Section of Medicine, Experimental Medicine & Therapeutics

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Meeting 21 May 1975

The Myocardium

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Structural and Ultrastructural Basis of Myocardial Disease

Signs and symptoms of diseases of the heart can often be explained on a structural basis. This may be obvious on macroscopic examination, as in cases of congenital abnormalities, and at histological level in, for example, myocarditis, in which case the changes may be characteristic, or they may be nonspecific. Much can also be learned from electronmicroscopic examination which can show structural changes not identifiable at light microscopic level (such as early changes of hypoxia). It can, however, reflect light microscopic changes and, by virtue of much higher magnification, may give greater understanding of the basic structural alteration such as in cases of hypertrophic obstructive cardiomyopathy. The structural changes may be secondary to a variety of stimuli, e.g. immunological, biochemical or viral, or they may be unknown. The change in the myocardial cell often appears to react to these different stimuli in a similar manner at light microscopic magnification but ultrastructurally qualitative, quantitative or chronological differences may be evident.

Ischæmia, hypoxia, anoxia or hypoxæmia have been extensively studied in experimental animals and in man. In ischæmia resulting in myocardial infarction, macroscopic changes do not become evident until approximately fifteen hours after the event. Perfusion with substances such as nitro blue tetrazolium, according to the method of Glagov *et al.* (1963), permits recognition after only eight hours (Nachlas & Shnitka 1963, Ramkissoon 1966).

At histological level, early changes cannot be recognized by conventional staining (hæmatoxylin and eosin) but special staining procedures, such as the hæmatoxylin basic fuchsin picric acid (HBFP) method (Lie *et al.* 1971, Nayar & Olsen 1974), has permitted recognition of earliest changes (approximately 30 minutes) in cells underperfused with oxygen or totally deprived of their blood supply. If these processes continue for too long a time, cell death occurs, which after a period of six hours can be recognized histologically. The changes of healing take place in a fairly regular manner, permitting dating in the early stages of myocardial infarction (Lodge-Patch 1951).

Early ultrastructural changes have included decrease of glycogen, cristolysis in mitochondria, swelling of the tubular system, and chromatin clumping in the nuclei. These early changes are reversible; later, irreversible changes include rupture of mitochondrial membrane or rupture of nuclear membrane and tearing of myofibrils (Caulfield & Klionsky 1959, Olsen 1973, Heggtveit 1969).

Although similar changes are seen irrespective of the experimental methods employed, certain differences do exist. For example, the distribution in ischæmia tends to be patchy, whilst changes of hypoxia and anoxia are more widespread.

The changes may also differ in sequence, depending on the experimental method chosen. For example, widening and increase in density of Z bands has been observed within 30 minutes after administration of isoproterenol, and mitochondrial swelling and cristolysis occur later (approximately after two hours), and glycogen decrease is not conspicuous (Ferrans *et al.* 1964). In experiments on dietary magnesium depletion, the events are slower and the earliest changes become evident only after five days. In potassium depletion, myofibrillar alteration is an early event. Mitochondria are better preserved and

nuclear loss is minimal (Molnar *et al.* 1962, Poche 1969).

Another nonspecific way for the myocardium to react is by hypertrophy, a reaction which occurs secondary to almost all disease processes involving the heart.

Macroscopically, appearances include increase in thickening of chamber walls, and the myocardium is firm.

Histologically, the diameter of the myocardial fibres exceeds the normal range (5–12 μm). Nuclei become hyperchromatic or vesicular, and abnormal forms such as horse-shoe shapes may also be found.

Ultrastructurally, an increase in the number of mitochondria, often without qualitative or quantitative signs of insufficiency, takes place (Laguens & Gomez-Dumm 1967). Enlargement of sarcoplasmic tubules, eventual accumulation of glycogen and prominence of the Golgi apparatus may be seen. Deposition of new myofibrillar tissue also occurs early (Büchner 1971).

Linzbach (1947) suggested that hypertrophy alone occurred even if the critical heart weight was exceeded. The subject of quantitative morphology has been excellently reviewed by Hort (1971). Meerson (1969) has argued that only hypertrophy occurs and not hyperplasia.

Hypertrophy may be accompanied by dilatation of the myocardium, resulting in normal measurement of wall thickness despite the hypertrophy. This is histologically recognized by attenuation of myocardial fibres, particularly in the inner layers of the myocardium. As a result of this dilatation, myocardial cell diameter falls within the normal range, but nuclear changes of hypertrophy remain. In chronic dilatation, the endocardium is thickened and the smooth muscle fibres may be prominent (Fisher & Davis 1958). Inter- and intra-cellular œdema may also be found.

The changes described above may also be found in heart failure following excess alcoholic intake. There is no doubt that alcohol has a deleterious effect on the myocardium (Burch *et al.* 1971) but resulting changes are nonspecific, though much has been written on characteristic appearances.

Structural changes with characteristic appearances include conditions such as myocarditis, the etiology of which may remain unknown. Aschoff nodules are the characteristic feature of rheumatic heart disease. Amyloid also has a characteristic appearance, although the exact mechanism is as yet not fully known; immunological processes seem likely (Stirling 1975).

Heart involvement of the group classified as neuromuscular disorders is similar, consisting in the later stages of the diseases of adipose or fibrous tissue replacing myocardial cells. The association of neuromuscular disorders and heart disease is well recognized but the exact mechanism is still not fully understood. The subject has been reviewed by Emanuel (1972) and Olsen (1972).

The pearly white, thickened fibrous endocardium in isolated endocardial fibroelastosis, involving each trabecula of the ventricular wall, gives it a characteristic appearance. Histologically the thickened endocardium contains regularly arranged elastic fibrils throughout its entire thickness.

The final group of myocardial disease is that of cardiomyopathy, defined as heart muscle disease of unknown cause or association (Goodwin 1973), and has been classified according to clinical manifestations (Goodwin 1970, Oakley 1976) into congestive, hypertrophic and restrictive (obliterative) groups.

The congestive group has no characteristic appearance and is indistinguishable from those changes described in the section of the dilated heart. There are no other abnormalities within the heart or elsewhere in the body.

By contrast, hearts in hypertrophic obstructive cardiomyopathy show macroscopically characteristic changes, consisting often of extreme asymmetric thickening, involving most frequently the interventricular septum. Histologically, extreme hypertrophy of myocardial fibres, which are irregularly arranged, is seen in this asymmetric area. Bizarre-shaped nuclei, often surrounded by clear zones, are found. In addition whorl formation is evident and varying degrees of fibrous tissue are present (Olsen 1971).

Histochemically, immense accumulation of glycogen, particularly in the perinuclear area, is part of the characteristic picture (Van Noorden *et al.* 1971). At ultrastructural level the irregular arrangement of the fibres is also reflected in the fibrillar structure of the myocardial cell (Olsen 1975). In addition there is an immense increase of mitochondria, patchily distributed. A characteristic feature is frequent cross-over of myocardial fibrils (Ferrans *et al.* 1972), which may explain failure of compliance in these patients. These frequent cross-overs are unfortunately not confined to hypertrophic obstructive cardiomyopathy (Olsen 1971, personal observation, Sekiguchi *et al.* 1972–73).

The final group of cardiomyopathy is that of the restrictive (obliterative) type; examples include conditions such as endocarditis parietalis fibroplastica (Löffler 1936, Weiss-Carmine 1957) and endomyocardial fibrosis (Davies 1948, Ball *et al.* 1954). The characteristic lesions are those of extreme thickening of the endocardium which, in the case of endomyocardial fibrosis, often ends in a thick rolled edge as the outflow tract of the

left ventricle is approached (Davies 1968). The possibility that Löffler's endocarditis and endomyocardial fibrosis form a spectrum of the same disease process, the origin of which can be traced back to the presence of eosinophils in the myocardium, has been suggested by Brockington & Olsen (1973).

The list of examples of the structural and ultrastructural appearances forming the basis of myocardial disease is by no means complete. Apart from the characteristic appearances at histological level, the myocardial cells appear to react in a limited way to many different stimuli, but ultrastructural examination permits us to study the structural changes in greater detail and permits some insight into the structural basis of myocardial disease.

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Myocardial Preservation during Cardiac Arrest

Replacement of the aortic valve raises the problem of the mutually antagonistic requirements of providing a good operative field for the surgeon and adequately preserving myocardial integrity during the period of aortic occlusion. The three main procedures used clinically under these circumstances are continuous coronary artery perfusion, no myocardial perfusion at all, and coronary 'infusion' – injection into the root of the aorta below the aortic clamp of various cardioplegic media prior to a period of ischæmia. Each technique may be combined with hypothermia. In an attempt to assess the effect of each of these procedures on a comparable experimental preparation, they were investigated using the isolated beating rat heart. Male rat (Sprague-Dawley 280–320 g) hearts were excised and the aorta and left atrium cannulated, leaving the pulmonary artery open. For 5 min they were perfused via the aorta in a Langendorff (1895) non-working preparation with Krebs-Henseleit bicarbonate buffer at a hydrostatic pressure of 65 cmH_2O (6.37 kPa). They were then converted to a working preparation (Neely *et al.* 1967, Hearse *et al.* 1974) for a 15 min control period. The left atrium was perfused at a pressure of 20 cmH_2O (1.96 kPa). At the end of the control period the left atrial inflow and the aortic outflow were stopped for a 30 min experimental period. During this time the myocardium was subjected to a variety of experimental procedures. Following this the heart was converted back to an atrially perfused working preparation. Aortic flow and other parameters of cardiac function were measured as the heart recovered from the effects of the chosen experimental procedure.