

AGE-RELATED SEROPREVALENCE STUDY FOR ST. LOUIS ENCEPHALITIS IN A POPULATION FROM CORDOBA, ARGENTINA

Lorena Ivana SPINSANTI(1), Viviana Elizabeth RÉ(1), María Pilar DÍAZ(2) & Marta Silvia CONTIGIANI(1)

SUMMARY

A serological screening was performed in 615 individuals aged 0-87 years, living in the city of Cordoba, Argentina to study the relationship between antibody prevalence for the SLE virus and age.

A 13.98% prevalence of neutralizing antibodies was obtained and its relation to age was significantly high ($p = 0.045$). The highest seroprevalence was noted on individuals over 60 years old (>20%), whereas no subject under 10 was seropositive for this virus.

Our results confirm that the agent is endemic in this area and neurological pathology studies should be performed on those individuals aged 60 since they represent the most susceptible group to SLE virus.

KEYWORDS: Seroprevalence; St. Louis encephalitis; Age; Córdoba, Argentina

INTRODUCTION

St. Louis encephalitis (SLE) is a human mosquito-borne viral disease. The causative agent, SLE virus, is a member of the Flaviviridae family. It was first isolated in Missouri in 1933, and it has been responsible for more than 4,000 reported human cases of infection in the United States since 1955¹⁰. Several reports have shown that SLE virus is widely distributed in the Americas; it has been isolated from Canada to Argentina¹. The results of several serological surveys suggest that SLE virus is able to infect man in Central and South America^{1,2,11}, and seems to cause sporadic illness as the few isolations obtained from human blood in Argentina, Panama, and Trinidad came from patients with a mild febrile illness¹. One case of fever and jaundice has been reported in Brazil, but cases of encephalitis caused by SLE virus have not been reported¹².

Three clinical syndromes are described associated with the infection: encephalitis, aseptic meningitis, and febrile headache. The occurrence and severity of SLE in human beings is strongly dependent on age. It has been reported that during epidemics, incidence of disease in persons older than 60 years was generally 5-40 times greater than in those less than 10 years old and the case-fatality rate increased with age, from 2% in young adults to over 22% in the elderly¹⁰.

In Argentina the distribution of SLE virus is very wide, the serological evidence (in some cases up to 50%) for transmission has been found practically wherever it has been sought. The virus is endemic in subtropical provinces, as well as in other regions located in the temperate

area of Argentina. However, the SLE infection has been rarely associated to human illness in Argentina and South America^{13,14}.

In the province of Buenos Aires in 1963, two clinical cases characterized by febrile syndromes, without signs of central nervous system infection were reported and diagnosed by virus isolation and serology⁷. Between 1966 and 1984 two viruses were isolated from unusual sources in the province of Córdoba: the strain Cba. An 9124 from the urine of a *Calomys* sp and the strain Cba. An 9275 from a brain-spleen pool of a juvenile *Mus musculus*¹³. Afterwards, 6 other strains of SLE were recovered from *Culex* sp mosquitoes in the province of Santa Fe^{8,9}.

In molecular studies of genetic variability and its relationship to virulence TRENT *et al.* established 5 genotypes and showed that none of the Argentine strains had the epidemic North American genotype from the Central and Atlantic Coast States of USA, represented by the MSI-7 strain¹⁵.

Until now no study was done in order to provide information about age-specific immunity rates or the prevalence of infection in rural vs. urban populations in Argentina. A high prevalence of immunity in the population over 60 years old, representing the age group most susceptible to clinical disease, could explain the absence of epidemics in the region¹³.

A serological screening was performed in 615 individuals age range 0 to 87 years, living in Cordoba, Argentina to evaluate the association

(1) Institute of Virology "Dr. J.M. Vanella", Faculty of Medical Sciences, National University of Cordoba, Argentina.

(2) Statistics and Biometrics, Faculty of Agronomy, National University of Cordoba, Argentina.

Correspondence to: Lorena Ivana Spinsanti, Instituto de Virología "Dr. J.M. Vanella", Facultad de Ciencias Médicas, UNC, Enfermera Gordillo Gómez s/n, Ciudad Universitaria. Córdoba 5016, Argentina. Tel: 54 - (351) 4334022; e-mail: contigia@cmefcm.uncor.edu

and functional relationship between the prevalence of SLE infections and the age of individuals.

MATERIAL AND METHODS

Area studied: This study was performed in the city of Cordoba which is the main inland province of Argentina (Fig. 1). This city has a sub-temperate climate (transition temperate) and it has all the characteristics corresponding to an urban system. The annual average rainfall is 727 mm and it had 1,179,067 habitants in 1991.

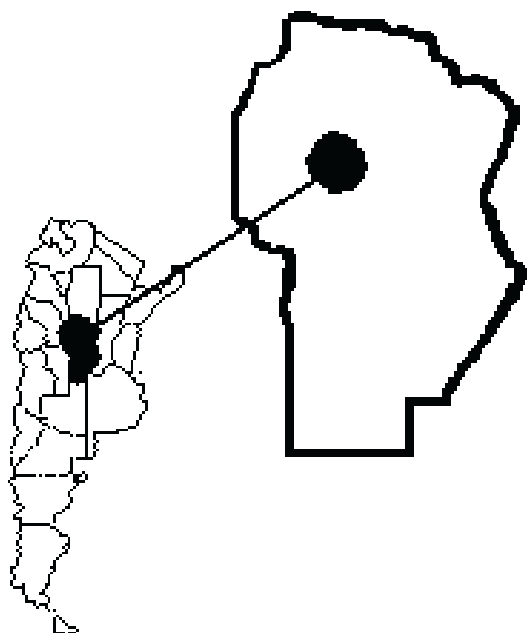


Fig. 1 - Insert shows the city of Cordoba in the Cordoba province, Argentina.

Human Serum: Between 1998 and 2000 period 615 serum samples were studied. They were taken from individuals belonging to the Blood Bank (aged between 16 and 55 year-old) Infantile Hospital and from the health care allocation for those people over 60 who had a check up at that moment. All the individuals coming from these places were considered representatives of the different neighborhoods of the city. The samples were preserved at -20°C until they were processed.

Serological test: Plaque reduction neutralization test was performed with an 80% plaque reduction endpoint³. The viral suspension was prepared using a mouse's brain infected by SLE 78V6507 strain to 10% (weight/volume) in Medium Essential Minimum (MEM) with Earle, L-glutamine salts, 10% of fetal bovine serum and antibiotic. It was centrifuged at 10,000 rpm during 30 minutes so as to decontaminate. In the screening was used a 1:5 dilution of each serum.

Statistical Analysis: Age distribution was studied and turned into a discrete variable with 16 intervals. The middle point of each class was taken to represent age covariate in the prevalence study. Generalized linear models, with logistic link, were fitted⁶ for the response variable: number of positive subjects, assuming linear and quadratic regression model for the linear predictor. The system GLIM (Generalized Interactive Linear Models)⁵ was used to do it. Several checking techniques were implemented for the diagnostic analysis in order to obtain a suitable model for predictions.

RESULTS

Table 1 summarizes all the information related to the study showing the prevalence values obtained according to the age turned into discrete 7 intervals. It can be noted that the prevalence gradually increases from 10 years and that it is higher in adults over 60.

Table 2 shows the deviance analysis results for the adjustments on the logistic models having $\eta = \alpha + \beta_1 \text{ Age} + \beta_2 \text{ Age}^2$ (quadratic linear regression) and the parameters estimated for the model as a systematic component.

Table 1

Prevalence of SLE neutralizing antibodies according to age in individuals from Cordoba, Argentina

Age (years)	Positive/tested	Prevalence (%)
0-9	0/91	0
10-19	3/51	5.9
20-29	14/148	9.5
30-39	11/76	14.5
40-49	9/63	14.3
50-59	6/26	23.1
>60	43/160	26.9
TOTAL	86/615	13.98

Table 2

Deviance analysis from logistic regression model, estimates and standard errors, p-value and fitted models

Parameter	Estimate	Standard error	p-value	Adjusted model
α (intercept)	-4.2010	0.6144	0.0000	$\ln(p/1-p)^{(1)} = -4.2010 + 0.0839 \text{ age} - 0.0055 \text{ age}^2$
β_1 (linear coefficient)	0.0839	0.0282	0.0000	
β_2 (quadratic coefficient)	-0.00055	0.0022	0.0455	

⁽¹⁾ p is the positive proportion (prevalence/100).

The p-values to covariable incorporation showed that prevalence was strongly related to individuals age. The determining coefficient value for the adjusted model $R^2 = 0.821$ show that approximately 80% of the prevalence can be explained by age relation.

Figure 2 shows the estimated prevalence for different ages.

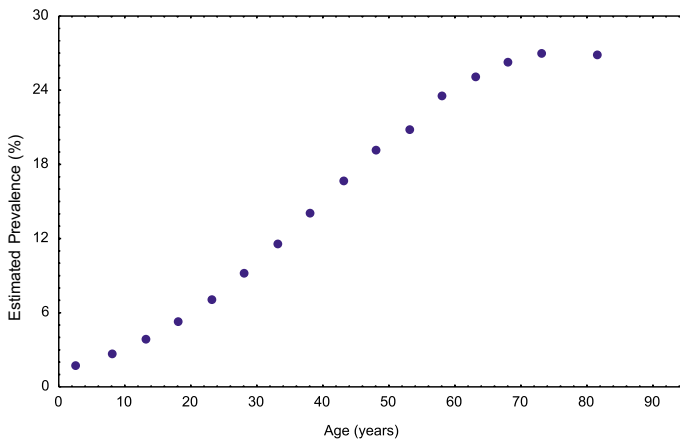


Fig. 2 - Estimated prevalence for different age values

DISCUSSION

In Cordoba the last records related to SLE virus circulation date from 1963-66 when EVANS *et al.* found 11% of antibodies in men aged 20⁴. The results of our study showed a seroprevalence slightly higher (13.98%) and a significant statistical relationship between the presence of neutralizing antibodies and the age of the subjects ($p = 0.045$).

The prevalence estimated by the statistical model according to age (Fig. 2) showed that the seroprevalence would be more than 20% in adults over 60. However, the absence of specific antibodies for the SLE in children under 10 (Table 1) (the statistical model predicts or estimates a low prevalence, Fig. 2) could be due that during the last decade the virus remained silent in the area studied. It would be interesting to extend this study among individuals under 10 from other areas to check if the same behavior is repeated.

Despite the relationship between SLE prevalence and age is statistically significant, the quantity of samples studied in the 50-60 years old group was lower than in the other age groups.

A higher number of samples would reinforce the significance of these results.

One explanation for the rarity of cases in humans in Argentina is the probable accumulation of immunity with age, thereby reducing the size of the susceptible elderly population. Furthermore, the cases of milder febrile syndromes and the Central Nervous System infections that could be associated to the SLE infection are not yet researched.

The highest antibody prevalence to the SLE virus found among individuals over 60 years old points out that the presence of neurological

pathologies in these subjects should be investigated to better understand the etiological role of SLE virus in Argentina.

RESUMO

Estudo da prevalência para vírus da encefalite St. Louis relacionado com a idade em uma população de Córdoba, Argentina

Com a finalidade de estudar a relação entre prevalência de anticorpo para o vírus da encefalite St. Louis (ESL) e a idade, foram analisados 615 soros de indivíduos entre 0-87 anos de idade, morando na cidade de Córdoba, Argentina.

Detectou-se uma prevalência de anticorpos neutralizantes de 13,98% e observou-se uma relação com a idade estatisticamente significativa ($p = 0,045$). A soroprevalência mais alta foi encontrada em indivíduos de mais de 60 anos ($> 20\%$), enquanto nenhum com menos de 10 anos foi soropositivo para este vírus.

Nossos resultados confirmam que o agente é endêmico nesta área. Por esse motivo os autores sugerem que deveriam se realizar estudos de patologias neurológicas nesses indivíduos de 60 anos de idade dado que eles representam o grupo mais suscetível ao vírus ESL.

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