

## RELATIONSHIP BETWEEN THE PREVALENCE OF ANTIBODIES TO HEPATITIS B CORE ANTIGEN AND ARBOVIRUS IN FISHERMEN FROM THE RIBEIRA VALLEY, BRAZIL

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### SUMMARY

Sera from 299 fishermen 16 to 80 years old, residents in Cananea and Iguape counties, southern coast of São Paulo State, Brazil, were studied in order to identify a possible association between the prevalence of specific antibodies to the hepatitis B virus (HBV) and exposure to haematophagous mosquitoes evaluated by the prevalence of arbovirus antibodies. This professional group presented the highest prevalence of arbovirus antibodies (54.1%) in past investigations carried out in this heavily forested region. Detection of antibody to hepatitis B core antigen (anti-HBc) in the sera was done by enzyme immunoassay (Roche).

Prevalence of anti-HBc antibodies in this group was 31.4% (94/299) which is very high compared with 7.2% to 15.0% for different groups of healthy adults in State of São Paulo. No significant difference is observed between the prevalences of HBV antibodies in Iguape and Cananea. Prevalence of anti-HBc and anti-arbovirus antibodies increases with age. There is a concordance in the distribution according to age groups of the frequency of anti-HBc and anti-arbovirus positive sera. Ag HBs was detected in 4% of the studied sera.

These results support the hypothesis that the transmission of the hepatitis B virus and the arboviruses may be due to the same factor, one of the possibilities would be by anthropophilic mosquitoes.

**KEY WORDS:** Hepatitis B Transmission; Hepatitis B Seroepidemiology; Arbovirus and hepatitis B.

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### INTRODUCTION

A previous study conducted in children, 6 to 14 years old, which lived in Ribeira Valley, Brazil, in four areas with different topographical and phytological characteristics (urban, periurban, cultivated and jungle zones), showed highest prevalence of HBV antibodies among the

jungle zone dwellers that have also had high prevalence of arbovirus antibodies<sup>33</sup>.

The possibility of mechanical transmission of the hepatitis B virus (HBV) through vectors was suggested by the detection of HBsAg in va-

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rious species of mosquitoes and other haematophagous arthropods captured in tropical regions<sup>6, 8, 18, 26, 34, 40, 41</sup> and even in urban areas of the United States<sup>12</sup> and by experimental studies that showed the persistence of this antigen in arthropods, when they were fed with blood containing HbsAg, until this blood was fully digested<sup>3, 7, 11, 25, 27, 30, 32, 35, 38</sup> or for a larger period<sup>37</sup>. If the transmission is in any way related to haematophagous mosquitoes, a high prevalence of immunological markers of the HBV should occur more frequently in tropical regions specially in people that live in forested areas where the activity of these insects is intense.

This fact has been consistently observed among Indian tribes in the Amazon area<sup>9, 10, 19, 20</sup>. In São Paulo, Brazil, a seroepidemiologic survey carried out in different population groups has recently brought out an unusual high prevalence of anti-HBs among children from 1 to 11 years old that were living in a camp near a forested area<sup>29</sup>. In order to study the possible role of wild mosquitoes in HBV transmission, serological surveys were conducted in a neighboring region with similar ecological characteristics, namely the Ribeira Valley, where abundant *Culicidae* fauna has been found<sup>16, 17</sup>.

We describe here the results of a research on HBV immunological markers in male adults, a group of fishermen of two coastal counties of Ribeira Valley.

In previous studies<sup>21-24</sup> this group presented the highest prevalence of arbovirus antibodies, specially to Ilhéus, Rocio, Caraparu viruses and a new sub-type of Venezuelan equine encephalomyelitis virus. This is explained by high outdoor exposition to anthropophilic mosquitoes during their work in a period that usually includes crepuscular hours.

The prevalence of arbovirus antibodies should thus provide an indirect indicator of the risk of infections transmitted by mosquitoes.

## MATERIAL AND METHODS

### The area.

Ribeira Valley (24° — 25°16'S, 46°50' — 49°20'W) is located in the south of São Paulo Sta-

te, southeastern Brazil. About 58% of the area is still covered by forest and presents a highly humid tropical climate<sup>36</sup>. The local population work mostly in agriculture, fishing and lumbering and has the lowest income of the State of São Paulo.

### Serological study

Anti-HBc and HBs Ag were determined by enzyme immunoassay in 299 sera collected from fishermen, 16 to 80 years old, residents in Iguape and Cananeia, coastal counties of Ribeira Valley. Most of these fishermen worked in a channel between the continent and an extense island, by day or by night, in a period that includes the dawn or the dusk when the activity of haematophagous mosquitoes increases. They frequently walked in neighboring forest, 63% reporting to do so at least once a week. The majority (92%) was born in Ribeira Valley and live there all the time. Drug addiction with exception of alcohol is not present; daily alcohol consumption in small quantities is a habit among male adults of this rural area. Promiscuous sexual habits are not reported; the cultural behavior of Ribeira Valley rural people is very traditional.

In a previous study these fishermen proved to have a high prevalence of haemagglutination-inhibition (HI) antibodies to the arboviruses Ilheus, Rocio, a new subtype of Venezuelan equine encephalomyelitis and Caraparu<sup>24</sup>.

Anti-HBc was determined by enzyme immunoassay based on competition principle<sup>5</sup> (Roche Laboratories). HBs Ag was researched according to procedure of VIEIRA et al<sup>39</sup>.

The statistical analysis was based in  $\chi^2$  and Kolmogorov-Smirnov tests, the last one used to study the concordance between the distribution of hepatitis B and arbovirus positive sera.

## RESULTS

Anti-HBc were detected in 94 of 299 sera (31.4%). No significant difference ( $p > 0.05$ ) was observed in the prevalence of anti-HBc antibodies from the residents in Iguape and Cananeia. The same was observed in regard to the prevalence of arbovirus antibodies (Table 1).

TABLE 1

Prevalence of antibodies to hepatitis B core and arbovirus antigens in fishermen, according to the country of residence in Ribeira Valley Brazil.

Residence	no. tested	no. positive (%) anti-HBc	no. positive (%) anti-arbovirus
Cananeia	139	48 (34.5)*	76 (54.7)
Iguape	160	46 (28.7)*	89 (55.6)
Total	299	94 (31.4)	165 (55.2)

\*  $\chi^2 = 1.15$   $p > 0.05$

The prevalence rates of anti-HBc and anti-arbovirus antibodies increases with age. A significant difference ( $\chi^2 = 16.56$   $p < 0.05$ ) was observed in the distribution according to age group of the positive and negative sera (Table 2). A concordance in the distribution of cumulative frequency of HBc and arbovirus positives sera

was observed in the Kolmogorov-Smirnov test (Table 3).

Only two of the 94 positives sera to anti-HBc are from fishermen that were not born in the region, but they have lived there for more than 10 years.

HBs Ag was detected in 12 men (4.0%).

## DISCUSSION

Anti-HBc, described in 1971 by ALMEIDA et al<sup>1</sup>, is early detected in the blood of infected people, before the presence of anti-HBs and stays for a long time. Consequently it is a very useful indicator of HBV past or recent infections.

TABLE 2

Prevalence of antibodies to hepatitis B core and arbovirus antigens according to age in fishermen from Ribeira Valley, Brazil.

age (years)	no. tested	no. negative	no. positive anti-HBc and anti-arbovirus	no. positive anti-HBc	no. positive anti-arbovirus
16 — 34	124 (100.0)	51 (41.1)	16 (12.9)	14 (11.3)	43 (34.7)
35 — 54	114 (100.0)	35 (30.7)	24 (21.0)	18 (15.8)	37 (32.5)
55 +	61 (100.0)	12 (19.7)	18 (29.5)	4 (6.5)	27 (44.3)
Total	299 (100.0)	98 (32.8)	58 (19.4)	36 (12.0)	107 (35.8)

$\chi^2 = 16.56$   $p < 0.05$

TABLE 3

Concordance between the cumulative distribution of anti-arbovirus and anti-hepatitis positive sera in age groups.

age (years)	no. anti-hepatitis sera	cumulative frequency	no. anti-arbovirus sera	cumulative frequency	D (difference)
16 — 24	12	12/94 (0.128)	23	23/165 (0.139)	0.011
25 — 34	18	30/94 (0.319)	36	59/165 (0.358)	0.039
35 — 44	23	53/94 (0.564)	34	93/165 (0.564)	0.0
45 — 54	19	72/94 (0.766)	27	120/165 (0.727)	0.039
55 — 64	13	85/94 (0.904)	28	148/165 (0.897)	0.007
≥ 65	9	94/94 (1.000)	17	165/165 (1.000)	0.0
Total	94		165		

D max = 0.039 < 0.175 (critical value of D,  $\alpha = 0.05$ )

The fishermen of Ribeira Valley present significantly higher prevalence of HBc antibodies than other healthy adult groups studied in the State of São Paulo. In these groups prevalence rates of HBc antibodies ranged from 7.2%<sup>13</sup> (hospital administrative employees), 14.8%<sup>14</sup> (blood donors) to 15.0% (blood donors and medical students). Only known hepatitis B risk groups, also residents in São Paulo, such as hospital medical staff, drug abusers, homosexuals, prostitutes, siblings of cases of hepatitis B and patients with AIDS have prevalence rates similar or higher than the investigated fishermen<sup>2, 9, 13, 15, 28, 31</sup>. Furthermore the fishermen present a prevalence of HBs Ag, 4.0%, higher than other healthy adults studied in other areas of the State of São Paulo, values from 0.3% to 1.5% with a median of 0.8%<sup>4, 9, 13, 14, 31</sup>. The 149,837 blood donors which were investigated from May 1981 to December 1984 at the Medical School Hospital, University of São Paulo, had HBs Ag prevalence of 0.9%<sup>4</sup>. The 599 blood donors which were examined at the Regional Hospital of Ribeira Valley, from January 1988 to July 1989, had HBs Ag prevalence of 2.0% (unpublished data).

This high prevalence of HBV markers is present in both Iguape and Cananeia counties. Prevalence increases with age as the infections that are dependent on cumulative environmental exposition. Obviously it is not possible to exclude vertical transmission, mainly because the women from rural zone are also exposed to the bite of anthropophilic mosquitoes. A research on HBV markers in the relatives of positive cases could shed light upon this aspect. A preliminary study on anti-HBc antibodies conducted in families of the rural area of Ribeira Valley did not show that vertical transmission has had an important role in hepatitis B transmission (IVERSSON et al, unpublished data). But other studies become necessary to evaluate the role of the vertical transmission in the high prevalence of HBV infection.

The results agree with the information obtained in previous research on children from the rural zone of Iguape, that is, the highest prevalence of hepatitis B specific antibodies is present in the group with highest exposition to wild mosquitoes.

They suggest the existence of an environmental factor in the dissemination of HBV and arboviruses, both in Iguape and Cananeia counties, one of the possibilities being that of transmission by anthropophilic mosquitoes.

Anyhow since the children residents in forested areas and the fishermen of Ribeira Valley presented a high prevalence of HBV markers, they must be included among the risk groups that should receive a priority attention in a hepatitis B vaccination program.

## RESUMO

**Relação entre as prevalências de anticorpos para hepatite B (anti-HBc) e arbovirus em pescadores da região do Vale do Ribeira, São Paulo, Brasil.**

Soros de 299 pescadores de 18 a 80 anos de idade, residentes nos municípios de Iguape e Cananeia, região sul do Estado de São Paulo, foram estudados com o objetivo de identificar uma possível associação entre a prevalência de anticorpos específicos para a hepatite B e a exposição a mosquitos hematófagos, avaliada indiretamente pela prevalência de anticorpos de arbovírus. Esse grupo profissional apresentou, em pesquisas anteriores nessa área extensamente coberta por mata, a mais alta prevalência de anticorpos de arbovírus (54,1%).

Utilizou-se teste imunoenzimático para detecção de anti-HBc e HBs Ag.

Em 31,4% dos soros estavam presentes anticorpos anti-HBc, proporção significativamente mais alta que as detectadas em adultos sadios examinados no Estado de São Paulo (7,2% a 15,0%). HBs Ag foi observado em 4% dos soros examinados. As prevalências de anticorpos de arbovírus e hepatite B aumentaram significativamente com a idade ( $p < 0,05$ ), ocorrendo concordância na distribuição das frequências segundo idade entre os soros positivos para hepatite B e para arbovírus. Os resultados sugerem uma forma semelhante de transmissão desses agentes apoiando a hipótese da transmissão de hepatite B por mosquitos antropofílicos.

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