ELETROCARDIOGRAPHIC CHANGES IN CHRONIC *Trypanosoma cruzi* INFECTED *Cebus apella* MONKEYS

CARLOS A. FALASCA, DANIEL R. GRANA, EDUARDO A. MARESO, ELENA GOMEZ, MONICA M. GILI
Buenos Aires, Argentina

Purpose *To evaluate electrocardiographic data of Cebus apella monkeys with Chagas disease.*

Material and Methods *53* ***Cebus apella*** *monkeys (juvenile and adult of both sexes) were used: 35 as control group and 18 inoculated fourfive years ago with 3 different Trypanosoma cruzi strains (CA1, n = 10; Colombian, n = 4; Tulahuen, n = 4).*

Results *The normal electrocardiogram (ECG) showed differences with that of man, a) high cardiac rate; b) presence of pulmonary p wave without pulmonary pathology. The ECT alterations found between 11 and 58 months after last inoculation were. right bundle branch block; intermittent right bundle branch block; left ventricle overload; repolarization disturbances; left anterior hemiblock; extra systole. These alterations resemble those found in humans, as well as clinical parasitological and immunological alterations. Their incidence and the time at which they appeared, seem to vary according to the route, strain, inoculum andfrequency of the inoculation. Three of the monkeys died spontaneously 46, 48 and 52 months after the infections due to the natural evolution of the disease, and six were sacrificed during thefollowup. In both cases histopathological lesions were found, and their intensity was directly related to the time and resembled the human disease.*

Conclusion *The Cebus apella, as it reproduces human electrocardiographic and histopathological alterations, a short time after experimental infection, is a suitable modelfor the study of the different aspects of the physiopathology.*

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Address: Carlos A. Falasca —Tucuman 1845 — 1050, Buenos Aires, Argentina.

ALTERAÇÕES ELETROCARDIOGRÁFICAS EM MACACOS *Cebus apella* COM INFECÇÃO CHAGÁSICA CRÔNICA

Objetivo *Avaliar aspectos eletrocardiográficos da doença de Chagas em macacos Cebus apella.*

Casuística e Métodos *53 macacos Cebus apella (jovens e adultos de ambos os sexos) 35 como grupo controle e 18 inoculados 4 ou 5 anos atrás com três cepas diferentes de Trypanosoma cruzi (CA1, n = 10; Colombian, n = 4, Tulahuen, n = 4).*

Resultados *O eletrocardiograma basal mostrou-se diferente do ser humano, com alta freqüência cardíaca e ondas p pulmonale sem patologia pulmonar. Alterações em ECG entre 11 e 58 meses depois da primeira inoculação foram: bloqueio do ramo direito; bloqueio intermitente direito; sobrecarga ventricular esquerda; alterações de repolarização; hemibloqueio anterior esquerdo; extra-sístoles. Estas alterações se assemelham às encontradas nos humanos, assim como também as alterações clínicas, parasitológicas e imunológicas.*

Sua incidência e o tempo de aparição parecem variar segundo a cepa, o inóculo e a freqüência de inoculação. Três dos macacos morreram espontaneamente 46, 48 e 52 meses depois da infecção, por causa de evolução natural da doença. Seis sacrificados durante o seguimento tinham lesões histopatológicas cujos intensidades se relacionavam diretamente com o tempo e se assemelhavam à doença humana.

Conclusão *O* ***Cebus apella*** *é modelo apropriado para o estudo dos diferentes aspectos da doença de Chagas, particularmente aqueles relacionados com as fases indeterminada e crônica, já que reproduz alterações eletrocardiográficas encontradas no ser humano.*

Palavras-chave *Tripanosomíase americana; alterações eletrocardiográficas; modelo animal.*

Immunopathology and therapeutics of the indeterminate and chronic phases of Chagas disease.

Key words *American trypanosomiasis, ECG changes; monkey model.*

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Chagas disease or American trypanosomiasis is one of the principal causes of death in many Latinamerican countries, from the south of the United States of America to Argentina, and affects at least 20 million people in the continent[1-3].

Many questions concerning the pathogenesis of the chagasic myocardial damage have not been answered yet. Different immunological[4,5], hypoxemic[6,7] and neurogenic[8] mechanisms have been proposed, but most of its aspects still remain unknown.

One of the main difficulties was to obtain an experimental model, able to develop electrocardiographic and their correlated anatomopathological alterations similar to those found in humans during the chronic stage, and the possibility of studying the natural evolution of the different phases of this disease in humans.

The aim of the present work is to describe in the **Cebus apella**, a New World primate from the Paraguayan Chaco, naturally infected[9], the normal ECG patterns and their modifications by the experimental infection with different *T. cruzi* strains during the first five-years follow-up and to emphasize that these alterations joined to those found in anatomopathological studies, would suggest that this primate is a suitable animal model for the study of this complex endemic disease. Partial clinical, parasitological[10], pathological[11] echocardiographic [9], immunologic al 12 and gastroenterological aspects (unpublished data), have been previously reported.

MATERIAL AND METHODS

Fifty-three Cebus monkeys, selected from the out-door colony, were utilized and lodged in individual cages, in our indoor colony. They had free water provision and their feeding was based on a standard pellet diet (25% protein, 290 calories/100 g. Cargill, Buenos Aires, Argentina).

The primates were free from natural chagasic infection and other diseases, as shown by serology, xenodiagnosis, electrocardiographic[13] and hematologic parameters and seric enzymes[14].

They were divided into four groups, one control and three infected with three different *T. cruzi* strains, CA1, Colombian and Thlahuen. The experimental design is summarized in table I.

TABLE I Experimental design.

Strain / rial and methods	CA1	Colombian	Tulahuen	Control Mate-
Number of animals	10	4	4	35
Sex	male	2 male - 2 female	male	male
Estimated age at first inoculation (years)	6-10	1,2-3	4-4,5	5-9
Weight (g)	210-3320	940-1800	1660-1950	2250-257
Date of first inoculation	06-80,07-81	09-82, 10-82	11-82, 12-82	–
Number of inoculation at 06-84	1/2	17/18	10/11	–
Number of *T. cruzi* (each inoculation)	$4X10^2$ to $1X10^6$	$3X10^6$	$3X10^6$	–
Route	conjunctival	i.p.	i.p.	–

The periodic reinfections were carried out in order to put the animals in conditions similar to those found in humans, living in endemic areas, where the periodicity of natural reinfection varies and the number of parasites entering the circulation along their lives is not known.

The parasitemia, fresh drop, Strout test[15] and xenodiagnosis[16], conventional serology (indirect hemagglutination IHA, Cellognost Chagas Bochringwerker, and IgG by Enzyme-Linked Immunosorbent Assay — ELISA)[17] and electrocardiographic modifications, were evaluated.

These studies were performed once a week during the first three months of infection and then twice a month, during the first two years post-infection. At present they are carried out once a month, in the surviving infected and control animals.

The handling of the animals was performed under 10 mg/kglw ketamine hydrochloride anesthesia (Ketalar, Parke Davis, Buenos Aires, Argentina). The animals were placed in a dorsal decubitus position on an adequate stretcher and after 5 min. rest, an ECG was recorded with a Fukuda FJC-7100 monitor at a speed of 25 and 50 mm/sec.

Needle electrodes were placed in he subcutaneous cellular tissue in order to reduce dermic impedance. The limb electrodes were placed on the anterior portion of the forearms and on the inner portion of both legs.

The precordial leads used were V1 to V6 as in humans, and V1R and V3R in order to obtain a better evaluation of the right cavities.

Six infected animals, sacrificed for anatomopathological studies, were selected at random from those inoculated that presented ECG disturbances and with a post-infection survival, ranging from 11 to 58 months. Simultaneously, four controls were also sacrificed for the same purpose.

All organs and tissues were examined for gross section and fixed in conventional formaldehyde buffer and Zamboni, for hematoxylin-eosin, and special technics (Massons's Tricromic).

RESULTS

No alterations in the parameters studied were observed in the control group, during the first five years of the experience.

None of the infected monkeys showed neither clinical signs of illness or fever nor loss of weight, during the course of the infection.

In the three groups of animals infected with *Trypanosoma cruzi*, positive parasitemia was detected by the fresch drop, the Strout's test and/or the xenodiagnosis, until week 21, 52 and 80 for the CA1, Colombian and Tulahuen strain respectively. Specific serology for anti *Trypanosoma cruzi*, determined by IHA and ELISA, became positive from week 4 on. The highest titers detected by IHA ranged from 1/128 for the CA1 and Colombian strains to 1/256 and 1/1050 for the Tulahuen strains, being lower from week 49 onwards.

The electrocardiographic parameters of the control group are summarized in table II and an ECG corresponding to one of these monkeys is represented in figure 1.

The cardiac rate at rest and under light anesthesia varied between 220 and 316 beats per minute. The P wave was narrow, of short duration (0.04 sec.) and with a relatively high voltage (1.46 mm), similar to the "P pulmonale" wave of the human.

In 3 out of 35 monkeys, a RSr' in V1 and V2, that was not accompanied by any other ECG alteration, was observed. The presence of a small and narrow Q (0.5 mm and 0.02 sec) in D2, D3 and a VF was detected in some snimals without QS morphology. A septal Q in V3, V4 was coincident with a gross anatomy in which the anterior face of the heart was formed by both ventricles. The presence of S1-S2-S3 was interpreted as an "apex behind" heart.

The medium electric axis ranged from +70° to -60°. The 59% of the 35 monkeys showed an average axis of +48.81° with a range of +30° to +70° whereas the 11% had their axis at -60º, the 11° between -30° and 45º, the 11% indeterminate, the 5% at 0° and the 3% on the right.

TABLE II Mean values of the electrocardiographic parameters in a control group of *cebus* monkeys (n = 35).

	HR beats/min	AQRS degrees	QRS sec	P(D2) sec	P mm	PR (D2) sec	QT (D2) sec	Int. Deflex sec	T (V1) mm
X	270.51	18.13	0.039	0.04	1.46	0.07	0.16	0.0193	1.31
S.D.	23.34	45.72	0.002	0.00	0.27	0.01	0.007	0.0011	0.33
S.E.	3.74	8.08	0.001	0.00	0.04	0.002	0.001	0.0003	0.11

TABLE III - Electrocardiographic disturbances in *cebus m*onkeys inoculated with different *T. cruzi* strains and examined during the chronic phase.

ECG characteristics	CA 1 (n = 10)			Colombian (n = 4)			Tulahuen (n = 4)		
	%	A/T	t	%	A/T	t	%	A/T	t
IIRBB	20	2/10	32-47	0	0/4	20	0	0/4	20
RBBB	40*	4/10	27-47	25	¼	18	25	1/4	18
LVO	10	1/10	47	0	0/4	20	25	1/4	58
RD	20	2/10	47-48	50	2/4	20-37	50	2/4	18
LAH	10*	1/10	27	25	1/4	13	0	0/4	18
SE	10	1/10	27	0	0/4	0	20	0/4	18
Total pathologic ECG	90	9/10	27-48	100	4/4	13-37	100	4/4	18-58

%: percent of animals
t: time in months elapsed after 1st inoculation
A/T: number of animals total
*: one animal had both pathologies associated
IIRBB: intermittent right bundle branch block; RBBB. right bundle branch block; LVO: left ventricle overload; RD: repolarization disturbance; LAH. left anterior hemiblock; SE supraventricular extrasystole.

The ST segment was isoelectric in all the leads and rectification was observed in V5-V6 in the 10% of the animals. The T wave was positive in the horizontal and frontal planes, except in aVR that was always negative and occasionally in aVL.

Supraventricular extrasystoles were not observed in any of the monkeys studied.

None of the animals died spontaneously during the acute phase of the infection that was subclinical and without basal ECG alterations.

In the chronic stage, alterations in the ECG pattern were recorded in the 94% of the monkeys. The incidence of the alterations found in each one of the groups in summarized in table III.

The main ECG alterations observed are shown in figures 2a, b, c and 3a, b, c. The electrocardiographic patterns in the group of infected animals resemble those described in the human chagasic cardiomyopathy in the chronic phase[18-20].

The electrocardiographic disturbances found in the last ECG, of the three animals that died spontaneously after infection, 52, 48 and 46 months later, were respectively: repolarization disturbances, sinus bradycardia with conduction disturbances of the right bundle branch and left ventricle hypertrophy with left auricle enlargement.

In the six monkeys sacrificed (3 with the CA 1 strain, 1 with the Tulahuen and 2 with the Colombian), between 11 and 47 months post infection, septal fibrosis was the predominant lesion in the animals with the longest time of evolution (fig. 4a, b, c). The overall results concerning this matter were described in another paper[11].

Fig. 1 - Normal ECG tracing in the Cebus apella monkey.

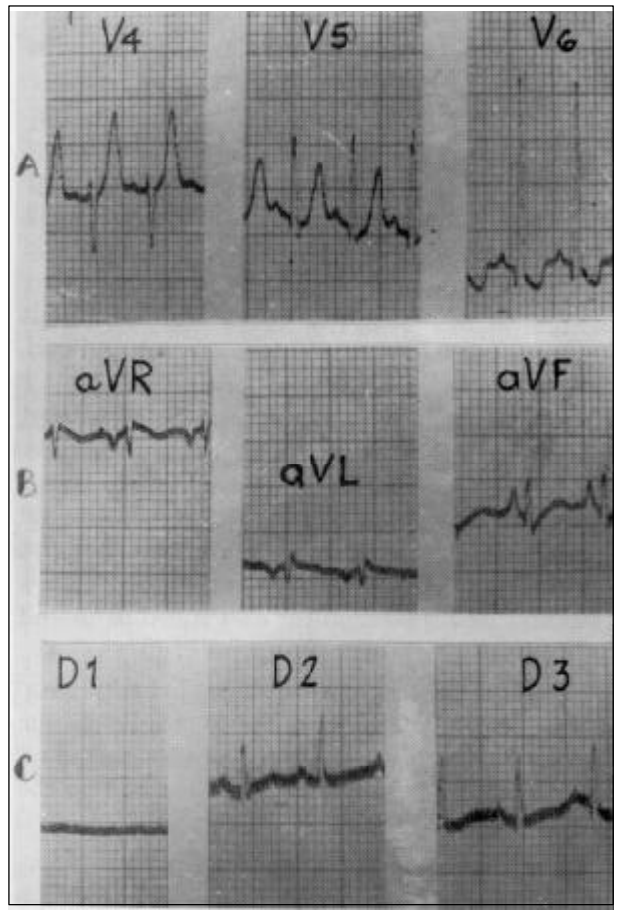

Fig. 2 - a) left ventricle systolic overload diaghragmatic fibrosis; b) left ventricle repolarization disturbances (upper lateral) face; c) sinus bradycardia (170 beats/ min).

DISCUSSION

The necessity of an experimental model of chronic Chagas' disease and particularly of the chagasic cardiomyopathology observed in the 90% of the autopsies[21] of patients dying for this reason, is a reality that the Program of Tropical Diseases of the World Health Organization has been studying for several years, in order to be able to progress in the knowledge of the immunopathogenesis of the chronic phase.

The electrocardiographic alterations produced by the experimental infection with Trypanosoma cruzi have been studied in mice[22], rats[23], dogs[24,25], rabbits[26]

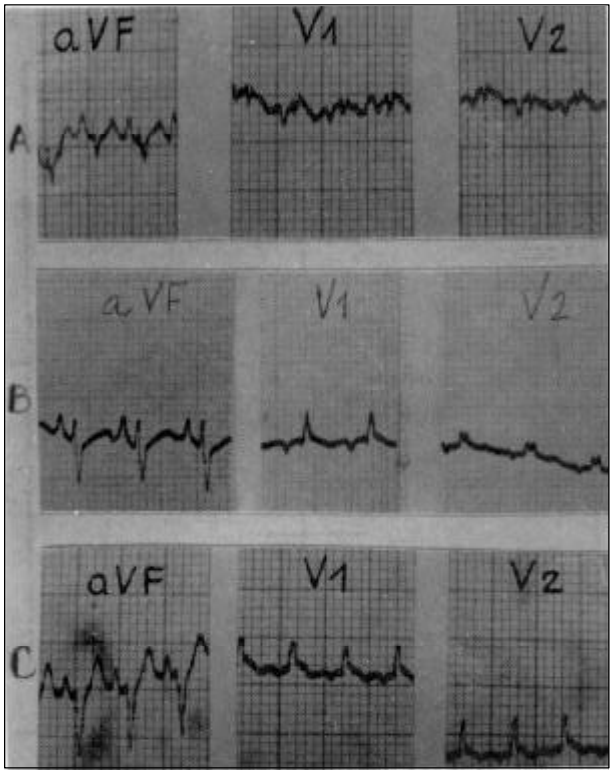

Fig.3 - a) right bundle block; b) right ventricle hypertrophy, presence of Q wave in V1 (enlargement of right auricle) with left anterior hemiblock associated, conductive disturbance of the right ventricle hypertrophy; c) left anterior hemiblock and right ventricle hypertrophy.

being the alterations non specific and difficult to interprete even in the non infected controls[27] and consequently they cannot be used as a good element to demonstrate, the cardiomyopathy in the chronic stage. Complementary studies of some of these models with auxiliary methods of diagnosis (echocardiography), scintigraphy) are almost impossible due to the anatomical size of the heart, whereas in the Cebas appela monkey, the echocardiography has been used by us with very good results[28], as well as the scintigraphy[29]. The course of experimental Chagas' disease in adult *Cebus sp* moneky and other primates, has been previously studied, by other groups, with not very clear results[30-34].

It must be emphasized, as a very important difference, with other experimental models, that in general, the ECG of the *Cebus apella* is similar to that of man and other non human primates, but there exist some differences to be recognized. The high cardiac rate that can be due to the small corporal size and probably, also, to the neurovegetative discharge produced by the preexam catching and the anesthesia (270.51 + 23.34 a min.), a narrow P wave, of short duration and a relatively high voltage of 1.46 mm, similar to the "P pulmonale" that was also described

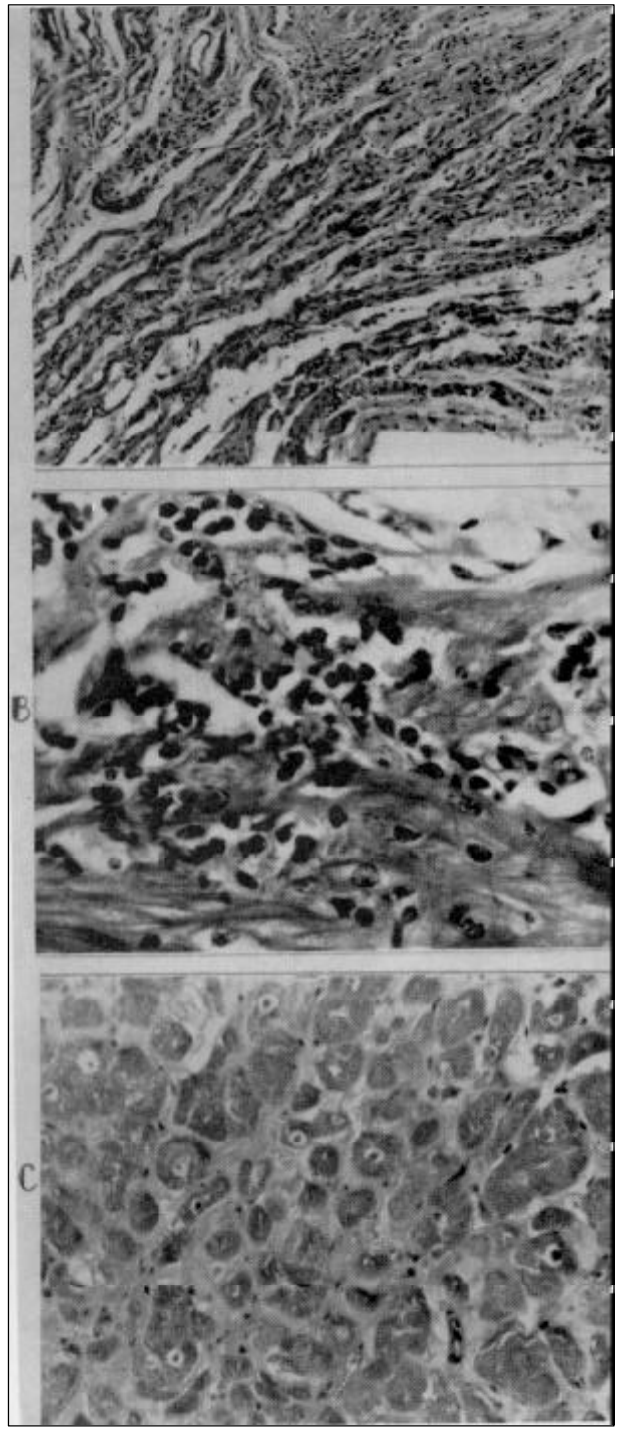

Fig. 4 - Atrioventricular node in which moderate infiltrates are observed among the specialized fibers, 67 month post-infection (HE 32x); b) ventricular wall with mononuclear infiltrates and myocitolysis 11 months post-infection (HE 400 x); c) interventricular septum with inersticial and substitution fibrosis 47 months post-infection (Masson's trichromic 400 x).

in the Old World primates[35,36] and that it is not associated to chronic pulmonary disease as it occurs in human pathology; a short PR interval with a mean value of 0.07 sec.

When analysing the changes produced by the experimental infection, in the conduction system, it

must be remembered that the 22% of the monkeys had in the ECG a AQRS between -30º and -60º and an "apex behind" heart (S1 - S2 - S3), in order to avoid wrong interpretations. It is also very important to point out that in the study of this pathology neither arrhythmias nor spontaneous right bundle branch block were observed in this monkey as they are in the rhesus monkey[37,38].

During the first five-years-follow-up, the animals of the control group did not present any kind of alterations of the ECG. This fact, makes this animal particularly different from other experimental models commonly used[22 27],

Chagas' disease in *Cebus* monkeys and in human share many similarities[8,10]. The acute phase of the infection in this host went unrecognized, and patent parasitemia usually existed independently of the parasite strain used.

The immunological test remained positive during the undeterminte phase, in which the parasite was difficult to demonstrate.

The absence of correlation between parasitemia and morbidity and/or mortality was as in human.

The electrocardiographic alterations in *Trypanosoma cruzi* infected monkeys were recorded from the 11th month post-inoculation up to the present. The 94% of the animals showed alterations of the electrocardiographic pattern after the experimental infection with different Trypanosoma cruzi strains, independently of the age and sex.

The electrocardiographic patterns more frequently observed (Table II) are those described in human chronic chagasic pathology[18,20], were permanent along the experience once appeared and of easy interpretation in the records.

These alterations correlated with he histopathological findings in the animals sacrificed, in which the lesions were of the myocarditis type with myocytolysis, and areas of scattered fibrosis, according to the evolution of the diseases[11].

CONCLUSIONS

According to Brener's requirements[39] this New World non human primate is an experimental model suitable for the study of chronic Chagas'disease. The precocity of the ECG and histopathological alterations favors the utilization of this primate for the study of the natural evolution of this disease, especially in the indeterminate and chronic phased[11].

The variation of the inoculum, the route, the frequency of the inoculations, in addition to the virulence of the strain utilized, might play an important role in the natural evolution of the disease, as well as in its pathogenesis and immunopathology.

This pathology, still unknown and difficult to treat, continues to be, as Chagas said, one of the most important and serious medico-social problems in the Latinamerican rural areas, in which millions of persons are infected. Nevertheless, at present, it is also a serious problem in the urban zones since the frequency of this pathology increases, due to the internal migratiosn and transfusional transmission by non detected chagasic donors. The treatment available is far from satisfactory and a vaccine may not be possible by conventional means, up to the present.

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REFERÊNCIAS

1. World Health Organization Teehnical Report Series 697 Chagas' disease, cardiomyopathies. Report of a WHO Committee. World Health Organization, Genebra, 1984; 50-5.
2. Chagas, C. Nova trypanosomiaze humana. Estudos sobre a morfologia e o ciclo evolutivo do Schizotripanum cruzi n. gen.. n. sp, agente etologico de nova entidade morbida do homem, Mem Inst Osw Cruz, 1909; 1: 159-218.
3. Mazza — Caso de Schizotripaniosis humana observado en Jujuy. Rev Univ Bs As Argentina, 1926; 4: 400-3
4. Cossio PM, Laguens RP, Diez C, Szarfman A, Segal A, Arana RM — Chagasic cardiopathy: antibodies reacting with plsma membrane of striated muscle and endothelial cells. Circulation, 1974; 50:1252-9.
5. Teixeira ARL, Teixeira L, Santos-Busch CA — The immunology of experimental Chagas'disease IV. The production of lesions in rabbits similar to those of chronic Chagas'disease in man. Am J Pathol, 1975; 80: 163-77.
6. Jorg ME — Tripanosomiasis cruzi: anarquia angiotopografica por descapilarizacion mesenquimorreactiva, cofactor patogenico e la miocardiopatia cronica. Pren Med Arg, 1974, 61: 94-106.
7. Torres CM — Miocitolise e fibrose do miocardio na doenca de Chagas. Mem Inst Oswaldo Cruz, 1960; 58: 161-82.
8. Koberle F — Chagas' disease and Chagas' syndromes: the pathology of American trypanosomiasis In: Dawes B, ed Advances in Parasitology Vol. 6 London: Academic Press, 1968; 63-116.
9. Falasca CA. Mareso C, Grana D, Merlo A, Gili M — Alteraciones ecocardiograficas (modo M) en un primate no humano, Cebus apella, con infeccion natural cronica por Tripanosoma cruzi Ultrasonografia Med Biol, 1989; 3:96-102.
10. Falasca CA, Grana D, Buccolo J, Gili M, Merlo A, Zoppi J, Mareso E Susceptibility of the Cebus apella monkey to different strains of T cruzi after single or repeated inoculation. PAHO Bulletin, 1986; 20:117-37.
11. Falasca CA, Gili M, Grana D, Gomez E, Zoppi J, Maresco E — Chronic myocardial damage in experimental T. cruzi infection of a new world primate. Cebus sp. monkey. Rev inst Med Trop São Paulo 1990; 32:151-61.

12. Eiguchi de Palmero K, Carbonetto CH, Malchiodi EL, Margni RA, Falasca CA — Humoral and cellular immunological profile in Cebus apella monkeys, a model for chronic Chagas'disease. Cross Reactivity with human immunoglobulins. Veterinary Immunology and Immunopathology, 1988; 19: 341-9.
13 Falasca CA — Parametos electrocardiograficos y ecocardiograficos en el primate Cebus apella, un modelo experimental cronico para la enfermdade de Chagas. Boletines de la Academia Nacional de Medicina, 1985; 63 (1 Sem): 179 82.
14. Grana D, Mino J, Merlo A, Gomez E, Sesma W, Falasca CA — Parametros normales de laboratorio de un primate del nuevo mundo Cebus appela. Revista de la Sociedad de Medicina Veterinaria Bs As, 1988; 69: 156-60.
15. Strout RG — J A method for concentrating hemoflagellates. J Parasitol, 1962; 48:100.
16. Brumpt EO — O xenodiagnostico. Aplicação do diagnostico e algunas infecciones parasitarias em particular tripanosomiase de Chagas. Ann Paul Med Cir, 1914; 3:97-117.
17. Voller A, Draper CC, Bidwell DF, Bartlett A—Microplate enzyme. Linked immunosorbent assay for Chagas' disease. Lancet, 1975; 1:426 7.
18. Rosembaum MB, Alvarez A — The ECG in chronic chagasic myocarditis. A M Heart, 1955; 50: 492-527
19. Rosembaum MB, Lazzari JO, Elizari MV—Los hemobloqueos. Ed Paidos, 1970.
20. Capris TA, Fernandez Moores AJ — Alteraciones electrocardiograficas en la cardiopatia chagasica cronica. Rev Arg Cardiol, 1967; 34:200-4.
21. Koberle F—Pathogenesis of Chagas' disease. CIBA Foundation Symp, 1974; 20.
22. Laguens RP, Meckert PC, Gelipi RJ — Chronic Chagas' disease in the mouse I. Electrocardiographic and morphological patterns of the cardiopathy. Medicina (Buenos Aires), 1981; 41: 35-9.
23. Revelli SS, Amerio N. Moreno HS, Valenti JL, Balbarrey H, Morini JC—Enfermedad de Chagas cronica en la rata. Caracteristicas serologicas, electrocardiograficas e histopatologicas. Medicina (Buenos Aires), 1980; 40 (supl. 1): 69-76.
24. Anselmi A, Pifano F, Suarez A, Dominguez A, Vazquez A, Anselmi G—Experimental Schizotrypanum cruzi myocarditis. Correlation between histopatologic and electrocardiographic findings in experimental Chagas' heart disease. American Heart Journal, 1965; 70:683-5.
25. Marsden P, Hagstron JWC—Experimental Trypanosoma cruzi infection in beagle puppies. The effect of variations in the dose and source of infecting trypanosomes and the route of inoculations on the course of the infection. Trans Roy Soc Trop Med Hyg, 1968; 62:816-24.
26. Teixeira ARL, Figueiredo F. Rezende FJ, Macedo B — Chagas' disease: a clinical, parasitological, immunological and pathological study in rabbits. American Journal of Tropical Medicine and Hygiene, 1983; 32: 258-72.
27. De Oliveira JSM de, Bestetti RB, Garcia Soares E, Marin Neto JA — Ajmaline induced electrocardiographic changes in chronic Trypanosoma cruzi infected rats, 1986; 80: 415-9.
28. Gili M, Grana D, Mareso E, Garcilazo E, Falasca CA — Ecocardiografia en Modo M normal y patologica en el mono Cebus, un modelo experimental cronico de cardiopatia chagasica. VIII Jornadas Rioplatenses de Cardiologia. Buenos Aires. Diciembre, 1986.
29. Perez Balino N, Gili M, Grana D, Sporn V, Meretta A, Falasca CA—Miocardiopatia chagasica cronica: estudio de la perfusion miocardica con RP30 Tc 99 m en un modelo experimental en primates del Nuevo Mundo (Cebus apella). Medicina (BS As), 1989; 19: 468.
30. Dorland JD—Infection in monkeys with strains of Trypanosoma cruzi isolated in the United States. US Pub Health Resp, 1943; 58:1006-10.
31. Torres CM, Tavares BM—Miocardite no macaco Cebus apos inoculações repetidas con Schizotrypanum cruzi. Mem Inst Oswaldo Cruz, 1958; 56: 85-152
32. Marsden P. Seach SKK, Draper CC, Pettitt LE, Miles MA, Voller A — Experimental T. cruzi infections in Rhesus monkeys. II. The early chronic phase. Trans Roy Soc Trop Med Hyg, 1976; 70: 247-51.
33. Bolomo NJ, Milei J, Cossio PM, Segura E, Laguens R, Fernandez L, Arana R—Experimental Chagas' disease in South American non human primate (Cebus sp. monkey). Medicina (Buenos Aires), 1980; 40:667-72
34. Bolomo NK, Milei J, Cossio P, Nagle C, Arana R, Del Prado CE, Segura EL—Infeccion cronica por Trypanosoma cruzi en el Cebus sp. adulto. Medicina (Buenos Aires), 1981; 41: 678-9.
35. Malinow MR, De Lannoy CW—The electrocardiogram of Macaca Fuscata. Folia primatol, 1967; 7: 284.
36. Malinow MR, De Lanney CWThe electrocardiogram of Cyno pithecus niger. Folia primatol, 1966; 4:66.
37. Klein HO, Sullivan TA, Hoffman BF—Ectopic atial rhythms in the primate. Am Heart J, 1974; 87: 750.
38. Bristow JD, Malinow MR—Spontaneous bundle branch block in rhesus monkeys. Circ Res, 1965; 16: 210.
39. Brener Z—Progres recents dans le domaine de la malade de Chagas. Bull Org Mond Sante, 1982; 60: 845-56.