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The T-tubule system in the specialized and general myocardium of the rat

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INTRODUCTION

Transversely oriented tubules invaginated from the sarcolemma were described in myocardial cells as early as 1957 by Lindner, who observed them in the ventricular musculature of the dog. However, the possible significance of these invaginations was not appreciated until similar tubules, which came to be known as T-tubules, were seen in skeletal muscle cells and were implicated in excitation–contraction coupling (for recent reviews see Endo, 1977; Fabiato, 1977). Since then the extent and nature of the T-system in mammalian myocardial cells has been studied by many investigators including Simpson & Oertalis (1961, 1962), Nelson & Benson (1963) Simpson (1965). Page (1967*a*, 1968), Rayns, Simpson & Bertaud (1968), Fawcett & McNutt (1969), Hibbs & Ferrans (1969), Forssmann & Girardier (1970), Nayler & Merrillees (1971) and Forbes & Sperelakis (1977).

It is now generally recognized that, in typical ventricular myocardial cells, tubular invaginations of the sarcolemma recur more or less periodically at or near the Z bands, where they are frequently coupled with elements of the sarcoplasmic reticulum (SR). Such primary T-tubules are usually substantial in size, often attaining a diameter of over 100 nm, and they characteristically possess a luminal coating of basement membrane. Early investigators had difficulty in determining the full extent of the T-system and they regarded all longitudinally oriented tubules as part of the SR because neither a connexion to the sarcolemma nor a coating of basement membrane could be discerned. In recent years, however, infiltration techniques using tracers such as ferritin, lanthanum salts or horseradish peroxidase have demonstrated that the T-system comprises more than transverse invaginations at Z band level. For instance, Forssmann & Girardier (1970), who studied the rat myocardium after peroxidase labelling, reported that several branches arise from the primary invaginations and course in different planes. These branches are generally narrower than parent tubules, but the presence of peroxidase enables them to be assigned as part of the T-system, rather than the SR which remains unlabelled. Some branches ramify transversely at Z line and I band levels, in planes different from that of the parent invagination. Other branches course along the long axis of the myofibrillae, forming an extensive network which intertwines with the SR and surrounds the A band region. Forssmann & Girardier drew attention to the presence of membrane coupling between the T-system and SR, not only in relation to the primary T-tubules, but also in association with the longitudinal elements. In addition, it has long been known that, unlike skeletal muscle, cardiac muscle exhibits extensive coupling between the surface sarcolemma and superficial SR cisterns (Page, 1968), and these are frequently seen near intercalated discs.

When the various reports in the literature are compared, it appears that the degree of differentiation of the T-system may depend on the region of the heart. For instance, in the rat Forssmann & Girardier (1970) found a T-system only in a small proportion of atrial cells. Moreover, there is much support for the view that T-tubules are poorly developed or absent in the specialized nodal and conducting systems. Sommer & Johnson (1968*a*, *b*) could find no T-tubules in the Purkinje cells of the mouse heart, nor could Hayashi (1971) find them in the conducting system of the dog, Kim & Baba (1971) in the guinea-pig, Viragh & Porte (1973) in the monkey or Mochet, Moravec, Guillemot & Hatt (1975) in the rat. Page (1967*b*) reported the presence of T-tubules in Purkinje cells of the cat heart, but they comprised primary invaginations only, and a similarly restricted system was reported by Challice (1965) in the rabbit sinus node, by Colborn & Carsey (1972) in the node of the squirrel monkey, James & Sherf (1968) in the human atrioventricular node, and by James, Sherf & Urthaler (1974) in the human bundle branches. The majority of the above-mentioned investigators, apart from Sommer & Johnson (1968*b*), did not employ tracer studies, and it is possible that the full extent of the T-system was not demonstrated.

In view of the likely significance of the T-system in excitation–contraction coupling it was thought desirable to make a systematic study, employing a tracer labelling technique, of its distribution and character in different parts of the heart, including general atrial and ventricular myocardium and representative areas of the specialized nodal and conducting systems.

MATERIAL AND METHODS

Ten adult rats of 200–300 g body weight were used for horseradish peroxidase infiltration studies. 0·3 mg/g body weight horseradish peroxidase (Sigma II) dissolved in 0·5 ml isotonic saline was injected intravenously through a tail vein or femoral vein under ether anaesthesia. The rats were killed with open ether 30–45 minutes later and immediately perfused through the left ventricle with 2 % glutaraldehyde in 0·075 M sodium chloride at pH 7·2 for 5–10 minutes, followed by perfusion with Karnovsky fixative in 0·1 cacodylate buffer (pH 7·2) for 10 minutes. During the course of the perfusion, the distended right atrium was cut in order to release the fluid. Excess fixative was washed out by perfusing with 0·1 M cacodylate buffer for 5 minutes.

With the heart still *in situ*, the aorta was clamped just above the coronary ostia. The heart wall was then perfused through the coronary arteries with a solution containing 0·1 % 3,3′-diaminobenzidine in 0·05 M tris buffer at pH 7·5 for about 30 minutes (perfusion with the diaminobenzidine solution was preferred to incubation because it improved the degree of penetration). A 1 % solution of hydrogen peroxide prepared from a 30 % stock solution was added to the perfusate just before it was

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Fig. 1. Ventricular myocardial cell showing two wide primary T-tubules (*T*) invaginated from the sarcolemma. × 28000.

Fig. 2. Ventricular myocardial cell showing primary T-tubules in cross section at I band level near the Z line. Note coupling with SR profile which is looping over a T-tubule and linking with SR in the next sarcomere. × 60000.

Fig. 3. Ventricular myocardial cell after peroxidase infiltration. High magnification view of a SR sac coupled with a T-tubule of the tertiary type (*T*) Note the densely stained bridges in the gap between the membranes. × 110000.


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T
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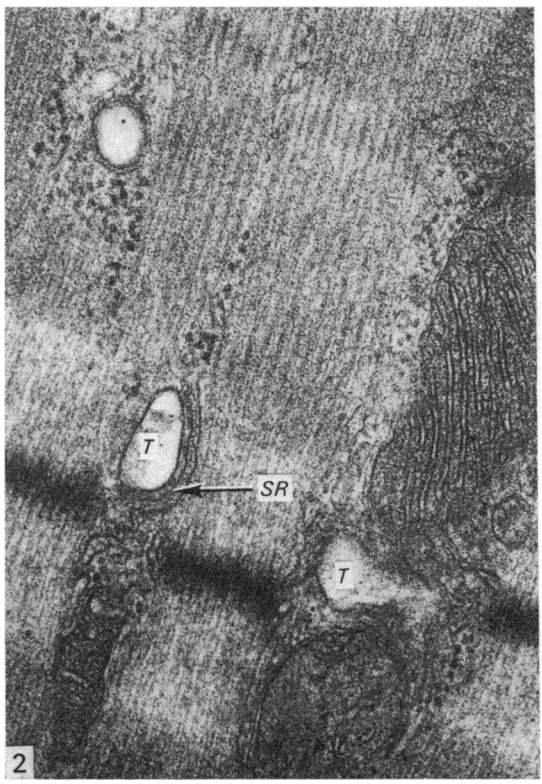
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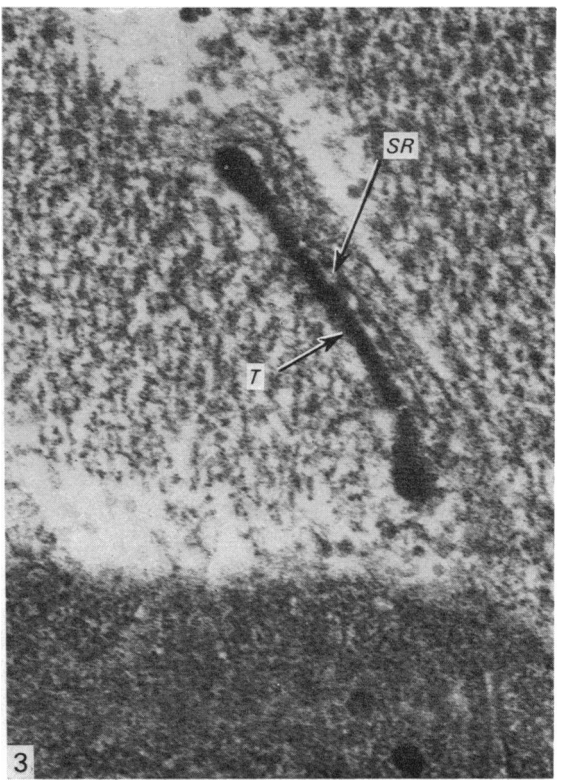
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allowed to run to produce a final concentration of 0·01 %. After perfusion the heart was rapidly removed and the relevant regions were dissected under a dissecting microscope in a bath containing a similar diaminobenzidine solution. Two samples each of atrial myocardium, right ventricle, and one sample from sinus node, atrioventricular node, bundle, and each bundle branch, were obtained from each heart.

The material was post-fixed for 2½ hours in 1 % osmium tetroxide in 0·1 M cacodylate buffer after three quick washes in buffer. This was followed by brief washing in distilled water and dehydration through the alcohol series. The specimens were embedded in four stages, beginning with a 50:50 solution of absolute alcohol and Spurr resin left overnight at room temperature; then in a mixture of 25:75 absolute alcohol and Spurr for 1 hour then in absolute Spurr at room temperature for 2 hours, and finally in fresh Spurr cured at 60 °C for 16 hours.

Thin sections (approximately 60–90 nm) were cut on a Huxley ultramicrotome and stained in uranyl acetate for 15 minutes followed by lead citrate for 10 minutes. Stained sections were examined in a Siemen's Elmiskop 1 and in a Philips EM 300.

Material not infiltrated with peroxidase from corresponding regions of the heart from a large series (over 30) of animals was prepared for electron microscopy.

OBSERVATIONS

General ventricular myocardium

In typical ventricular myocardial cells it is possible to discern several components in the T system, namely: (*a*) primary transverse tubules, (*b*) secondary transverse tubules, (*c*) longitudinal tubules and (*d*) tertiary transverse tubules.

(*a*) Primary transverse tubules are direct invaginations of the sarcolemma (Fig. 1). They are oriented at right angles to the long axis of myofibrillae and are usually located at I band level. These tubules have a mean diameter of about 130 nm and, in material prepared for electron microscopy without peroxidase infiltration, a luminal coating of basement membrane can often be discerned in them.

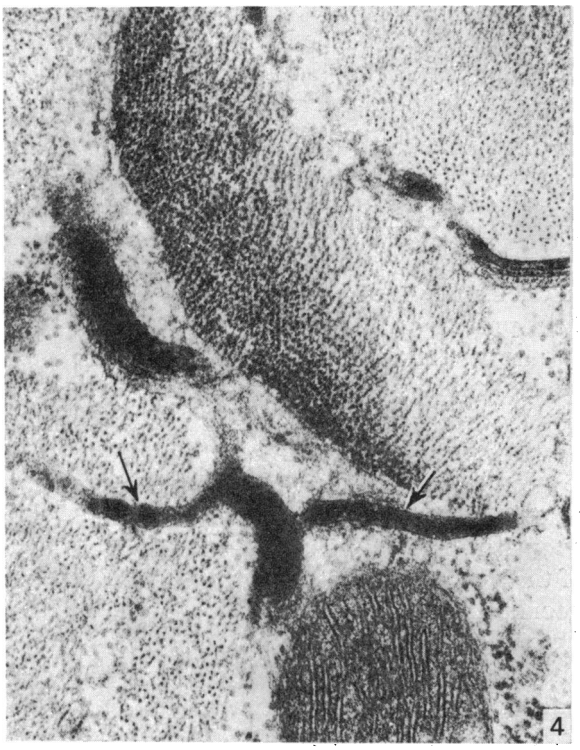
Closely associated with the primary transverse tubules are profiles of SR: these cannot be accurately described as terminal sacs since they are not dilated in comparison with other parts of SR (approximate diameter 38 nm) and, moreover, very often they curve over the T-tubule to extend from one sarcomeric region to the next (Fig. 2). As a result it is possible to find several morphological arrangements, including some resembling dyads or triads, in ventricular myocardium. The cytoplasmic gap between T-tubule and SR membranes at such couplings is usually in the 10–12 nm range, which is slightly wider than dyad or triad couplings in skeletal muscle. There is no discernible specialization in the membranes themselves at these couplings, but in many instances the cytoplasmic gap seems to be occupied by a series of densely stained bridges similar to those seen in Figure 3.

Fig. 4. Cross section through ventricular myocardial cell after peroxidase infiltration. Shows wide primary T-tubules with narrower secondary branches (arrows) which also run in the transverse plane. × 50000.

Fig. 5. Longitudinal section through ventricular myocardial cell after peroxidase infiltration. Note longitudinally aligned tubules labelled with peroxidase staining. × 48000.

Fig. 6. Cross section of ventricular myocardial cell. Note peroxidase labelled longitudinal tubules (*L*). × 40000.

Fig. 7. Ventricular myocardial cell. Shows tertiary T-tubule (3) linking longitudinal tubules (*L*) at A band level. Note coupling with SR and typical cytoplasmic densities. × 56000.



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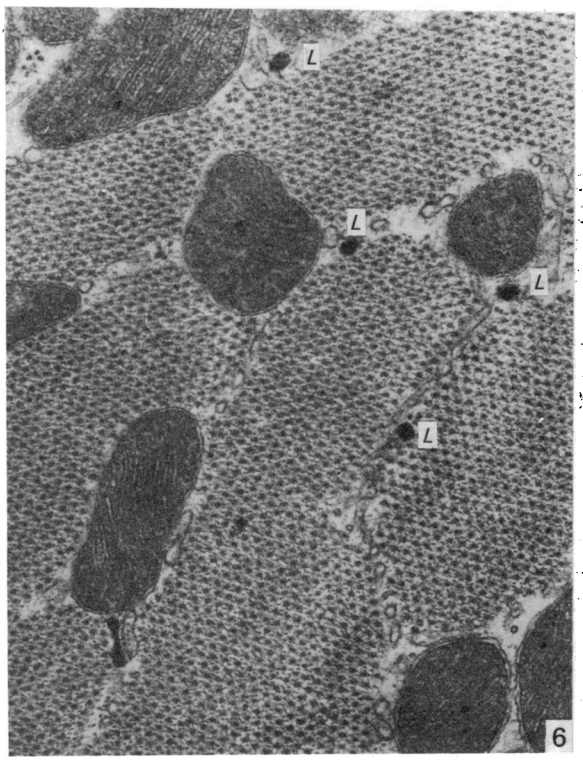

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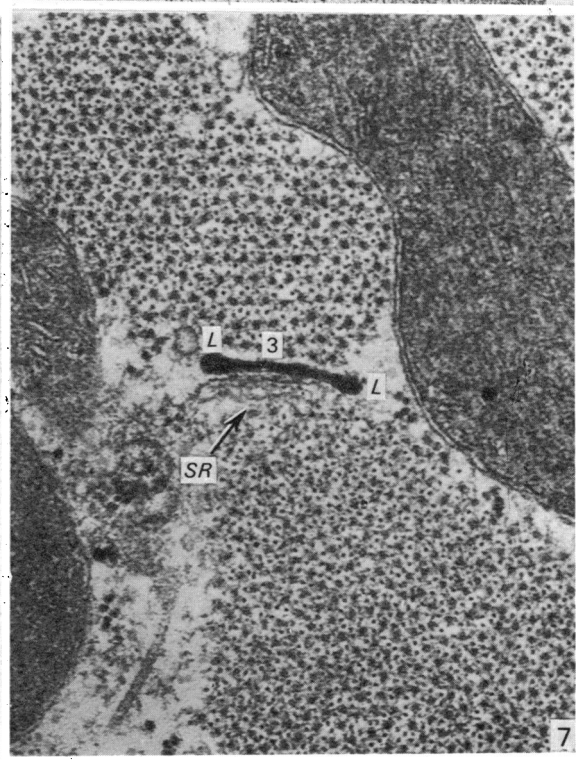

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(*b*) Secondary T-tubules arise directly from the primary invaginations (Fig. 4) and run across the long axis of the cell, retaining the level of the I band and Z line; they are fairly narrow (51 nm diameter) and do not appear to give off any branches. Typical couplings occur between the secondary T-tubules and adjacent profiles of SR, but they are less frequent than couplings in relation to primary tubules.

(*c*) Longitudinal tubules of the T-system are also direct branches of the primary invaginations. They run parallel to the myofibrillae (Figs. 5, 6) and many can be followed over the entire length of a sarcomere. There is no evidence to suggest that any of these longitudinal tubules transgress Z lines. Longitudinal tubules are narrower than the parent transverse tubules, usually measuring about 64 nm in diameter. Typical couplings with adjacent SR profiles can be observed.

(*d*) Tertiary transverse tubules are very narrow (25–30 nm diameter) channels occasionally found linking adjacent longitudinal tubules at the A band level of the myofibrillae (Fig. 7); within the A band they are absent from the H zone, i.e. they occur where the thick filaments are overlapped by thin filaments. Couplings between these elements and the SR have been observed in several preparations.

The sarcoplasmic reticulum (SR) in ventricular myocardial cells comprises a system of channels widely ramified in the cytoplasm. These SR channels are narrower (about 38 nm in diameter) than any part of the T-system except perhaps the tertiary T-tubules. Most of the SR forms an interconnecting system more-or-less investing the myofibrillae throughout their length, there being continuity in the system by way of narrow connexions across Z line level. The SR forms numerous couplings with the T-system, especially in relation to the primary tubules. In addition, flattened sacs of SR frequently form similar couplings (i.e. the gap is about 10–12 nm and may contain dense bridges) with the surface sarcolemma (Fig. 8) and with the sarcolemma along intercalated discs (Fig. 9). In the disc region the SR sacs are invariably related to the 'non-specialized' lengths of sarcolemma where the intercellular gap measures about 20 nm (i.e. they are not related to nexuses, interfibrillary regions or desmosomes). Most often the associated sac of SR is found on only one side of the apposed membranes but, not infrequently, sacs may be seen on both sides.

A noteworthy feature of ventricular myocardial cells is the frequent presence of a narrow tubule (30–40 mm) immediately adjacent to the myofibrillar Z line (Fig. 10). It lacks a clear connexion with the surface sarcolemma, and a connexion with SR has not been identified in this study. In the majority of specimens peroxidase infiltration does not stain these structures which are similar to the Z-tubules described by Simpson (1965). In exceptional instances stained material has been identified in relation to a Z-tubule, but it is possible that this appearance is created by a superposition artefact, and it should not be accepted too readily as indicating that the Z-tubule is part of the T-system.

Atrial myocardium

In typical atrial cells the presence of numerous darkly stained granules (80–360 nm in diameter) may obscure the identification of peroxidase-filled tubules, especially

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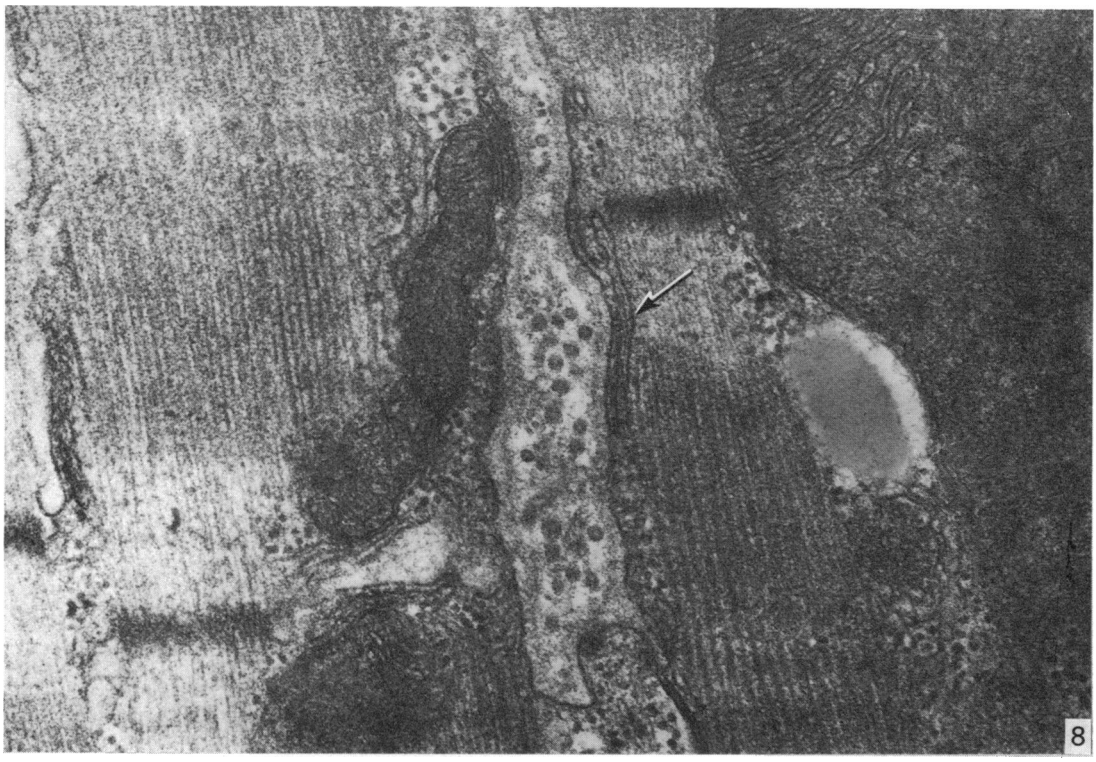
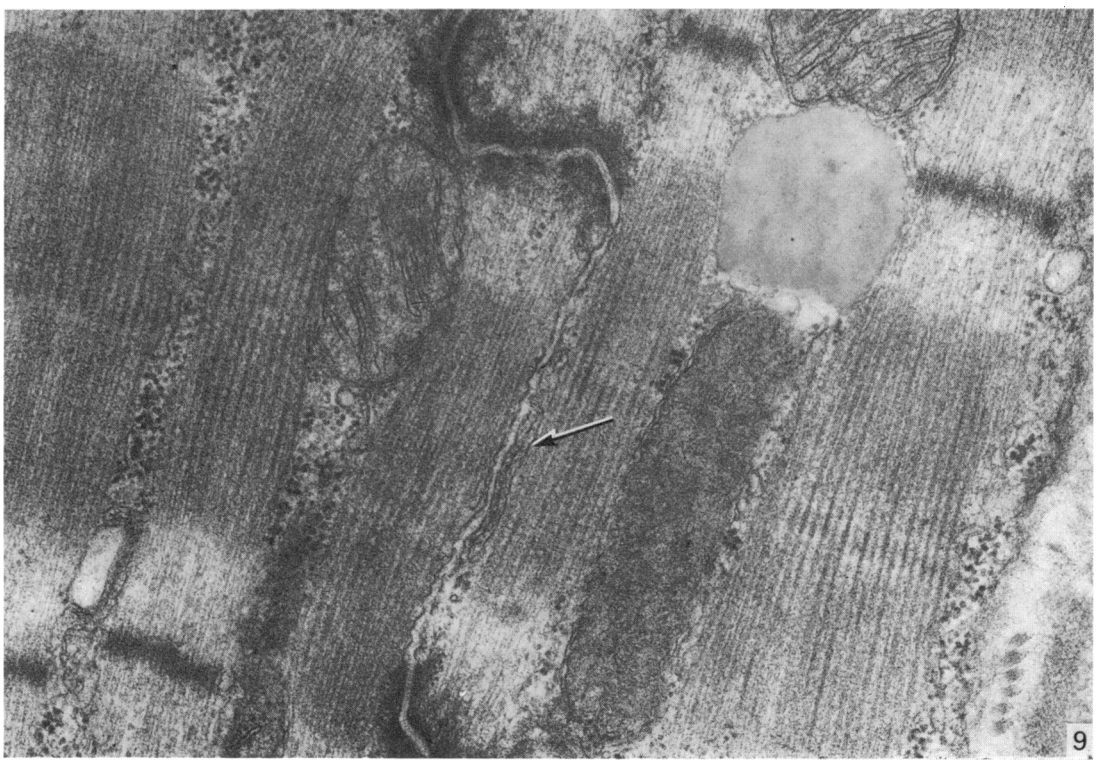
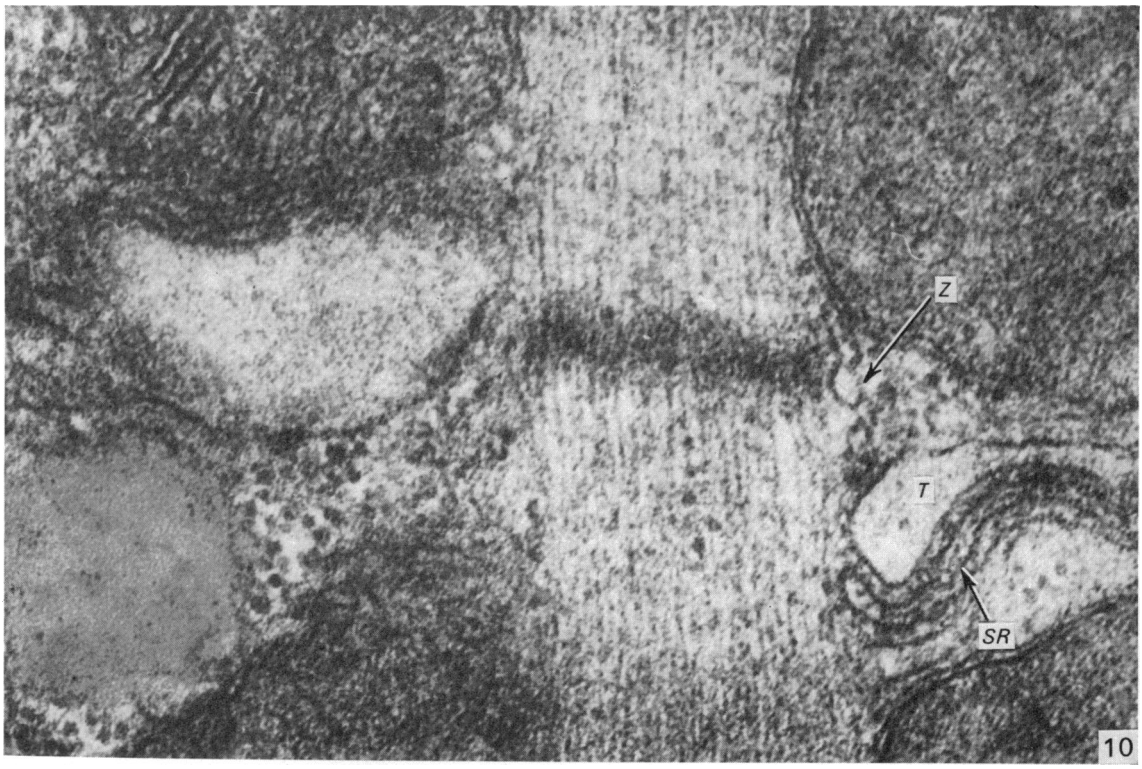
Fig. 8. Ventricular myocardial cell showing coupling between surface sarcolemma and peripheral SR cistern (arrow). × 28000.

Fig. 9. Intercalated disc in ventricular myocardium showing coupling with an SR cistern (arrow). × 44000.







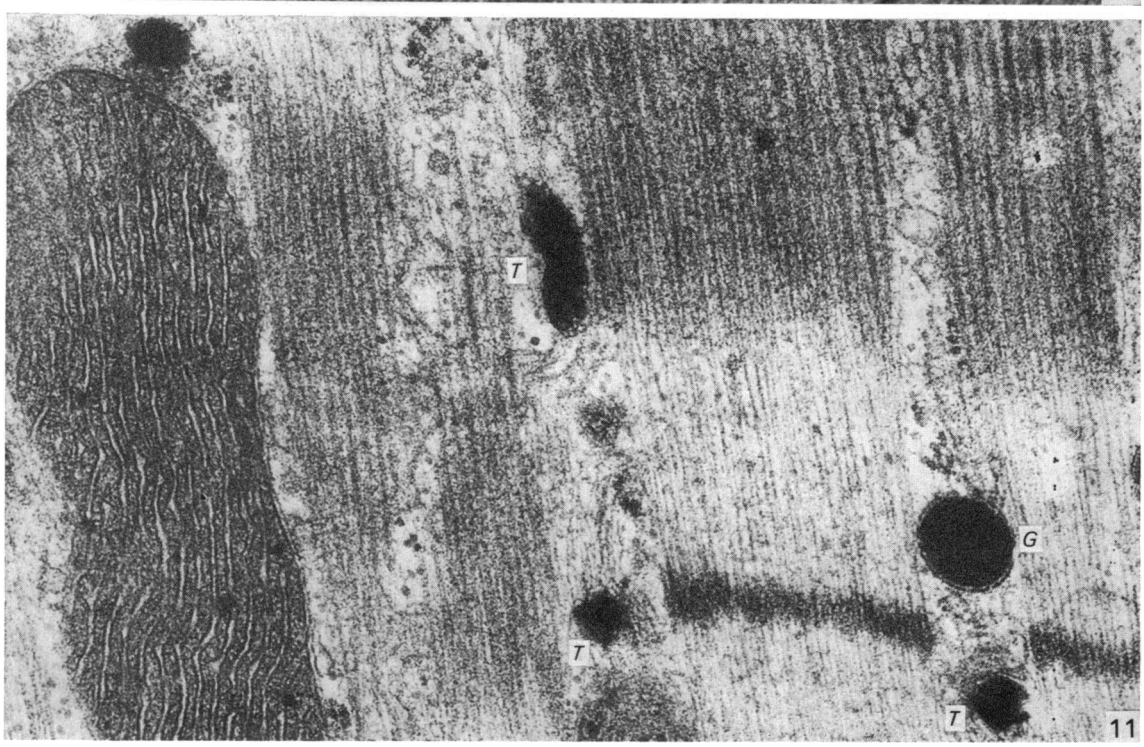
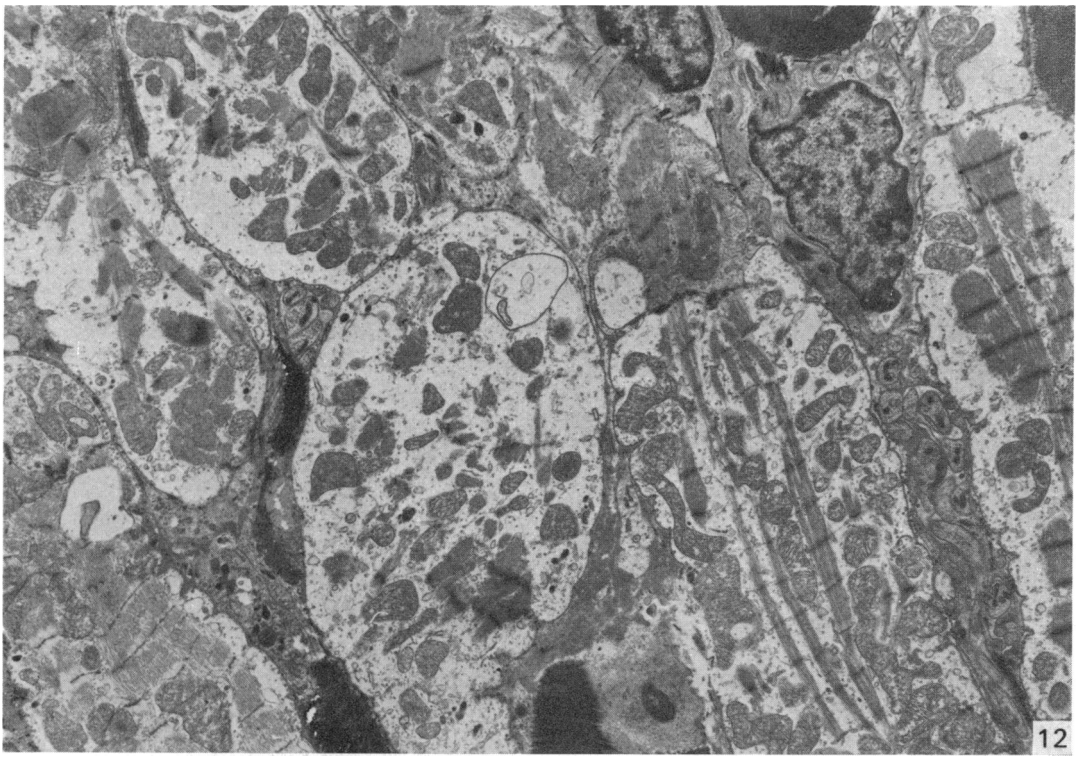


Fig. 10. Longitudinal section of ventricular myocardial cell showing a so-called Z-tubule. × 150000.

Fig. 11. Atrial myocardial cell showing peroxidase-labelled T-tubule profiles (*T*) and specific atrial granules (*G*). × 66000.



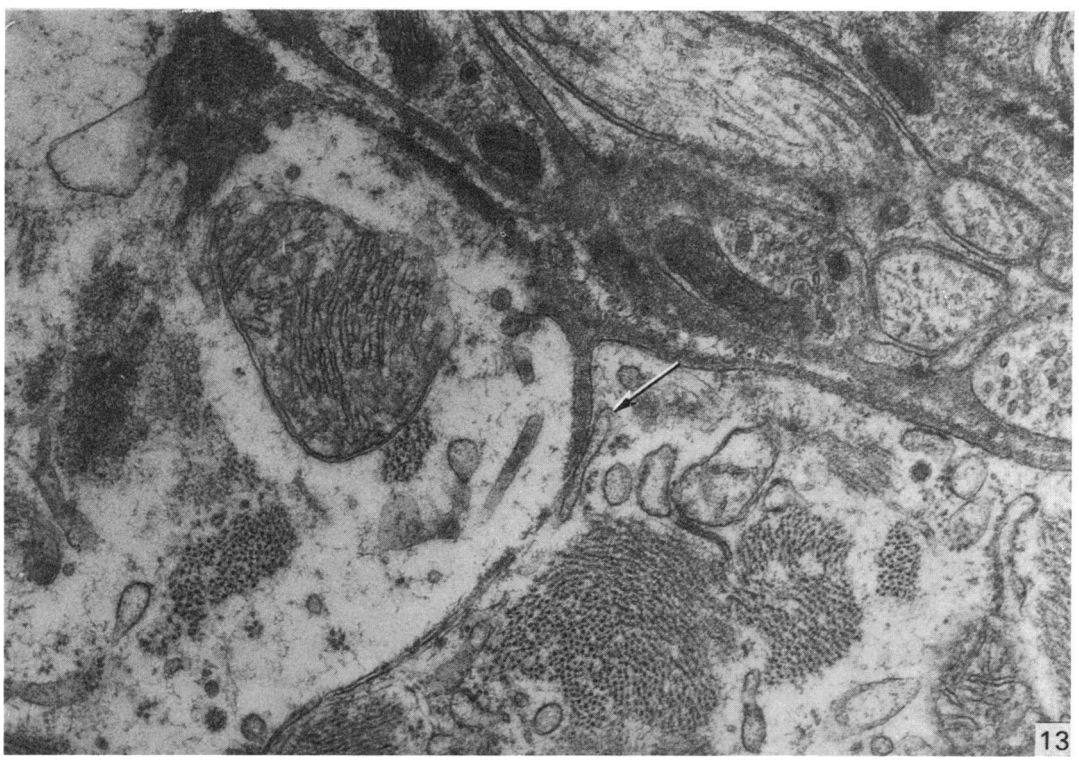


Fig. 12. Section through sinuatrial node showing small cells with poor myofibrillar content and no specific atrial granules. × 4000.

Fig. 13. SA node cell showing coupling between SR cistern (arrow) and surface sarcolemma. × 40000.

where these tubules are transected (Fig. 11). However, peroxidase staining is usually identifiably darker than the granule material and, moreover, by studying preparations where lengths of tubules appear, it is possible to gain a comprehensive picture of the T-system.

As far as can be determined under these circumstances, the T-system in atrial cells is less extensive than that in general ventricular myocardium. It comprises primary T-tubules and longitudinal tubules, but very few secondary or tertiary T-tubules can be identified. The primary tubules measure about 105 nm in diameter, which is narrower than those in ventricle cells. They are invaginated from the surface sarcolemma and lie at I band level close to the Z band. There are numerous couplings with the SR at this level.

Longitudinal tubules in atrial cells are similar in size and distribution to those in the ventricle in that they measure about 67 nm in diameter and run along the length of a single sarcomere from one set of primary invaginations towards the next; several couplings with the SR can be identified along the way. Couplings between SR sacs and sarcolemma are also present at the surface of the atrial cell and at intercalated discs. The appearance of all couplings involving the SR are very similar to those seen in the ventricle; the cytoplasmic gap between the constituent membranes is about 10–12 nm and it lodges darkly stained bridges. Z-tubules are present in many atrial cells.

Sinuatrial node

Sinuatrial (SA) nodal cells are distinguished from general atrial myocardium by their small size, by their poor content of myofibrillae, which moreover are irregularly arranged, and by the absence of specific atrial granules in the cytoplasm (Fig. 12). Moreover, compared to general myocardial cells, the T-system is poorly defined even in peroxidase-treated material. There are short, narrow (about 60 nm in diameter) invaginations of the sarcolemma which probably correspond to primary T-tubules, but they are far less regular than in typical atrial or ventricular cells, and even those that are present do not usually penetrate sufficiently far to contact the myofibrillae. The primary invaginations do not give off any branches and, moreover, couplings between them and the SR have not been identified. The SR itself is not a prominent feature, but some peripherally lying sacs coupled with the surface sarcolemma can be identified (Fig. 13). Typical intercalated discs do not occur in the SA node; most of the cell-to-cell contact is made by unspecialized sarcolemma (there are occasional desmosomes, but no nexuses at all) and SR couplings with sarcolemma occur as frequently in relation to cell contacts as anywhere else on the surface.

There are several transitional cells within the SA node and immediately surrounding it which, though they resemble nodal cells in diminutiveness of size and lack of atrial granules, possess a better-developed T-system, SR and myofibrillae. They

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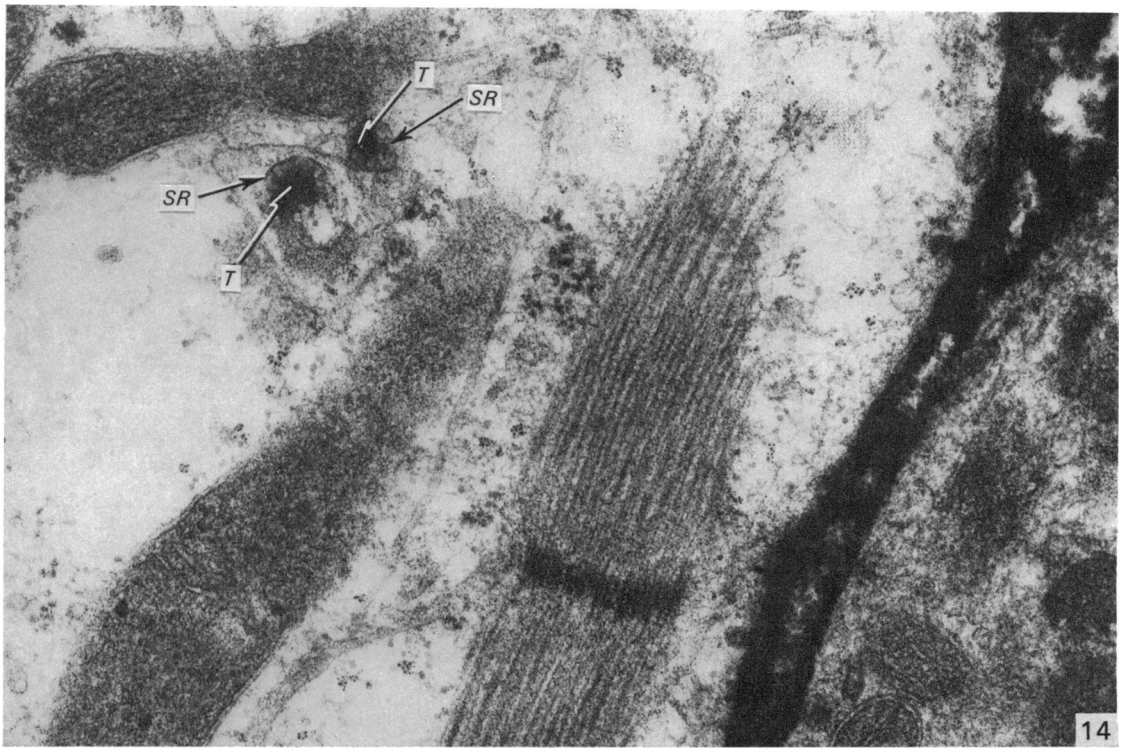
Fig. 14. AV node cell showing two peroxidase-labelled T-tubules (*T*) each coupled with an SR cistern. × 60000.

Fig. 15. AV bundle cells showing longitudinal elements of T-system (arrows). × 9000.

Fig. 16. Longitudinal section through Purkinje-type cell in terminal ramifications of right bundle branch. Shows an extensive T-system with numerous primary tubules at I band level as well as longitudinal elements (arrows). × 10000.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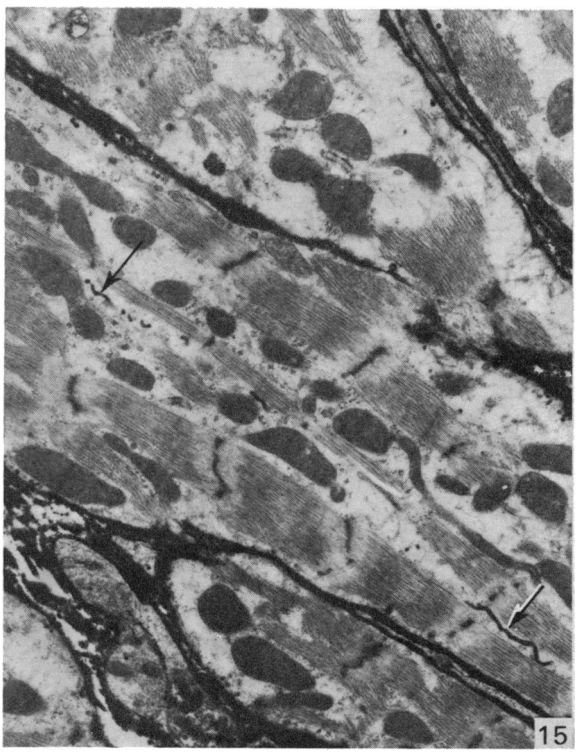
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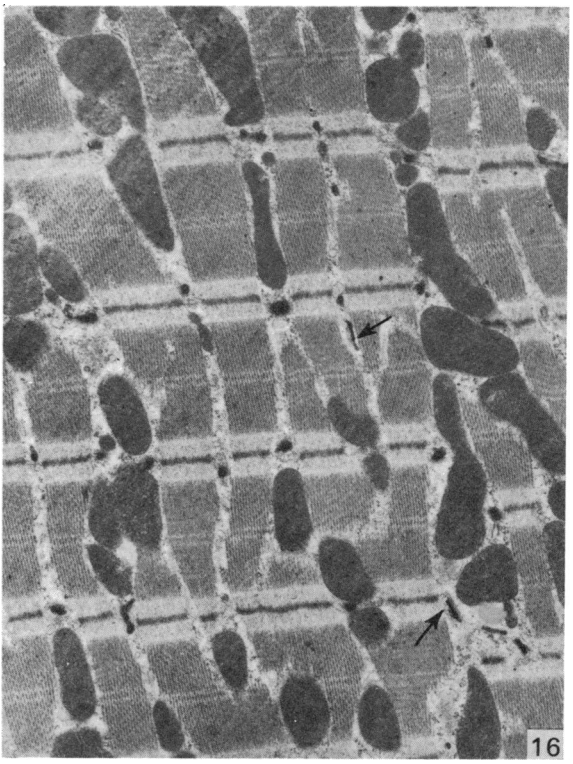
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contain primary T-tubules and longitudinal tubules with SR couplings similar to those found in general atrial myocardium.

Atrioventricular node

The ultrastructural features of typical cells in the atrioventricular (AV) node are in many ways similar to those of SA node cells in that they are small and spindle-shaped with sparsely distributed myofibrillae and no atrial granules. Moreover they lack typical intercalated discs, the cell contacts being predominantly of unspecialized membrane interspersed with the occasional desmosome.

The T-system in these cells is limited to short, narrow invaginations; they enter into couplings with the SR (Fig. 14) a feature which has not been seen in the SA node. There are no longitudinal or other branches of the primary tubules, and the SR itself is poorly differentiated apart from sub-sarcolemmal sacs which have typical couplings with the surface membrane at cell junctions and elsewhere.

In the postero-inferior part of the interatrial septum, just behind the AV node, there are cells which are intermediate in appearance between typical atrial myocardial and typical nodal cells. Like nodal cells they are small, spindle-shaped and lack specific atrial granules, but they contain a recognizably better developed T-system and SR. Such cells are generally termed atrionodal cells, or prenodal cells, and their T-system comprises primary T-tubules and longitudinal branches similar to the arrangement found in atrial myocardium, though the longitudinal elements are narrower (44 nm). Couplings between the SR and T-system are frequently encountered. In many respects therefore atrionodal cells resemble transitional sinuatrial cells.

Atrioventricular bundle and bundle branches

There is no sharp demarcation between the AV node and the bundle. Nevertheless, as one passes from node to bundle and then to bundle branches, the cells increase in diameter, the myofibrillae become more prominent and more regularly arranged, while typical intercalated discs, which include nexuses and interfibrillary regions as well as desmosomes and unspecialized membrane, can be recognized with increasing frequency. Bundle cells have a moderately well developed T-system (Fig. 15) which comprises primary T-tubules measuring about 72 nm in diameter and longitudinal branches of approximately 46 nm diameter; both these components, but particularly the primary invaginations, are associated with SR couplings. Couplings of SR with sarcolemma along the free surface of the cell, and along unspecialized parts of intercalated discs, are also frequently encountered. No secondary or tertiary T-tubules have been observed in bundle cells.

Terminal ramifications of bundle branches

It is not always possible to distinguish between terminal muscle fibres of the AV conducting system and general ventricular cells. However, as a rule, the former cells are considerably thicker and may have a diameter of almost 26 μm, compared with the mean diameter of 14 μm for ventricular myocardial cells. SR and myofibrillae are generally well developed so that these features may not be useful in the recognition of terminal ramifications. However, a consistent feature is the presence of longer nexuses in intercalated discs within these ramifications when compared with the surrounding myocardium.

The T-system is less well developed than in other parts of the ventricle wall. The

primary invaginations (102 nm in diameter) are not quite as wide and there are few secondary T-tubules and, where these occur, they are very narrow (about 40 nm in diameter). On the other hand, longitudinal branches are frequent (Fig. 16) and they are fairly wide (62 nm). There are no tertiary T-tubules. Couplings between T-tubules and SR are common, especially at primary tubules and, in addition, frequent couplings occur between peripherally lying sacs of SR and the sarcolemma, including the intercalated disc regions. In many cells profiles similar to Z-tubules can be recognized; as in general ventricular myocardial fibres, continuity of such Z-tubules with either SR or the T-system could not be established.

DISCUSSION

From the present study on peroxidase infiltrated material it does appear that the T-system is a general feature of all myocardial cells, but its degree of differentiation varies in different parts of the heart. It is modestly, or even poorly, represented in the specialized impulse generating and conducting tissues, especially in the nodes, while it is a prominent feature of working myocardium, particularly in the ventricular wall.

These observations are interesting in view of the known differences in capacity for contraction and conduction shown by these regions of the heart.

The sequence in excitation–contraction coupling in muscle cells is believed to entail transmission of excitation from the surface membrane to the interior along primary T-invaginations. The mechanism whereby the signal is conveyed along the T-tubule membrane is not entirely clear, early evidence favouring the view that it occurs by passive cable-like transmission rather than by propagation of action potential. More recently, however, results from voltage clamp experiments have indicated that in skeletal muscle regenerative propagation of action potential along T-tubule membranes does occur, and that it is probably essential for the initiation of contraction (see Bastian & Nakajima, 1974). Similar evidence is not yet available for cardiac muscle. Girardier and his colleagues (see Girardier, 1965) have observed that myocardial tissue, in which T-tubules were reversibly widened by immersion in hypertonic solution, exhibits a correspondingly reversible reduction in conduction velocity along the surface membrane, thus indicating that the T-system shunts an appreciable fraction of current into the cell. If this is so, the propagation of action potential along the surface of each individual cell should be faster in the specialized impulse generating and conducting tissues of the heart where the T-tubules are much narrower than in working myocardium; similarly, conduction in atrial muscle will be expected to be faster than in ventricular muscle. That the effective speed of conduction through the SA and AV nodes is slower than anywhere else in the heart is probably a reflexion of the absence of low-resistance nexuses in the cell-to-cell contacts in these regions. In the AV bundle system, which contains fast conducting cells as well as many nexuses, conduction is particularly quick.

In its turn T-tubule activation is believed to trigger the SR by way of membrane coupling to release Ca^{2+} into the sarcoplasm and, when the sarcoplasmic concentration of Ca^{2+} is sufficiently high, the troponin sites on actin filaments are activated, thus initiating the contractile mechanism. The SR at couplings has certainly been shown to contain Ca^{2+} ATPase activity as well as Ca^{2+} itself (see Forbes & Sperelakis, 1977), but the precise manner in which calcium release from the SR is effected is not clear. Two possible mechanisms have been suggested, namely: (1) depolari-

zation of the SR membrane – this is regarded as the most likely physiological mechanism in skeletal muscle (Endo, 1977); (2) the slow entry of Ca^{2+} into the cell from the extracellular space, which is known to occur during activation of the sarcolemma, is thought to trigger the release of larger quantities of Ca^{2+} from the SR.

Forssmann & Girardier (1970) argued that the membrane separation at T–SR couplings is so wide relative to the area of contact that practically all the current leaving the T-tubule at such couplings will bypass the SR "unless there exist between the two structures low resistance bridges which our present techniques of fixation and preparation fail to preserve". They claimed that each coupling of myocardial cells has a smaller contact area and a wider cytoplasmic gap than in skeletal muscle fibres. Nevertheless, the demonstration of bridge-like structures by Rayns, Devine & Sutherland (1975), as well as in the present study, does recall the possibility that low-resistance channels, whereby an electrical signal can be transmitted to the SR, may exist. In skeletal muscle fibres, depolarization of SR during physiological contraction has been strongly indicated by birefringence changes (Baylor & Oetliker, 1975), fluorescence changes in muscle stained with Nile blue A (Bezanilla & Horowicz, 1975), and by electrode recording studies (Natori, 1975). Corresponding evidence is not easy to elicit in cardiac muscle, but the possibility of a similar mechanism must be kept in mind.

It is clear that cardiac muscle SR is more sensitive to the effects of Ca^{2+} than skeletal muscle, and that calcium-induced calcium release can be more easily evoked in cardiac muscle, even with a low level of free calcium. Moreover, there is a fair amount of circumstantial evidence (Endo, 1977) favouring the view that this is the predominant mechanism, if not the only one. Unfortunately, however, the exact calcium requirement for calcium release in cardiac SR has not been defined, and more evidence is necessary before this problem can be resolved.

Another ultrastructural feature of cardiac muscle cells not shared by skeletal muscle is the presence of numerous couplings between surface sarcolemma and peripherally lying SR sacs. Whether or not this feature is designed to boost the effects of slow calcium entry through the sarcolemma during depolarization is arguable, but it is striking that such peripheral couplings are present in all types of myocardial cell, even in SA nodal cells which lack deeper coupling arrangements.

As indicated above, most investigators consider that couplings are designed to release Ca^{2+} into the sarcoplasm on activation of the muscle cell. On the other hand, Langer (1971) suggested that the primary function of a coupling, at least in myocardial cells, is the uptake and sequestration of Ca^{2+} prior to its release into the extracellular fluid. He thus argued that couplings participate primarily in the relaxation, and not in the contraction, of the myocardial cell. There is little in the morphological arrangement of couplings in cardiac cells to provide information on this problem one way or the other. Nevertheless, for skeletal muscle there is good evidence that the SR at couplings is involved in the excitation–contraction sequence, rather than in relaxation. For instance Huxley & Taylor (1958) showed that contraction and local activation of the SR via a single T-invagination was confined to the adjacent half sarcomeres, which suggested that calcium was released only from the adjacently coupled terminal cisternae; this interpretation received support from the autoradiographical studies of Winegrad (1970), which indicated that calcium is released physiologically mainly from the terminal cisternae, where the main part of calcium is stored in resting muscle. On the other hand, for cardiac

muscle Müller (1966) showed that localized surface stimulation does not elicit localized contraction, but instead causes widespread contraction of practically all the sarcomeres in the field of observation. A possible explanation of Müller's finding is that the distribution of couplings in typical cardiac muscle is far more extensive than in skeletal muscle, for it includes some at every level of an extensive T-system, as well as others in relation to the surface sarcolemma. Moreover, the SR is not separated into units corresponding to each sarcomere, but extends across Z-line levels, possibly by way of the so-called Z tubules. Such observations do not go very far towards explaining long known but ill-understood contraction characteristics of myocardium, such as its failure to exhibit tetanic contraction, but they give some insight into the working of these remarkable cells.

SUMMARY

The T-tubule system in cardiac muscle cells has been investigated with the electron microscope in 10 adult rats after infiltration with horseradish peroxidase.

All cardiac muscle cells possess a T-system, but its complexity varies according to the region of the heart. It is most extensive in the general ventricular myocardium where there are primary, secondary and tertiary transverse tubules as well as longitudinal elements, and there are numerous couplings between the T-system and the sarcoplasmic reticulum (SR). The T-system and associated SR couplings are less extensive in the atrium and in the atrioventricular conducting system. It is least well developed in the nodes, particularly in the sinuatrial node, where it is restricted to primary T-tubules.

There are numerous SR couplings with the sarcolemma in all types of cardiac muscle cells. Where intercalated discs occur, SR couplings are associated with non-specialized parts of the disc.

The possible significance of these ultrastructural features in regard to the speed of conduction by myocardial tissues, and in the excitation–contraction sequence is discussed.

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