already noticed nerves freely communicate with the node.

THE ATRIO-VENTRICULAR NODE AND THE BUNDLE OF HIS.

The A.V. node was first described by Tawara in 1906, but later Aschoff distinguished two parts of it so that the designations Tawara-Aschoff or simply Tawara are frequently used as synonyms.

In the horse this node is not easily dissected out and thorough examination of the same structure in the heart of an ox or sheep is recommended before attempting to isolate it in the horse.

If the right atrial wall to a depth of about 3 mm. is removed below and to the right of the ostium of the sinus coronarius as far up as the foramen ovale a plexiform confused mass of pale grey fibres will be revealed, wider posteriorly than in front, and generally irregular in shape, though definite descriptions have been given of its form, e.g. deltoid, oval or spindle, etc. It measured, in 20 cases, between .6 cm. - .8 cm. long by .4 - .7 cm. wide. It is smaller than the corresponding structure in the bovine heart.

It is enveloped in a fibro-fatty sheath and is provided with a well marked artery. At its origin it is more open and difficult to follow as it is formed by the convergence of pale and delicate fibres running in from all parts of the atrial wall.

These fibres may be traced back to the atrial musculature but are soon lost. Very many of them arrive from the interatrial septum near the foramen ovale and from about the sinus coronarius. Anteriorly the node becomes greatly confused with the fibrous tissue of the A.V. ring and even with the cartilago cordis.

In this part of its course macroscopic sections must be made to demonstrate the relations of cartilage, connective tissue, and bundle. At the cartilago cordis the node is continued without any sharp demarcation in the equine heart by the main stem of the bundle of His, which in the horse, is very short, has a swollen appearance and is generally less than 1 cm. long and about .5 cm. broad.

Keith has emphasised the relation of the bundle of His to the central fibrous body of the human heart and has shown that this body often becomes diseased under certain conditions when the bundle also becomes affected.

The relation between the two structures may be so intimate that the bundle may be said to run through the groove in the fibrous body, the groove being converted into a canal by the attachment of the septal cusp of the tricuspid valve.

In the dissections of this region in the equine heart the relation of the septal cusp of the tricuspid valve, the fibrous ring of the atrioventricular orifice and the cartilago cordis is very apparent, and it requires very great care to demonstrate intact the main stem of the bundle. A section of this region showing the three structures described above is given at the end of this paper.

The bundle now descends into the septum membranaceum of the interventricular septum and bifurcates into its two main terminals, i.e. right and left septal limbs or divisions. During this short course it detaches numerous pale filaments which enter the myocardium and become quickly lost to macroscopic examination.

The septum membranaceum is a difficult structure to demonstrate in the equine heart. It may be recognised by holding up the intact interventricular septum to the light when it appears as a feebly translucent area, roughly triangular in shape, in the region below the aortic valves. This part of the interventricular septum separates the left ventricle from the right atrium and right ventricle.

The main bundle and its two terminal bifurcations are enveloped in a strong connective tissue sheath which may be injected and shown up by Berlin-blue method (Aagaard and Hall). Numerous ganglia and nerves are generally found in connection with the node of Tawara and the bundle, especially at its division.

It is not intended here to enter into the controversy as to whether the heart beat is myogenic or nervous in origin, but many anatomists have emphasised the rich nerve supply and ganglionisation of the A.V. system. Retzer and De Witt both regard the bundle as a neuromuscular end organ.

Embryologically the system appears early. Keith found it differentiated and well formed in the human embryo of 45 mm. (11 weeks), while Zimmerman has demonstrated Purkinje fibres in a 12 cm. horse embryo.

To demonstrate this part of the bundle it is essential to entirely remove the septal cusp of the tricuspid valve when a small irregular shaped cartilaginous nodule—the cartilago cordis—will be exposed. Often in old cases the cartilage has become enlarged and ossified and this will make the dissection more difficult than usual.

At this stage either the left or right septal division should be carefully traced upwards in the myocardium to the septum membranaceum and thus reveal the main bundle of His, which may be laid bare by piecemeal dissection of the dense fibrous or fibro-cartilaginous tissue of the atrio-ventricular ring.

The Right Septal Division is long and narrow. In 20 cases it averaged 5.90 cm. long and 1.2 mm. wide. It has a rounded cord-like appearance, is enveloped in a connective tissue sheath, and is paler than the surrounding heart muscle. During its downwards course it gives off several collateral branches which enter the myocardium and are soon lost to view.

This division, branch or limb, lies just under the endocardium, being only covered by a superficial stratum of heart muscle about 1.5 mm. thick. Its course is not straight but somewhat flexous. Soon after its origin from the main bundle it descends sharply forming a knee-shaped bend (called by Keith "the genu.") It maintains the downward and slightly cranial direction for about 2 cm. It then inclines more cranially for another 2 cm. and finally becoming nearly vertical is continued into the moderator band of the ventricle. Near its termination it gives off several collaterals which enter the substance of the septum and ramify subendocardially.

The best way to expose this division is to follow the moderator band found at the entrance to the conus arteriosus into the septum and with a little practice its course will be rapidly dissected out.

The Left Septal Division is 3.60 cm. long and averages 1.5 mm. wide. It is thus broader than the right division but it is more indistinct, being more open in texture and less clearly marked off from the heart muscle. Leaving the main stem in the septum

membranaceum it inclines steadily downwards and slightly cranially and passes through the thickness of the subaortic musculature (which may be as much as 3.50 cm. thick) to become subendocardial in The exact location of this part of the bundle lies just below a cranial-caudal plane, passing through the apices of the tricuspid valves, about 5-6 cm. below the point of contact between the cranial and caudal medial aortic semilunar valves. The division terminates by dividing into cranial and caudal fasciculi, which, after a brief subendocardial course, pass mainly into the two largest moderator bands. Numerous collateral filaments may be traced into the neighbouring myocardium as they arise from this division, from its bifurcation and from terminal fasciculi. The cranial and caudal moderator bands are 5-5.25 cm. and 3.5-4.5 cm. long respectively. They terminate in the large compound lateral and caudal papillary muscles, in which, however, they cannot be easily dissected as they break up into delicate microscopic network. caudal band gives off from 2-6 secondary branches which may fuse inter se and then plunges into the ventricular septum. while the cranial fasciculus detaches at least one collateral. These small branches ramify and anastomose subendocardially as Purkinje's fibres, which will be noticed later. Towards the apex of the left ventricle a number of small thread-like filaments (about 1 cm. and less in length) may be seen passing from one columna carnea to another. These are structurally moderator bands, and their histology is quite typical as will be seen later.

THE TERMINAL DISTRIBUTION OF THE A.V. SYSTEM.

As already mentioned in the introduction this part of the A.V. system has long been known, though it was only in 1906 that Tawara realised the significance of the apparatus still generally called the fibres of Purkinje, as being the terminal distribution of the A.V. system and the connection between it and the ordinary ventricular muscle fibres. Tawara insisted that the A.V. system was a conducting and not contracting system and he made his observation on the heart of the ruminant where the terminal apparatus is best developed.

In the equine heart, as in many mammals, the fibres of Purkinje are not visible to the naked eye, but they can be demonstrated both histologically and by means of a simple chemical test.

If the endocardium of both ventricles of the equine is removed without any tearing of the myocardium and the exposed surfaces are flooded with Lugol's solution of iodine there will be a rapid appearance of a mahogony brown plexus which differs in macroscopic features in the two ventricles.

In the left ventricle the plexus will present large wide meshes and long comparatively narrow fibres which freely anastomose with adjacent fibres and produce a typical open network. The phenomenon is best marked on the septum between the diverging cranial and caudal fasciculi where the secondary branches of the two cranial moderator bands ramify. It will be also observed, but less prominently, in the subaortic region and on the lateral wall of the cavity. It is very well marked at the apex of the ventricle, over the numerous small columnae carneae there.

In the right ventricle and conus arteriosus the same reaction to the iodine is noted, but the appearance of the subendocardial plexus is different. The meshes are here much narrower and smaller with the result that the fibres are short and thick. The mahogony colour is more diffuse and more difficult to discern than in the left ventricle where the spaces of the large meshes make it plainer.

The explanation of this phenomenon is that the Purkinje fibres are richer in glycogen than the surrounding heart muscle and thus stain characteristically brown with Lugol's iodine. By this very simple test the terminal apparatus of the A.V. system may be demonstrated in the heart of the horse.

According to Zimmerman this test is much more effective in the equine heart than in any other, and its efficacy is more enhanced if the endocardium is removed before applying the iodine. Similar results, however, can be obtained when the endocardium is left intact though the time for the typical picture to appear is considerably longer.

The A.V. bundle and Purkinje's fibres have been reconstructed in the calf's heart by Miss De Witt. The task would be more formidable in the equine heart where the bundle would require artificial or histological aid to reveal its ramifications.

Comparison between the Purkinje system of the equine heart. as revealed by the iodine test, and the model of the same apparatus in the bovine heart, as worked out by De Witt, would show a remarkable similarity in arrangement and distribution in both species. It has often been remarked how analogous the entire system is not only in animals of the same or allied species but in mammalia in general.

THE HISTOLOGY OF THE A.V. SYSTEM.

Sections may be preserved in the 10 per cent. formalin in the usual way or in Zenkers fluid. They are preferably stained by Van Giesen or any other preparation which shows up the connective tissue particularly well, e.g. Mallery's connective tissue stain. As has already been pointed out on several occasions in this paper the A.V. bundle and the nodes stand out prominently macroscopically because of the abundant pale coloured connective tissue which envelopes them. In Van Giesen stained sections the vivid red of the connective tissue offers a very marked contrast between the specialised fibres of the A.V. system and the ordinary myocardium.

In the Keith-Flack node the fibres are narrow, average 10 μ wide, and are multinucleated. They stain pale olive green in Van Giesen preparations, and are very rich in sarcoplasm. The fibres run in all directions and are held together by dense white fibrous tissue. The deeper the fibres run into the usual atrial musculature, the more regular becomes their course until they are finally arrayed in parallel bundles. Histologically no special apparatus can be demonstrated connecting this node with the A.V. node.

Numerous nerves, ganglia and blood vessels may easily be found.

Sections of the node of Tawara show the plexiform arrangement of the fibres which run together forming a characteristic network with meshes of different sizes. There is the usual strong connective tissue envelope in which there may be much fat.

The fibres of the node are narrow and rich in sarcoplasm. Transverse striation is generally observable but may not be noticed in some fibres, in all of which, however, is a lighter central area occupied by the nucleus.

The A.V. bundle itself and its main septal divisions are readily recognised microscopically as they are at once revealed by the brilliant red colour of their connective tissue envelope. The fibres, however, are now running in a parallel direction, and have a fibrofatty sheath of their own. This is particularly well seen in the left septal division as it runs down through the subaortic musculature. The two septal limbs are composed of a number of bundles of fibres which may be between 15-40 μ in width. As in other parts of this system numerous blood vessels and nerves may be seen and also, especially after the use of orcein or Weigert's stain, much elastic tissile

Purkinje's fibres are large, pale, coarse, poorly branched and peripherally striated structures, which may be easily isolated from pieces of the subendocardial heart muscle in the ruminant after maceration in equal parts of glycerine, 95 per cent. alcohol, and distilled water.

In the horse it is much more difficult to obtain these fibres after maceration, but they are readily spotted in ordinary sections of heart muscle.

On transverse section these fibres are wider than the ordinary heart muscle fibre, they contain more sarcoplasm but fewer peripheral myofibrillae.

In the equine heart they are probably most easily demonstrable on sectioning the various columnae carneae, and staining by Van Giesen. Their appearance is then quite characteristic.

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Although some of the domestic animals are regarded as being ideal for the Although some of the domestic animals are regarded as being ideal for the study of the A.V. system, most of the present-day standard textbooks in veterinary anatomy and physiology practically ignore the structure altogether, so that the investigator is forced to rely largely on the original works of Tawara and others, which are difficult or quite impossible to obtain in South Africa, or on descriptions given in more or less detail in the well-known physiology textbooks by Bayliss, Starling, Howell, and others.

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MacCallum.

PLATES.

PLATE No. 1.—Left ventricle of equine heart with lateral wall removed and exposing septum. Endocard not removed.

(a) Bifurcation of the left division of the A.V. bundle.
(b) The cranial fasciculus.
(c) The caudal fasciculus, c' = caudal moderator band.
(d) The caudal papillary muscle.
(e) Secondary moderator band.

PLATE No. 2.—Right ventricle showing course of right division of A.V. bundle.

(a) Main stem of the A.V. bundle.

(b) Right septal division.(c) Moderator band.

(d) Cartilago cordis.(e) Ostium of coronary sinus.

For explanation of asterisk see Plate No. 4.

PLATE No. 3.—Left ventricle with wall and atrium removed.

(a) Main left septal division.

(b) Bifurcation of left septal division.

(c) Cranial fasciculus.

(d) Caudal fasciculus.(e) Caudal moderator band.

(f) Secondary branches of caudal fasciculus entering septum.

PLATE No. 4.—Section from region marked with an asterisk on Plate No. 2. $\times 50.$

It shows portion of the cartilago cordis in top left hand corner. The rest of the field is mainly occupied by a dense mass of connective tissue with lighter stained isles. These are characteristic fibres belonging to the A.V. system.

PLATE No. 5.—Shows the left septal division running through the subaortic musculature. $\times 50$.

(a) Connective tissue envelope of the division.

(b) Individual fibres of the division separated by a small amount of connective tissue.

(c) Ordinary heart muscle fibres.

PLATE No. 6.—Section (longitudinal) through a moderator band of the left ventricle. ×55.

(a) Connective tissue envelope.

(b) Purkinje's fibres.

The thicker layer of connective tissue will be noticed on the concave side of the band in contrast to the subendocardial position of some of Purkinje's fibres on the convex side.

PLATE No. 7.—Section (longitudinal) over the site of the node of Tawara.

(a) Fibres of the A.V. system with well marked connective tissue envelopes.

- (b) Connective tissue.
- (c) Atrial muscle fibres.
 (d) Adipose tissue.