Ultrastructural Features of the Endomyocardium in Patients with Eosinophilic Heart Disease. An Endomyocardial Biopsy Study

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In a series of studies with endomyocardial biopsy, 7 adult cases with cardiac disease and eosinophilia were studied clinically and electromicroscopically. Degranulation of the eosinophils in the peripheral blood was observed ultrastructurally in 3 of the 4 cases studied. The clinical expression of the 7 patients were, restrictive cardiomyopathy in 2, dilated cardiomyopathy in 2 and sick sinus syndrome in 1 and others in 2. Endocardial thickening was observed in 5 cases, one of whom showed a marked cellular infiltration with macrophages, plasma cells, lymphocytes and mast cells. One other case showed cell debris of degranulated eosinophils. Degeneration of the myocytes was manifested by an increase in Z-bands of the myofibrils as well as streaming (1 case), disarrangement of the myofibrils, and mitochondrial change (1 case) which was characterized by giant mitochondria (1 case) as well as a numerical increase in mitochondria.

Increase of atrial granules and mitochondria in a right atrial biopsy of a case with sick sinus syndrome was noteworthy. Basal lamina layering of the capillaries of the myocardium and pyknosis of an endothelial cell cytoplasm were also noted in each case. These observations may reveal that various disease processes are taking place in the endocardium and adjacent myocardium.

Since Löffler first described two cases with the term endocarditis parietalis fibroplastica with blood eosinophilia in 1936, the association of hypereosinophilia and heart disease and especially endomyocardial fibrosis has been the subject of study for nearly a half century.1,3 Brockington and Olsen4 proposed that Löffler's endocarditis and endomyocardial fibrosis should be included in the same category. They stressed that inflammation and fibrosis were the manifestations of the course of the disease.

A possibility was raised that the eosinophils release their toxic substance and injure the endocardium and adjacent myocardium.5 Some immune mechanism is considered to be an important pathogenetic factor in this setting.2,5 One of the important morphological features is degranulation and vacuolization of the eosinophils in the peripheral blood.5 They are seen in the endocardial and myocardial tissue which are damaged. Some histopathological and ultrastructural features of this condition have been

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In our series of studies using endomyocardial biopsy in more than 1300 cases a review of the material and current clinicopathological studies have shown that there are varieties of cardiovascular manifestations of the disease! We are inclined to believe that the disease can be termed “eosinophilic heart disease.” Clinical and histopathological aspects of the cases have been presented and it is the purpose of this study to describe essential clinicopathological features with special reference to the ultrastructural findings of the endocardium and myocardium obtained by the endomyocardial biopsy.

**MATERIALS AND METHODS**

A breakdown of 7 cases in which the criteria of eosinophilia with blood eosinophil counts of over $1.0 \times 10^9$ were met and a distinct cardiac disease was found is presented in Table I. Endomyocardial biopsies employing Konno-Sahakibara’s biopuncture from the right ventricle in all cases and from the right atrium in one case (Case 3) was carried out while the disease was manifested. One or two specimens from the right ventricle were halved and submitted for light and electron microscopic analysis, respectively. The paraffin-embedded specimens were processed in a routine manner and a histopathological examination was carried out. Ultrastructural observation was made using conventional tissue processing using glutaraldehyde-Oso4 fixation, and the specimen was stained with uranyl acetate and lead citrate. For the assessment of both the histopathological and ultrastructural findings we used our own criteria for a semiquantitative recognition of the pathology.

As we will later report the clinical and histopathological aspects of the 7 cases, the details are not given in this paper. An outline of the clinical and histopathological findings are tabulated in Table I.

**RESULTS**

The essential clinical and ultrastructural findings in representative cases are described as follows:

Case 1: This 49-year-old man showed a degranulation process of the eosinophils in the peripheral blood (Fig. 1A). Among the 4 cases where the eosinophils were observed ultrastruc-
Fig.1. A. Ultrastructure of an eosinophil of the peripheral blood in a patient with restrictive cardiomyopathy and eosinophilia (Case 1). Note that some granules show a degrading process and vacuoles are formed (arrows). N = nucleus. The bars in each figure indicate 1 μ.

B. Ultrastructure of thickened endocardium of the right ventricle which was infiltrated with numerous mononuclear cell infiltrations. A lymphocyte (L), a plasma cell (Pl), a degraded plasma cell (DPl) and a fibroblast (FBl) are observable.

C. Ultrastructure of a mast cell which was observed in the endocardium of case 3. Same case as in Fig. 3B.

D. Low power ultrastructural view of thickened endocardium (E) in a patient with dilated cardiomyopathy (Case 4). The endocardium is filled with collagenous fibers. Cell infiltrations are scarce and cardiac myocytes (My) are observed beneath the endocardium.

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Fig. 2. A. Ultrastructure of a degraded eosinophil in the thickened endocardium in a case with restrictive cardiomyopathy (Case 4). Note that the cell border of the eosinophil is not clear and granules are scanty. As one of the granules (arrow) shows a high density in the central zone, it indicates that the cell is an eosinophil.

B. High electron density of a part of a capillary endothelium in a patient with dilated cardiomyopathy (Case 5) with eosinophilia (arrows).

C. Basal lamina layering of a capillary in the myocardium in a case with dilated cardiomyopathy with eosinophilia (Case 4). Note that up to 5 layers are observable at one side of the capillary (arrow).

naturally, 3 cases (cases 1, 4 and 6) showed the degranulation process of peripheral blood while one case (Case 3) did not.

Case 3: This 38-year-old woman with sick sinus syndrome showed a marked endocardial thickening of the right ventricle which was infiltrated with numerous and variable mononuclear cells. In spite of 3 or 4 attempts to obtain myocardial tissues at the time of the right ventricular biopsy, only endocardial tissue could be obtained. It was presumed that the endocardium was so thick that the tip of the bioptome had slipped off. Electronmicroscopically, the infiltrated cells were composed of plasma cells, fibroblasts and lymphocytes. There were many plasma cells which showed a degradation process (Fig. 1B). Also noteworthy was the intermixture of mast cells in the infiltrated cell group (Fig. 1C). In this particular patient, a right atrial biopsy was carried out in order to clarify

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Fig. 3. A. Ultrastructure of a cardiac myocyte in a case with dilated cardiomyopathy with eosinophilia (Case 6). Note that the mitochondria are generally swollen and there is a great increase in rough surfaced endoplasmic reticulum (rER).

B. Ultrastructure of giant mitochondria (GM) in a case with dilated cardiomyopathy with eosinophilia (Case 7). The 2 giant mitochondria are surrounded by mitochondria of normal size. Swelling of the mitochondrial matrix is also seen.

C. Z-band streaming observed in a case with dilated cardiomyopathy with eosinophilia (Case 6). Also note that the Z-band electron density is high.

D. Ultrastructure of a right atrial myocyte in a case with sick sinus syndrome (Case 3). Atrial granules (AG) as well as mitochondria have increased in number. Also, dilatation of sarcoplasmic reticulum (SR) is noteworthy.

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that the basic pathology in the right atrium was a cause of the sick sinus syndrome. Atrial cardiac myocytes showed mitochondriosis, an increase in atrial granules and numerous and slightly dilated sarcoplasmic reticulum (Fig. 3D).

Case 4: This 41-year-old man showed a structureless endocardial thickening which was infiltrated with a small amount of cells (Fig. 1D). In some parts of the endocardium, cell debris which was thought to consist of degranulated eosinophilic was recognized partially within the myocardium (Fig. 2A). Another characteristic change was the presence of basal lamina layering (BLL) of a capillary in the myocardium. The BLL showed a close proximity to a pericyte (Fig. 2C).

Case 5: This 59-year-old man with dilated cardiomyopathy had dark cytoplasm in an endothelial cell (Fig. 2B).

Case 6: This 51-year-old male patient showed disarrangement of the muscle bundles as well as an increase in large mononuclear cells suggesting the presence of postmyocarditic change at the histopathological level. Ultrastructurally, disarrangement of myofibrils which was associated with an increase in Z-band material showed an advanced steaming appearance (Fig. 3C). Swelling of the mitochondrial matrix and an increase in rough-surfaced endoplasmic reticulum were observed as well (Fig. 3A). In this patient, eosinophiles in the peripheral blood showed a degranulation process which was similar to that seen in Fig. 1A.

Case 7: This 49-year-old man showed a striking feature; giant mitochondria (GM) in a myocyte. Only two GMs were observed. They were 4 x 3 microns in size (Fig. 3B). In this case, mitochondriosis was also prominent.

**DISCUSSION**

This paper is a preliminary report of our study of eosinophilic heart disease regarding the ultrastructure of the endocardium and myocardium. Some important profiles of the disease were noticed and comments on each of the findings are as follows:

1. Endocardial change

   In 2 of the 7 cases studied ultrastructurally, endocardial thickening was characteristic. Three cases showed endocardial thickening which was composed of a small number of macrophages, collagenous fibers and ground substance. Various cell debris was also observed. The above described pictures may demonstrate that endo-
carditis was present and the healing process has begun. Case 3 showed numerous cell infiltrations, composed mostly of macrophages, plasma cells and lymphocytes. Eosinophils were not identified within the endocardium in this case. However, it should be noted that infiltration had occurred once but the eosinophilic granules had since disappeared due to the degranulation process. In Case 4, degraded eosinophils were identified within the slightly thickened endocardial tissue (Fig. 2A) indicating that endocardial change was manifested prior to the biopsy.

2. Degeneration of myocytes:

Swelling of the mitochondria as well as numeral increase in SR which were observed in case 6 may signify the effect of hypoxia of myocytes. Giant mitochondria in case 7 was a unique feature. An analysis of 270 biopsied cases of various heart muscle diseases was performed at the author's laboratory10-12 Giant mitochondria were observed in 2 other cases, both of whom were observed in a group of 40 dilated cardiomyopathy cases. It is generally known that giant mitochondria are observable in suppressed metabolic conditions in various organs such as myopathy18 starvation or hypoxia19 intoxication20 and in the left atrium of mitral valvular disease21.

Furthermore, in this particular case, an eosinophilic condition may have caused the impaired metabolic condition of the myocardium. Z-band abnormalities with streaming and disarrangement of myofibrils which were seen in case 6 are considered to be one of the common regressive forms of myofibrillar change. According to our experience, such changes are observable in cases in whom degenerative and regenerative disease processes are taking place concomitantly as in myocarditis22. The increase in rough surfaced endoplasmic reticulum observed in cases 5 and 6 may also suggest regenerative change of the myocytes.

Increased atrial granules as well as increase in mitochondria in the right atrial muscle in case 3 may represent a hyperfunction of the myocytes. We have previously studied right atrial myocardium in cases with atrial septal defect and found that in the early stage of the disease, such a proliferative phenomenon was observable23. In case 3, right atrial stimulation which is caused by eosinophilia may also have played a role.

Ferrans6 has stressed the importance of endothelial changes such as markedly pyknotic cytoplasm, increase in thickness of the cytoplasm and of ribosomes within the endocardial tissue but not in the myocardial tissue. We also observed the pyknotic cytoplasm of capillary endothelium within the myocardium in a case with dilated cardiomyopathy (Case 5). In this particular case, a histopathological picture suggesting postmyocarditic change was observed, though it was not so prominent. Also, in this case, the degranulation process was also observed ultrastructurally in the eosinophils of the peripheral blood. It is therefore suggested that some tissue damage processes are also taking place in the myocardium.

Basal lamina layering (BLL) of the capillaries in the myocardium was detected in 14 out of 18 cases with cardiac sarcoidosis and it may be a factor related to the cause of the myocardial damage which occurs at the microvascular level24. This particular pathology was evidenced in the skeletal muscles of patients with diabetes mellitus and we proposed an additional concept that BLL is an expression of microangiopathy. Some immune mechanisms in relation to the BLL are to be taken into consideration and the above observation in a case with eosinophilia is worthy of further documentation.

The authors made a national survey in Japan of cardiac diseases which included 89 cases of eosinophilia and the results indicated that the disease also manifests itself as a systemic disease by affecting numerous organ systems which are caused by still unknown toxic agents. As Take of our group has suggested, the relation between eosinophilia and the wide spectrum of heart disease should be considered within the framework of eosinophilic heart disease.

Most of the above described ultrastructural features are regarded as being non-specific in nature. They are pathognomonic of myocardial damage. However, marked endocardial thickening which is infiltrated with active cells, the presence of their debris, as well as an increase in collagen fibers characterized the disease.

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