Drugs for the Etiologic Treatment of Chagas Disease: Myths and Truths

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Abstract

Chagas disease is a life-threatening illness caused by the protozoan parasite *Trypanosoma cruzi*. One of the characteristics of chronic chagasic infection is the parasitic persistence in cardiac and smooth muscle tissues. For this reason etiologic treatment of Chagas disease in all phases of infection is highly recommended. Despite the high number of trypanocidal drugs that have been discovered in the last years, only two compounds, Benznidazole and Nifurtimox remain as the unique drugs approved for Chagas treatment. Far from ideal, these drugs display low sensitivity and specificity resulting in limited applications, mainly in the onset of the acute phase. Thus there is an urgent need to validate new anti- *T. cruzi* drugs that can be applied even in the cases of chronic patients, those who today have no safe and effective treatment available. This paper reviews the most important compounds that have been tested in clinical trials and the results obtained to date.

Key words: Chagas disease, treatments for chronic phase, Benznidazole, Posaconazole, Cruzipain inhibitors.

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Introduction

Chagas disease (CD) or American trypanosomiasis is a vector-borne parasitic disease belonging to the group of neglected tropical diseases. Many have been neglected in terms of funding, research and policy. According to the most recent WHO estimates there are presently around 6 to 8 million people infected with *T. cruzi*, mostly in Latin America, where the insect vector is prevalent[1]. Each year 56,000 new cases of infection and 12,000 deaths linked to CD are seen[2] in endemic areas. CD is mainly transmitted through a blood-sucking insect commonly known as the "kissing bug". During the vector meal, parasites present in the insect faeces are deposited in the mucosa or damaged skin followed by parasite penetration into host cells. Infection may also be produced by congenital transmission, blood transfusion, organ transplantation, or be transmitted orally (by contaminated food or drinks). This parasitism common in underdeveloped countries has in the past been a marker of poverty because the bugs grow in adobe houses with thatched roofs. Nowadays, with the globalization process, CD has expanded to several countries around the world, mainly related to organ transplants and blood transfusions, especially from Latin American donors.[3]

Chagas disease involves two phases:

i - The initial, acute phase is characterized by a high number of parasites circulating in the blood and mild nonspecific symptoms (fever, headache, enlarged lymph glands, pallor, muscle pain, swelling and others) or no symptoms at all. Only 10% of people bitten by a triatomine bug develop typical visible signs of skin lesion or the painless periorbital swelling of one eye, called "signo de Romaña". Acute infection is a self-limited illness and lasts 2 months after infection.

ii - The chronic phase develops in approximately one third of patients two or three decades after the acute infection. It is asymptomatic in around 70% of the patients. The chronic phase also has two phases, most frequently an indeterminate phase, characterized by positive serology without organ impact and the properly chronic phase, which shows both positive serology and organ impact. During the chronic phase, the parasites are hidden mainly in the heart and digestive muscles. Up to 30% of patients suffer cardiac disorders and up to 10% suffer digestive (typically enlargement of the esophagus or colon), neurological or mixed alterations. In later years the infection can lead to sudden death or heart failure caused by progressive destruction of the heart muscle or nervous system. The kind of clinical manifestation mainly depends of the particular *T. cruzi* strain. In Argentina the most frequent are *T. cruzi* genetic subtypes V and VI, with prevailing heart infection, in Brazil *T. cruzi* subtype II prevails that typically produces digestive infections.

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The acute disease can be cured with trypanocidal treatments. In Argentina the National Program of Chagas recommends specific treatment in the following cases.[4]

- all patients in the acute phase,
- patients under 15 years,
- reactivations in immunocompromised patients (HIV, post transplantation, autoimmune diseases),
- laboratory or surgery accidents,
- to evaluate treatment of chronic patients, since there is evidence
- that etiologic treatment prevents progression or complications of the disease in this group of patients.

Etiologic treatment in the chronic phase of the infection has always been controversial because the belief of the autoimmune origin of chronic myocarditis and the absence of the parasite in tissues. In current medical practice, most physicians only prescribe palliative treatment for adult Chagas patients with dilated cardiomyopathy. In contrast to this old paradigm, specific antiparasite treatment for all chronic-phase *Trypanosoma cruzi*-infected individuals is mandatory.[5] There is an important body of evidence that strongly suggests the involvement of parasitic presence in the disease progression toward cardiomyopathy.[6,7,8]

Approved treatments for Chagas disease

As mentioned above, only two approved drug treatments are available, Benznidazole (BZN) and Nifurtimox (NTX), and more than four decades have gone by since their discovery. Nifurtimox's action mechanism is not completely understood. Reduction of its nitro group to unstable anionic radicals that leads to the formation of reactive oxygen species is the most probable effect, since *T. cruzi* lacks efficient detoxification mechanisms for such metabolites and is very sensitive to oxidative stress.[9-10] Additional molecular mechanisms involving cytotoxic nitrile metabolites have also been reported.[11-12] NTX is indicated in doses of 8-10 mg/kg/day in adults and adolescents and 10 to 12 mg/kg/day in infants and children, divided in three doses for 60 or 90-120 days depending to the countries specifications.[13-14] The maximum dose is 700 mg in 24 hours.

Benznidazole's mode of action involves covalent modification of biomolecules owing to reactive intermediates from nitroreduction.[9,15] BZN doses are 5-7 mg /kg/day orally, divided into two daily doses for 60 days. Both treatments are contraindicated in patients with severe liver, renal or neurological disease and also in pregnancy and lactation. BZN is considered the first-line treatment, while NTX is reserved for those patients that do not tolerate the former.[14-16]

The main limitations of these drugs are their very frequent side effects, which occur in up to 40% of the patients[1] with adults being more prone than children.[10,13,17] During the first two weeks of treatment nausea, abdominal pain, headaches, dizziness and rashes are common but are usually benign. Paresthesiae or symptoms of peripheral polyneuritis are dose-related effects that can cause discontinuation of treatment. Other adverse effects include leukopenia or agranulocytosis. These drugs should not be combined with alcohol, because they produce unpleasant symptoms immediately after alcohol consumption similar to those of Disulfiram. Patients should be strictly monitored for early detection of the occurrence of adverse effects from the first week of treatment. Other issues related to these medications include the length of the treatment regimes,[13, 14] accessibility problems and the low cure rate in chronic stage (10-20%).

Clinical Trials for Chagas disease

To determine the real efficacy of Benznidazole to produce parasitological cure and to interrupt the disease progression in the chronic phase of infection, two randomized clinical trials have been conducted.

The Etiologic Treatment With Benznidazole in Adult Patients With Chronic Chagas Disease (TRAENA) was a randomized, double-blind clinical trial conducted in Argentina with the purpose of studying whether BZN will be able to modify the natural evolution of chronic Chagas disease in adult patients (ClinicalTrials.gov NCT02386358). The study included 713 patients in the chronic phase of Chagas disease, who were treated with BZN (at a dose of 5 mg/Kg/day until 60 days have been completed or development of non-acceptable toxicity) or Placebo and were followed-up over 10 years. The results available to date show a high percentage of negative values for serum antibodies (by ELISA F29 and qPCR) in patients treated with BZN, through the years.[18.] In a sample of 243 patients of TRAENA a low grade of adhesion to treatment in some patients was also observed, especially in men, young people and in some cases of adverse effects, although this last characteristic was seen only in 16% of patients. Clinical results are currently concluding and final results not have been published to date.

The Benznidazole Evaluation for Interrupting Trypanosomiasis Trial (BENEFIT) is a prospective multicenter, randomized, double blind, placebo-controlled clinical trial of 3,000 patients with Chagas' cardiomyopathy enrolled in 35 centers from Argentina, Brazil, and Colombia (ClinicalTrials.gov NCT 00123916). Patients were randomized to receive benznidazole (5 mg/kg per day) or matched placebo, for 60 days. The average follow-up time will be 5 years.[19] The results of the study involving 2854 patients with Chagas' cardiomyopathy who received BNZ or placebo for up to 80 days show that trypanocidal therapy with benznidazole in patients with established Chagas' cardiomyopathy significantly reduced serum parasite detection but did not significantly reduce cardiac clinical deterioration through 5 years of follow-up.[20] In other words, in patients with established Chagas' cardiomyopathy, BZN treatment significantly reduced the detection of circulating parasites but did not reduce cardiac clinical progression. For this reason, they recommend treatment with trypanocidal therapy in the early stages of chronic Chagas' infection.

The most promising new treatments for Chagas disease currently being investigated in clinical trials are repurposed drugs. Drug repurposing consists in finding new uses or applications for drugs currently used for other pathologies. These strategies have the potential advantage of facilitating rapid and cost effective drug development because preclinical and clinical development can build on already available data (safety and other pharmaceutically relevant data). Among them, a number of azole antifungal drugs are maybe the most promising candidates.[21]

Posaconazole (POS) inhibits ergosterol biosynthesis; and, like fungi, *T. cruzi* depends on endogenous ergosterol and its derivatives which are crucial for normal functioning of the parasite membranes, cell division, growth and development.[22,23] Moreover, inhibition of the ergosterol biosynthetic pathway leads to cytotoxic accumulation of abnormal amounts of sterol precursors.[22] Posaconazole has demonstrated efficacy on several *T. cruzi* strains that are resistant to BZN and NTX such as Y and Colombian strains.[24] Back in 2012, it entered clinical trials in Argentina, Bolivia and Spain to assess the efficacy and safety of POS as compared with the efficacy and safety of BZN in adults with chronic *Trypanosoma cruzi* infection.

Randomly assigned patients received POS at a dose of 400 mg twice daily (high-dose posaconazole), POS at a dose of 100 mg twice daily (low-dose posaconazole), or BZN at a dose of 150 mg twice daily; all the study drugs were administered for 60 days. The results of this study demonstrated that POS was clearly inferior to the standard therapy, possibly due to low systemic bioavailability[21,25] Following that study, Merck started a second trial to investigate the combination therapy of POS and BNZ, which was concluded in 2014; results are still awaiting publication.[26] It has been pointed out that even if these trials were successful, POS is highly expensive due to the low yielding and costly synthetic scheme, which threatens its widespread use in endemic countries.[27] It is interesting to mention in this context, however, that POS has been proved to be very effective in at least one recent case of compassionate treatment. A patient with systemic lupus erythematosus, after being immunosuppressed, developed an acute infection from a hidden Chagas disease, and was successfully cured with Posaconazole.[28]

Ravuconazole and E-1224 (a Ravuconazole prodrug), two other antifungal azoles, have also displayed potent in vitro activity against *T. cruzi*. Despite the unfavorable pharmacokinetic profile of Ravuconazole in animals (characterized by a very short elimination half-life), its pharmacokinetic parameters in humans prompted a proof-of-concept clinical trial of E-1224.[29] E-1224 failed to develop sustained efficacy one year after the treatment in comparison with BZN and presented some safety issues at high doses.[28] Further trials of E-1224 as a combination therapy with BZN have been announced.[21]

Fexinidazole, is a nitroimidazol drug candidate currently under study for the treatment of human African trypanosomiasis. This compound has also been evaluated in experimental models of acute and chronic CD caused by different strains of *T. cruzi*. The study showed comparative effects between BZN and higher doses of Fexinidazole on Benznidazole-susceptible CL *T. cruzi* strain, and on the partially resistant Y strain, while a superior effect of Fexinidazole was observed on Benznidazole-resistant VL-10 and Colombian strains in mice models of acute and chronic infection.[30.] In the same study, Fexinidazole was also shown to reduce myocarditis in animals infected with VL-10 and the Colombian strain. These results set the basis for the beginning of a Phase II proof-of-concept trial to determine the efficacy of the compound in adults with chronic indeterminate disease, which is being held in Bolivia.

The vinylsulfone K777 is an inhibitor of cruzipain, the major cysteine-protease of *T. cruzi* and a good target of trypanocidal therapy due to its participation in parasite invasion and reproduction.[31] It has been proven to be effective in animal models of Chagas' disease, but discarded for clinical trials due to high hepatotoxicity in rats.[32] Other approved drugs chemically related to K777 are being studied nowadays. In virtual screening campaigns for computer-assisted drug repurposing, Bellera et al. have recently propose two new cruzipain inhibitors, Benidipine and Clofazimine. Both candidates significantly reduced the parasitemia and the number of *T. cruzi* cardiac nests in a murine acute model of *T. cruzi* infection.[33] Benidipine and Clofazimine were also able to reduce the parasite burden in cardiac and skeletal muscles of chronically infected mice compared with untreated mice as well as diminish the inflammatory process in these tissues.[34]

Concluding remarks

Although a better therapeutic alternative that improves the classical BNZ treatment for Chagas disease is still being sought, some useful conclusions have been derived from the clinical trials to date. Up to date, it is important to point out:

- Etiologic antiparasitic treatment during the chronic phase of *T. cruzi* infection is highly recommended,
- Benznidazole therapy in the early stages of chronic Chagas infection reduces the progression to cardiac complications,
- Posaconazole did not show more efficacy that conventional BZN treatment, although producing fewer side effects,
- Cruzipain inhibitors could be good candidates for acute and chronic Chagas treatment,
- Therapeutic options for symptomatic Chagas patients are still absent.
- Combination of BNZ (at low doses) with other trypanocidal compounds is a clever strategy whose results will be made clear in the next few years.

Finally, new research that proposes the use of BZN in combination with compounds directed to other targets (ergosterol biosynthesis, cruzipain activity, etc) are beiing studied in ongoing trials. The results of these studies are expected to be published in the ensuing years.

Declarations of Interest

The authors declare no conflicts of interest.

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