Clinical And Histopathologic Study Of Surgically Excised Mitral Valves In Children
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Citation

Abstract
The present study aims to interpret the histological changes observed in surgically excised mitral valves, and to how those changes relate to the clinical criteria of valve failure. A retrospective study was performed on 25 children patients with RHD, who had mitral valve surgery between 2000 and 2004 at the GOTHI hospitals. A highly significant association was present between absence of Aschoff nodules and atrial fibrillation (p=0.0070). Mitral calcification was significantly associated with atrial fibrillation (p=0.0050), followed by double mitral dysfunction (p=0.0142). Valvular endothelial ulcerations were significantly associated with mitral stenosis (p=0.0129) and double mitral dysfunction (p=0.0100). Rheumatic heart disease continues to be an important cardiac problem afflicting the young population of Egypt. The disease has a definite effect on mitral valve microanatomy. Clinical-pathologic correlation is important in evaluating the extent of valvular affection.

INTRODUCTION
Rheumatic fever is an acute, systemic, partly autoimmune disease triggered by a beta-hemolytic streptococcal throat infection that cross-reacts with human tissues and may stimulate or modify immunologic responses. It is characterized by a constellation of findings that include as major manifestations: migratory polyarthritis of the large joints, carditis, subcutaneous nodules, erythema marginatum of the skin, and Syndeham chorea; the latter is a neurological disorder with involuntary purposeless, rapid movements [1,2].

Rheumatic heart disease (RHD) occurs in 30 to 45% patients with rheumatic fever (RF). The incidence of RF and RHD in the tropics remains high, with a high proportion of children suffering from carditis with the first episode. The prevalence of RHD in school-age children varies considerably throughout the world. It has been reported as 0.6/1000 in the USA and 0.7/1000 in Japan, while in developing countries the prevalence rates range from 0.3/1000 to 18.6/1000. In an Egyptian study by Refat and his colleagues, the prevalence of RHD in the primary school children was 3.4/1000 [3]. Approximately 20 million cases of rheumatic fever occur in third world countries annually, with a correspondingly high incidence of advanced mitral stenosis later in life. A genetic predisposition to develop RHD appears to be important in certain countries like India, Egypt and Turkey [4].

Chronic rheumatic heart disease is the most important consequence of rheumatic fever. It is characterized principally by deforming fibrotic valvular disease (particularly mitral stenosis), which can produce permanent dysfunction and severe, sometimes fatal, cardiac dysfunction decades later [5,6]. Clinical manifestations may not be seen for years after the initial episode of rheumatic fever. The signs and symptoms of valvular disease depend on which cardiac valve(s) is(are) involved. In addition to cardiac murmurs, cardiac hypertrophy and dilation, and heart failure, patients with chronic rheumatic heart disease may suffer from arrhythmias (particularly atrial fibrillation in the setting of mitral stenosis), thromboembolic complications, and infective endocarditis. Long-term prognosis is highly variable. There may be a relentless cycle of valvular deformity yield hemodynamic abnormality, which begets further deforming fibrosis. In addition to affecting the cardiac valves, rheumatic heart disease is a pancarditis affecting to various degrees the endocardium, myocardium, and pericardium. In some cases, rheumatic myocarditis results in cardiac dilation and progressive heart failure [7].

Mitral valve leaflets are the most common structures involved in rheumatic disease; typical pathological features of rheumatic mitral disease come from acute and recurrent inflammation [8]. The rheumatic process includes leaflet thickening, calcification and retraction, perianular calcification with limitation of annular motion, leaflet fusion, chordal thickening, shortening and fusion as well as
papillary inflammation [9], but the specific immunologic and inflammatory mechanisms leading to the valvulitis are unknown. The normal mitral valve consists of two major leaflets, the roughly semicircular anterior cusp, which makes up about one third of the annular circumference, and the semilunar posterior cusp, which consists of three scallops, one middle and two commissural ones. The commissures, in contrast to the semi lunar valves, do not extend completely to the annulus and are supported by commissural chordae. Histologically, the normal mitral valve is composed of three layers. The fibrosa layer, which is continuous into the chordae tendineae and the tip of the papillary muscle, faces predominantly toward the ventricular side of the valve and is covered by the ventricularis layer [10,11]. The ventricularis consists mainly of elastic tissue and is covered by a single layer of endothelium. On the atrial side of the valve is the spongiosa, which consists mainly of proteoglycans, mesenchymal cells, and elastic and collagen fibers. It also contains cardiac muscle cells, which are continuous with the atrial myocardium; the auricularis layer and its endothelium cover the atrial side of the valve; the collagen fibrils and elastic fibers of the chordae are continuous with the mitral valve leaflets. By far the most common cause of mitral stenosis is post inflammatory scarring (rheumatic heart disease); the valve leaflet shows fibrosis, calcification, ossification, and neovascularization, sometimes with a prominent chronic inflammatory infiltrate. Aschoff nodules are seen in the papillary muscles, but they are more frequently found in atrial appendages removed at surgery.

The surface of the valve, particularly the line of closure, may be covered with fibrinous exudates [12].

AIM OF STUDY
The aim of the present study is to interpret the morphological and histological changes observed in surgically excised mitral valves with rheumatic heart disease, and to how those changes relate to the clinical criteria of valve failure.

MATERIALS AND METHODS
A retrospective study of 25 children patients with rheumatic heart disease, who had mitral valve surgery between 2000 and 2004 at the GOTHI (General Organization for Teaching Hospitals and Institutes) hospitals.

CLINICAL EVALUATION
Before surgery, all patients were clinically evaluated for mitral dysfunction; mitral valve lesions were classified as purely regurgitant, purely stenotic, or mixed according to recognized clinical and radiologic/echocardiographic criteria. Mitral regurgitation was considered to be pure when associated with unrestricted valve leaflet excursion and a normal mitral orifice area, as assessed by two-dimensional echocardiography. Pure mitral stenosis was diagnosed when no clinical or echocardiographic evidence for regurgitation was found. Mixed mitral valve disease was diagnosed when features of both regurgitation and stenosis were present. Atrial fibrillation was diagnosed when a wide notched P wave in lead II and a biphasic P wave in lead V1 were detected during ECG.

HISTOLOGICAL EVALUATION
Mitral valves selected for histological examination were fixed in 10% neutral formalin for 18 hours, absolute methanol for 24 h, and then embedded in paraffin. Paraffin sections were stained by hematoxylin-eosin (HE). The light microscopic findings used as criteria of rheumatic activity were the presence of inflammatory infiltrate, Aschoff bodies, neovascularization, calcification and the valvular lining ulceration.

STATISTICAL EVALUATION
Histopathologic findings of the surgically excised mitral valves were statistically studied with the Chi-square test to determine any possible correlation with the preoperative clinical criteria of mitral valve failure. A p value of <0.05 was considered significant.

RESULTS

CLINICAL FINDINGS
In the present study, mitral stenosis was diagnosed in 14 cases (56%), regurgitation in 3 cases (12%), and double mitral dysfunction in 8 cases (32%). Atrial fibrillation was clinically diagnosed in 17 cases (68%).

HISTOPATHOLOGIC FINDINGS
In all the microscopically examined mitral valve specimens the basic structural change was collagen fibrinoid degeneration. The interstitial connective tissue was seen edematous and eosinophilic, with fraying, fragmentation, and disintegration of collagen fibers seen associated with lymphocytic infiltration (Fig. 1 & 2), that occurred in several patterns: perivascular lymphocytic aggregate tightly surrounding small to medium vessels in 4 cases (16%), diffuse streaming of lymphocytes into the surrounding interstitium in 7 valves (28%), and a mixed pattern of infiltration in 14 valves (56%). Aschoff nodules were detected in 3 valves (12%) (Fig 3).
Histologically, they are granulomatous lesions localized mainly in the endocardium, subendocardium, or perivascular regions of myocardial interstitium. The Aschoff nodule has different stages, which involve different cells (Anitschkow cells, multinucleated cells, few lymphocytes, macrophages, plasma cells, and polymorphonuclear leukocytes [2]). Neovascularization in 13 valves (52%) (Fig 3 & 4), interstitial calcification in 15 valves (60%) (Fig 4 & 6), and valvular endothelial surface lining ulceration in 16 valves (64%) (Fig 5 & 6).

**Figure 1**
Figure 1: Valvular collagen fibrinoid degeneration associated with lymphocytic infiltrate

**Figure 2**
Figure 2: Valvular interstitial edema

**Figure 3**
Figure 3: Aschoff nodule in RHD

**Figure 4**
Figure 4: Neovascularization, lymphocytes infiltration and calcification in RHD
ASCHOFF NODULES IN RELATION TO VALVULAR DYSFUNCTION

Aschoff nodules were present in only 3 valves (12%), all belonging to cases presented clinically with mitral stenosis. Interestingly, a high significant association was present between absence of Aschoff nodules and atrial fibrillation (p=0.0070) (Table 1).

NEOVASCULARIZATION IN RELATION TO VALVULAR DYSFUNCTION

Neovascularization was microscopically evident in 2 valves (8%) clinically diagnosed as mitral regurgitation; 7 (28%) valves clinically diagnosed as mitral stenosis and 4 valves (16%) with double mitral dysfunction. Moreover, microscopic neovascularization was seen in 7 cases (28%) clinically presented with atrial fibrillation. No significant association was detected with regard with the neovascularization and clinical criteria of mitral valve failure (Table 1).

MICROSCOPIC CALCIFICATION IN RELATION TO VALVULAR DYSFUNCTION

Microscopic mitral calcification was present in 3 valves (12%) clinically diagnosed as mitral regurgitation; 10 valves (40%) clinically diagnosed as mitral stenosis and 2 valves (8%) with double mitral dysfunction. On the other hand, microscopic calcifications were seen in 7 cases (28%) presented with atrial fibrillation. Microscopic calcification was significantly associated with atrial fibrillation (p=0.0050), followed by double mitral dysfunction (p=0.0142) (Table 1).

VALVULAR ENDOTHELIAL ULCERATIONS IN RELATION TO VALVULAR DYSFUNCTION

Valvular endothelial ulcerations were microscopically present in 2 (8%) valves that presented clinically with mitral regurgitation, 6 valves (24%) with mitral stenosis, and 8 valves (32%) with double mitral dysfunction. With regard to atrial fibrillation, 11 valves (44%) presented with valvular endothelial ulceration. Valvular endothelial ulceration was significantly associated with mitral stenosis (p=0.0129) and double mitral dysfunction (p=0.0100) (Table 1).

DISCUSSION

Rheumatic heart disease is the most common cause of valvular disease in developing countries [13], where predisposing factors to RF persist and prophylactic therapy is often inadequate [14]. The disease is endemic in poor nations, most patients are children, and acute rheumatic
In the current study, clinically detected mitral valvular stenosis accounted for 56% of all cases; the same finding was reported in Hanson et al study (54%). Jaya et al mentioned in their study that the time period between the onset of RF and symptoms due to mitral stenosis was less than 3 years in the majority of their patients [11]. Mitral regurgitation was detected in 3 cases (12%); a diagnosis of pure mitral regurgitation may have been incorrectly excluded in patients with a mitral mid-diastolic murmur in that and other studies that relied solely on clinical data [1]. Double mitral dysfunction was present in 8 cases (32%). Mixed mitral stenosis and incompetence was the most frequent malfunction in others studies [12-14]. Atrial fibrillation in patients with rheumatic valvular heart disease results in significant morbidity and possibly mortality [15, 16]. In the present study, atrial fibrillation was clinically diagnosed in 68% of cases. Atrial fibrillation is common when the left atrium enlarges and is almost always present when the atrium is extremely large [17].

The pathological changes in the valvular specimens from patients with rheumatic heart disease have been defined in the past, but these have been mainly descriptive postmortem studies [18]. Our study emphasizing the surgical specimen study of the excised valves indicates that the main pathological process in chronic rheumatic heart disease is a progressive fibrosis particularly affecting the heart valves. The basic structural change in collagen is fibrinoid degeneration. The interstitial connective tissue becomes edematous and eosinophilic, with fraying, fragmentation, and disintegration of collagen fibers. This is associated with infiltration of mononuclear cells including large modified fibrohistiocytic cells (Aschoff cells) some of them of multinucleated forms. In RHD patients there is a persistent inflammatory process in the heart tissue, in the absence of the infectious agent [12, 19]. Once damage has developed on a valve, the altered hemodynamic stresses on the valve perpetuate and extend the damage, even in the absence of a continuing rheumatic process [1].

In the present study Aschoff nodules were present in only 3 valves (12%), all belonging to cases presented clinically with mitral stenosis. None of the cases with Aschoff nodules had atrial fibrillation. Aschoff bodies are characteristic lesion of unclear etiology and can be taken as an indicator of a recent episode of RF, although there are no clear data to determine how long Aschoff bodies persist after a clinical episode [19].

Normally, the valves cusps in non-inflammatory conditions are non-vascular and sufficiently thin to allow complete nutrition by diffusion [20]. Neoangiogenesis or new blood vessel formation is defined as an increased number and concentration of thin delicate vascular spaces. The presence of neovascularization probably represents a sequel of the rheumatic process, after release of angiogenic growth factors such as fibroblast growth factor, VEGF, and transforming growth factor-β released from mast cells, that are likely to induce angiogenesis in calcified cardiac valves [11].

Rheumatic valve calcification is not a random passive process but rather a regulated, inflammatory cellular process associated with the expression of osteoblastic markers and neoangiogenesis [12]. Microscopic calcification was highly significantly associated with atrial fibrillation (p=0.0050), followed by double mitral dysfunction (p=0.0142). In the present study calcifications appeared as large amorphous basophilic areas surrounded by a variable amount of fibrous tissue, frequently associated with inflammatory changes. These changes varied from a few dilated vascular channels with scanty lymphocytes and plasma cells to dense cellular infiltration, including numerous PMNs. Calcification of heart valves is an active process; the local cells secrete osteopontin, a protein that results in bone deposition. Calcium deposits, neoangiogenesis, and inflammation appear to be relevant biological features resulting in calcification [20].

The most vulnerable part of the valve is the endocardium, which lines the valves. When valvular endothelial damage occurs, there is micro thrombus formation. Moreover, since the valves are continually flexing, it is not surprising that they are subject to damage, particularly in the presence of other factors (rheumatic Heart Fever, calcification, myxomatous degeneration) [15].

CONCLUSIONS

Rheumatic heart disease continues to be an important cardiac problem afflicting the young population of Egypt; the disease has a definite effect on the mitral microanatomy. Double mitral dysfunction is associated with endothelial ulcerations and calcification. In our series, atrial fibrillation was associated with mitral calcification and absence of Aschoff bodies.
References

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