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HUMAN NEUROCYSTICERCOSIS

IgE IN CEREBROSPINAL FLUID

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ABSTRACT - The detection of IgE is technically difficult because of its reduced concentrations in serum, and even lower concentrations in cerebrospinal fluid (CSF). In the present investigation we studied 86 CSF samples using an immunoenzymatic method with an anti-IgE-alkaline phosphatase conjugate and a fluorigenic substrate. The samples were from three groups: A) 29 patients with neurocysticercosis (NC), B) 36 patients with different neurologic disorders (neurosyphilis, neurotuberculosis, meningitis, tumors, hemorrhage) and C) 21 discharged individuals who had been hospitalized for bacterial meningitis. The results obtained were: A) 0.05 to 3.00 IU/ml (0.76±0.79), B) 0.00 to 1.50 IU/ml (0.23±0.34) and C) 0.05 to 1.25 IU/ml (0.34±0.34). The present results suggest that IgE appears to play a role in the pathogeny of NC and that efforts should be made to standardize a test for the detection of specific IgE antibodies.

KEY WORDS: neurocysticercosis, cerebrospinal fluid, IgE, immunoenzymatic assay.

Neurocisticercose humana: IgE no líquido cefalorraquiano

RESUMO - A detecção de IgE apresenta dificuldades técnicas pela reduzida concentração que se encontra no LCR e no soro. Utilizando método imunoenzimático com conjugado anti-IgE-fosfatase alcalina e substrato fluorigênico, foram estudadas 86 amostras de LCR de três grupos: A) 29 pacientes com NC, B) 36 pacientes com afecções neurológicas diversas (neurossífilis, neurotuberculose, meningites, tumores, hemorragias) e C) 21 indivíduos de pós-alta médica de meningites bacterianas. Os resultados obtidos foram: A) 0,05 a 3,00 UI/ml (0,76±0,79), B) 0,00 a 1,50 UI/ml (0,23±0,34) e C) 0,05 a 1,25 UI/ml (0,34±0,34). Os resultados obtidos sugerem que a IgE parece ter papel na patogenia da NC e esforços devem ser feitos para padronização de teste para detecção de anticorpos IgE específicos.

PALAVRAS-CHAVE: neurocisticercose, líquido cefalorraquidiano, IgE, teste imunoenzimático.

Human neurocysticercosis (NC) is caused by the presence of the larval form of *Taenia solium, Cysticercus cellulosae*, in the central nervous system (CNS) and by the biological parasite-host interactions, leading to severe symptoms and high morbidity and lethality.[14] The CNS has been considered to be a privileged site with its own defense system, which usually maintains its integrity even in the presence of systemic infections. The accidental invasion by *Taenia solium* embryos causes local defense events that have not been fully elucidated. Extracellular changes can be indirectly investigated by the study of the cerebrospinal fluid (CSF)[7,14]. When parasite degeneration occurs, there is exacerbation of the immune response, with pleocytosis and proteinorrachia related to the intrathecal production of immunoglobulins representing specific antibodies[6]. Some investigators

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consider the production of antibodies to be a determinant of parasite death[8], others have observed that resistence to infection coincides with the maturation of the cell immune response[1]. Host genetic differences affect both mechanisms and the integrity of the cell response is probably important for the production of antibodies, regardless of the damaging effects it will have on the occasion of parasite death.

The production of high levels of specific IgE has been demonstrated in parasitic infections[10,19]. Flisser et al.[3] detected serum antibodies of the IgE class in 37% of 116 sera that were positive for precipitating anti-*Cysticercus cellulosae* antibodies, whereas Gorodezky et al.[5] only observed an increase in the total IgE in 52% of 50 serum samples from patients and absence of specific IgE. In a CSF study, Goldberg et al.[4] reported higher total IgE levels in 19 patients with NC compared to 34 samples from a control group. Spina-França et al.[15] detected higher IgE levels in CSF and serum of 30 patients with NC compared to 15 samples from a control group. In contrast, Short et al.[12] did not detect differences in total serum IgE nor in CSF IgE in 21 patients with NC.

Radioimmunoassay has been used for the detection of total IgE, a costly procedure that requires scintillation counters [4,5,12,13]. In the present study we used the UMELISA-IgE test (SUMA, Cuba) to determine the concentration of total IgE in CSF samples of patients with NC and of a control group.

MATERIAL AND METHODS

We analyzed 86 CSF samples obtained from patients seen at the University Hospital, Faculty of Medicine, University of São Paulo, and at Emilio Ribas Institute. The patients were divided into three groups: A) 29 patients with NC; B) 36 patients with various neurologic disorders (neurosyphilis, neurotuberculosis, meningitis, tumors, hemorrhage); and C) 21 patients from a group who had been discharged from the hospital after treatment for bacterial meningitis and with normal CSF. The samples examined in this study were collected and assayed previously and only the CSF samples from group A were reactive to anti- *Cysticercus cellulosae* IgG by ELISA.[17]

The immunoenzymatic test for IgE determination employed was a capture test [16] using microtiter plates with 10µl wells previously coated with anti-IgE antibodies. The conjugate used was anti-IgE-alkaline phosphatase, and the fluorigenic substrate was 4-methyl-umbelliferyl-phosphate. The calibration curve used correspond to a linear range up to 20 IU/ml. Fluorimetric readings were obtained with a series SUMA-321 apparatus (SUMA, Cuba).

The mean and standard deviations were calculated for each group and the data were analyzed statistically by the Student t-test.

RESULTS

Figure 1 shows the distribution of the IgE results (IU/ml) obtained for the 86 CSF samples according to group. The following results were obtained: Group A) 0.05 to 3.00 IU/ml (0.76±0.79); Group B) 0.00 to 1.50 IU/ml (0.23±0.34); and Group C) 0.05 to 1.25 IU/ml (0.34±0.34). A significant difference ($p<0.05$) was observed between the group of patients with NC and the control group (B + C).

DISCUSSION

The complex parasite-host relationship, the physiopathogeny of the immune response and the difficulty in establishing similar experimental models for investigation prevent a precise understanding of the immunobiology of NC during the initial phase of infection. The major mechanisms of the immunologic response, and antibodies production in particular, have been studied during the active phase of the disease, parasite death and degeneration. Antibodies of the IgG class are the most frequent in the CSF of patients with NC and are present in high concentrations and therefore are detected in the diagnosis of the disease [6,18].

In parasitic infections the level of total IgE is often elevated, many times without the activity of a specific antibody. The mechanisms of IL-4 induction by the parasites and the role of the immunoglobulins are not well understood, although macrophages appear to participate in this isotype-

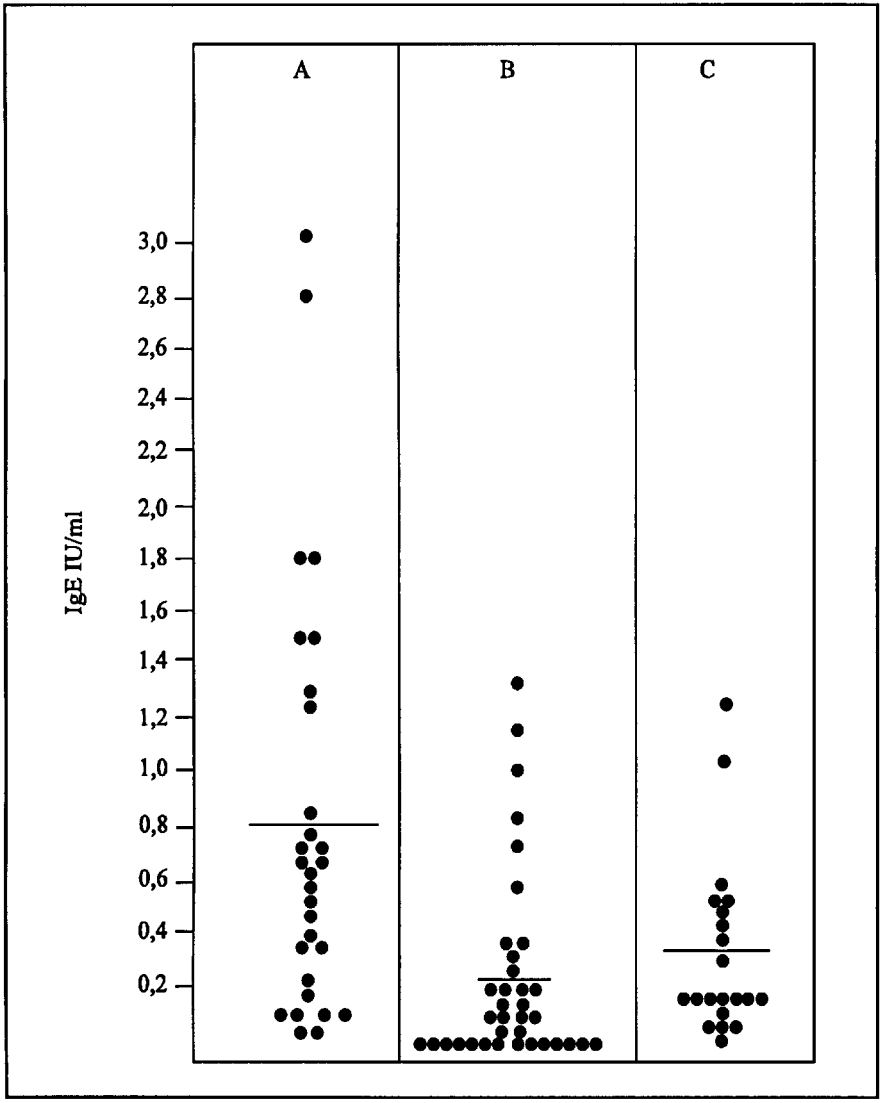

Fig 1. IgE concentration (IU/ml) in the CSF samples from 29 patients with NC (A), 36 patients with non-neurocysticercosis neurologic disorders (B) and 21 normal individuals (C).

restricted response [19]. The correlation between IgE and aspects of the parasitic infection is also obscure, although it seems to be related to the chronic aspect of the disease [9].

For the determination of IgE concentration in the CSF we used the ultrasensitive immunoenzymatic test with a fluorigenic substrate, standardized for the detection of minimal IgE levels [16], a test easier to perform than radioimmunoassay [12]. The results obtained (Fig 1) demonstrated a significant difference ($p<0.05$) between group A and the control groups.

According to the method, reference values for serum are up to 90 IU/ml for adults, and no standardized values are available for CSF. Considering the results of the group C for the calculation of the cut-off point, a 5% error (mean + 2 SD), we obtained 1.02 IU/ml for the normal CSF. Eight

(27.6%) of the 29 CSF samples from patients with NC and 8,8% of the 57 CSF samples from the control groups presented values above 1.0 IU/ml.

Goldberg et al,[4] applying the PRIST to 19 CSF samples from patients with NC, obtained results differing from those for the 34 CSF samples from a control group (0.44±0.19 versus 0.07± 0.02 IU/ml). Spina-França et al.[15] established reference values of IgE in CSF as ≤ 0,1 IU/ml and related median of 1.2 IU/ml for CSF samples from 30 patients with NC. Otherwise, Short et al.,[13] using a radioimmunoassay with 3.0 IU/ml sensitivity, did not detect a difference between the CSF of 21 patients with NC and 17 controls.

In contrast to Espinoza et al.,[2] who detected specific IgE in 2 of 60 CSF samples studied, we were unsuccessful in our previous attempt to detect specific anti-*Cysticercus cellulosae* antibodies by indirect ELISA[11] using an anti-IgE-peroxidase, possibly due to competition with IgG (data not shown). Although a similar unsuccessful attempt has been reported by others,[5,13] certain hypotheses should be better evaluated, such as the study of specific antigens in the induction of IgE production.

Studies of the specific IgE humoral response appear to be useful to a better understanding of the complex parasite-host relationship in NC and perhaps establish new diagnostic and prognostic markers for the infection, with possible differences in terms of clinical manifestations and course.

REFERENCES

1. Anderson MJD, Griffin JFT. *Taenia crassiceps* in rats: differences in susceptibility to infection and development of immunocompetence in relation to age and host strain. Int J Parasitol 1979;9:229-233.
2. Espinoza B, Ruiz-Palacios G, Tovar A, Sandoval MA, Plancarte A, Flisser A. Characterization by enzyme-linked immunosorbent assay of the humoral response in patients with neurocysticercosis and its application in immunodiagnosis. J Clin Microbiol 1986;24:536-541.
3. Flisser A, Woodhouse E, Larralde C. Human cysticercosis: antigens, antibodies and non-responders. Clin Exp Immunol 1980;39:27-37.
4. Goldberg AS, Heiner DC, Firemark HM, Goldberg MA. Cerebrospinal fluid IgE and the diagnosis of cerebral cysticercosis. Bull Los Angeles Neurol Soc 1981;46:21-25.
5. Gorodezky C, Diaz ML, Escobar-Gutierrez A, Flisser A. IgE concentration in sera of patients with neurocysticercosis. Arch Invest Med 1987;18:225-227.
6. Livramento JA. Síndrome do líquido cefalorraqueano na neurocisticercose. Arq Neuropsiquiatr 1987;45:261-275.
7. Livramento JA, Machado LR, Spina-França A. Immunobiology of neurocysticercosis. In Fejerman N, Chamoles NA (eds). New trends in pediatric neurology. New York: Elsevier, 1993:307-312.
8. Molinari JL, Tato P, Lara-Aguilera R, White AC Jr. Effects of serum from neurocysticercosis patients on the structure and viability of *Taenia solium* oncospheres. J Parasitol 1993;79:124-127.
9. Ottesen EA, Poindexter RW, Hussain R. Detection, quantitation and specificity of antiparasite IgE antibodies in human schistosomiasis mansoni. Am J Trop Med Hyg 1981;30:1228-1237.
10. Parkhouse RME, Harrison LJS. Antigens of parasite helminthes and diagnosis, protection and pathology. Parasitology 1989; 99 (Suppl.):S5-S19.
11. Pialarissi CSM, Vaz, AJ, Souza AMC, Nakamura PM, Camargo ED, Silva MV,Ueda M. Estudo comparativo de testes sorológicos no diagnóstico imunológico da neurocisticercose. Rev Inst Med Trop S Paulo 1987;29:367-373.
12. Polmar SH, Waldmann TA, Terry WD. A comparison of three radioimmunoassay techniques for the measurement of serum IgE. J Immunol 1973;110:1253-1261.
13. Short JA, Heiner DC, Hsiao RL, Andersen FL. Immunoglobulin E and G4 antibodies in cysticercosis. J Clin Microbiol 1990; 28:1635-1639.
14. Spina-França A. Patogenia das infecções do SNC e LCR: análise crítica da contribuição diagnóstica. Rev Paul Med 1989;107: 169-174.
15. Spina-França A, Kleine TO, Livramento JA, Machado LR. IgE determination in cerebrospinal fluid and serum of patients with neurocysticercosis by a modified enzyme immunoassay. Protides in Biological Fluids 1986;34:657-660.
16. Urquiza HD. Algunas consideraciones sobre la determinación de immunoglobulina E en el suero del cordón y la prevención de enfermedades alergicas. Rev Cub Med Gen Integral 1988;4:28-36.
17. Vaz AJ, Ferreira AW. Imunodiagnóstico da neurocisticercose: teste imunoenzimático com antígenos quimicamente ligados a suportes para pesquisa de anticorpos em soro e líquido cefalorraquiano. Rev Inst Med Trop S Paulo 1988;30:1-10.
18. Vaz AJ. *Cysticercus longicollis:* caracterização antigênica e desenvolvimento de testes imunológicos para pesquisa de anticorpos em líquido cefalorraquiano no imunodiagnóstico da neurocisticercose humana. Tese de Doutorado, Instituto de Ciências Biomédicas da Universidade de São Paulo. São Paulo, 1993.
19. Zakroff SGH, Beck L, Platzer EG, Spiegelberg HL. The IgE and IgG subclass responses of mice to four helminth parasites. Cell Immunol 1989;119:193-201.