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ORIGINAL ARTICLE

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Chronotopic and Blood Pressure Response to Oral Glucose Load in Chagas' Disease

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Cardiac chronotropic and pressor responses after an oral load of glucose were assessed in sixteen Chagasic subjects and 28 controls by means of blood pressure and pulse rate measurements. Cardiovascular response was correlated with serum insulin and glucose levels. The experiment identified a subgroup of Chagasic subjects (n=8) with a hypoinsulinemic behavior presenting less chronotropic and pressor responses than controls. This may indicate a lower insulin activity and/or an early Autonomic Nervous System dysfunction in this subgroup.

UNITERMS: Cardiovascular response, insulinemic response, Autonomic Nervous System, South America Trypanosomiasis.

INTRODUCTION:

Ever since the first report on South American Trypanosomiasis in 1909, this syndrome has been extensively studied with the aim of understanding the physiopathogenic link underlying damage to the Autonomic Nervous System (ANS) and dysfunction of the cardiovascular system. (1,5,6,15). However, metabolic changes in this disease have not receive a great deal of attention to date.

In a recent study, variations in the glucose tolerance test (GTT) were found to relate to changes in gastric emptying rate (18). Moreover, pancreatic involvement by Trypanosome cuzi has been documented and pancreatic islet size increase has also been documented in Chagasic subjects when compared with control subjects (27,35). Regarding pancreatic endocrine function in Chagas' disease, a delay in blood glucose recovery after the insulin tolerance test, as well as lower insulin levels after oral glucose ingestion, has been observed in subjects with the intestinal type of Chagas' disease when compared with control subjects (11,17).

Additionally, insulin has been proposed to affect the cardiovascular system directly or by means of enhancing the activity of the Sympathetic Autonomic Nervous System through changes in ionic cell transport (3,7,21,29,33,36). However, only a few and contradictory studies have been carried out on the correlation between metabolic, arterial blood pressure and chronotropic activity change responses in Chagasic patients.

The objective of the present investigation was to evaluate the behavior of the cardiovascular responses to serum insulin and to glucose load in subjects with the indeterminate form of Chagas' disease — ICF (8).

MATERIAL AND METHODS

Sixteen Chagasic subjects (15 males and 1 female) aged 20 to 45 years were included. All subjects reacted positively in at least two serological tests for Chagas' disease (complement fixation — 1/4; and indirect Chagas immunefluorescence test — 1/40) and had strong personal

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epidemiological foregoing indicating the presence of the disease. All subjects were asymptomatic ICF with unremarkable physical exams, normal resting EKG, as well as unremarkable chest, esophagus and colon roentgenograms. None of the subjects had history of either diabetes mellitus or hypertension. No subjects presented a glucose curve indicative of intolerance after an oral glucose overload (23). A normal blood pressure measure was obtained in 3 different occasions for all subjects and body weight fell within the 20% of the ideal body weight as defined by the Metropolitan Life Insurance tables (1959). No subjects were taking any medications 7 days before testing.

Results obtained from the 16 Chagasic patients (Ch) were compared to a control group (C) consisting of 28 subjects (18 males and 10 females) aged 20 to 45 years. The control group fulfilled exactly the same criteria as the Chagasic subjects except for an absence of serological reactions and personal epidemiological history. The women were submitted to testing during the follicular phase of the menstrual cycle. Informed consent was obtained from both groups of subjects (Ch and C) to participate in the study that was conducted in accordance with the guidelines of the Declaration of Helsinki (1964) for human experimentation.

Chagasic and control subjects were submitted to an oral glucose tolerance test — GTT.*(13). One hundred grams of glucose were given orally at 0700 hours after an all-night 12 - to -14 - hour fasting period. Blood samples were drawn 30 minutes before and at 0, 30, 60, 90 and 120 minutes after glucose intake. Blood pressure (BP) was measured from the right arm employing a standard Mercury manometer in keeping with the recommendations made by the American Heart Association *1(12) (references for the GTT and American Heart Association are desirable). Heart rate (HR) was obtained and measured from pulse palpation for 30 seconds. Blood pressure was measured at 10-minute intervals from 0700 to 1100 hours.

Biochemical Tests

Serum glucose was measured by means of the glucose oxidase method (19). Insulin level was measured by means of radioimmuneassay employing the double antibody method for human insulin (34).

The following parameters were calculated:

Mean + - SEM of the baseline blood pressure.

Mean + - SEM of the baseline heart rate.

Integrated insulin secretion, i.e., the total area under the insulin secretion curve - TAUC. This was calculated taking into account the trapeziodal area under the curve. Confidence limits of the controls for single values were used for TAUC comparisons.

Double cross index — DCI that corresponds to the systolic blood pressure (SBP) value multiplied by HR. Percentage variation - var % for SBP - - SBP var %..

Percentage variation - var % for diastolic blood pressure DBP - - DBP var %..

The latter three measures relate to values obtained before glucose intake.

Statistical Analysis

Serum glucose, insulin, HR, SBP, DBP, DCI were statistically analyzed employing the one-way analysis of variance. Significance level was set at $p < 0.05$.

RESULTS

In the control group the pre-glucose load insulin level was 10.86 +- 5.18uU.ml-^{1} with a significant insulin peak at 60 minutes after GTT onset (112.29 +- 41.47uUml-^{1} $p< 0.05$ against baseline) followed by a continuous reduction up to 120 minutes (57.57 +- 29.05 uUml-^{1}). In this group, the TAUC for the different time intervals agreed with plasma insulin response (1560.00 +- 872.82 for 0-30 minutes; 3081.43+-1266.59 for 30-60 minutes; 3020.36 +- 1001.89 for 60 to 90 minutes; 2199.64 +- 973.81 for 90-120 minutes).Serum insulin curve after GTT in the Chagasic patients displayed a lower fasting insulin level for this group as compared to controls (5.94+-5.37uUml-^{1}). Maximum values were reached at 30 minutes followed by some oscillations and a discrete reduction starting at 60 minutes (see table 1).

There was no significant difference when the time distribution of the Chagasic group was compared to that of the mean values for the control group ($F = 2.55 < F = 5.32$).

However, the analysis of the confidence interval for 2 SD regarding the control group (5268uUml-^{1} . 120 min-^{1}1 < 10123uU.ml-^{1}. 120 min -^{1} < 1539uU.ml-^{1}. 120 min-^{1}) demonstrated a TAUC sharply differing between two subgroups of Chagasic subjects and the control group.In the first subgroup of eight Chagasic subjects, the mean TAUC for the 120 minute period was 9220 +- 825 uU.ml-^{1} (1561.88 +- 467.38 for 0-30 minutes; 2409.38 +- 742.54 for 30-60 minutes; 2184.38 +-1166.00 for 60-90 minutes and 1818.75 +- 708.17 for 90-120 minutes).

In the second subgroup of eight Chagasic subjects, the mean TAUC for the 120 minute period was 2976 +- 447.6 uU.ml-^{1} (551.25 +- 381.23 for 0-30 minutes; 802

+-454.35 for 30-60 minutes; 800.63 +- 266.32 for 60-90 minutes and 871 +- 449.59 for 90-120 minutes). The first subgroup was considered to show a normoinsulinemic pattern (NICh) and the second subgroup was hypoinsulinemic (HICh) (see figure 1b). The NICh subgroup showed a TAUC variation behavior that was similar to the control group (F^a =1.33 < F^{TM} =5.99). By contrast, there was a statistically significant difference from the control group at 5% level (F= 21.4 > F^a) for the HICh subgroup.

The NICh subgroup insulin curve (7.88 +- 7.08 at 0 minute; 96.25 +- 28.74 at 30 minutes; 64.38 +- 44.36 at 60 minutes; 81.25 +- 38.05 at 90; 40.00 +- 30.67 at 120 minutes) did not differ significantly from the control group (10.86 +- 5.18 at 0 minute; 93.14 +-54.37 at 30 minutes; 112.29 +- 41.47 at 60 minutes; 89.07 +- 40.71 at 90; 57.57 +- 29.05 at 120 minutes) with an F = 0.38 <. F^a = 5.32). On the other hand, the HICh subgroup insulin curve (4.00 at 0 minute; 32.75 +- 25.42 at 30 minutes; 20.75 +- 8.57 at 60 minutes; 32.63 +- 13.04 at 90 minutes; 25.50 +- 19.47 at 120 minutes) differ from the control group insulin curve (F = 7.13 > F^a).

The levels of the glucose curve in the control group presented discrete and insignificant oscillations around the baseline value (75.15 +- 9.89 mg%). During the GTT, serum glucose levels increased, reaching the highest value between 30 and 60 minutes (135.30 +-19.86 mg %). None of the control subjects demonstrated a clear-cut glucose intolerance response (fig 1a). The response of the Chagasic subjects (74.25 +- 9.56 at 0 minute; 139.63 +- 22.27 at 30 minutes; 139.19 +- 26.01 at 60 minutes; 99.63 +- 28.83 at 90 minutes; 88.44 +- 22.00 at 120 minutes) was not significantly different from controls (F=0.22 < F^a). There was no statistically significant difference in the glucose curve between the two Chagasic subgroups — NICh and HICh and controls (F= 0.28 and F= 0.003 < F^a).

HR Var percentage (fig 2 c) of the NICh subgroup (6.89 +- 12.15 at 30 minutes ; 15.45+-14.19 at 60 minutes ; 13.70 +-11.80 at 90 minutes and 11.87 +- 12.31 at 120 minutes) was similar to the control group (9.77 +- 9.88 at 30 minutes ; 9.41 +- 8.34 at 60 minutes ; 10.90 +-7.56 at 90 minutes and 10.13 +- 9.51 at 120 minutes) with an F value = 1.06 < F^a (see figure 2 c).

HR Var % (fig 2 c) of the HICh subgroup (3.39 +- 3.68 at 30 minutes ; 6.60 +- 8.03 at 60 minutes ; 5.82 +- 7.08 at 90 minutes and 6.85 + -5.45 at 120 minutes) was significantly lower than the control group (F= 30.92 < F^{TM}). The SPB % for the NICh subgroup was (figure 2 a): 0.13 +- 6.29 at 30 minutes ; -2.25 +- 5.24 at 60 minutes ; 0.30 +- 8.66 at 90 minutes ; and -1.30 +- 6.24 at 120 minutes and the SPB % for the HICh subgroup was 0.85 +- 5.02 at 30 minutes ; -3.60 +-7.13 at 60 minutes; -3.93 +- 6.09 at 90 minutes ; and -1.62 +-8.22 at 120 minutes. Both values were significantly lower than the control group (2.91 +- 6.59 at 30 minutes ; 4.60 +- 7.84 at 60 minutes ; 2.25 +- 5.36 at 90 minutes ; and - 2.38 +- 6.51 at 120 minutes with an F value = 24.96 > F^a for the NICh subgroup and a F value = 18.56 > F^a for the HICh subgroup.

The DBP Var % for NICh (fig 2b) (-10.94 +- 12.91 at 30 minutes ; -14.16 +- 8.41 at 60 minutes ; -11.70 +- 11.85 at 90 minutes ; and -6.59 +- 6.89 at 120 minutes) was significantly lower than the control group (-3.55 +- 8.45 at 30 minutes ; -3.97 +- 8.53 at 60 minutes ; -4.56 +- 8.28 at 90 minutes ; -6.34 +- 7.56 at 120 minutes; F = 13.6 > F^a). The DBP Var % for HICh group (-4.83 +- 5.90 at 30 minutes ; -3.94 +- 8.39 at 60 minutes ; 0.41 +- 10.70 at 90 minutes ; -0.90 +- 7.21 at 120 minutes; F = 2.75 < F^a).

The DCI Var % of the control group (fig 2 d) (13.30 +- 15.26 at 30 minutes ; 14.64 +- 13.96 at 60 minutes; 13.52 +- 11.17 at 90 minutes ; and 13.01 +- 14.47 at 120 minutes did not differ in a significant way from the NICh subgroup (6.99 +- 14.44 at 30 minutes ; 11.66 +- 11.58 at 60 minutes ; 14.02 +- 16.20 at 90 minutes ; and 10.59 +- 16.16 at 120 minutes) with a F value = 3.44 < F^a but it was significantly higher than the HICh subgroup (4.01 +- 4.33 at 30 minutes ; 2.00 +- 9.89 at 60 minutes; 1.40 +- 7.89 at 90 minutes ; and 5.12 +- 12.18 at 120 minutes) with an F value = 2636 > F^a.

DISCUSSION

The analysis of the confidence limits for the control TAUC permitted the authors to further separate the ICF Chagasic subjects in two subgroups, namely normoinsulinemic (NICh) and hypoinsulinemic (HICh). Contrasting to the report by Long et al, the response obtained in the HICh in this present study could not be associated to digestive tract changes undetected in the physical exam or by the evaluation methods employed herein this study (17). Additionally, absorption or intestinal transit disorders are not commonly seen in this form of Chagas' disease (8). Rocha et al demonstrated in chronic Chagas' disease subjects a clear tendency for a considerable dispersion of the glucose values after an oral GTT (32). However, glucose levels similar to normal subjects were obtained in the same subjects after an EV GTT, thus demonstrating a defective glucose absorption mechanism in Chagas' disease (32).

Other studies with Chagasic subjects not classified as ICF demonstrated an increase rather than a decrease in glucose absorption that was associated with an intolerance pattern of the oral GTT (4, 22, 31).

These studies made clear that changes in the glucose absorption related to the pattern of the blood glucose curve.The study herein with ICF subjects (NICh and HICh) produced blood glucose curves similar to controls and without any tendency for dispersion. This is suggestive that our subjects had no abnormality in the glucose absorption mechanism.. On the basis of other literature reports, the authors judged that the presence of abnormalities of the entero-insular axis seemed to be unlikely in the subjects reported herein. Nonetheless, no objective evaluation of the axis was performed in this study.

On the other hand, the NICh subgroup may include subjects whose pancreatic islet may be increased in area (35). This indicates a compensatory mechanism on the part of the pancreas to counteract the secretory deficiency caused by the ANS dysfunction. Yet, this phenomenon occurs independently of an abnormal oral GTT blood glucose response as all 8 NICh subjects in this report had no intolerance response to the test (fig 1 a).

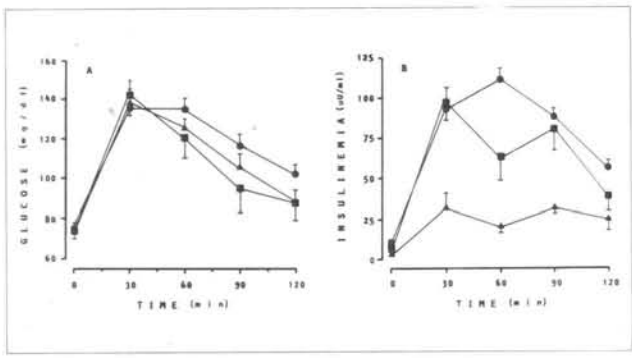

Figure 1:
Plasma glucose (A) and insulin (B) level measured at 30 minutes intervals after the oral glucose load ($\bar{x}$+SE)
l = control n = normoinsulinemic Chagasic subjects - NICh
s = hypoinsulinemic Chagasic subjects - NICh

This NICh subgroup may well represent an imbalance between the tonic inhibitory activity of the sympathetic alpha-receptors and the beta-adrenergic stimulatory activity at the cellular level in the pancreatic islet. This has been reported elsewhere (2, 28).There is substantial experimental evidence showing that insulin and/or glucose increase blood pressure and pulse rate (HR) by different mechanisms. Sympathetic activation, sodium reabsorption by the kidney and a direct action upon the myocardium have been described elsewhere (9,10,16,18, 33,36). A surge in the arterial blood pressure was confirmed in both the control and NICh subjects in this study (figure 2). By contrast, the HICh subgroup showed a significant decrease of the chronotropic response and of the systolic blood pressure.

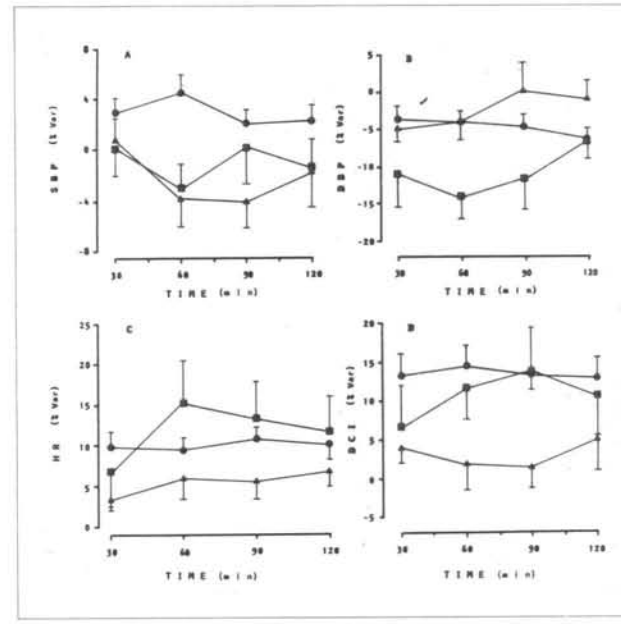

Figure 2
Var % : SBP (A), DBP (B), HR (C), DCI (D) after the oral glucose load ($\bar{x}$+SE)
l = control; n = normoinsulinemic chagasic patients
s = Hypoinsulinemic chagasic patients

These two findings may be explained by the hypoinsulinemia, a possible reduction in the action of the insulin receptors or a change in the myocardial insulin receptor present in these HICh subjects. These two findings may also indicate a decreased pressoreceptor activity caused by a reduced vasodilating response due to the action of peripheral insulin as demonstrated by the lower drop in DBP shown by HICh subjects when compared to control and to NICh subjects (figure 2 b) (23,18).

The DCI has been interpreted as an indirect measure of myocardial oxygen consumption and cardiac sympathetic reflex activation (14). The study herein demonstrated a simultaneous decrease of DCI and insulinemic response after the GTT in HICh subjects only.

This was not documented in NICh and control subjects whose chronotropic and SBP responses were followed by an increase in DCI and in serum insulin levels. This cardiac response seen in HICh subjects, may be due to a decreased insulin action on the myocardium or to a reduced insulin-induced activation of the ATPase (3,7,16,20).

In conclusion, the data presented herein suggest that HICh subjects have a significant decrease in both chronotropic and pressor responses possibly because of a lower insulin activity and/or an early dysfunction of the Autonomic Nervous System.

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RESUMO

Neste estudo foram avaliados 16 pacientes chagásicos e 28 indivíduos normais, em termos de respostas pressórica e cronotrópica pós sobrecarga oral de glicose. Essas respostas foram apreciadas através das medidas de pressão arterial e frequência de pulso. A resposta cardiovascular foi correlacionada com os níveis de insulinemia e glicemia. O experimento detectou um subgrupo de chagásicos (N=8) com padrão hipoinsulinêmico com atividades cronotrópicas e pressórica deprimidas quando comparadas ao grupo controle, o que indica um possivel déficit nas atividades insulínicas e/ou disfunção autonômica precoce neste subgrupo.