Prevalence of clinical forms of Chagas disease: a systematic review and meta-analysis – data from the RAISE study

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Summary

Background There is a lack of up-to-date estimates about the prevalence of Chagas disease (ChD) clinical presentations and, therefore, we aimed to assess the prevalence of clinical forms of ChD among seropositive adults, pooling available data.

Methods A systematic review was conducted in Medline, Embase, *Biblioteca Virtual em Saúde* and Cochrane databases looking for studies published from 1990 to August 2023, which investigated the prevalence of ChD clinical forms among seropositive adults, including: (i) indeterminate phase, (ii) chronic Chagas cardiomyopathy (CCM), (iii) digestive and (iv) mixed (CCM + digestive) forms. Pooled estimates and 95% confidence intervals (CI) were calculated using random-effects models. Studies quality and risk of bias was assessed with the Leboeuf-Yde and Lauritsen tool. Heterogeneity was assessed with the I^2 statistic. The study was registered in the PROSPERO database (CRD42022354237).

Findings 1246 articles were selected for screening and 73 studies were included in the final analysis (17,132 patients, 44% men). Most studies were conducted with outpatients (n = 50), followed by population-based studies (n = 15). The pooled prevalence of the ChD clinical forms was: indeterminate 42.6% (95% CI: 36.9–48.6), CCM 42.7% (95% CI: 37.3–48.3), digestive 17.7% (95% CI: 14.9–20.9), and mixed 10.2% (95% CI: 7.9–13.2). In population-based studies, prevalence was lower for CCM (31.2%, 95% CI: 24.4–38.9) and higher for indeterminate (47.2%, 95% CI: 39.0–55.5) form. In meta-regression, age was inversely associated with the prevalence of indeterminate ($\beta = -0.05$, $P < 0.001$) form, and directly associated with CCM ($\beta = 0.06$, $P < 0.001$) and digestive ($\beta = 0.02$, $P < 0.001$) forms. Heterogeneity was overall high.

Interpretation Compared to previous publications, our pooled estimates show a higher prevalence of CCM among ChD seropositive patients, but similar rates of the digestive form.

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Keywords: Chagas disease; Prevalence; Clinical forms; Meta-analysis

Research in context

Evidence before this study

Chagas disease is a potentially fatal multisystemic condition caused by the protozoan parasite *Trypanosoma cruzi*, which has two phases: acute and chronic. There is paucity of up-to-date data on Chagas' chronic clinical forms (indeterminate, cardiac, digestive, or mixed (cardiac + digestive)), and current estimates rely on individual epidemiological studies from the past 3 decades. We conducted a comprehensive systematic review looking for studies published from 1990 to August/2023, which investigated the prevalence of clinical forms of Chagas disease among seropositive adults, to update current estimates. The methodology and diagnostic modalities for evaluating clinical forms of ChD have evolved over the decades and, in a scenario of great heterogeneity, this allows for more accurate and reproducible metrics to be incorporated into epidemiological studies. The systematic search was performed in Medline, *Literatura Latino-Americana e do Caribe em Ciências da Saúde* (LILACS), through *Biblioteca Virtual em Saúde* (BVS), Cochrane, and Embase databases, and the MeSH terms were *("Chagas Disease" OR "Trypanosoma cruzi") AND (Prevalence OR Epidemiology) AND ("Clinical forms" OR "Indeterminate phase" OR "Acute phase" OR "Acute form" OR "Chronic phase" OR "Chronic form" OR "Arrhythmias, Cardiac" OR "Heart Failure" OR Thromboembolic OR Thromboembolism OR Thrombotic OR "Digestive manifestations" OR "Gastrointestinal Diseases" OR "Form associated" OR "Congenital form")*.

Added value of this study

Our findings suggest a higher prevalence of the cardiac form compared to previous data, however in population-based and blood-donor studies our results reflect previous estimates. This reinforces the need for resource allocation for the treatment of late sequelae of ChD cardiac form, including the wider availability of advanced therapies, such as implantable cardiac devices and catheter-based therapies for arrhythmias. In addition, our findings point towards the need for improvement of death registration systems, especially in endemic areas, for considering ChD as the underlying cause of several cardiovascular or ill-defined fatal events, and for refining mortality estimates. Finally, the results reinforce the progressive incidence of clinical forms with age in seropositive individuals. Thus, studies assessing strategies and specific therapies for refining the early identification and halting the progression of ChD clinical forms are of utmost importance.

Implications of all the available evidence

Our findings will allow for the incorporation of up-to-date pooled prevalence data into existing Chagas disease estimates, by applying meta-analytic methods to address the limitations of individual studies. These pooled data will contribute to the update of current estimates about global disease burden, associated sequelae, and mortality.

Introduction

Chagas disease (ChD), also known as American trypanosomiasis, is caused by the protozoan parasite *Trypanosoma cruzi*, named after the Brazilian physician and scientist Carlos Chagas, who discovered and almost fully described the disease—from basic science to epidemiology and clinical features—in 1909.[1] Despite being potentially fatal and with considerable morbidity associated, ChD is still part of the group of neglected tropical diseases (NTDs), by the definition of the World Health Organization (WHO).[2,3]

The main transmission pathway of ChD is via contact with the droppings of hematophagous insects of the subfamily Triatominae infected with *T. cruzi*. The disease can also be transmitted vertically, by blood transfusion, organ transplantation, accidents with biological materials, or by ingestion of food or drinks contaminated with the parasite—an occurrence that is progressively emerging as the main route of transmission in regions where vector control has improved, mainly due to socioeconomic development and better housing conditions.[1,2,4]

ChD is multisystemic and represented by two phases, acute and chronic. The acute phase tends to manifest with mild and nonspecific symptoms such as fever, fatigue, hepatosplenomegaly, and cutaneous nodules. Rare cases progress to myocarditis and meningoencephalitis. The chronic phase can last for years and is mainly characterized by indeterminate (asymptomatic), cardiac, digestive, or mixed forms.[3–5] The incidence of clinical forms of chronic ChD depends on a complex interaction between host and parasite, mostly driven by chronic inflammatory pathways in different tissues and organs.[6,7] Progression is frequently unpredictable. For some clinical presentations, notably chronic Chagas cardiomyopathy (CCM), the occurrence seems to be associated with age and time lived with the disease.[4,8]

WHO estimates a worldwide prevalence of 6–7 million individuals infected, mostly in Latin America, where the disease is still highly endemic.[1,3,7] These estimates, however, are highly uncertain, mainly due to underreporting and underdiagnosis of the disease, which predominantly affects areas of social vulnerability and poor health. In addition to prevalence, mortality information also seems to be unprecise, with very few reports in endemic areas such as Central America.[3]

Moreover, with increases in migration, ChD has been progressively registered in non-endemic countries such as the US, Japan, Australia, and European countries, posing new challenges to local health systems.[9,10] ChD sequelae represent a significant burden for endemic regions, raising the alert for non-endemic countries with recent records of the disease, especially among immigrants.[1,6,11]

There is a paucity of up-to-date data on ChD clinical forms, and current estimates rely on individual epidemiological studies from the past 3 decades.[12] Efforts to quantify the total health burden of ChD, like in the Global Burden of Disease (GBD) study,[12] are substantially limited by suboptimal incidence and prevalence data, a lack of data on the frequency of individual sequelae, imprecise reporting of deaths—even in endemic settings—and an inability to separately quantify the burden of Chagas-associated mortality due to other causes of death, such as atrial fibrillation and embolic events. Understanding ChD epidemiological patterns helps to update such disease burden and mortality estimates and can guide the development of more efficient control measures. Here, we assessed the prevalence of the different clinical forms of ChD in seropositive adult patients, compiling available published data.

Methods

This study is part of 'The buRden of ChAgas dISEase in the Contemporary World (RAISE) project, a partnership among the Brazilian UFMG with the World Heart Federation, Novartis Global Health, and the University of Washington's Institute of Health Metrics and Evaluation. Upon reasonable request to the corresponding author, study data and analytical methods may be made available to other researchers to reproduce the results or replicate the procedures of this study. This work follows the PRISMA statement for systematic reviews[13] and is in accordance with specific guidelines for non-randomized studies. The systematic review and meta-analysis was registered in the PROSPERO database, under the number CRD42022354237.

Search strategy

A systematic search was performed in Medline, *Literatura Latino-Americana e do Caribe em Ciências da Saúde* (LILACS), through *Biblioteca Virtual em Saúde* (BVS), Cochrane, and Embase databases with the merged MeSH terms *("Chagas Disease" OR "Trypanosoma cruzi") AND (Prevalence OR Epidemiology) AND ("Clinical forms" OR "Indeterminate phase" OR "Acute phase" OR "Acute form" OR "Chronic phase" OR "Chronic form" OR "Arrhythmias, Cardiac" OR "Heart Failure" OR Thromboembolic OR Thromboembolism OR Thrombotic OR "Digestive manifestations" OR "Gastrointestinal Diseases" OR "Form associated" OR "Congenital form"*), without language restrictions, looking for studies, published until August 2022, that evaluated the prevalence of the classic clinical forms of ChD (indeterminate, CCM, digestive, thromboembolic) among seropositive patients. Bibliographic citations from hand-search of texts and associated citations, from the Medline "*related articles*" section, and email contact with authors were used to further identify potential articles. After the systematic review, the search was refined from a comprehensive review of existing population-based and cohorts involving patients with ChD, after discussion with experts in the area, and the searches were updated until August 2023.

Eligibility criteria

In summary, the paper selection criteria were: 1. Time frame: until August 2023; 2. Types of studies: population-based cohorts, surveys, or cross-sectional studies; 3. Sampling: including exclusively patients with serological diagnosis of ChD or with data of ChD patients presented in separate, with detailed subgroup information, enrolled in the following settings: a. population-based studies; b. outpatient clinics; c. inpatient wards; d. blood-donor studies; 4. Population: demographic and clinical variables reported, ideally with presentation of disease-related data (time living with ChD, time under clinical follow-up), associated morbidities and risk factors, and preexisting cardiovascular disease; 5. Prevalence data: systematically reported (separate prevalence of clinical forms); 6. Data stratification: ideally with minimal stratification by age group and sex; 7. Stratified sampling procedures (sample adjustment for population characteristics, such as age, gender and sociodemographic composition) and losses (eligible patients not presenting for evaluation) reported when applicable; 8. Geographical data (endemic vs. non-endemic countries for ChD); 9. Any language.

We screened studies for overlapped populations based on centers, authors' affiliations, and assessment period. In the case of overlapped populations, studies with larger sample sizes were included unless different studies presented different data.

Study selection and data extraction

The titles returned were peer-reviewed by 2 researchers (A: BRN and B: ADNN), according to the previously established criteria. Exclusion by title, abstract, and full-text analyses were independently performed and discrepancies in each stage were solved by consensus after discussion with a senior author (IM). The selected articles were read in full to confirm eligibility and their data was tabulated in a Microsoft Excel pre-populated spreadsheet and reviewed for statistical analysis. The second investigator independently double-checked data extraction. The prevalence of each specific clinical form of ChD (indeterminate, CCM, digestive, thromboembolic) was tabulated per study definition, according to pre-established criteria. For the definition of CCM, specific abnormalities observed in the ECG, echocardiogram, X-ray, clinical examination, or other

complementary tests were considered. For the digestive form, the diagnosis could be based on clinical examination, esophagogram, barium enema, endoscopy, colonoscopy, or other specific tests. For the thromboembolic form, reports of ischemic stroke, in the absence of other reasonable causes, with or without imaging tests were considered, as well as other documented systemic embolic phenomena. When "mixed" forms (CCM + digestive) were reported, the absolute numbers and rates were also counted in both forms (generally cardiac and digestive) separately.

Study quality was assessed by 2 authors (A: BRN and B: ADNN) using the modified Leboeuf-Yde and Lauritsen tool[14] developed by Hoy and colleagues, a method for assessing the risk of bias in prevalence studies (cross-sectional, cohort studies and time-interrupted series). Studies were graded separately by the investigators, and discrepancies were resolved by consensus before the final grading.

Statistical analysis

Data analysis was performed using Comprehensive Meta-Analysis Software v. 2.2.048 (Biostat Inc., Englewood/NJ, USA). Heterogeneity was explored with the I^2 statistic (inconsistency measure) from Cochran Q[15] that describes the percentage of variability of the effect that is due to heterogeneity rather than chance, and by the Tau^2 estimate (variance of the effect size parameters across the population of studies). Heterogeneity was considered substantial if $Tau^2 > 0$ and $I^2 > 50\%$ or P-value <0.10 in the chi-square test. Given the small number of large population-based cross-sectional or cohort studies, and to overcome the risk of bias due to the intrinsic heterogeneity of the included publications, the results were pooled by random effects model for the outcomes of interest (prevalence of ChD clinical forms). Pooled prevalence data were presented as mean percentages, computed as weighted sums considering the sample of seropositive patients as the denominator, and 95% confidence intervals, meaning the best-achieved estimate of the average rates.[2] Publication bias was assessed through the Funnel Plot and Egger's test.

To confirm the results, sensitivity analyses were done. Analyses by meta-regression (unrestricted maximum likelihood) or by subgroups were done, when possible, in order to explore heterogeneity sources.[15] Prespecified variables for meta-regression included mean age, gender (% male) and year of publication of the study (every 1 year). A separate subgroup analysis was performed by separately pooling data from studies conducted in the 4 main settings: population-based cross-sectional and cohort studies, blood donors, outpatients, and inpatients. In addition, a subgroup analysis was carried out for ChD endemic and non-endemic countries, according to the World Health Organization definition. To evaluate the possible impact of low-quality studies, we also did a sensitivity analysis by excluding studies in the lower bound (lowest quartile) of the modified Leboeuf-Yde and Lauritsen tool. Two additional analyses were carried out, one excluding studies conducted with outpatients and inpatients and one including exclusively recent publications, from 2010 to 2023. In all cases, central estimates and 95% CI different non-paired groups were indirectly compared, and a P-value for comparison <0.05 was considered significant.

Ethics statement

As this study utilized exclusively secondary data from published manuscripts, without primary data collection, no specific ethical approval was required. The project was registered in the Universidade Federal de Minas Gerais.

Role of funding source

The funder did not have any relationship with the conduct of the study, the collection, analysis, and interpretation of the data. Novartis Pharma AG employees (YG, CD and MQ) participated in the review of this manuscript as coauthors.

Results

Study selection and characteristics

The initial systematic search returned 1246 titles, and 846 were evaluated after the exclusion of duplicates. After peer review, 58 abstracts were selected, and 41 articles were read in full, resulting in 33 studies included in the first phase. Subsequently, related articles and references from the included studies were revised in full, resulting in 71 additional abstracts and 40 additional articles included in the meta-analysis, totaling 73 studies reporting the prevalence of ChD clinical forms (Fig. 1, Supplementary Table S1). In 3 cases, more than one arm (subgroup) of a study population was considered in the analysis.

The 73 studies included for the final analyses were published from 1966 to 2022; 58 were from endemic countries (45 from Brazil and 13 from other Latin American countries: Argentina, Bolivia, Colombia, Mexico, Nicaragua, and Venezuela). In total, they report the prevalence of ChD clinical forms for 17,132 seropositive patients. The average mean age of the patients was 43.6 years and 44% were males. Regarding the setting in which patients were enrolled, 50 studies were conducted among outpatients with ChD, 15 were population-based studies and/or surveys, 5 recruited blood donors and 3 reported data from inpatients with ChD Table 1.

Overall, the included studies systematically reported the prevalence of CCM, digestive, and mixed forms, and only one study reported the prevalence of thromboembolic events (namely stroke). For this reason, an analysis of the prevalence of the thromboembolic form was not possible Table 1.

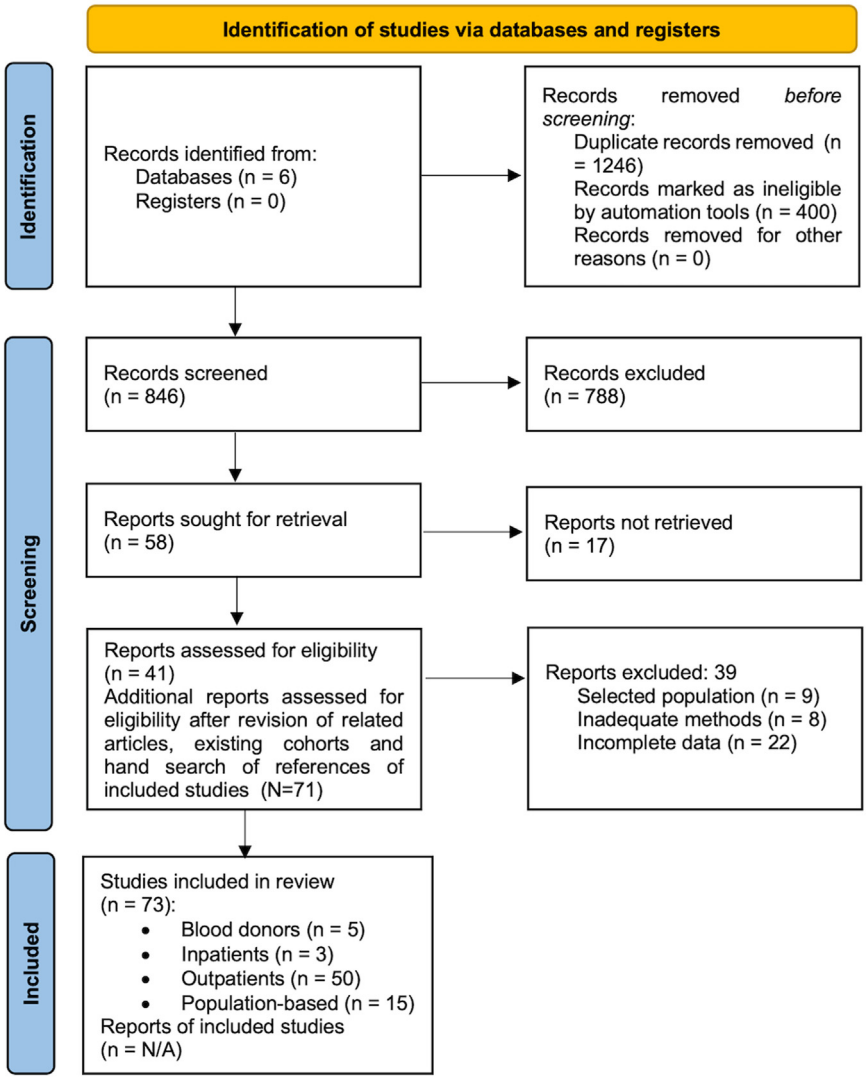

Fig. 1: PRISMA flowchart of article exclusion during peer-review.

Quality assessment

The quality of included studies was overall moderate, based on the applied criteria, being higher for population-based studies and surveys. None of the studies utilized stratified sampling procedures to make the sample representative of the national population (Criteria 1). In the qualitative analysis of data, the diagnostic criteria for each clinical form varied widely, given the long period frame of the included studies and the great differences among populations and study settings. The quality assessment of the included studies, using the modified Leboeuf-Yde and Lauritsen tool is presented in Supplementary Table S1, and the criteria utilized for defining the clinical forms are detailed in Supplementary Table S2.

Indeterminate form

The prevalence of the ChD indeterminate form was reported in 46 studies. Pooled prevalence in the overall population (7,559, considering all study settings) was 42.6% (95% CI 36.9–48.6, I^2 = 95.1%) (Fig. 2a). In the subgroup analysis by study setting, the prevalence of the indeterminate form was higher in population-based studies (4 studies, 421 participants, 47.2%, 95% CI 39.0–55.5, I^2 = 62.0%) and among blood donors (2 studies, 139 participants, 48.2%, 95% CI 40.0–56.5, I^2 = 0%) as compared to outpatients (38 studies, 6951 participants, 41.8%, 95% CI 35.3–48.5, I^2 = 95.8%), while among inpatients the estimates were discrepant and not reliable due to wide confidence intervals. However, there was no statistically significant difference

Author. year	Country	Sample	Age (mean/median)	N Men	% Men	Type of population	Rural/Urban	N Indeterminate	% Indeterminate	N Cardiac	% Cardiac	N Digestive	% Digestive	N Mixed	% Mixed
Puigbó. JJ. 1966	Venezuela	729				Population	Rural			168	23				
Pimenta. J. 1982	Brazil	44	49.9	26	59.1	Blood Donors	Urban			19	43.1				
Coura. JR. 1985	Brazil	110			0.0	Population	Urban and Rural	57	51.8	50	45.5				
Breniere. SF. 1989	Bolivia	131				Population	Urban and Rural	59	45	36	27.5	46	35.1	10	7.6
Zicker. F. 1990	Brazil	624	42.6	479	76.8	Population	Urban			277	44.4				
Rivera. BT. 1995	Nicaragua	39				Population	Rural			14	35.9				
Gontijo. ED. 1996	Brazil	626	37.78	444	70.9	Outpatient	Urban	350	56						
Modesto Santos. V. 1998	Brazil	362	57	0	0.0	Outpatient	Rural	125	34.5	179	49.4	58	16.0		
Nisida. IV. 1999	Brazil	57	32	0	0.0	Outpatient	Urban	27	47.4	25	43.8	5	8.8		
Dantas. RO. 1999	Brazil	68	41.5	63	92.6	Blood Donors	Urban	33	49	28	40.6	17	25	10	15
Bestetti. R. 2000	Brazil	79	56	45	57.0	Outpatient	Urban			69	87				
Borges-Pereira. J. 2001	Brazil	134		48	35.8	Population	Urban			33	24.6				
Pinto Dias. JC. 2002	Brazil	38	55.13	11	28.9	Outpatient	Rural	14	36.84	22	57.89	4	10.52		
Storino. R. 2002	Argentina	214	47.3	88	41.1	Outpatient	Urban			115	53.7				
Borges-Pereira. J. 2002	Brazil	189	54	84	44.4	Population	Rural			58	30.4	63	33.3		
Silveira. HJ. 2003	Brazil	48		41	85.4	Inpatient	Urban	31	64.5	15	31.0	8	16	3	6.25
Gazin. P. 2004	Brazil	57	52	28	49.1	Outpatient	Rural	32	56.1	21	36.8	5	8.7	1	1.75
Oliveira-Marques. DS. 2005	Brazil	163	42.95	106	65.0	Outpatient	Urban			62	38.0				
Pompilio. MA. 2021	Brazil	113		59	52.2	Outpatient	Urban			24	20.2	12	10		
Oliveira-Marques. DS. 2005	Brazil	106	43	51	48.1	Outpatient	Urban			31	29.2				
Bozelli. CD.2006	Brazil	55	61.2	36	65.5	Inpatient	Urban	0	0	33	60	33	60	11	20
Bozelli. CD. 2006 (2)	Brazil	40	50.1	26	65.0	Outpatient	Urban	19	47.5	18	45	6	15	3	3.75
Sanchéz-Guillén. MC. 2006	México	71	39.31	39	54.9	Outpatient	Rural	34	49	25	35	15	21	3	4.2
Oliveira-Mar. A. 2006	Mexico	19	58	8	42.1	Outpatient	Urban	4	21.0	15	79.0				
Sanchez-Guillen. MC. 2006	Mexico	71	48.3	39	54.9	Blood Donors	Urban and Rural	34	47.9	25	35.2	15	21.1	3	4.2
Borges-Pereira. J. 2007	Brazil	261	57.9	115	44.1	Outpatient	Rural			108	41.37				
Geraix. J. 2007	Brazil	66	49.6	35	53.0	Outpatient	Urban	47	71.2	11	16.7	13	19.7	5	7.6
Williams-Blangero. S. 2007	Brazil	722	49.1			Population	Rural			314	43.5				
Almeida. EA. 2007	Brazil	61	66.03	20	32.8	Outpatient	Urban	1	1.6	54	88.5	22	36		
Almeida. EA. 2007	Brazil	61	39.3	28	45.9	Outpatient	Urban	11	18	48	76.7	20	32.8		
Borges-Pereira. J. 2008	Brazil	17		7	41.2	Outpatient	Rural	10	59	7	41	1	6		
Cruz. OA. 2009	Mexico	39	31.6	18	46.2	Outpatient	Rural	19	48.7	20	51.3				
Angelis Alves. RM. 2009	Brazil	90	67.7	40	44.4	Outpatient	Urban	9	10	42	46.7	12	13.3		
Alves. RMA. 2009	Brazil	90	67	40	44.4	Outpatient	Urban	9	10	69	76.6	39	43	27	30
Munoz. J. 2009	Spain[a]	202	36	44	21.8	Outpatient	Urban	114	56.4	30	14.9	15	7.4		
Lescure. FX. 2009	France[a]	60	33	24	40.0	Outpatient	Urban			14	23.6	13	22		
Yves. J. 2010	Switzerland[a]	124	41	16	12.9	Outpatient	Urban	109	87.9	11	11.3	1	0.8		
Silva. EM. 2010	Brazil	14	67	4	28.6	Outpatient	Rural	3	21	11	79				
Silva-Greco. RL. 2010	Brazil	151	38.5	71	47.0	Population	Urban and Rural	58	38.4	83	54.9	21	13.9	11	7.3
Moretti. E. 2010	Mexico	325				Population	Urban and Rural			95	29.2				
Roca. C. 2011	Spain[a]	22		10	45.5	Outpatient	Urban	17		4	19.0	2	9.5		
Valerio. L. 2011	Spain[a]	100	38.2	35	35.0	Outpatient	Urban			49	49				
Jackson. Y. 2011	Switzerland[a]	258	41	43	16.7	Outpatient	Urban	178	69	52	20.2	4	1.6	1	0.4
Ramos. JM. 2012	Spain[a]	128	35	47	36.7	Outpatient	Urban	101	78.9	27	21.1	2	1.6	1	1.6
Salvador. F. 2013	Spain[a]	1274	37.7	414	32.5	Outpatient	Urban			190	16.9	154	14.8		

(Table 1 continues on next page)

Author. year	Country	Sample	Age (mean/median)	N Men	% Men	Type of population	Rural/Urban	N Indeterminate	% Indeterminate	N Cardiac	% Cardiac	N Digestive	% Digestive	N Mixed	% Mixed
(Continued from previous page)															
Ribeiro. AL. 2013	Brazil	499	48	261	52.3	Blood Donors	Urban			120	24				
Matos. CS. 2014	Brazil	101	48	68	67.3	Outpatient	Rural	14	13.9	80	79.2	57	56.4	50	49.5
González. B. 2014	Venezuela	115		56	48.7	Outpatient	Rural	34	29.5	81	70.4				
Pinazo. MJ. 2014	Spain[a]	71	36	12	16.9	Outpatient	Urban					15	21.1		
Perez-Ayala. A. 2014	Spain[a]	252	36		0.0	Outpatient	Urban	196	77.8	47	18.7	13	5.2	4	1.6
Mesquita Andrade. C. 2015	Brazil	186	49	92	49.5	Outpatient	Rural	96	51.6	75	40.0	30	16.0		
Pereira. LS. 2015	Brazil	95	67	48	50.5	Outpatient	Urban	14	14.7	75	78.9	19	20.0	13	13.7
Fernandez. AB. 2015	Bolivia	398	38	128	32.2	Population	Rural			55	13.8				
Garcia. MN. 2015	United States[a]	17	51	10	58.8	Blood Donors	Urban			5	29.4				
Ribeiro. AL. 2015	Brazil	557	68	181	32.5	Population	Urban			312	56				
Diaz-Cardoso. EM. 2017	Colombia	19	47.8	13	68.4	Outpatient	Urban	13	73.7	6	32.3				
Bruscato. A. 2017	Brazil	80		52	65.0	Outpatient	Urban	19	24	53	66	20	25	12	15
Yasuda. MAS. 2017	Brazil	18				Outpatient	Rural	13	71.42	5	28.57				
Vizzoni. AG. 2018	Brazil	619	60	267	43.1	Outpatient	Urban	180	29.1	415	67.0	106	17.1	72	10.0
Gasparim. AZ. 2018	Brazil	270	67.1	97	35.9	Outpatient	Urban	82	30.3	151	55.9	90	33.3	53	19.6
Antinori. S. 2018	Italy[a]	29	37.5	7	24.1	Population	Urban	18	62.1	5	17.2	6	20.7	1	3.6
Arruda. HMBS. 2019	Brazil	171	45	59	34.5	Outpatient	Urban	114	66.7	33	19.3	25	14.6	8	4.7
Nielebock. M.A. 2020	Brazil	139	47.4	57	41.0	Outpatient	Urban	48	34.5	84	60.0	24	17%	17	12.2
Borges-Pereira. J. 2020	Brazil	298	51	114	38.3	Outpatient	Rural			212	71.14				
Gonzalez-Sans. M. 2020	England[a]	60		18	30.0	Outpatient	Urban			12	20	3	5		
Lidani. KCF. 2020	Brazil	237	57.5	97	40.9	Outpatient	Urban	85	35.9	126	53	57	24.1	31	13.1
Sanz. MG. 2020	United Kingdom[a]	60	41	18	30.0	Outpatient	Urban	28	47	16	26.7	11	18.3	5	8.3
Xavier. IGG. 2021	Brazil	361	60.7	159	44.0	Outpatient	Urban	97	26	251	69.5	58	16		
Lima. NA. 2021	United States[a]	2037	51.8	1028	50.5	Inpatient	Urban			1316	64.6				
Resende. BAM. 2021	Brazil	283	57.5	108	38.2	Population	Urban			49	17.3				
Echalar. JC. 2021	Bolivia	122	53.3	66	54.1	Population	Urban and Rural			24	19.7				
Medeiros. CA. 2022	Brazil	801	62	244	30.5	Outpatient	Rural	182	22.5	733	91.0	114	14.0	114	14.0
Portela. LF. 2022	Brazil	985	65.2	399	40.5	Outpatient	Urban	303	30.8	630	63.9	201	20.4	149	15.1

Abbreviation: N: number of patients (total sample). [a]Non-endemic countries for Chagas Disease, according to the World Health Organization.

Table 1: **Detailed characteristics of the included studies.**

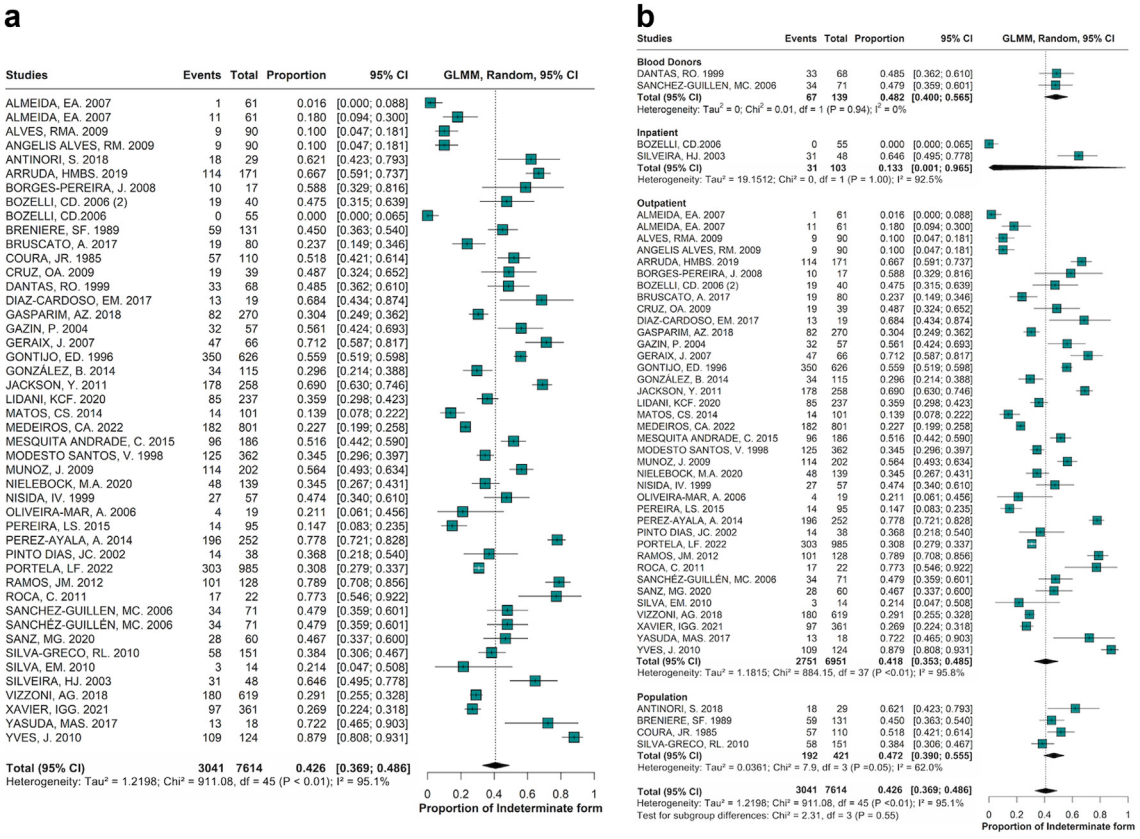

Fig. 2: Pooled prevalence plots: prevalence of the Chagas Disease indeterminate form in **a:** the overall population; **b:** by study setting (blood donors, inpatients, outpatients and population-based studies).

between settings, with overlapping CIs (P-value = 0.55) (Fig. 2b, Table 2).

In the subgroup analysis of prevalence by endemicity, the pooled prevalence of the indeterminate form was significantly lower in endemic (38 studies, 6484 participants, 36.8%, 95% CI 31.9–42.0, I^2 = 92.8%) compared to non-endemic (8 studies, 1075 participants, 70.8%, 95% CI 61.1–79.0, I^2 = 88.7%) countries, P-value<0.001 (Supplementary Figure S1). Overall, heterogeneity was high for estimates of the indeterminate form, being lower for blood donors/population studies and non-endemic countries in separate (Table 2).

Chagas chronic cardiomyopathy

The prevalence of CCM was reported in 71 studies (16,435 participants; Table 1). The prevalence of CCM in the overall population was 42.7% (95% CI 37.3–48.3), I^2 = 96.1% (Fig. 3a, Table 2). In the subgroup analysis by study setting, CCM prevalence was considerably higher among outpatients (48 studies, 9053 participants, 47.1%, 95% CI 39.5–54.9, I^2 = 97.5%) and inpatients (3 studies, 2140 participants, 64.6%, 95% CI 62.5–66.7, I^2 = 89.9%) as compared to population-based studies (15 studies, 4543 participants, 31.2%, 95% CI 24.4–38.9, I^2 = 96.1%) and blood donors (5 studies, 699 participants, 29.4%, 95% CI 12.8–54.2, I^2 = 75.1%), P-value = 0.01 (Fig. 3b, Table 2).

In the subgroup analysis by endemic and non-endemic settings, the pooled prevalence of CCM was significantly higher in endemic countries (57 studies, 11,812 participants, 48.0%, 95% CI 42.3–53.8, I^2 = 96.8%) vs. non-endemic countries (14 studies, 4623 participants, 23.1%, 95% CI 12.7–38.1, I^2 = 98.7%), P = 0.004 (Table 2, Supplementary Figure S2). Heterogeneity was also high for CCM estimates, being lower for blood donors in separate and similarly high between endemic and non-endemic countries (Table 2).

Digestive and mixed forms

The digestive form of ChD was reported in 44 studies, and fewer publications (26 studies) simultaneously reported the prevalence of the mixed form. The overall pooled prevalence of the digestive and mixed forms was, respectively, 17.7% (8421 participants; 95% CI 14.9–20.9), I^2 = 90.2% and 10.2% (5073 participants; 95% CI 7.9–13.2), I^2 = 87.5% (Figs. 4a and 5a).

Type of analysis:	Indeterminate form, N = 46 (%, 95% CI):	Chronic Chagas cardiomyopathy, N = 71 (%, 95% CI):	Digestive form, N = 44 (%, 95% CI):	Mixed form, N = 26 (%, 95% CI):
Overall:	42.6% (36.9–48.6), I^2 = 95.1%	42.7% (37.3–48.3), I^2 = 96.1%	17.7% (14.9–20.9), I^2 = 90.2%	10.2% (7.9–13.2), I^2 = 87.5%
Study setting:				
Blood donors:	48.2% (40.0–56.5), I^2 = 0.0% (2 studies)	33.9% (24.9–44.1), I^2 = 75.1% (5 studies)	23.1 (16.8–30.8), I^2 = 0.0% (2 studies)	8.7% (2.5–26.3), I^2 = 75.0% (2 studies)
Inpatients:	13.3% (0.1–96.5), I^2 = 92.5% (2 studies)	52.9% (34.5–70.6), I^2 = 89.9% (3 studies)	35.8 (7.2–80.1), I^2 = 94.4% (2 studies)	12.4% (3.8–33.8), I^2 = 73.1% (2 studies)
Outpatients:	41.8% (35.3–48.5), I^2 = 95.8% (38 studies)	47.1% (39.5–54.9), I^2 = 97.5% (48 studies)	16.0 (13.3–19.2), I_2 = 89.7% (36 studies)	10.7% (7.9–14.4), I^2 = 89.8% (19 studies)
Population studies:	47.2% (39.0–55.5), I^2 = 62.0% (4 studies)	31.2% (24.4–38.9), I^2 = 96.1% (15 studies)	25.4% (16.0–37.7), I^2 = 85.4% (4 studies)	7.2% (4.8–10.7), I^2 = 0.0% (3 studies)
P-value for comparison:	P = 0.55	P = 0.01*	P = 0.07	P = 0.44
Endemic vs. non endemic countries:				
Endemic:	36.8% (31.9–42.0), I^2 = 92.8% (38 studies)	48.0% (42.3–53.8), I^2 = 96.8% (57 studies)	21.7% (18.4–25.4), I^2 = 88.6% (38 studies)	12.6% (9.8–16.0), I^2 = 86.6% (21 studies)
Non-endemic:	70.8% (61.1–79.0), I^2 = 88.7% (8 studies)	23.1% (12.7–38.1), I^2 = 98.7% (14 studies)	8.5% (5.5–12.8), I^2 = 83.4% (8 studies)	2.0% (0.6–6.1), I^2 = 69.4% (5 studies)
P-value for comparison:	P < 0.001*	P = 0.004*	P < 0.001*	P < 0.001*

Abbreviation: CI: confidence interval. * P-value <0.05.

Table 2: **Prevalence of the clinical forms of Chagas disease, in all studies included, by study setting and by endemicity, with between-group comparison.**

Subgroup analyses by study setting and endemicity were limited for digestive and mixed forms, considering the smaller number of publications reporting such estimates (Table 2, Figs. 4b and 5b, Supplementary Figures S3 and S4). Thus, discrepant point estimates and wider confidence intervals were observed. In general, the prevalence of the digestive form was lower among outpatients, compared to population-based studies and blood donors (Table 2), whereas for the mixed form the lowest prevalence estimates were observed in population studies and blood donors—similarly to CCM. In the subgroup analysis by endemicity, the prevalence of digestive and mixed forms was significantly lower in non-endemic countries (Table 2, Supplementary Figures S3 and S4).

Sensitivity analyses–study quality and publication period

In the sensitivity analysis excluding studies with grades ≤7 points (11 studies) in the modified Leboeuf-Yde and Lauritsen tool, the prevalence of ChD clinical forms in a final sample of XX, were: indeterminate: 45.7% (95% CI 39.6–51.9), I^2 = 94.7%; CCM: 38.8% (95% CI 33.7–44.2), I^2 = 96.7%; digestive: 16.5% (95% CI 13.4–20.0), I^2 = 91.1%; and mixed: 9.0% (95% CI 6.4–12.6), I^2 = 89.9%.

In the sensitivity analysis including only studies published from 2010 to 2023, the prevalence rates of the indeterminate and CCM forms were similar to the overall analysis of all included papers, in the endemic and non-endemic settings. However, the prevalence of the digestive and mixed ChD forms was slightly lower. Detailed prevalence data for this sub-analysis are presented in Table 3 and Supplementary Figures S5–S8.

Meta-regression analyses

In meta-regression, the mean age of the population was inversely associated with the prevalence of the indeterminate (β = –0.053, 95% CI –0.057 to –0.048, P < 0.001) form, and directly associated with CCM (β = 0.060, 95% CI 0.056–0.063, P < 0.001) and digestive (β = 0.021, 95% CI 0.016–0.027, P < 0.001) forms. The percent of male patients was directly associated with the digestive (β = 0.018, 95% CI 0.014–0.022, P < 0.001) form and CCM (β = 0.006, 95% CI 0.005–0.0090, P < 0.001), but not with the indeterminate presentation (β = 0.002, 95% CI –0.0008 to 0.0045, P = 0.18). Finally, the year of publication of the studies was not associated with the prevalence of the indeterminate (β = –0.007, 95% CI –0.046 to 0.032, P = 0.73), CCM (β = 0.010, 95% CI –0.014 to 0.033, P = 0.41) and digestive (β = –0.011, 95% CI –0.048 to 0.026, P = 0.55) forms.

Publication bias

The Funnel plot and Egger's test did not indicate the presence of publication bias for the prevalence estimates of the indeterminate, CCM and digestive forms of ChD. However, there was asymmetry of the Funnel plot for the mixed form, with Egger's test P-value = 0.014, suggesting an underestimation of prevalence rates (Table 2, Supplementary Figure S9).

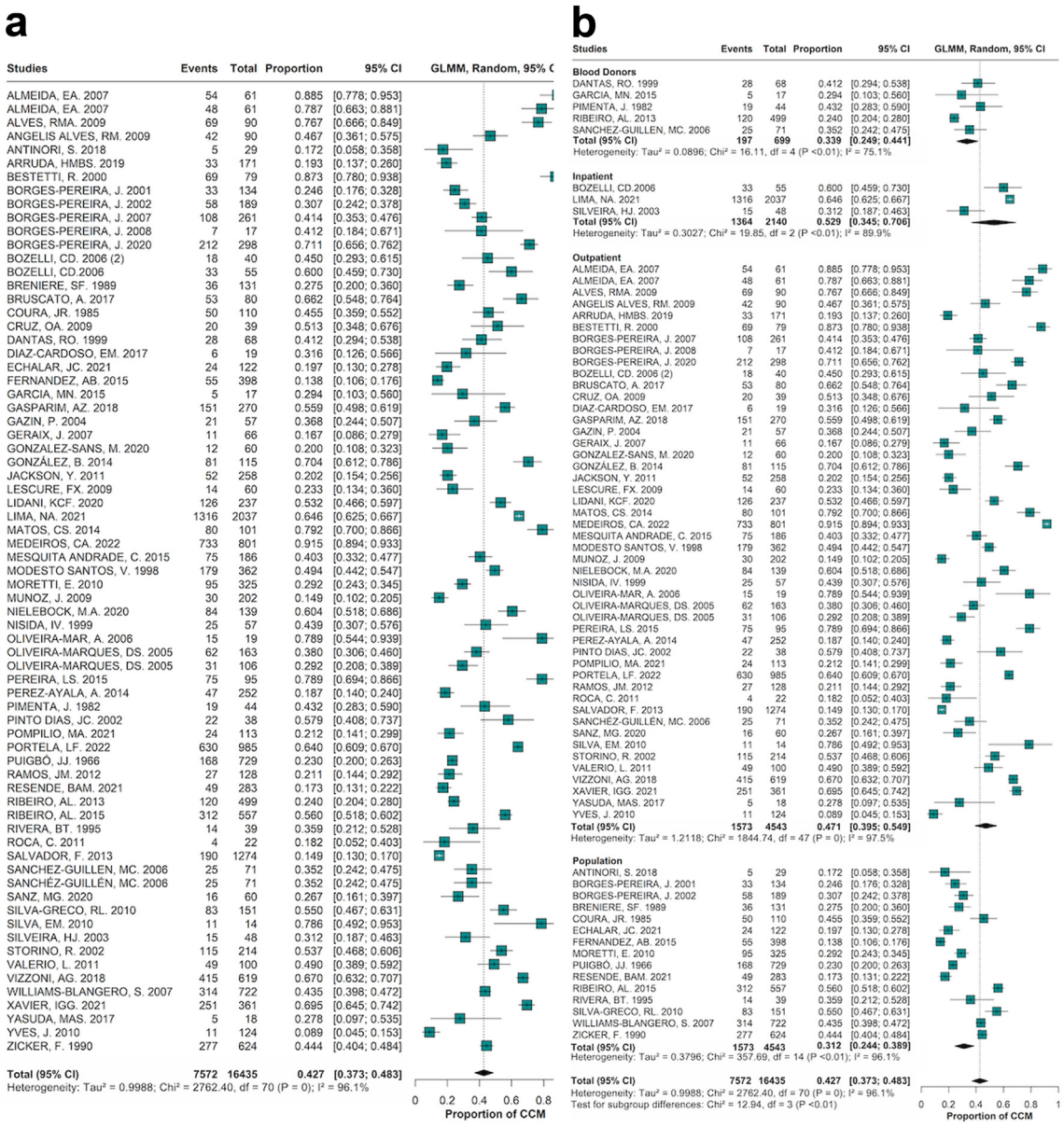

Fig. 3: Pooled prevalence plots: prevalence of the Chagas Disease cardiac form in **a:** the overall population; **b:** by study setting (blood donors, inpatients, outpatients and population-based studies).

Discussion

In this systematic review and meta-analysis, we compiled up-to-date prevalence data about the clinical forms of Chagas disease, suggesting that the prevalence of CCM among ChD seropositive patients is higher than suggested by old individual studies (ranging from 31% in population studies to 52% among inpatients), while rates of the digestive form were similar to literature (ranging from 16% in outpatients to 36% in inpatients).[7,16] It was notable, however, that data quality was suboptimal and limited to certain geographic areas, and heterogeneity was considerable, especially due to different study settings and allocation methodologies. When population-based and blood-donor studies were pooled separately, the findings tended to overlap with existing literature.[9,16]

The clinical course of ChD has been widely investigated since the description of the disease, in the early 1900's, with special interest in understanding the predictors and determinants of progression to its clinical forms and, ultimately, its prognostic markers.[16,17] When the disease became more prevalent in high-income non-

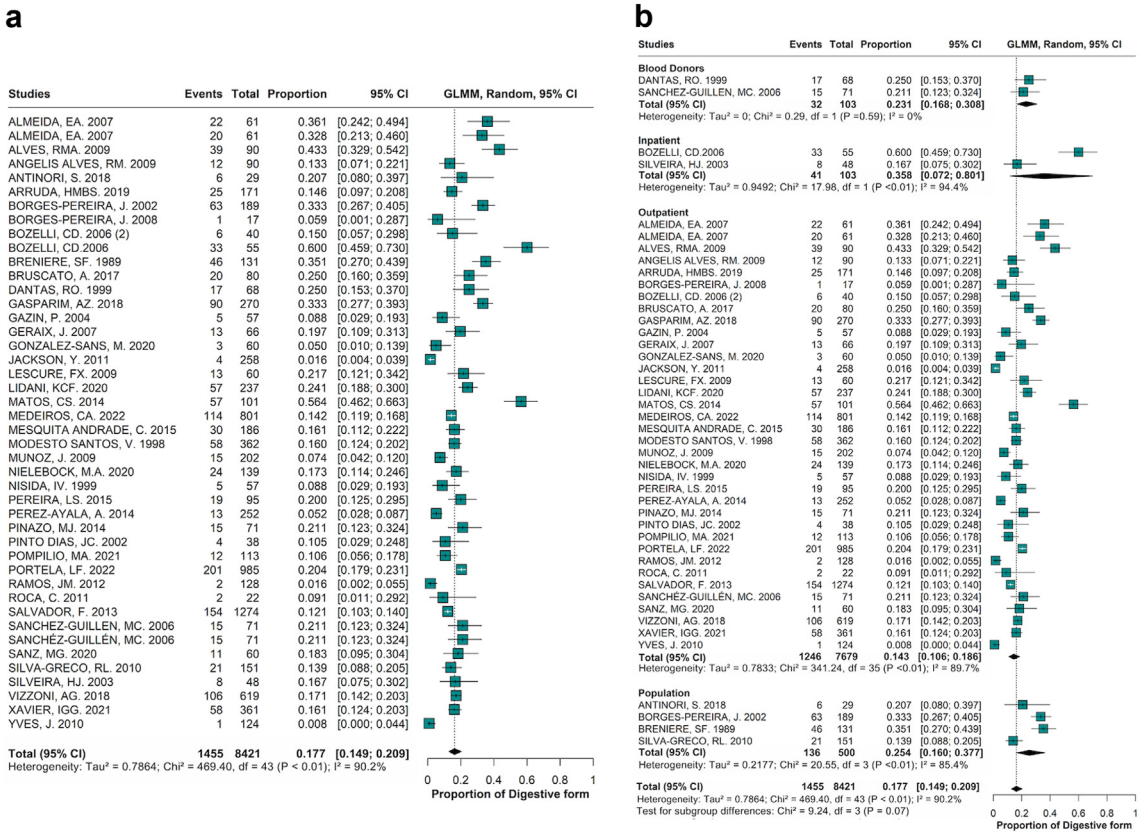

Fig. 4: Pooled prevalence plots: prevalence of the Chagas Disease digestive form in **a:** the overall population; **b:** by study setting (blood donors, inpatients, outpatients and population-based studies).

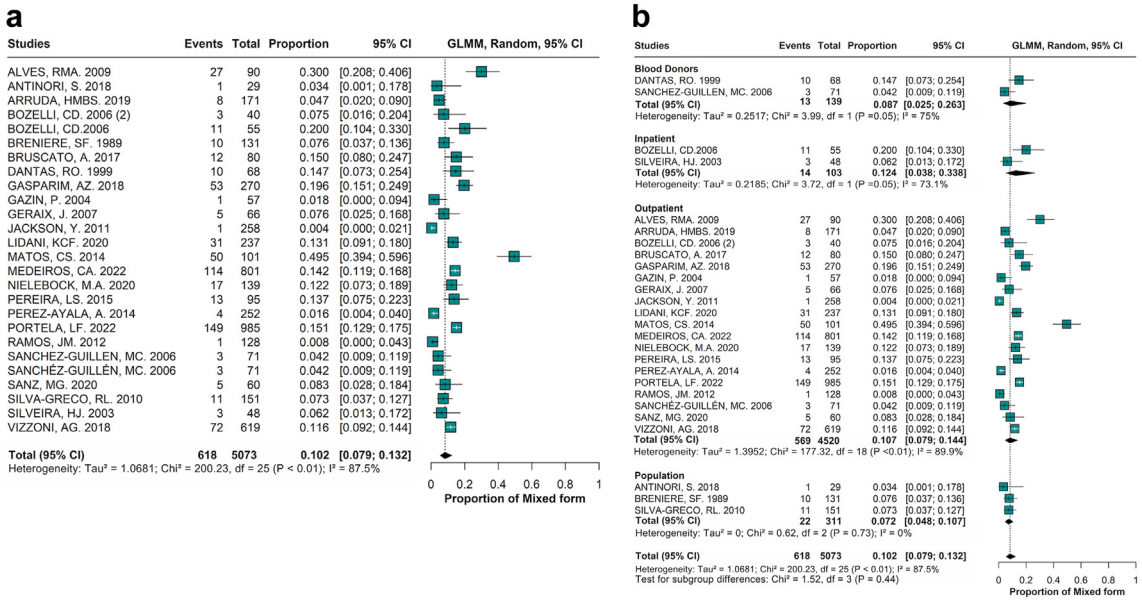

Fig. 5: Pooled prevalence plots: prevalence of the Chagas Disease mixed (cardiac + digestive) form in **a:** the overall population; **b:** by study setting (blood donors, inpatients, outpatients and population-based studies).

Type of analysis:	Indeterminate form, N = 24 (%, 95% CI):	Chronic Chagas cardiomyopathy, N = 35 (%, 95% CI):	Digestive form, N = 23 (%, 95% CI):	Mixed form, N = 16 (%, 95% CI):
Overall:	45.6 (35.4–56.1), I^2 = 96.6%	41.7% (32.8–51.1), I^2 = 98.4%	13.3% (9.1–19.0), I^2 = 91.4%	8.1% (4.6–14.1), I^2 = 90.4%
Study setting:				
Blood donors:	No studies included.	24.2% (20.7–28.1), I^2 = 0% (2 studies)	No studies included.	No studies included.
Inpatients:	No studies included.	64.6% (62.5–66.7), I^2]N/A (1 study)	No studies included.	No studies included.
Outpatients:	45.2% (34.3–56.6), I^2 = 96.8% (22 studies)	46.3% (35.1–57.8), I^2 = 98.4% (26 studies)	13.0 (8.5–19.2), I_2 = 92.2% (21 studies)	8.6% (4.5–15.6), I^2 = 91.2% (14 studies)
Population studies:	47.1% (31.1–63.7), I^2 = 81.3% (2 studies)	27.8% (17.2–41.5), I^2 = 97.16% (7 studies)	15.0% (10.5–21.0), I^2 = 0% (2 studies)	6.7% (3.8–11.4), I^2 = 0.0% (2 studies)
P-value for comparison:	P = 0.85	P < 0.01*	P = 0.59	P = 0.56
Endemic vs. non endemic countries:				
Endemic:	33.8% (26.5–42.0), I^2 = 92.1% (17 studies)	51.8% (40.9–62.5), I^2 = 98.1% (24 studies)	20.8% (16.3–26.2), I^2 = 90.9% (13 studies)	14.1% (9.8–19.8), I^2 = 90.5% (11 studies)
Non-endemic:	73.2% (63.0–81.5), I^2 = 85.4% (7 studies)	23.8% (16.4–33.2), I^2 = 98.7% (12 studies)	6.2% (3.1–12.2), I^2 = 84.3% (10 studies)	1.6% (0.6–4.6), I^2 = 69.4% (5 studies)
P-value for comparison:	P < 0.01*	P < 0.01*	P < 0.01*	P < 0.01*

Abbreviation: confidence interval, N/A: not applicable. * P-value <0.05.

Table 3: **Prevalence of the clinical forms of Chagas disease, in studies published from 2010 to 2023, by study setting and by endemicity, with between-group comparison.**

endemic areas, due to migration waves,[10] additional research efforts were put together, aimed at investigating the mediators of the complex host–parasite interaction that leads to end-organ damage. After a vector transmission event, incubation usually lasts no longer than 1–2 weeks, and this critical period is marked by microscopically detectable parasitemia.[6,7] Diagnosis and initiation of specific treatment with antiparasitic drugs in this phase are challenging, considering that symptoms are usually mild and nonspecific, such as fever, mild hepato-splenomegaly, and lymphocytosis.[1,4,6,7]

After the resolution of the acute phase of ChD, individuals transition into an indeterminate phase characterized by the persistence of infection, verified through serological tests, but frequently without detectable parasites in blood sample microscopy.[6,7] This phase is further defined by the absence of organic manifestations, as denoted by the absence of typical findings in the standard ECG and normal radiological assessments of the chest, esophagus, and colon.[1,7] Thus, according to the classification criteria, individuals in this phase exhibit chronic ChD without discernible pathological abnormalities, making diagnosis even more challenging.[2,6] Notably, approximately one-third of these individuals are anticipated to progress to chronic symptomatic ChD in the two decades following the initial infection,[1,2] and progression is associated with a complex host–parasite interaction, mediated by factors such as age, gender, parasite load, inflammatory response and individual susceptibility.[1,6,7]

In medical literature, there has been a greater interest in the study of CCM compared to other clinical forms of ChD, given its complex pathophysiology and immunology, as well as the sometimes unpredictable evolution and prognosis.[1] Among blood donors without known heart disease, ChD seropositivity was independently associated with a 40% higher chance of developing cardiomyopathy.[18] Some features of CCM are unique, such as the complex pattern of rhythm disturbances, the associated thrombogenicity, and variable forms of structural involvement, notably the coexisting involvement of the left and right ventricles.[1,7,19] Moreover, while anti-parasitic drugs have no consistent effects in advanced stages of CCM,[20] the prognostic benefit of heart failure pharmacotherapy seems to extend to ChD patients with cardiac involvement.[19] Thus, investigations about the prevalence, classification, diagnosis, and prognosis of CCM emerged after the disease was first described. From our pooled data, the diagnostic criteria for CCM was one of the key sources of heterogeneity of the analyses: as the heart is the most commonly involved organ in ChD, inaccurate diagnosis of CCM will substantially impact the estimates of indeterminate and mixed forms.

From the inception of studies about CCM until the mid-1990s, the diagnostic criteria relied almost exclusively on typical ECG findings, chest R-ray, and clinical manifestations.[8] Some ECG findings seem to be the earliest signs of cardiac involvement and the inflammatory and fibrotic processes triggered by ChD[8]—and continue to be used as the key guideline-based criteria for reclassification from indeterminate to CCM. However, more recent studies have often used cardiac imaging as an additional part of the diagnostic criteria. The introduction of echocardiography and the development

of its more modern modalities–such as tissue strain and 3D imaging—have led to multiple areas of research in ChD, with a special focus on refining existing diagnostic and prognostic criteria.[21] In addition, advanced imaging, such as cardiac magnetic resonance and computed tomography, have been used for the management of CCM in high-resource settings.[21] Cardiac imaging has presumably added accuracy to the diagnosis of CCM, especially when ECG and R-ray are not typical for a definite diagnosis,[22] and improving specificity in epidemiologic studies. These shifts in diagnostic criteria for CCM, however, limit the comparability of older and more recent studies, likely driving much of the observed heterogeneity in this meta-analysis.

The positive association between mean study age and the prevalence of CCM and the subsequent negative association observed for the indeterminate form are expected, as cardiac involvement seems to progressively develop in seropositive individuals.[16,23] This has been notably demonstrated in ECG studies, in which the development or worsening of typical ECG abnormalities occur over the decades, especially in untreated patients, and several findings, such as widening of the QRS complex, lowering voltage, new onset of ventricular ectopias, and the number of new major abnormalities, seem to have prognostic impact.[18,24] Notably, this trend is not as clear for other organ involvements.

Similar findings were observed for the digestive form of ChD, in terms of increasing prevalence with age. First, given its lower prevalence and overall clinical impact—especially compared to CCM—the ChD digestive form has been given less priority in medical research. This results in considerably fewer studies reporting its prevalence, with a wide range and less standardized diagnostic criteria.[25] Second, the predictors of progression to the digestive form are not clear, and some case series only suggest a higher risk for women.[26] Early ChD studies that reported the digestive form relied basically on clinical manifestations and contrasted esophagograms and enema. Clinically, manifestations of the ChD digestive form, especially in its early stages, overlap with common gastrointestinal conditions, such as chronic constipation, inflammatory bowel diseases, esophagitis, and diverse motility disturbances of the upper digestive tract.[26] Therefore, clinical diagnosis alone tends to be imprecise. On the other hand, even the availability of contrasted X-ray is relatively limited in clinical practice and is often impractical in the research setting, especially in large health surveys. Recently, invasive imaging studies of the digestive tract became an additional tool, especially for symptomatic ChD patients. However, its applicability is impacted by cost—especially in low-income endemic regions—and by the practicality of implementing resource-demanding modalities in large research samples. Moreover, both contrasted X-ray and invasive imaging modalities detect advanced stages of the digestive form, underestimating its true prevalence.[27] Despite the recent advances in digestive nuclear imaging for ChD patients,[28] currently, a definite early diagnosis would only be possible through anatomopathological tissue sampling.[26] Our data, thus, reflect this heterogeneous scenario, with more discrepant estimates, wide dispersion, and limited conclusions from subgroup analyses.

Despite the categorization of the included articles into 4 main groups (outpatients, inpatients, blood donors, and population-based studies), it is notable from our systematic review that the sampling methodology varied widely between studies. From the pooled results, there is a clear selection bias associated with the enrollment of outpatient and especially inpatient samples. Most of these studies were conducted in specialized institutions, with established protocols for patients with positive ChD serology, in which several tests—including those for early diagnostic of clinical forms such as CCM—are routinely performed in clinical care or research protocols. Furthermore, patients with any clinical manifestations of ChD tend to present more frequently to specialized health institutions, and those severely ill are more prone to hospitalizations. In contrast, investigations based on population-based methodologies and surveys involving seropositive candidates for blood donation inherently have a better capacity to mirror the actual prevalence of ChD clinical forms. However, pivotal studies with such designs have predominantly been conducted in populations characterized by elevated prevalence, in endemic regions or locations marked by specific socioeconomic and demographic traits. Consequently, the direct generalization of findings to an entire country or region is circumscribed by these contextual limitations.

Regarding the differences between studies conducted in endemic and non-endemic countries, some hypotheses can be drawn to explain the observed trends. There is, in general, an evident economic gap between endemic and non-endemic regions, which ultimately impacts patient's access to healthcare. Although no evidence-based interventions change the clinical course of ChD, access to anti-parasitic drugs in pre-clinical (indeterminate) stages may slow progression.[29] This hypothesis, however, emerged from non-randomized observational studies[29] and requires further assessment in clinical trials. Access to cardiovascular medication and control of coincident risk factors can also retard symptom onset and need for medical care. Also, although nearly all individuals included in non-endemic countries are immigrants from endemic areas, it is hypothesised, from animal models, that recurrent contact and reinfection with different Trypanosoma strains may be associated with disease severity.[30] Moreover, a considerable number of studies in non-endemic countries recruited candidates for blood donation, a population with lower age and less prone to the selection bias of specialized centers. Early diagnosis in pre-clinical

stages is also favoured by recommendations for routine screening of individuals at risk in such settings.[21,22]

Finally, the continuous assessment of epidemiological data about the clinical forms of ChD is crucial for the refinement of global disease burden models. Despite the growing number of publications, the literature about ChD is heterogeneous and spread among different regions, with particular sociodemographic and epidemiological backgrounds, requiring specific research approaches. Heterogeneity of data is an intrinsic characteristic of ChD literature, and pooled analyses with detailed sensitivity approaches may improve not only the understanding of the disease and its trends over the decades but also the development of structured strategies to improve patients' access to optimal care.

Our study has several limitations. The primary limitations are related to the available data, including heterogeneity—mainly driven by the absence of stratified sampling procedures (adjustment for population characteristics) and standardized methodology for systematic data collection in the different study settings—and study quality, especially in terms of very limited population-wide data. The settings where the studies were conducted varied widely, from small municipalities with high prevalence of ChD and restricted endemic areas to specialized outpatient clinics and infectious diseases wards. While these data provide a wider view of how prevalence may vary in different populations, this variation results in uncertainty in the pooled estimates. Second, the time frame of the included studies was large, from classic epidemiological studies to contemporary series and ongoing cohorts. In addition to contributing to heterogeneity due to methodological issues, changes in the use of diagnostic methods over time in research studies (especially imaging) may have resulted in variable diagnostic accuracy over time. In addition, changes in age composition are known to impact the incidence of sequelae such as CCM. Third, available evidence does not allow for robust inferences about progression/development of ChD clinical forms over time, as few studies present longitudinal data. Fourth, reporting and definition of clinical forms were also heterogeneous between studies and varied according to setting. While CCM was almost universally reported, fewer studies reported the digestive form, and rarely mixed forms were presented separately. Moreover, data was markedly scarce for thromboembolic events, including stroke. Finally, the results of the meta-analysis essentially reflect the epidemiology of Brazil and Latin America, given the paucity of population-wide data from other endemic countries and, especially, from non-endemic areas.

Our analytic method is also subjected to limitations, such as the choice to pool heterogeneous results from different settings and populations, and the impossibility of analyzing prevalence by age range, as contact with authors for primary data requests was not feasible. Despite the aforementioned limitations, our study provides important up-to-date estimates about the prevalence of ChD clinical forms, pooling data from a wide range of publications, and covering different settings in a large time frame. By using meta-analytic methods to address the limitations of individual studies, our pooled data will contribute to the update of current estimates about global disease burden, associated sequelae, and mortality. Specific data about the prevalence of clinical forms in different settings (endemic and non-endemic), based on different study designs and with temporal analysis across decades, provide invaluable insights about attributable mortality and disability weights for the refinement of global estimates. The understanding of disease burden, especially in high-prevalence areas, may help improve the development of public policies and guide prioritization of care.

Conclusions and future research

Compared to previous epidemiological studies, our pooled data shows a higher prevalence of CCM among ChD seropositive adult patients, but a similar prevalence of the digestive form. Heterogeneity, however, was overall high, mainly due to limited data quality and the small number of population-based studies. Improving quality of data on the prevalence of ChD clinical forms, notably the development of broad epidemiological surveys, is crucial for the update of global estimates of disease burden and mortality.

Our study also emphasizes the need for continued development of epidemiological projects aimed at refining estimates of the burden of disease and mortality due to ChD. Parallel efforts are being made in the RAISE study group to: (*i*) incorporate up-to-date prevalence data from meta-analyses (prevalence of clinical forms, overall ChD prevalence in endemic and non-endemic countries) into existing estimates; (*ii*) update mortality data from mortality information systems in Brazil and other countries; (*iii*) improve the redistribution of garbage codes in death certificates, to refine ChD estimates; (*iv*) remodel the economic burden of ChD; and (v) investigate the incidence and mortality associated with specific sequelae of ChD, especially arrhythmias and embolic events.

Contributors

Conception and design of the research: BRN, ALPR, CD, MQ, PAP, EC, JFM; Acquisition of data: BRN, ADNN, BMPS; IM, FRM; Analysis and interpretation of data: BRN, ALPR, ADNN, BMPS; Statistical analysis: BRN; Obtaining financing: YG, CD, MQ, PAP; Writing of the manuscript: BRN, ADNN, FRM, IEM; Critical revision of the manuscript for intellectual content: All authors; Authors responsible for the overall content as guarantors: BRN, ALPR, GY, CD, MQ, PAP.

Data sharing statement

Data analytic methods and study materials will be made available to other researchers for purposes of reproducing the results or replicating the procedure, from the corresponding author upon reasonable request.

Declaration of interests
The authors have no conflicts of interest to disclose regarding this manuscript. Yvonne Geissbühler, Caroline Demacq, and Monica Quijano are Novartis employees and declare stocks of the company.

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Appendix A. Supplementary data
Supplementary data related to this article can be found at https://doi.org/10.1016/j.lana.2024.100681.

References

1 Nunes MCP, Beaton A, Acquatella H, et al. Chagas cardiomyopathy: an update of current clinical knowledge and management: a scientific statement from the American heart association. *Circulation*. 2018;138(12):e169–e209.
2 Pereiro AC. Guidelines for the diagnosis and treatment of Chagas disease. *Lancet*. 2019;393(10180):1486–1487.
3 Chagas disease in Latin America: an epidemiological update based on 2010 estimates. *Wkly Epidemiol Rec*. 2015;90(6):33–43.
4 Bern C. Chagas' disease. *N Engl J Med*. 2015;373(19):1882.
5 Chagas C. A short chronicle of the discovery of Chagas' disease. *Pacing Clin Electrophysiol*. 1988;11(7):1108–1113.
6 Ribeiro AL, Nunes MP, Teixeira MM, Rocha MO. Diagnosis and management of Chagas disease and cardiomyopathy. *Nat Rev Cardiol*. 2012;9(10):576–589.
7 Nunes MC, Dones W, Morillo CA, Encina JJ, Ribeiro AL. Chagas disease: an overview of clinical and epidemiological aspects. *J Am Coll Cardiol*. 2013;62(9):767–776.
8 Brito BOF, Ribeiro ALP. Electrocardiogram in Chagas disease. *Rev Soc Bras Med Trop*. 2018;51(5):570–577.
9 Dias JC. [Globalization, inequity and Chagas disease]. *Cad Saúde Pública*. 2007;23(Suppl 1):S13–S22.
10 Pinto Dias JC. Human Chagas disease and migration in the context of globalization: some particular aspects. *J Trop Med*. 2013;2013:789758.
11 Guhl F, Jaramillo C, Vallejo GA, Cardenas AAF, Aufderheide A. Chagas disease and human migration. *Mem Inst Oswaldo Cruz*. 2000;95(4):553–555.
12 Diseases GBD, Injuries C. Global burden of 369 diseases and injuries in 204 countries and territories, 1990-2019: a systematic analysis for the global burden of disease study 2019. *Lancet*. 2020;396(10258):1204–1222.
13 Liberati A, Altman DG, Tetzlaff J, et al. The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate health care interventions: explanation and elaboration. *J Clin Epidemiol*. 2009;62(10):e1–e34.
14 Hoy D, Brooks P, Woolf A, et al. Assessing risk of bias in prevalence studies: modification of an existing tool and evidence of interrater agreement. *J Clin Epidemiol*. 2012;65(9):934–939.
15 Dinnes J, Deeks J, Kirby J, Roderick P. A methodological review of how heterogeneity has been examined in systematic reviews of diagnostic test accuracy. *Health Technol Assess*. 2005;9(12):1–113.
16 Chadalawada S, Sillau S, Archuleta S, et al. Risk of chronic cardiomyopathy among patients with the acute phase or indeterminate form of Chagas disease: a systematic review and meta-analysis. *JAMA Netw Open*. 2020;3(8):e2015072.
17 Chadalawada S, Rassi A Jr, Samara O, et al. Mortality risk in chronic Chagas cardiomyopathy: a systematic review and meta-analysis. *ESC Heart Fail*. 2021;8(6):5466–5481.
18 Nunes MCP, Buss LF, Silva JLP, et al. Incidence and predictors of progression to chagas cardiomyopathy: long-term follow-up of trypanosoma cruzi-seropositive individuals. *Circulation*. 2021;144(19):1553–1566.
19 Rassi A Jr, Marin JAN, Rassi A. Chronic Chagas cardiomyopathy: a review of the main pathogenic mechanisms and the efficacy of aetiological treatment following the BENznidazole Evaluation for Interrupting Trypanosomiasis (BENEFIT) trial. *Mem Inst Oswaldo Cruz*. 2017;112(3):224–235.
20 Morillo CA, Marin-Neto JA, Avezum A, et al. Randomized trial of benznidazole for chronic Chagas' cardiomyopathy. *N Engl J Med*. 2015;373(14):1295–1306.
21 Nunes MCP, Badano LP, Marin-Neto JA, et al. Multimodality imaging evaluation of Chagas disease: an expert consensus of Brazilian cardiovascular imaging department (DIC) and the European association of cardiovascular imaging (EACVI). *Eur Heart J Cardiovasc Imaging*. 2018;19(4):459–460n.
22 Ralston K, Zaidel E, Acquatella H, et al. WHF recommendations for the use of echocardiography in chagas disease. *Glob Heart*. 2023;18(1):27.
23 Ribeiro AL, Marcolino MS, Prineas RJ, Lima-Costa MF. Electrocardiographic abnormalities in elderly Chagas disease patients: 10-year follow-up of the Bambui cohort study of aging. *J Am Heart Assoc*. 2014;3(1):e000632.
24 Brito BOF, Lima EM, Soliman EZ, Silva EF, Lima-Costa MF, Ribeiro ALP. The evolution of electrocardiographic abnormalities in the elderly with Chagas disease during 14 years of follow-up: the Bambui Cohort Study of Aging. *PLoS Negl Trop Dis*. 2023;17(6):e0011419.
25 Pinazo MJ, Canas E, Elizalde JI, et al. Diagnosis, management and treatment of chronic Chagas' gastrointestinal disease in areas where Trypanosoma cruzi infection is not endemic. *Gastroenterol Hepatol*. 2010;33(3):191–200.
26 Nunes da Costa EAP, Victoria C, Fortaleza C. Predictors of development of cardiac and digestive disorders among patients with indeterminate chronic Chagas Disease. *PLoS Negl Trop Dis*. 2021;15(8):e0009680.
27 Barros F, Soares JCA, Barros FP, et al. Agreement between upper endoscopy and esophagography in the diagnosis of megaesophagus in Chagas disease. *Rev Soc Bras Med Trop*. 2019;52:e20180258.
28 Dumont SM, Costa HS, Chaves AT, Nunes M, Marino VP, Rocha M. Radionuclide esophageal transit scintigraphy in chronic indeterminate and cardiac forms of Chagas disease. *Nucl Med Commun*. 2020;41(6):510–516.
29 Gonzalez NL, Moscatelli G, Moroni S, et al. Long-term cardiology outcomes in children after early treatment for Chagas disease, an observational study. *PLoS Negl Trop Dis*. 2022;16(12):e0010968.
30 Andrade SG, Campos RF, Sobral KS, Magalhaes JB, Guedes RS, Guerreiro ML. Reinfections with strains of Trypanosoma cruzi, of different biodemes as a factor of aggravation of myocarditis and myositis in mice. *Rev Soc Bras Med Trop*. 2006;39(1):1–8.