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CHAGAS' HEART DISEASE: CLINICAL-PATHOLOGICAL CORRELATION

Marcos A. Rossi, Simone G. Ramos and Reynaldo B. Bestetti

Department of Pathology, Faculty of Medicine of Ribeirão Preto, University of São Paulo, Ribeirão Preto, S.P., Brazil

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1. ABSTRACT

This review provides an overview of the clinicopathological aspects of chronic Chagas' heart disease, and a comprehensive view of predictors of mortality for chagasic patients with Chagas' cardiopathy in an attempt to help physicians with the management of their patients.

2. INTRODUCTION

Chagas' disease (*American trypanosomiasis*), caused by transmission of the protozoan *Trypanosoma cruzi*, is one of the major causes of morbidity and mortality in many countries of Latin America and in areas ranging from the southern United States to Argentina (1). Cardiac disease is by far the most important clinicopathological consequence of this disease (2), with a significant negative social impact (3). Moreover, since several million immigrants from countries with endemic disease are now living in the United States and Europe, and epidemiologic studies point out to a significant prevalence of infection, there has been a recent burst of interest on Chagas' disease all over the world (4,5). In addition, the study of this illness may have important impacts on other cardiac diseases due to the fact that "…the similarity of Chagas' heart disease to other dilated congestive cardiomyopathies, particularly those due to viral etiology, should make awareness of this South and Central American disease relevant to investigators outside endemic areas" (6).

3. HISTORICAL ASPECTS

In 1909, in a remarkable investigative analysis, Carlos Chagas (Figure 1) described the outstanding features of the disease, identified the causative parasite, and characterized its life cycle. The merit of his research, the circumstances surrounding it, and subsequent development of the field represent one of the most important pages in the history of medical science (7).

In 1908, the Brazilian Government, when trying to build a railroad from Rio de Janeiro to Belem (in the Amazon Basin), had to halt construction midway in Minas Gerais due to a severe malaria outbreak suffered by the railroad workers. Oswaldo Cruz, then Director of the Manguinhos Institute, commissioned Carlos Chagas and Belisario Pena to that region to attempt to control the outbreak. They settled their headquarters in Lassance in a railroad car, that served as consultation room, laboratory, and sleeping room. After one year of intensive work, Carlos Chagas was told by a railroad engineer about the

Figure 1. Photograph of Carlos Chagas.

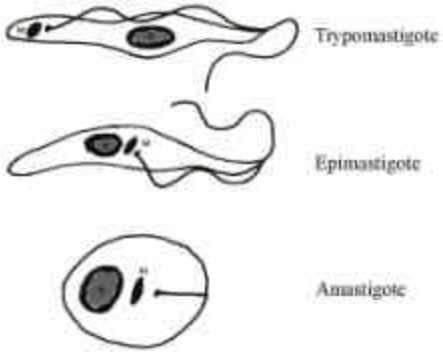

Figure 2. Different morphological forms of *Trypanosoma cruzi* (N, nucleus; kt, kinetoplast; um, undulating membrane).

Figure 3. *Triatoma infestans*, one of the vectors of *T. cruzi.*

existence of hematophagous bugs which were known as "barbeiros" (barbers) or "kissing bugs" due to their typical behavior of biting human beings while sleeping at night on the uncovered face. Chagas became interested in investigating the possibility of this bug transmitting parasites to human or other vertebrates. He soon detected flagellates resembling crithidiae in the hindgut of the bugs. Intrigued by the possibility that this parasite could represent an evolutionary stage of *Trypanosoma minasense*, which he had previously described infesting marmosets (*Callithrix*) in 1908, he sent some bugs to Manguinhos to be fed in primates free of infection. After some weeks, the same flagellates seen in the hindgut of the bugs were recovered from the blood stream of the animals and a new species different from *T. minasense* or "any other species of the same genus" was recognized. The parasite first named as *Schyzotrypanum cruzi* in honor of Oswaldo Cruz was subsequently renamed *Trypanosoma cruzi.*

Carlos Chagas returned to Lassance looking for the presence of vertebrate hosts of this newly discovered parasite. After several tests in human beings and animals, he found a cat with parasite in the bloodstream. A few weeks later, he was asked to investigate the possibility of an acute malarial episode in a 2 years old girl named Berenice living in the same house where the cat was found. He had previously examined this girl and no parasites had been observed. This time, however, several parasites were detected which suggested the possibility of an acute phase of a new disease. Upon further examinations it was discovered that the flagellates disappeared as the symptoms vanished, thus raising the possibility of a chronic phase of this new disease. On April 1909, Oswaldo Cruz announced Carlos Chagas discovery in a session of the Brazilian National Academy of Medicine. The genius of Carlos Chagas enabled him to describe, when he was only 29 years old, the agent, vectors, clinical signs in man and animals, and the existence of animal reservoirs of a new disease. Although his findings were hotly disputed during his lifetime, later investigators confirmed them. Never before or since has a single scientist, a clinician and clinical investigator, fully characterized a new disease in all its aspects and done so in this manner.

4. ETIOLOGIC AGENT

Trypanosoma cruzi is a flagellate of the *Kinetoplastida* Order, Family *Trypanosomatidae,* characterized by the presence of one flagellum and a single mitochondrion in which is situated the kinetoplast, a specialized DNA-containing organelle. The identification of this parasite by morphological and biological features does not offer difficulties and differential is only required for *Trypanosoma rangeli*, a non-pathogenic flagellate which infects humans in some areas of Central and South America and is transmitted by some of the same vectors that transmit *T. cruzi* (8).

Three morphological forms of *T. cruzi* can be identified: (1) metacyclic trypomastigotes, in the feces of the vectors and in the bloodstream or the tissues of the vertebrate host; (2) amastigotes, the result of transformation of the trypomastigote forms in the cytoplasm of tissue cells of the vertebrate host, when they replicate by binary fission and, (3) epimastigotes, within the midgut of the triatomine, resulting from differentiation of trypomastigotes taken up during blood feeding, undergoing multiple rounds of binary fission inside the triatomine gut and, finally, differentiating back into trypomastigotes in hindgut (Figure 2).

5. LIFE CYCLE, TRANSMISSION AND PORTAL OF ENTRY

The most important mechanism of transmission of *T. cruzi* to humans is throughout the feces of infected triatomines. The vectors of Chagas' disease are insects of the Order *Hemiptera*, family *Reduviidae* and subfamily *Triatominae* (Figure 3). Of the 118 species of triatomines, a relatively small number are epidemiologically significant as vector of *T. cruzi.* These are species that colonize poorer quality rural houses, where colonies of hundreds of

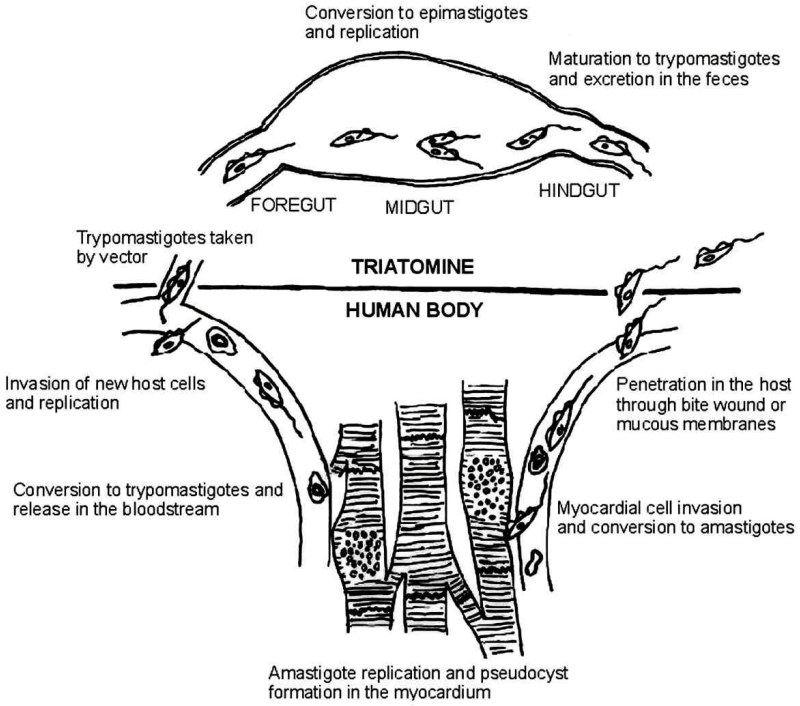

Figure 4. The life cycle of *T. cruzi*, beginning from the insect vector.

individuals (or even thousands) can be found. Other species are strictly residents of different wild ecotopes and never invade houses, thus not representing any problem to the man. The main domiciliated species are responsible for more than 80% of the cases of human Chagas' disease in endemic areas. These species, *Triatoma infestans, T. brasiliensis, T. dimidiata, T. sordida, Panstrongylus megistus*, and *Rhodnius prolixus* are characteristics of the so called "open" spaces of South and Central America, chiefly those poorer ones that were submitted to anthrop action. The vectorial transmission of *T. cruzi* to man and other mammals is almost always due to the contact of these vertebrates with the feces of the infected vector. After blood feeding act (mainly in exposed areas of the body during the night) the bug expels semiliquid feces contaminated with the metacyclic forms of the trypanosomes, near the wound of the bite. As the wound itches, rubbing or scratching follows and the parasites reach the subcutaneous tissue through the puncture produced by the insect or into an abrasion of the skin. They may also be transmitted through the mucous membrane, commonly the ocular conjunctiva. They enter a variety of host cells, including the reticuloendothelial cells of spleen, liver and lymphatics and also the smooth, skeletal and cardiac muscles. The method of entry is still uncertain although it is thought to be via receptor-ligand binding proteins. The actual method of entry is also a puzzle as there are no obvious pseudopods or cell surface alterations. Some studies have suggested that *T. cruzi* recruits host lysosomes which move toward and gradually fuse with the cell plasma membrane at the site of the parasite entry. The membrane of the parasitophorous vacuole, which contains the parasite, is very similar to that of the lysosomes. Once inside the cell the parasite changes to an amastigote form, necessary for reproduction. The amastigotes divide many times forming a pseudocyst that eventually ruptures. The amastigotes convert to trypomastigotes and are released into the bloodstream and can then invade new host cells and tissues. Once taken by the vector, the trypomastigotes pass into the midgut and transform into epimastigotes where they divide. Finally, 8-10 days after infection they appear in the insect hindgut as short stumpy trypomastigotes (Figure 4).

In human Chagas' disease, after the penetration, a short period of 7 to 9 days elapses until the beginning of the classical symptomatic acute phase (incubation period), in which the parasite undergoes an intensive process of tissue multiplication and invades the bloodstream and several organs. For practical purposes (in laboratory accidents, for instance), a short treatment with the available drugs at the period will be sufficient to prevent the setting of the infection.

Infection acquired through blood transfusion is the second most important mechanism of transmission of *T. cruzi.* In some endemic regions, such as the State of São Paulo, Brazil, where the vectorial transmission was controlled through an intensive and continuous Public Health Program, blood transfusion is now the main route of transmission. Furthermore, human migration from endemic areas to urban centers has created a rising risk of transfusional Chagas' disease in all Latin America and even in non-endemic countries. Considering the high number of infected blood donors in all Continent, it is estimated that thousands of new cases of transfusional Chagas' disease may occur yearly.

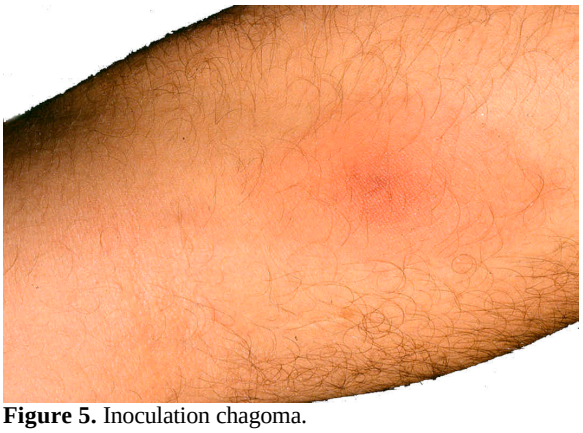

Figure 5. Inoculation chagoma.

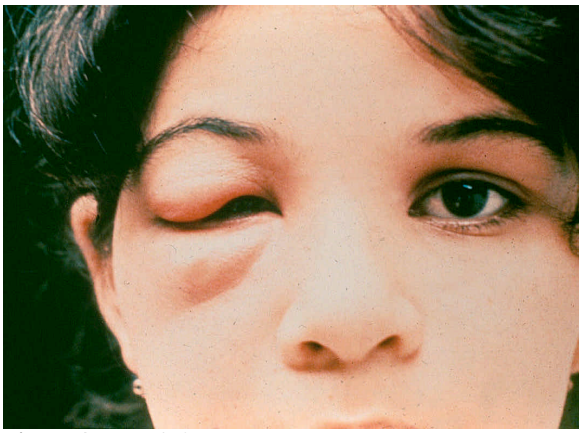

Figure 6. Romaña's sign.

6. EPIDEMIOLOGY

T. cruzi can be detected over a wide area of America, from latitude 42° N to latitude 46° S. Although the distribution of wild vectors and reservoirs is much greater than that of the human disease, the "domiciliation" of the triatomines exposes at least 100 million persons at risk of infection, from the south of the United States of America to the province of Chubut, Argentina. However, the better living standard of the population and the conditions of the local species of triatomines make the human infection by the vector extremely rare in the United States. In endemic countries it is estimated that 16-18 million people are infected by the parasite, excluding Mexico and Nicaragua, from which adequate data are not available (9). Classically considered as a typical rural disease of Latin America, the growing urbanization has changed the spectrum of human *American trypanosomiasis* in the Americas. Profound economic and social changes in the last four decades are stimulating rural-urban migration in most of endemic areas, with more than 60% of the population presently settled in urban centers. It is estimated that today, due to migration more than 500.000 infected individuals are living in big cities. In addition, chagasic patients are migrating northward to the USA and even eastward to Europe: it is estimated that currently around 100.000 infected individuals live in the USA, most of them immigrated from Mexico and Central America. The medical and social impact of Chagas' disease is high. For instance, it is estimated that about 752.000 of working days per year are lost due to premature deaths caused by disease in the seven southernmost American countries, at a cost of 1.21 US$ billion/year (9).

In addition, in Brazil alone, considering that at least 10% of infected people develop severe cardiac or digestive chronic involvement, the medical costs for their necessary treatment could reach an estimated US$ 250 million/year (9). Chagas' disease in blood transfusion is also in increasing problem in Latin America. Since the 1950's, many studies have demonstrated that the transmission of *T. cruzi* by blood transfusion from infected donors is extremely frequent in some endemic areas. Natural non-vector transmission of the parasite involving congenital and oral routes can also occur in both sylvatic and domestic cycles, while transfusional transmission is an artificial mechanism depending on socio-epidemiological situations and directly linked with the quality of the health system in endemic countries (10).

7. PATHOLOGY

The disease is characterized by three phases: acute, indeterminate, and chronic (10,11). The heart is the most severely and frequently involved organ. The degree of cardiac involvement during the acute phase varies from mild (asymptomatic or olygosymptomatic) to severe. The later involvement may be fatal, but this complication is infrequent and occurs in only 3% to 5% of the cases. The chronic Chagas' heart disease is the most serious and frequent manifestation of Chagas' disease, the highest incidence being found between 15 and 50 years of age (2).

7.1. Acute phase

The acute stage of the disease is most frequently seen in children of less than 5 years old. During the acute phase of the infection there is local inflammation at the side of the bite, often with the formation of a small red nodule or chagoma (Figure 5). In many cases the portal of entry is the ocular conjunctiva resulting in unilateral edema, conjunctivitis and pre-auricular lymphadenitis, the so-called Romaña's sign (Figure 6). As the infection progress there is an increase in the numbers of pseudocysts being formed, particularly in the heart muscle. The heart is globular and flabby. Foci of myocytolytic necrosis and degeneration are seen microscopically with an intense mononuclear inflammatory infiltrate associated with exudative phenomena and parasitism of myofibers (Figure 7). Other findings are nervous disorders, chills, pain in the bones and muscles followed by death, usually within 3-4 weeks after infection, due to heart failure (10).

7.2. Chronic phase

In the chronic phase, most of the hearts show marked alteration in size and form (Figure 8), although some of them appear to be normal in size and form. All degrees of enlargement of the heart may be found, mainly affecting the right-sided chambers of the heart. The mean weight ranges between 400 and 600g. Gross cardiomegaly is not unusual but a heart weight greater than 1000 g is

extremely rare (10). This fact suggests that dilatation is generally more pronounced than hypertrophy. The heart may appear globular in shape, but a separation between the left and right apex is usually found giving the heart the aspect of "cor bifidum" (Figure 9). Bulging of the conus arteriosus is always evident and sometimes enormous. The epicardium shows small plaques or tiny white granules along the subepicardial coronary vessels, the so-called pericarditis "in rosary" (Figure 10). Fifty-to-sixty-percent of Chagas' hearts show a peculiar lesion consisting of thinning and bulging of the apical region mainly of the left ventricle, but also of the right ventricle, called by various denominations such as apical aneurysm, atrophic lesion of the apex, thinning of the apex (Figure 11); its wall may consist of the endocardium and pericardium only with a few myocardial fibers interposed (12) (Figures 12 and 13). Similar localized parietal thinning may occur in the left and right ventricular free walls (Figure 14). Transillumination of the heart reveals areas of marked thinness of the ventricular walls (Figure 15). Fibroadipose or adipose replacement of the right ventricular myocardium, particularly at the apical area of the right ventricle, can be frequently seen, occasionally associated with bulging of the apex. This attenuation of the myocardium implicates mainly the subepicardium, although the entire right ventricular free wall can be involved (Figure 16). Thrombosis of the aneurysm is common (Figure 17). Even without aneurysm, extensive mural thrombosis in the lower part of the left ventricle and in the dilated right auricle may be seen (Figure 18). The presence of thrombi explains the frequent occurrence of thromboembolic phenomena in the pulmonary and systemic circulation (13). The myocardium of the lower parts of both ventricles shows diffuse small stellate fibrotic scars. Atrophy and fibrosis of the trabeculae carneae of both apical regions as wells as of the papillary muscle is a notorious and frequent finding. The coronary arteries are usually dilated with an increased capacity as compared with normal controls and showing a low incidence of atherosclerotic plaques (14). As well, it has been demonstrated that chagasic patients with chronic cardiopathy have abnormal endothelium-dependent coronary vasodilatation (15). The micropathology reveals focal and diffuse chronic fibrosing myocarditis (Figure 19). Foci of myocardial micronecrosis are present and associated with an inflammatory infiltrate composed predominantly of lymphomononuclear cells and interstitial fibrosis (Figure 20). Residual myocytes are present in the subepicardium, entrapped within the fibroadipose tissue, in the areas of right ventricular attenuation or replacement. The interstitial fibrosis is one of the most prominent features, mainly perimysial (16,17) (Figures 21 and 22). The increase in myocardial fibrosis could be directly related to an inflammatory reaction mainly composed of T lymphocytes and macrophages (17). These inflammatory cells could promote fibrosis by releasing mediators such as growth factors and cytokines, which act on fibroblasts. The pattern of myocardial fibrosis in chronic Chagas' heart disease probably reflects the pathogenic mechanisms involved. In addition, this remodeling of the collagenous matrix leads to progressive myocardial decompensation by decreased cardiac output, combined with an increased workload due to myocardial stiffness. The conduction system shows the same inflammatory and fibrotic lesions found in the myocardium (18). Although parasites and *T. cruzi* antigens can be immunohistochemically detected in the myocardium in chronic Chagas' disease (19,20), the frequency in each particular case is very low. Importantly, there is a lack of correlation between their presence and the myocardial changes, particularly the foci of micronecrosis and the inflammatory infiltrate. These observations suggest a role for a persistent antigenic stimulation throughout the chronic phase in the pathogenesis of myocardial changes. Moreover, destruction of the parasympathetic nervous system in the heart has been demonstrated. Recently, chronic inflammatory lesions of the mediastinal paraganglia very likely contributing as arrhythmogenic factor were described in chronic chagasic hearts (21).

8. PATHOGENESIS

The pathogenesis of chronic chagasic cardiopathy is still not totally understood. Different mechanisms have been proposed to explain its etiology.

8.1. Direct tissue destruction by *Trypanosoma cruzi*

The existence of different clinical forms of the disease was soon identified, which was at first thought to be associated with differences in the parasites implicated. Indeed, even Chagas noticed a peculiar dimorphism, the so-called slender and stout forms of the parasite in the bloodstream, observations later confirmed by many others (22,23). Today, these two morphological forms are believed to emerge from epigenetic phenomena and their pathological relevance is obscure (24). At a very early stage, the idea of a major role of differential tissue tropism in the pathogenesis of Chagas' disease was proposed (25,26). This idea has persisted in spite of only tenuous evidence based mainly on the parasite distribution in different tissues in the acute phase of experimentally infected animals (27-29).

In chronic Chagas' disease, parasites were rarely found in tissues examined by routine techniques (10,30). However, parasites antigens have disclosed in the myocardial tissue by application of immunohistochemical techniques (20) and of sensitive polymerase chain reaction (PCR) to the study of Chagas' disease (31-33). These observations give support for a role of a persistent antigenic stimulation throughout the chronic phase in the pathogenesis of myocardial changes. New studies may highlight the primary role of *T. cruzi* in the pathogenesis of Chagas' disease and set the stage for establishing the notion that genomic variation of *T. cruzi* could influence the course of the disease.

8.2. Autonomic abnormalities

The autonomic nervous system of patients with Chagas' disease has been extensively studied (10,14,34,35). The chronic cardiomyopathy of Chagas' disease could be a neurogenic form of heart disease promoted by the destruction of the parasympathetic ganglions cells in the heart (34). Early morphologic investigations revealed a conspicuous reduction in the number of cardiac parasympathetic neurons of patients who had died from

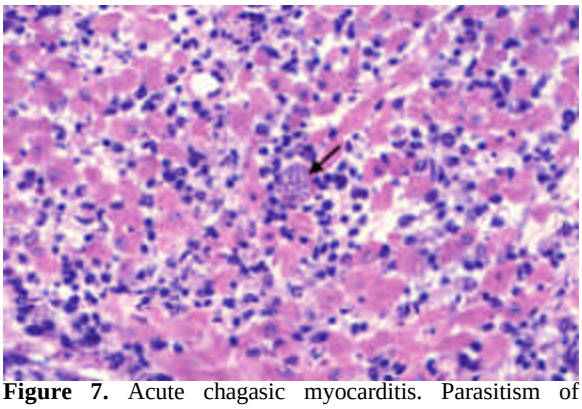

Figure 7. Acute chagasic myocarditis. Parasitism of myofibers is an outstanding feature (arrow).

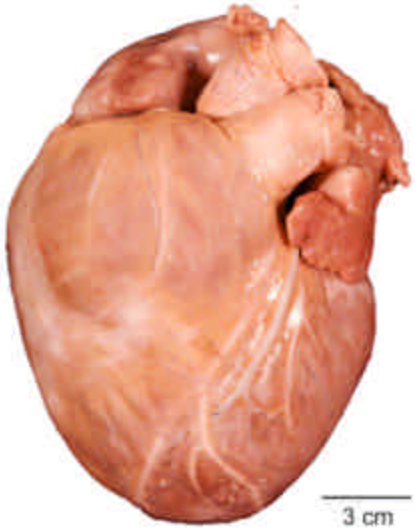

Figure 8. Globally enlarged chronic chagasic heart (anterior external view).

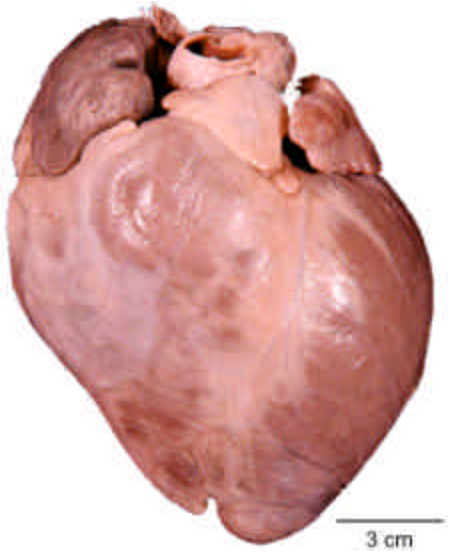

Figure 9. Globally enlarged chronic chagasic heart with a separation between the left and right apex giving the heart the aspect of "cor bifidum'.

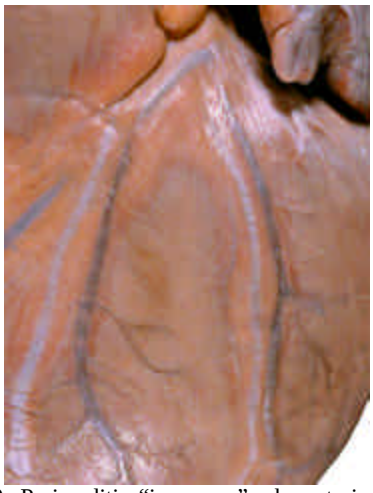

Figure 10. Pericarditis "in rosary", characterized by tiny white granules along the subepicardial coronary vessels.

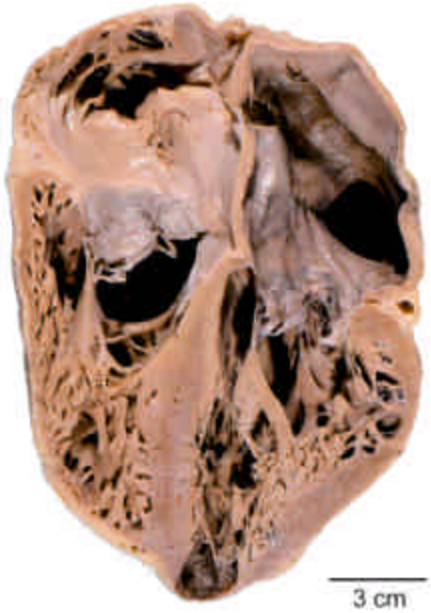

Figure 11. Four-chamber frontal section of globally enlarged chronic chagasic heart (dilatation and hypertrophy) with an apical aneurysm of the left ventricle.

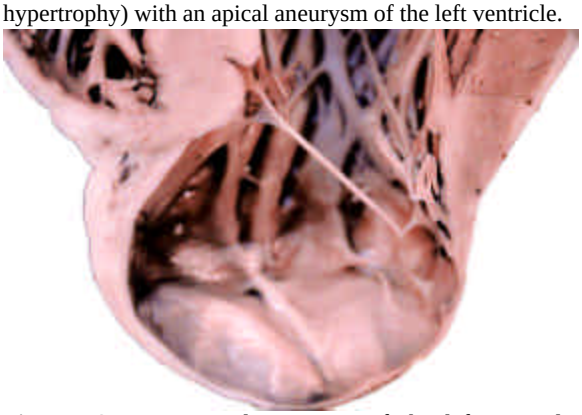

Figure 12. Huge apical aneurysm of the left ventricle, which wall consists mainly of endocardium and epicardium.

Figure 13. Stereomicroscopic view of an apical aneurysm of the left ventricle which wall is mainly composed of endocardium and epicardium with a few myocardial cells interposed.

Figure 14. Thinning of the right and left ventricular apices.

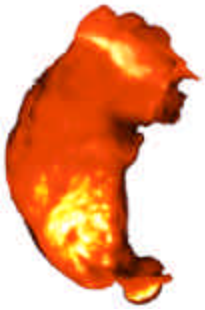

Figure 15. Transillumination of a chronic chagasic heart showing thinning of the muscle wall, "cor bifidum" with aneurysm at the left apex, and marked thinning of the anteroapical region of the right ventricle.

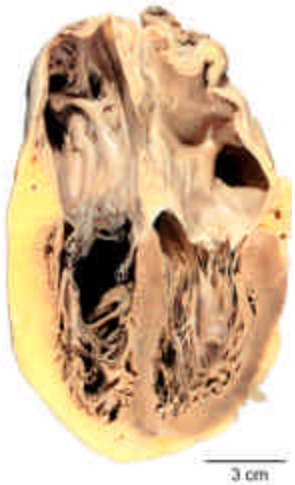

Figure 16. Four-chamber frontal section of globally enlarged chronic chagasic heart with dilatation mainly affecting the right-sided chambers. Adipose replacement of the right ventricular myocardium, particularly at the apical region, associated with bulging can be seen.

Figure 17. Thrombosis of a left ventricular apical aneurysm.

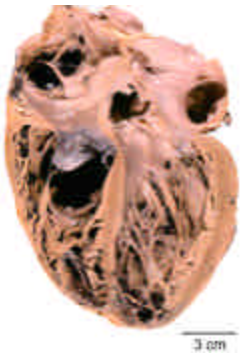

Figure 18. Four-chamber frontal section of chronic chagasic heart showing cardiomegaly with thinning and thrombosis at apices of both ventricles.

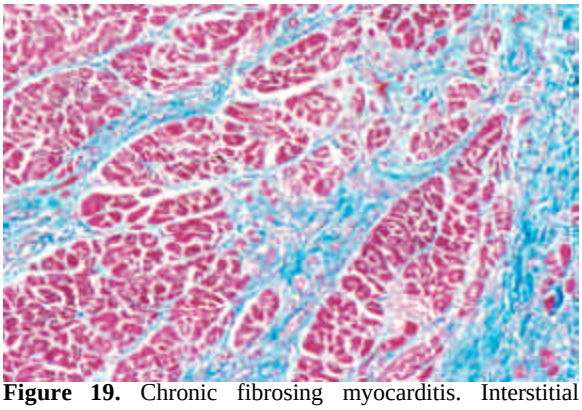

Figure 19. Chronic fibrosing myocarditis. Interstitial fibrosis associated with lymphomononuclear infiltrate (Gomori trichrome staining).

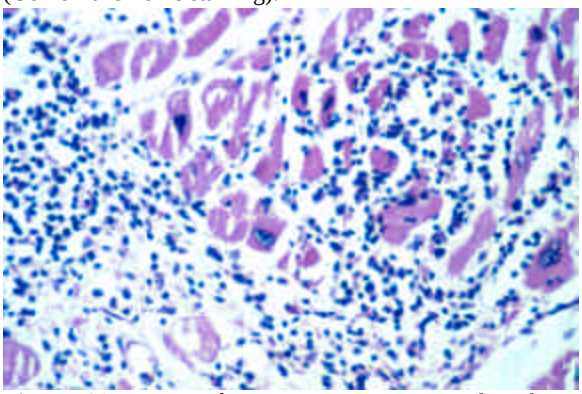

Figure 20. Focus of micronecrosis associated with an inflammatory infiltrate mainly composed of lymphomononuclear cells (H&E).

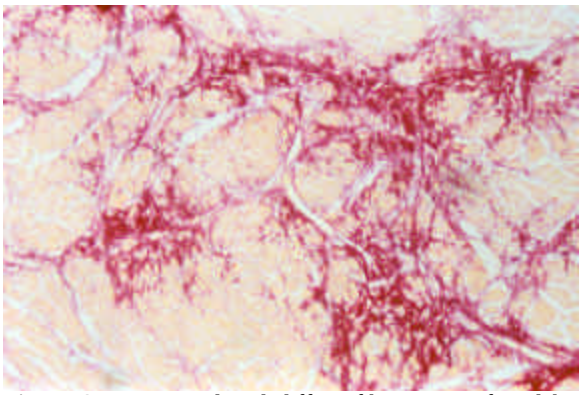

Figure 21. Interstitial and diffuse fibrosis manifested by increased amount of thick collagen fibers surrounding muscle fiber bundles (perimysial matrix) and around intramural coronary vessels, combined with less pronounced increase in the endomysial matrix (Picrosirius red staining).

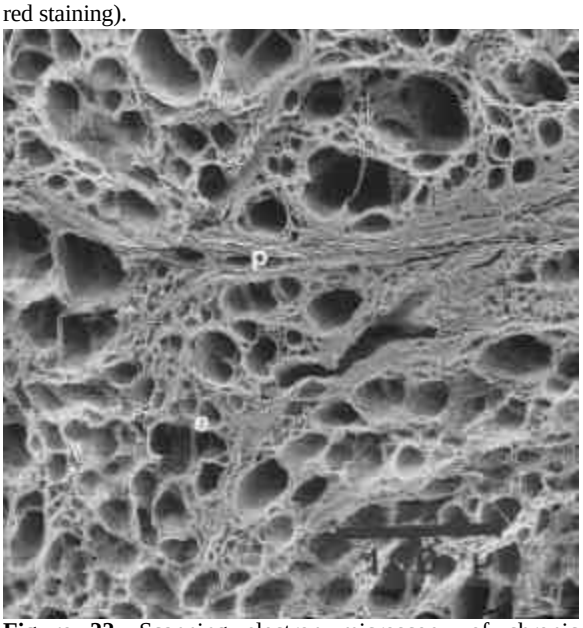

Figure 22. Scanning electron microscopy of chronic chagasic myocarditis showing broad bundles of perimysial collagen fibers (p) surrounding muscle fiber bundles. The increase of pericellular collagen matrix (endomysial matrix) occurs mainly in the periphery of the muscle bundles (scanning electron microscopy after removal of the myocardial tissue non-fibrous elements).

intractable congestive heart failure (10,14). The extent of heart denervation seen in Chagas' disease has not been detected in any other cardiopathy so far studied, though a number of cardioneuropathies have been described (36). Abnormalities of autonomic heart rate control were also described in asymptomatic patients with cardiac enlargement on chest X-rays (37-39). These functional abnormalities were subsequently confirmed by numerous clinical studies (40-42). Therefore chagasic patients clearly have abnormalities of the cardiac autonomic nervous system (43). On the other hand, a close relationship between altered autonomic tone and sudden cardiac death is well demonstrated (44,45). Malignant ventricular tachyarrhythmias (ventricular tachycardia, ventricular fibrillation) are major causes of sudden death among patients with Chagas' heart disease (46,47). The destruction of the parasympathetic innervation could induce an increased sympathetic tone that may have either a direct effect in arrhythmogenesis via altering the electrophysiologic properties of the heart or an indirect effect via other mechanisms such as increased oxygen demand by catecholamines, increased coronary vasomotor tone, and augmented platelet adhesiveness (45). Besides, the possible sympathetic overdrive would cause hypoperfusion and consequent myocytolytic necrosis. In fact, myofibrillar degeneration and extensive loss of contractile elements have been shown in the atrial myocardium of parasympathetically denervated monkey heart (48). The main dilemma of the neurogenic theory remains in the uncertainty about its physiopathologic mechanism. What remains to be determined is why and when do these abnormalities appear in the natural history of Chagas' disease (49).

8.3. Role of autoimmune mechanisms

The participation of autoimmune mechanisms in the genesis of the chronic myocarditis of Chagas' disease has been postulated (50-52). The lymphomononuclear infiltration in the absence of detectable parasites in the affected heart tissue and the demonstration that sensitized T cells from chronically infected mice were able to transfer myocarditis into normal recipients (53) strongly suggest the involvement of autoimmunity in the pathogenesis of chronic chagasic myocarditis. Organ-specific autoimmunity is often related to the outcome of many parasitic infections caused by virus, bacteria, and parasites (54,55). However, the establishment of a true organ-specific autoimmune nature for Chagas' disease myocarditis had been depending on an experimental model that could provide evidence in support of the hypothesis and allow a reliable readout of immune manipulations by which different sets of lymphocytes could be implicated in the generation of the disease. This has been provided by recent investigation undertaken to study the autoreactivity against syngeneic heart tissue *in vivo* by grafting newborn hearts into the pinna of the ear of mice chronically infected with *Trypanosoma cruzi* (56). Mice chronically infected were able to reject syngeneic newborn heart grafts in 20-30 days. The rejection of syngeneic hearts in chronically infected adult mice was faster when compared with the full allogeneic situation. This was in striking contrast to organs grafted into normal syngeneic recipients, which can persist and beat for up to one year (57). The histologic evaluation of heart tissue grafted into chagasic mice revealed intense mononuclear inflammatory infiltrate associated with myocytolytic necrosis, quite similar to the pattern obtained when hearts are transplanted in allogeneic conditions. Besides, before grafting the heart tissue, depletion of CD4 but not CD8 T cells neutralized rejection. Finally, CD4 T lymphocytes from chronically infected mice injected *in situ* were able to promote rejection, whereas CD8 or non-T cells were not effective. These findings suggest that autoimmunity is the major mechanism implicated in the rejection of syngeneic heart tissues grafted into the pinna of the ear of mice chronically infected with *T. cruzi.* The bulk

of the results establishes that autoreactivity is restricted to the CD4 T cell compartment, which is clearly different from allogeneic skin graft rejection that has been attributed to both subsets of lymphocytes (58,59). This is in agreement with other experimental models of organ-specific autoimmune diseases, in which CD4 T cells play a major role in the induction of tissue lesions (60). The analysis of T lymphocyte populations in the inflammatory infiltrates in Chagas' disease have shown that T cells represent about 5% of the total mononuclear cells present in the chronic inflammatory infiltrates, the majority of them being CD4 (61). Although the cellular response is T lymphocyte-dependent, the macrophages appear to be the most important effector cells in a chronic chagasic myocarditis (62), where they form the bulk of the cellular infiltrate (51,62,63). CD4 lymphocytes have been reported to modulate antibody production (64), macrophage activation (65), peripheral nerve injury (66), and to perform the recruitment of monocytes into the tissue to constitute inflammatory infiltrates (67).

One question that has arisen from these data refers to the triggering of the autoreactive CD4 lymphocytes. This could be the result of a response to the constituents of *T. cruzi* cross-reactive with myocardial tissue (68) or could be dependent on sensitization to parasite antigens (69,70). Since polyclonal activation of T and B cells is an important component during the acute phase of Chagas' disease, it has been proposed that such activation could play an important role in the development of autoimmune disease *a posteriori* (71). So far, we favor the hypothesis that extensive tissue damage and immune dysfunction during the acute phase are absolutely necessary for the appearance of organ-specific autoimmunity during the chronic phase. In this context, important issues to be considered are the factors that contribute to the progression from the acute to the indeterminate phase of Chagas' disease and the factors implicated in the progression from the indeterminate to the chronic phase of the disease. It has been proposed that impairment of the immunologic supressor network could play a role in the pathogenesis of myocardial lesions in both acute and chronic phase of Chagas' disease (72,73).

8.4. Role of microvascular changes

Microvascular changes have been proposed to play a significant role in the pathogenesis of the chronic cardiomyopathy of Chagas' disease (74-80). The focal nature of the myocytolic necrosis with associated interstitial fibrosis and inflammatory mononuclear infiltrate in chronic chagasic myocarditis suggests that the microcirculation could be involved, i.e., the primary site of disease could be at a level capable of causing focal necrosis of cells in discrete groups. Platelet aggregates and thrombi in the coronary microvasculature and histochemical evidence of the presence of foci of myocardial hypoxia were observed in mice chronically infected with the Colombia strain of *T. cruzi* (74,75). These observations led to the hypothesis that the consequences of infection converge on the myocardial microvasculature, resulting in areas of hypoperfusion, foci of myocytolytic necrosis, replacement fibrosis and surrounding cellular hypertrophy. Since then, a variety of abnormalities of the coronary microcirculation have been demonstrated in both human and experimental Chagas' disease. Focal vascular constriction, microaneurysm formation, dilatation and proliferation of microvessels, have been demonstrated in acute experimental Chagas' disease in mice (76), similar to those found in other congestive cardiomyopathies (81-83). These vascular lesions were observed early in the evolution of experimental Chagas' disease, even before there was significant myocardial degeneration or fibrosis. Marked basement membrane thickening in myocardial capillaries (up to 20 times the normal thickness) has been detected in chronic chagasic human hearts (84,85), similar to the thickening, either with or without multilayering, reported to occur in basement membranes of capillaries of patients with diabetes mellitus (86) and myxedema (87), in KK mice with genetically-transmitted cardiomyopathy associated with diabetes mellitus (88), and in renovascular hypertensive streptozotocin diabetic cardiomyopathic rats (89). Long-term administration of verapamil, a calcium channel blocker drug, markedly decreased myocardial inflammation and fibrosis in *T. cruzi* chronically-infected mice, producing a ten times decrease in mortality rate during a 70-day period of infection (90), probably through the maintenance of vascular perfusion due to its vasodilatatory action on smooth muscle cells and to its inhibitory effect on platelet aggregation (82,91). *In vitro* studies of infection demonstrated increased platelet adherence and aggregation related to infection-associated endothelial cell dysfunction (92). Furthermore, the elevated plasma levels of thromboxane A_2 detected in mice infected with *T. cruzi* have been implicated in increased intravascular platelet aggregation and microvascular spasm (92). A decrease in myocardium perfusion could be detected by thallium-201 scintigraphy in human chronic Chagas' heart disease (93,94), and in hearts of mice after 15 and 30 days of infection with *T. cruzi* (79). On the other hand, in the general population of chagasic patients, epicardial coronary arteries are usually clear of obstruction. Atherosclerotic coronary artery disease has been seen in patients after an episode of acute myocardial infarction, but the patterns of lesions in these patients are similar to those seen in the nonchagasic study population (95). Therefore, it is not clear that link, if any exists between *T. cruzi* infection and these changes.

9. HYPOTHESIS

The evidence provided by both human and animal studies on chronic Chagas' heart disease suggests that the pathogenesis of the cardiomyopathy occurs as a consequence of several physiopathological processes occurring after infection interacting with unidentified host factors. The development of the chronic fibrosing myocarditis is related to progressive and additive focal cellular necrosis, and associated inflammatory lymphomononuclear infiltrate and reactive and reparative myocardial fibrosis and surrounding myocyte hypertrophy. These processes may be initiated and perpetuated by alterations in the myocardial microcirculation and by autoimmune factors. The intrinsic and extrinsic cardiac nervous system impairment and/or the chronic fibrosing

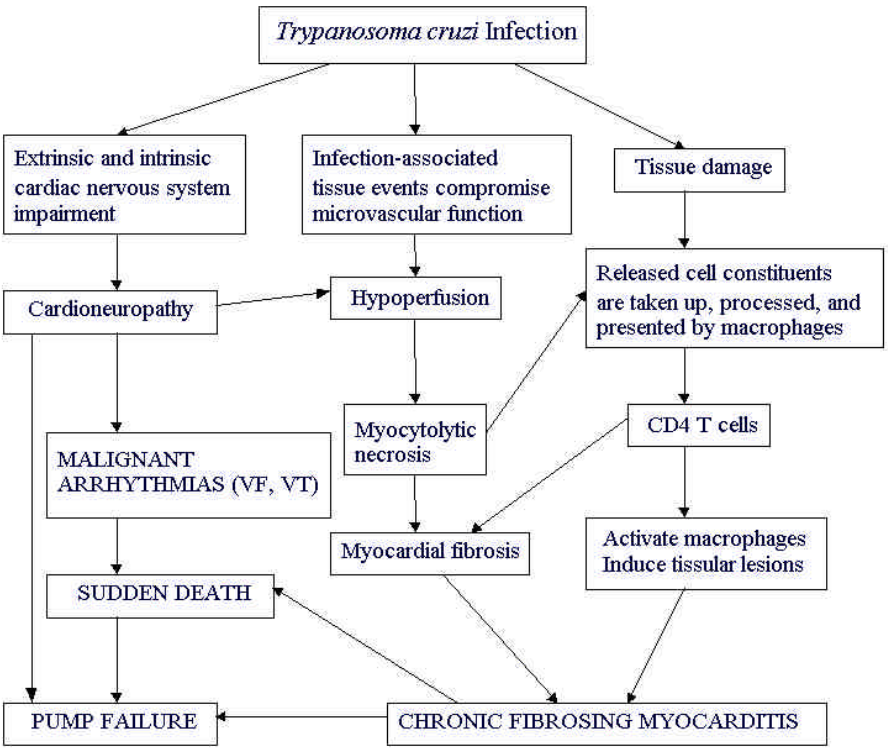

Figure 23. Diagrammatic representation of pathogenic mechanism of chronic Chagas' heart disease.

myocarditis and the left ventricular dysfunction could act as factors predisposing one to an increased risk of sudden cardiac death (Figure 23).

10. CLINICAL MANIFESTATIONS

The clinical picture of Chagas' cardiomyopathy is characterized by congestive heart failure, dysrhythmias, atypical chain, thromboembolic phenomena and sudden cardiac death. In patients with congestive heart failure, right-sided and left-sided heart failure are usually found, but isolated left-sided heart failure may be also observed. Isolated right-sided heart failure is virtually non-existent in chagasic patients. In most cases, congestive heart failure is mild to moderate in intensity. Nevertheless, patients with the severest form of the disease may also be seen on ambulatory basis. In the vast majority of cases, congestive heart failure is secondary to left ventricular systolic dysfunction (96). Left ventricular diastolic dysfunction may be observed in some patients with Chagas' disease at the early stages of the disease when patients are asymptomatic, but overt congestive heart failure secondary to diastolic left ventricular dysfunction has never been detected in chagasic patients thus far. The prognosis of patients with heart failure is unrelenting and worse than that of patients with dilated cardiomyopathy (97), perhaps reflecting the extensive myocardial fibrosis detected in chagasic patients (98). In chagasic heart failure, systolic blood pressure is an independent predictor of mortality (99,100), with patients having a systolic blood pressure less than 120 mmHg at higher risk (98).

Dysrhythmias, clinically manifested by palpitations, are frequently detected in patients with Chagas' cardiomyopathy. When a resting ECG is taken, isolated premature ventricular contractions predominate, although, bigeminal, multiform, pairs of premature contractions and nonsustained ventricular tachycardia are frequently seen in the resting ECG. A study using 24-h Holter monitoring has suggested that the complexity of ventricular arrhythmias depends on the severity of left ventricular dysfunction (101). Nonetheless, complex ventricular arrhythmias may be detected in chagasic patients without myocardial as well (102). Sustained ventricular tachycardia is uncommonly observed in cohorts of patients with chronic Chagas' disease (103). When present, sustained ventricular tachycardia has been associated with either mild left ventricular dysfunction (104) or the presence of left ventricular apical aneurysm (105). Exercise stress testing is also useful to disclose latent sustained ventricular tachycardia (106). Another frequent arrhythmia is atrial fibrillation, which may be detected in about 20% of patients followed at a tertiary referral center (96).

Despite the potential to herald a lethal outcome, the impact of such arrhythmias on the prognosis of patients with Chagas' disease is still obscure. High-degree AV blocks may be manifested by spells, syncopal episodes or self-perception of bradycardia. Because trifascicular block may be found in about 45% of patients in a hospital derived-cohort (96), physicians should be alert for this potentially lethal condition, which can be easily be treated with pacemaker implantation.

Atypical chest pain is usually found in about 15% of patients with chronic Chagas' disease (107). Although in the vast majority of cases the chest pain is not associated with emotional distress or physical exercise and not alleviated by sublingual nitrate, in some occasions the pain may stimulate a clinical picture of unstable angina. The mechanism underlying atypical chest pain is still obscure, but vasospasm is an attractive possibility because abnormalities in the coronary artery tonus have been detected in patients with Chagas' cardiomyopathy (15). The presence of atypical chest pain has been associated with overt acute myocardial infarction (107); therefore, such patients should be closed followed.

Thromboembolic phenomena, particularly pulmonary embolism and cardioembolic stroke, are frequently found in autopsied patients (13). In the clinical setting, pulmonary embolism is usually seen in patients with advanced heart failure. In patients with less severe forms of the disease, pulmonary embolism is seldom observed (96). The same can be said for cardioembolic stroke, whose prevalence is low in hospital-derived cohort of patients with Chagas' cardiomyopathy (108). Sudden cardiac death is another complication for patients with Chagas' cardiomyopathy. In most instances, sudden cardiac death occurs in patients with overt heart disease, but patients in indeterminate form of the disease may experience sudden cardiac death as well (109). Risk factors for sudden cardiac death determined by multivariate analysis are the atypical aneurysm and left ventricular diastolic dimension on echocardiography (100). Surprisingly, the role of premature ventricular contractions as a precursor of sudden cardiac death has never been established in patients with Chagas' cardiomyopathy. In chagasic patients, sudden cardiac death usually occurs as a result of ventricular fibrillation, which might be the consequence of an autonomic imbalance due to intracardiac or extracardiac parasympathetic denervation (98). Alternatively, left ventricular dilatation might elicit abnormal cardiac reflexes (110) leading to asystole and sudden cardiac death.

11. DIAGNOSIS

The diagnosis of the acute disease is made by detecting parasites in fresh blood, and serologic tests for anti-*T. cruzi* IgM. The diagnosis of the chronic form of the disease is made by detecting IgG antibodies that bind specifically to parasite antigens. Several sensitive serologic tests are used widely in Latin America. These tests often show cross-reactivity with illnesses such as malaria, leishmaniasis, and syphilis among others. PCR-based tests are currently under development.

12. TREATMENT

Symptoms of congestive heart failure are usually treated with diuretics. When ventricular dilatation is detected, even in the absence of overt heart failure, angiotensin converting enzyme inhibitors at maximal tolerated dose may be given. Nontheless, it should be stressed that no randomized clinical trial using such drugs has been carried out in patients to determine their impact on survival. Importantly, chagasic patients tolerated lower doses of angiotensin converting enzyme inhibitors in comparison to nonchagasic patients (97). Thus, the beneficial effect of such drugs in chagasic patients has not yet been established. Digitalis is useful in improving symptoms in patients with advanced heart failure, with no unfavorable impact on prognosis (96). The treatment of premature ventricular contractions is empiric. Amiodarone, in doses of 400 to 600 mg daily, has been found to be useful to suppress complex premature ventricular contractions (111). The impact of amiodarone on the prognosis of chagasic patients is unknown. Sustained ventricular tachycardia can be treated with either lidocaine or direct coutershock with good result (104). Marked bradycardia or advanced AV block is effectively treated with permanent or temporary pacemaker implantation; atropine, on the other hand, is of little value for chagasic patients because of marked nodal derangement (112). The automatic implantable defibrillator has been used in patients with sustained ventricular tachycardia or ventricular fibrillation with good preliminary results, but its impact on prognosis, particularly on the sudden cardiac death rate of chagasic patients remains to be determined.

There is no adequate treatment for atypical chest pain of chagasic patients. Anticoagulation may be recommended to patients with atrial fibrillation to avoid cardioembolic stroke. A recent randomized clinical trial has shown a beneficial effect of benznidazole in patients in the early stages of the disease (113). However, taking account the potential detrimental effects of benznidazole (114), the small number of patients enrolled and the obscure clinical significance of serological changes observed in that study, the use of this drug to treat chagasic patients is still disputable (115).

13. PREDICTORS OF MORTALITY IN CHRONIC CHAGAS' HEART DISEASE

A rational approach for stratifying chagasic patients according to risk factors for mortality has been proposed (Figure 24). In general unselected populations living in areas where the disease is endemic, a simple resting ECG should be performed in all patients with a positive serological test for Chagas' disease. Patients in NYHA class II or greater and asymptomatic patients who present abnormalities related to pathological Q waves and/or intraventricular conduction defects, specially left anterior fascicular block, should be sent to tertiary referrals centers for full evaluation. The remaining patients, because of the minimal risk of mortality, should be followed up annually or biannually, and allowed to work without restriction. At referral centers, patients should undergo 2-D echocardiogram. Those with a normal or mildly depressed left ventricular ejection fraction should be followed up annually, without performing more detailed tests. However, those with moderately depressed left ventricular ejection fraction should undergo 24h Holter monitoring to assess ventricular premature contractions complexity. In the presence of complex ventricular dysrhythmias, such patients should be followed up closely because of the risk

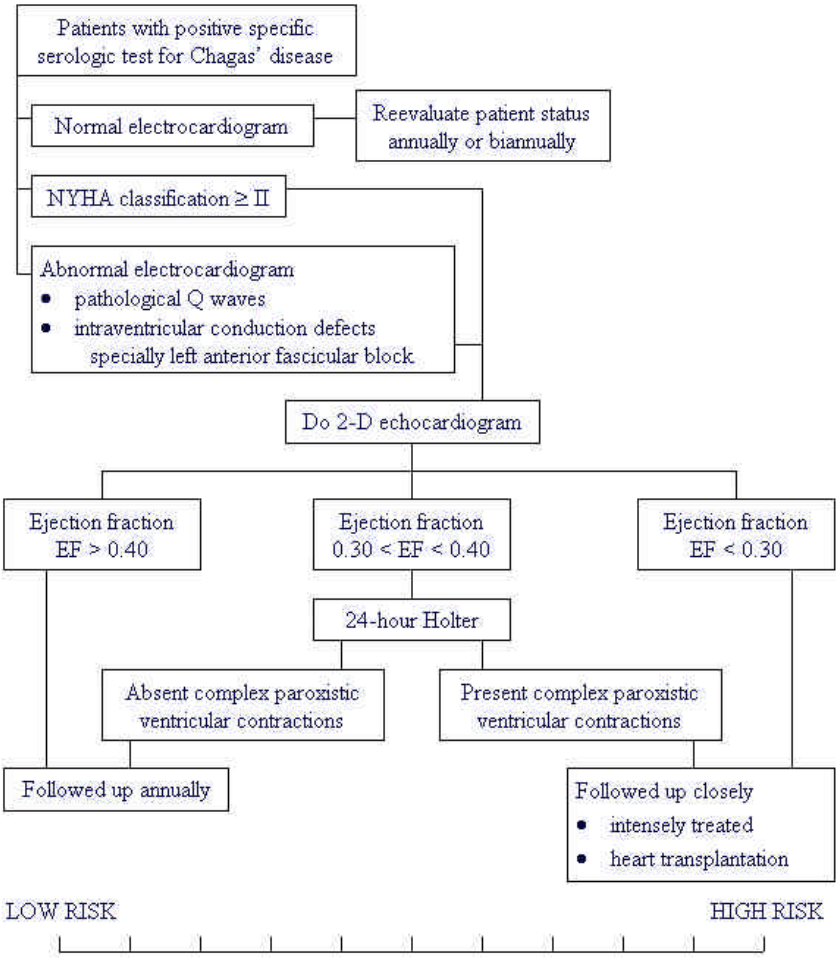

Figure 24. Prognostic stratification of chagasic patients according to risk factors for mortality.

of sudden death. Moreover, the same can be said with regard to patients with left ventricular dilatation, especially those with left ventricular diastolic dimension greater than 70mm, and apical aneurysm. Unfortunately, the best treatment for this subset of patients is not known at present. Patients with left ventricular ejection fraction <0.30, even in the absence of congestive symptoms, should be treated intensely and be considered for transplants, taking into account the good current results of heart transplantation in chagasic patients (114). By doing this, we can adequately select chagasic patients who really need treatment. Future studies contemplating the treatment of heart failure and sudden cardiac death should focus on such patients.

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Send correspondence to: Marcos A. Rossi, MD, Ph.D., Professor of Pathology, Department of Pathology, Faculty of Medicine of Ribeirão Preto, University of São Paulo, 14049-900 Ribeirão Preto, SP, Brazil, Tel/Fax: +55 16 602 3130, E-mail: marossi@fmrp.usp.br