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MODIFICATIONS OF ARBOVIRUS ECO-EPIDEMIOLOGY IN TUCURUI, PARA, BRAZILIAN AMAZONIA, RELATED TO THE CONSTRUCTION OF A HYDROELECTRIC DAM

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The Brazilian part of the Amazon River and tributaries hydrographic basin covers 3,984,467 km², or 63.1% of the total area (6,315,217 km²); maritime and land borders of the region represent 55.1% of the total borders of Brazil; approximately 3,373,000 km² (84%) of Brazilian Amazonia are covered by dense equatorial forest (Sioli 1973; Marlier 1973; Paiva 1977 and Guillaumet 1987).

Since the discovery of its numerous natural resources more than 100 years ago, this region is developing rapidly, with an ever increasing need for energy, for both agriculture and the extraction of minerals (Odinetz-Collart 1987). However this region is sparsely inhabited, with less than 10% of the total Brazilian population and, excluding the state capital cities, it has one of the world's lowest population densities, of <2 inhabitants/km². With its equatorial or humid tropical climate, its enormous potential for employment and its natural resources, Amazonia is very attractive especially for people from regions which suffer from severe droughts like the Northeast.

The area has many large rivers and their energy has been harvested since the 1950s by the construction of various dams. Besides evaluating the ecological problems which may arise from cutting and flooding large forestal areas (Sioli 1973; Lovejoy 1985; Odinetz-Collart 1987), the appraisal of the health situation of the local human populations was among the development agencies' priorities.

Thus, an agreement was made between the company Eletronorte (Eletrobras) and the Evandro Chagas Institute (found. SESP), with the objective of studying the possible impact of the construction of a large dam on the epidemiology of arboviruses. As these 8-years-duration studies, which were initiated in 1982, are not yet completed, and much of the collected material remains to be studied, only preliminary results will be presented here.

The Amazonian region of Brazil is the world's richest reservoir of certain infectious agents. Thus, the 163 different types of arboviruses so far detected in the Amazonian region (Hervé *et al* 1986; Pinheiro *et al* 1986; Travassos da Rosa *et al* 1986, 1987 and unpublished data), accounts for nearly one third of the world's arboviruses (533: Karabatsos 1985). Of these, 136 (83.4%) are endemic.

Methodology

The objective of the project was to evaluate the effects of the construction and flooding of a dam on the transmission and epidemiology of sylvatic arboviruses, so as to determine the potential health hazards to which human populations entering the area would be subjected.

Comparisons were made between a number of surveys, 1) inside the dam region, before, during and after the flooding period; 2) outside the dam region, but in an area with a similar climate, landscape and vegetation, during the total time of the studies. The reasons for using the latter as a permanent control were: a) the much lower number of isolations obtained in the dam area prior to flooding, and b) that the dam area used during the pre-flooding period, and the neighbouring area could not be monitored because of flooding or inaccessibility and, c) the annual variability of inter-tropical climatology (Nimer 1977) which probably greatly influenced the seasonal and/or annual pattern of arbovirus transmission.

Two chosen areas, Altamira and Tucurui, were studied from 1974, and from September, 1982, respectively. The former, and the latter before the outset of flooding (6 September 1984) were considered as a control or reference for comparisons with data obtained in Tucurui during flooding and thereafter.

MATERIAL AND TECHNIQUES

The surveys were orientated to obtain large possible sample pools of vertebrate and hematophagos diptera. Human sera were collected from febrile cases and random serological surveys.

Wild vertebrates were collected with fruit-baited traps or selective hunting in the case of monkeys, and mist nets in the case of bats and birds. Blood and viscera were usually collected from each animal. Insects, mostly mosquitoes, phlebotomine sandflies and ceratopogonid midges, were sampled during human-baited canopy or floor diurnal collections, nightly "CDC miniature light-trap" canopy or floor runs, and active search in the understorey vegetation. In the case of the terrestrial tortoises in the dam area, it was possible to do a marking — release — recapture experiment.

The collected material was preserved in liquid nitrogen in the field, transferred to freezers at the laboratory, before being identified and inoculated IC into 48 h-old Swiss suckling mice. Sera were tested against 19 antigens, most of them pathogenic for man, by HI. In some instances, neutralization tests (NT) were also performed, usually in mice.

For the interpretation of the temporal distribution of isolations, either minimum infection rates (number of positive pools/total inoculated) or "real" infection rates as developed by Walter *et al* (1980), were estimated.

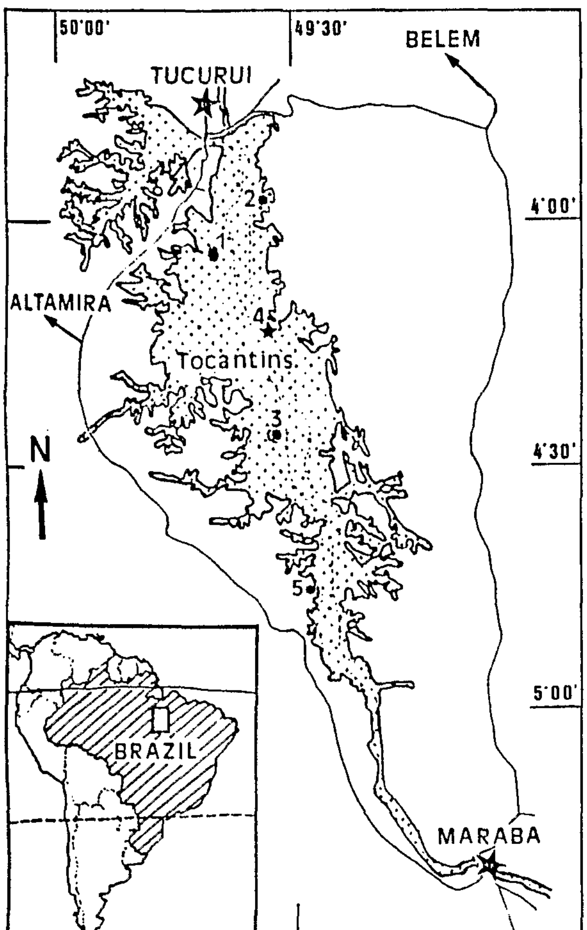

Figure 1. Situation map of the dam impoundment area, Tucurui, Par, Brazilian Amazonia. The flooded area is dotted and the locations of collecting are numbered 1 through 5.

DESCRIPTION OF STUDY AREAS

The Dam Area

Tucurui, Pará : 3°45'S – 49°41'W, is located on the low portion of the river Tocantins basin, at an altitude of 40 m above sea level. Though Marlier (1973) and Paiva (1977) have considered this tributary as somewhat independent from the Amazon river basin, it does not seem to be different in relation to our studies.

The following are some technical and physical data about the dam and the artificial lake, and the human population living nearby (Anon 1984; 1987; Pinguelli Rosa and Shaeffer 1988):

- — the dam extension – 7.5km; max height – 100 m; extension of accessory dams – 6.25 km; height – 98 m;
- — the lake: surface at the height +72 m – 2,430 km^2 with 75% of flooded area; length – 170 km; max width – 40 km (mean = 14.3 km); limits : 3°43' – 5°15'S/49°12' – 50°00'W; max depth = 75 m (mean – 18.9 m); total volume of water = 45.8 billions m^3; electrical production (half of projected) – 3980 Mw;
- — the human population: 17,319 persons, or 3,350 families were dis-lodged and relocated in new villages. Inhabitants of 2 villages were sampled during ther post-flooding serological surveys.

During the pre-flooding period, a field station, 67 km from Tucurui was used for collection of vertebrates and insects. During the 6 months duration of the flooding period, various small localities, situated in the area of the lake, were visited and their populations sampled. Of these, "Arumateua", "Vale do Caraip", "Canoal", "Jacunda", "Jatobal" survive only as virus names. Of the 5 field stations used during the rescuing of the fauna, 3°N and 4°N were also occasionally used and 4°N was assigned for all post-flooding studies (Fig 1)

The Reference Area

Altamira, Pará : 3°12'S – 52°12'W is located on the lower portion of the river Xingu basin, at an altitude of 74 m. The Xingu basin, a tributary of the Amazon, is not as extensive as the Tocantins/Araguaia basin. Four field stations were used in the same region, along the Itaituba — Altamira — Maraba road (Trans-Amazonian highway).

From 1974 to 1982, the surveys were done 10 km, 46 km, 80 km, 90 km, 95 km and 190 km from Altamira and 8 km, 21 km, 30 km, 55 km, 76 km, and 90 km, from Altamira on the Altamira-Maraba section.

The results of the studies conducted during the 1974-76 period, have been published by Dixon *et al* (1981); Roberts *et al* (1981); Peterson *et al* (1981), and Pinheiro *et al* (1977) have been incorporated and dealt with in the following discussions.

COMPARISON OF THE TWO AREAS

The reference and study areas, approximately 300 km from each other, are geologically and climatically similar, and have similar vegetation (Anon 1984; Nimer 1977; Moreira 1977; Kuhlmann 1977; SUDAM-PNUD/ Belém pers comm). The climate in the 2 regions is classified as warm (mean T° > 22°C), of tropical humid type with 3 dry months (< 60 mm). The mean annual relative humidity is between 85% and 90%. The mean monthly rainfalls (Fig 2) in the 2 localities are well correlated (Spearman correlation coeff. = 0.96 with $p < 0.01\%$). However, there are differences between the annual means of the 2 counties, which are situated on either side of the 26° isotherm. Altamira has a mean maximal annual temperature 2°C lower than Tucurui. Similarly, the mean annual rainfall is slightly lower in the Altamira region, being between 1500 and 2000 mm, while in Tucurui it is between 2000 and 2500 mm. The mean number of rainy days is also a little lower for the Altamira region, 120-180 versus 180-240 for Tucurui.

During the period from 1972 to 1988, the monthly rainfalls of the 2 counties were highly correlated (Spearman correlation coeff. = 0.85 with $p < 0.006\%$). The forests covering the 2 areas are classified by Kuhlmann (1977) into the same category of vegetation: hilean hygrophilous perennifolian Amazonian forest, or dryland ("terra firme") forest.

RESULTS

The Reference Area

Collected material

At the field stations in the Altamira region, annual totals of collected animals and human

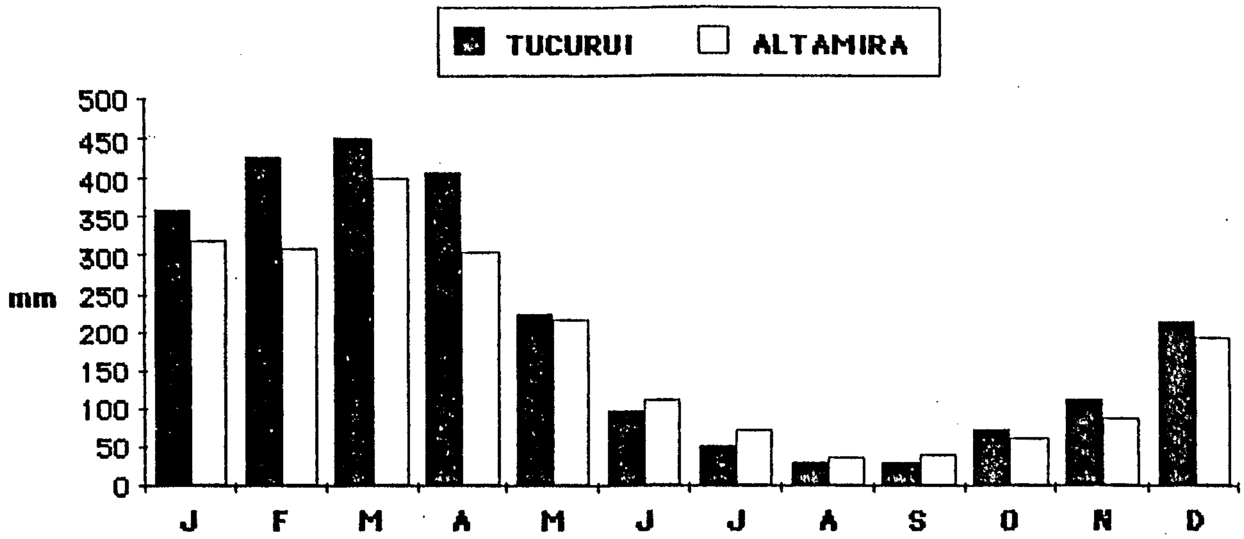

Figure 2. Mean monthly rainfall (in mm) at Tucurui (black) and Altamira (white), Par, Brazilian Amazonia, 1972-1988.

sera were as shown in Table 1. The mean annual numbers of collected/inoculated material were 21, 437, 125 (including sentinel animals), and 239 for Diptera, vertebrates and human sera, respectively.

Virus isolations

A total of 77 strains were isolated from the Altamira area, of which 15 were from humans, 4 from sentinel vertebrates, 5 from wild vertebrates, 42 from mosquitoes (1 from Simulium sp), and 11 from phlebotomine sandflies (Table 1).

Animal serology

A total of 33, 807 and 136 wild vertebrates were bled for serological testing during 1984, 1985, and 1986, respectively. Twenty were positive for at least 1 type of arbovirus.

Human serology

In the reference area (Altamira), we collected and tested 13,383 sera (annual mean = 955) for antibodies. The results for specific arboviruses are presented in Figure 3 and will be discussed in a separate section.

The Dam Area Before Flooding

Collected material

Table 2 shows the total collections in the dam region before flooding. The material collected during this period represented 13.8%, 40.9%, and 8.6% of the total Diptera, vertebrates, and human sera collected respectively.

Virus isolations

A total of 33 strains were isolated from the Tucurui region, before flooding, of which 15 were

TABLE 1

Hematophagous Diptera, wild and sentinel (s) vertebrates' viscera and sera, and human sera, collected and inoculated, from the Altamira region, Para, Brazilian Amazonia, with, in parentheses, the number of arbovirus isolations, 1974–1988.

Year/Origin	*Diptera*	*Vertebrates*	*Human*	
			Collected	*Inoculated*
1974	15 079 (6)	47 s (4s)	3 667	442 (3)
1975	4 746	54 s	5 852	471 (1)
1976	3 167	69 s, 66	1 914	220 (1)
1977	12 920 (1)	70 s, 40	1 244	325 (1)
1978	9 347 (4)	64 s	594	257 (1)
1979	24 329 (2)	69 s	666	484 (3)
1980	24 237 (6)	72 s	454	393 (2)
1981	26 733 (1)	34 s	322	301 (1)
1982	14 107 (4)	17 s, 14	245	232 (1)
1983	70 556 (11)	241 (2)	213	141 (1)
1984	8 389 (9)	30 (1)	467	168
1985	28 210 (1)	450	750	87
1986	47 942 (5)	216 (2)	437	25
1987	—	—	352	30
1988	10 361 (4)	154	13	11
Totals	300 123 (54)	496 s, 1 184 (4s,5)	17 190	3 587 (15)

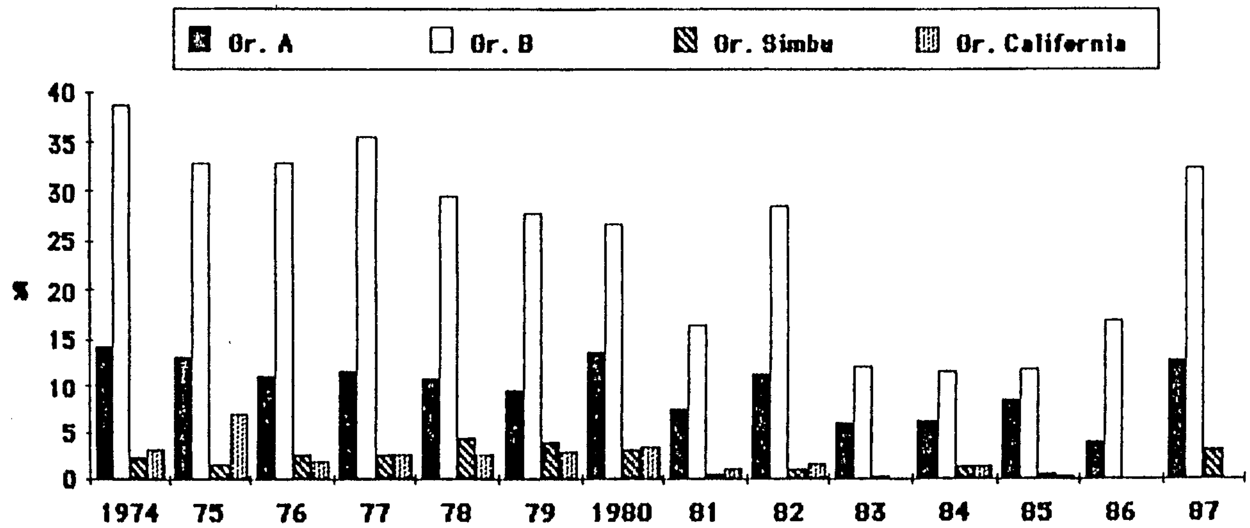

Figure 3. Yearly percents of positive HI antibodies against the 4 most prevalent groups of arboviruses, in human sera from Altamira, Par, Brazilian Amazonia, 1974-1987.

TABLE 2

Hematophagous Diptera, wild vertebrates' viscera and sera, and human sera, collected and inoculated, from the Tucurui hydroelectric dam area, Para, Brazilian Amazonia, with, in parentheses, the number of arbovirus isolations. The period of study, 1982–1988, is divided according to the phases of flooding of the lake. An * indicates that the collected material is yet under identification and/or inoculation.

Period	*Date*	*Diptera*	*Vertebrates*	*Human* Collected	*Human* Inoculated
	1982	53 118 (2)	818 (1)	163	59
PRE-FLOODING	1983	30 672 (4)	507 (4)	—	—
	Jan.1, 1984 to Sept. 6, 1984	90 361 (12)	679 (10)	352	7
Sub-Totals		174 151 (18)	2 004 (15)	515	66
DURING FLOODING	Sept. 7, 1984 to Dec, 31, 1984	617 526* (43)	590 (1)	1 165	72 (2)
	1985	265 114 (94)	915	1 764	68
POST-FLOODING	1986	146 576* (26)	695 (2)	1 678	71
	1987	60 087* (12)	357	384	23 (1)
	1988	*	333	476	32 (2)
Sub-Totals		471 777 (132)	2 300 (2)	4 302	194 (3)
Grand Totals		1 263 454 (193)	4 894 (18)	5 982	332 (5)

from wild vertebrates, 14 from mosquitoes, and 4 from phlebotomine sandflies (Table 2).

Animal serology

Before the flooding, a total of 457 wild animals were tested for the presence of antibodies. 132 were positive for at least 1 type of arbovirus.

Human serology

A total of 511 human sera were sampled and tested for arbovirus antibodies in the dam before the outset of flooding. Figure 4 shows trends in the serological responses during the 3 phases of flooding (see below).

The Dam Area After Outset of Flooding

Collected material

Although many mosquito pools collected during this period still have to be processed, approximately half of the total catch has been inoculated into mice. The number of animals which were collected during the 4 months flooding period represented only 12% of the total (Table 2).

A total of 5,467 human sera were collected, of which 266 were inoculated (80.1% of total inoculated). They were mostly from rural residents rather than people working on the dam construction.

Virus isolations

Of the 183 virus strains isolated since the outset of flooding (Table 2), 5 were from human, 3 from wild vertebrates, 114 from mosquitoes, and 18 from phlebotomine sandflies. During the flooding period the following strains were isolated: 1 from wild vertebrate, 2 from human, 41 from mosquitoes, and 2 from phlebotomine sandflies.

Animal serology

During the flooding period, 251 wild vertebrates were bled, of which 195 were positive for at least 1 type of arbovirus. During the post-flooding period, a total of 1816 animal sera were tested for antibodies, which were present in 100 sera.

Human serology

During the flooding period, 1,165 (19.6% of total) sera were sampled, including at least 126 local residents and 331 immigrants. During the post-flooding period, these figures were 4,246 (71.6% of total), with no data available about the origin of the sampled people.

A total of 1940 sera were obtained from the same donor more than once. Of these, 132 (6.8%) have serologically converted for at least 1 arbovirus. However the great majority (99/132 or 75%) were either group responses to toga viruses and flaviviruses or specific responses to YF virus. In these 2 cases, no conclusions can be drawn about the nature and time of human contaminations with sylvatic arboviruses. Thus, only serological conversions specific for one virus will be dealt with in the following accounts.

EVOLUTION OF HUMAN SEROLOGY

Changes in the profile of human serology and in the rate of virus isolations in people may be helpful indicators to assess the impact of ecological transformation on arbovirus transmission to man. Unfortunately, many data on historical and social aspects of the colonization are missing, for they are rarely collected at the same time as the ecological data. Despite these limitations, we collected data about age, sex, birthplace, residence (but not the date of entering the area) and occupation, thus allowing us only to infer about the infection inside or outside the region.

Results are shown in Figure 3 and 4 for the reference region and for the dam area, respectively. For the latter, the proportions are grouped according to the 3 periods: before, during and after flooding. Among the Togaviridae, Mayaro virus gave the greatest proportion of positive reactions. The few other positive reactions in the same group were due to Mucambo virus (member of Venezuelan Equine Encephalitis - VEE - complex), and to Eastern Equine Encephalitis (EEE) virus.

With respect to the prevalence of Flaviviridae antibodies, 2 facts shuld be emphasised. First, most of the positive results were probably due to YF vaccination. Secondly, among the specific reactions, Ilheus virus (ILH) and SLE viruses were responsible for 355 and 56 positive results, respectively. Antibodies to Oropouche (ORO) virus, a Bunyavirus of the Simbu group, were not very prevalent, despite the large impact in Amazonia during past epidemic episodes (Pinheiro *et al* 1986). Antibodies to the other groups were < 2%.

Few variations seem to have occurred between the 3 phases of the study, with a maximum of positive serologies during the flooding period, but the differences were not statistically significant ($chi^2 = .818$; $df = 6$; $p = .99$).

Virus-specific serological conversions (except for YF: vide supra) have been shown for the arboviruses ORO (12 conv.), ILH (8), Mayaro (6), Guaroa (3), Mucambo (2), and EEE (1), demonstrating few contacts of this population with these primarily sylvatic agents. The case of ORO virus is different, for it can cause large urban and rural epidemics.

DISCUSSION OF SELECTED ECO-EPIDEMIOLOGICAL TOPICS

In order to emphasise what may have happened in the transmission patterns of the arboviruses in relation with ecological modifications in the dam area, we will discuss the results under somewhat arbitrary but convenient categories.

First, these will concern evident epizootics caused by 1) new or already known arboviruses,

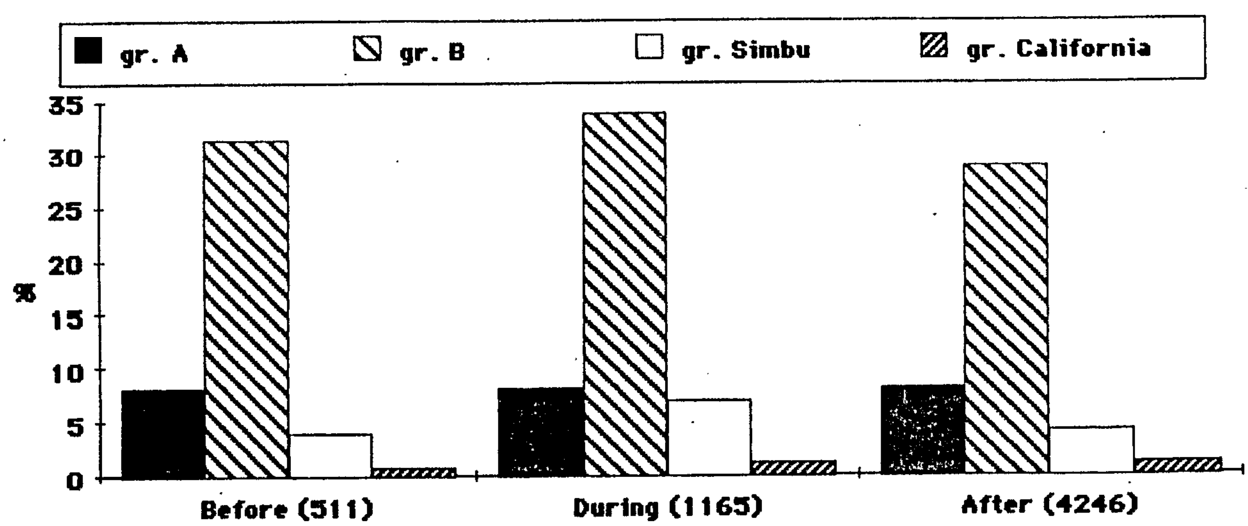

Figure 4. Percents of positive HI antibodies against the 4 most prevalent group of arboviruses, in human sera from the Tucurui dam area, Par, Brazilian Amazonia, grouped according to the three flooding periods, 1982-1988.

2) arboviruses from possible exogenous origin, and 3) endemic ones. Next, we will discuss the case of arboviruses: a) which have showed at most a little enhancement of their transmission level, whether or not caused by the new ecological conditions, and b) which have showed no apparent modification in their transmission patterns (referring to the control area).

Finally, we will briefly sumnmarise new ecological and virological data obtained as by-products of these studies, but which may reveal themselves important for planning complementary studies in the future.

Epizootics of New Arboviruses

The Anopheles A group (Bunyaviridae: Bunyavirus) arboviruses have shown a very distinctive transmission pattern as a consequence to the dam impoundment. Until 1984, only 3 different types were known in Brazilian Amazonia: Tacaiuma, Lukuni and Trombetas, the first 2 being pathogenic for man (Karabatsos 1985). These arboviruses, and 3 more new types appeared in the dam area during flooding or a little before, as a probable result of the proliferation of their mosquito hosts, mainly Anopheles (Nyssorhynchus) spp (Fig 5). *An nuneztovari* and/or *An triannulatus* seem to be the normal hosts of these viruses. However, it is the first time that Tacaiuma virus has been isolated from nocturnal anopheline, known only from the diurnal *Haemagogus janthinomys*, *Ae scapularis* and *An (Kerteszia)* sp. Similarly, Lukuni virus was known only from the latter mosquito and the diurnal-crepuscular *An nimbus*.

Considering the group as a whole, the numbers of isolated strains were 3 for Altamira region, 2, 26, and 54, for the pre-, during-, and post-flooding periods in the dam area, respectively. This suggested relationship between the flooding periods and intensity of transmission was confirmed when we considered the real infection rates of the mosquitoes for each arbovirus of that group ($chi^2 = 21.34$; df = 10; p = .0188).

Serological data were routinely available only for the Tacaiuma virus, which is the only 1 of these 6 Anopheles A viruses, from which an hemagglutinin has been obtained. Despite the 2 isolations obtained from man in the Altamira region, yearly antibody prevalence never exceeded 1.5%. In the dam area, the yearly prevalence was < 1.2%. The sera of 5 birds (different species), 3 monkeys (*Alouatta belzebul*) and 2 armadillos (Dasypus novemcinctus), collected either before or after flooding, had antibodies for this arbovirus. Some neutralization tests have shown that carnivorous and ungulate mammals are probably involved as vertebrate hosts of the Tucurui virus.

Epizootics of Exogenous Arboviruses

Gamboa virus (GAM: Bunyaviridae, Bunyavirus), although widely distributed in central and south America (Panama, Ecuador, Argentina: Calisher *et al* 1981; Karabatsos 1985), is new to Brazil. Epizootic-level transmission of this virus (probably including various related types) occurred in Panama during the period from April 1985 to July 1987 (Dutary *et al* 1989).

It is noteworthy that our first 2 isolated strains were from the blood of birds (Geotrygon montana: Columbidae, and Xenops minutus: Furnariidae) collected on August 2 and 3 1984, 1 month before the outset of flooding. These 2 species are very common from tropical Central America to Argentina (Dunning 1982) and the former is known as a possible although not proved migrant (F Novaes pers comm). Thirty three strains were isolated from mosquitoes *Aedeomyia squamipennis*, 1 from *An triannulatus*, and 2 more from the birds *G montana* and *Glyphorhynchus spirurus* (Dendrocolaptidae), all collected in the dam area during the post-flooding period, or at least 1 year after the initial isolations (Fig 6).

Two hypotheses may be formulated to explain what happened in this region: 1) the virus was

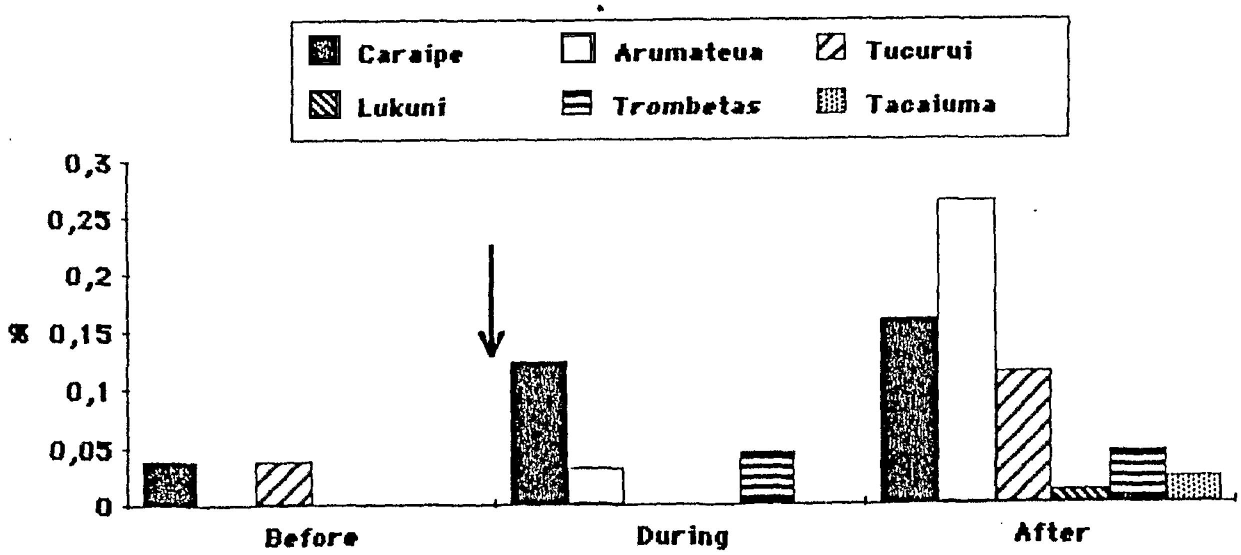

Figure 5. Percents of mosquito (*Anopheles* spp) pools, positive for 6 different Bunyaviridae of the Anopheles A group, from the Tucurui dam area, Par, Brazilian Amazonia, grouped according to the 3 flooding periods, 1982-1988.

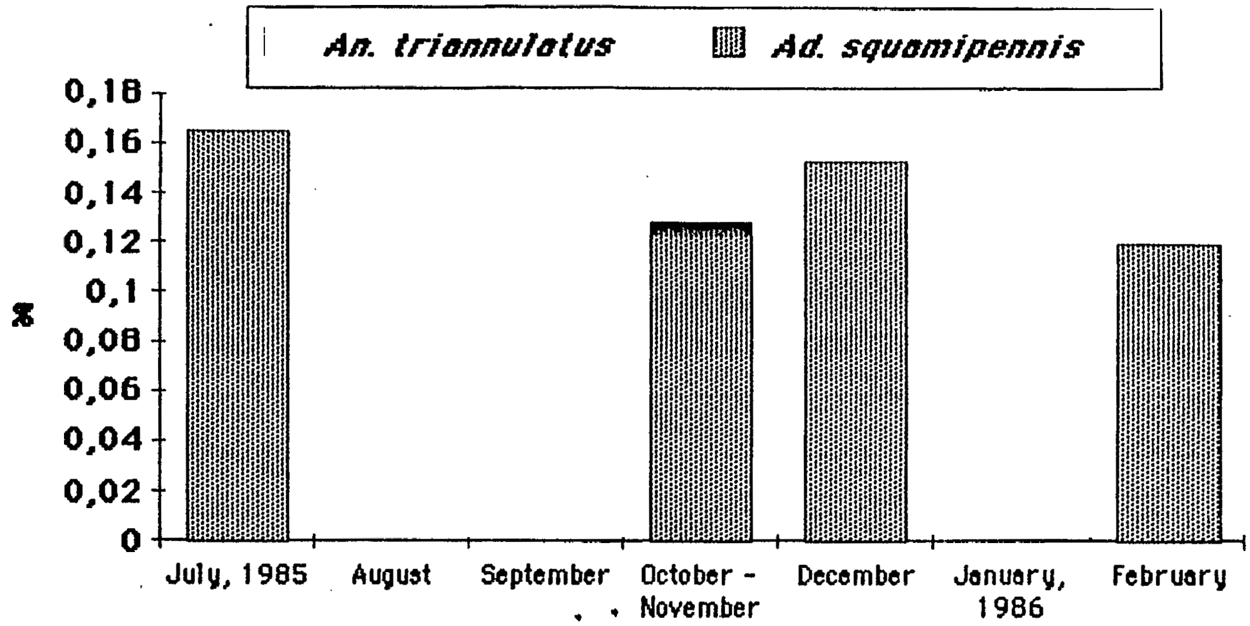

Figure 6. Monthly real infection rates of the mosquitoes *An triannulatus* and *Aedeomyia squamipennis* for arboviruses of the Ga group from the Tucurui dam area, Par, Brazilian Amazonia, during the post-flooding period, 1985-1986.

present in the area, but was never encountered despite the good sample size of vertebrates and mosquitoes that was examined; 2) the virus has been newly introduced in the area by a migrating bird from Central America, where the agent was circulating since its first isolation in 1962, and with greater intensity during the period from 1985 to 1987. Further, a delay of 8 months was necessary for the virus being detectable in the mosquito host. *Ad squamipennis* is known to breed in waters invaded by floating plants, thus it is probable that such favourable conditions were not met until some months after completion of the flooding.

As it was already noted by the authors, the vertebrate hosts of this virus are birds living in the understorey of humid tropical forests. Birds of the same families as those from Tucurui were found with antibodies in Argentina (Calisher *et al* 1981). The unique strain from *An triannulatus*, although probably not a laboratory artefact, is not thought to be of ecological importance.

Epizootics of Endemic Arboviruses

Guaroa (GRO: Bunyaviridae: Bunyavirus, group California)

Until the present studies, GRO virus has been isolated only from man in Brazil (Hervé *et al* 1986) and from anopheline mosquitoes in Colombia, Panama, and French Guiana (Karabatsos 1985; Dégallier 1982). In Brazilian Amazonia, it is restricted to the eastern part of Par State and has only been isolated once (August 1976) in Altamira, its westernmost station. Antibody prevalence in human sera is generally low and was zero in the reference region in 1986 and 1987 (Fig 3). In the dam area of Tucurui, it has been isolated twice from man and 4 and 3 times from *An nuneztovari* and *An triannulatus*, respectively. The virus was present probably during the flooding period (November 1984) and an epizootic occurred 1 year later between October and December 1985 (Fig 7). Although 3 human seroconversions occurred between

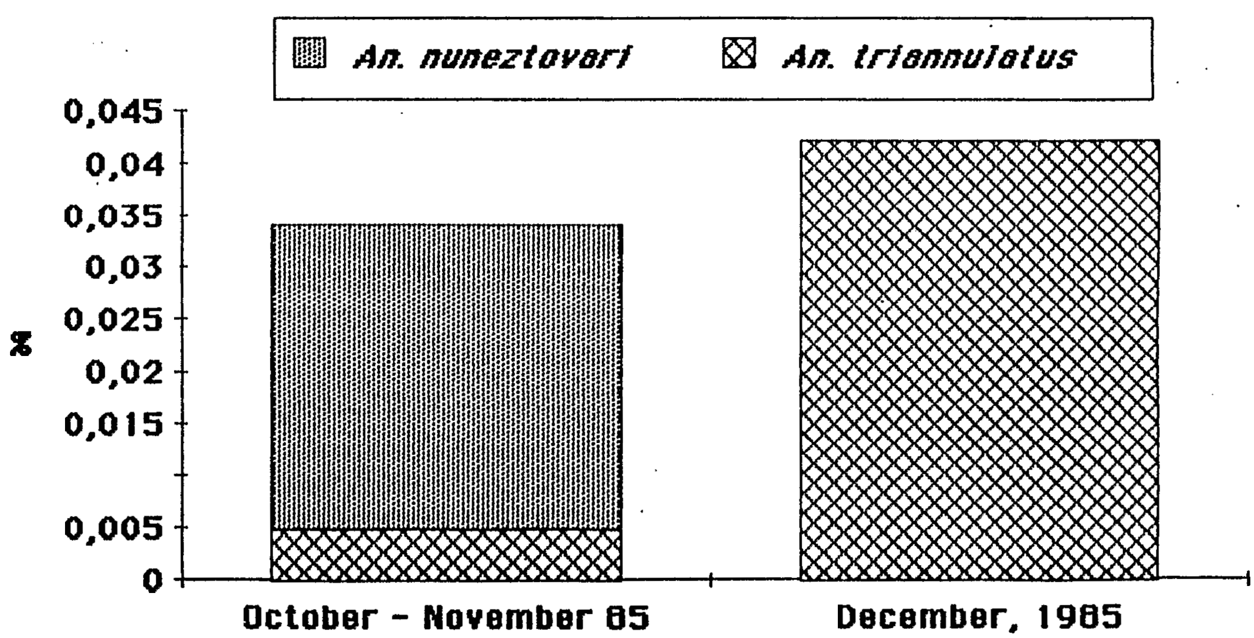

Figure 7. Monthly real infection rates of the mosquitoes *An nuneztovari* and *An triannulatus* for Guaroa virus from the Tucurui dam Pará, Brazilian Amazonia, during the post-flooding period, 1985.

June 1985 and November 1988, and 1 strain was isolated in July 1987, its serological prevalence in human sera remained negligible. The only wild vertebrates which had shown antibodies were 3 monkeys (*Alouatta belzebul*), captured during the flooding period.

Available data idicates that this arbovirus has been favoured by the new eco-epidemiological conditions due to the dam construction and flooding. It is also probable that human cases have been missed because of the lack of reporting of febrile cases.

Temporary Enhancement of Transmission of Enzootic Arboviruses

Six arboviruses have shown transsmission patterns which were thought to be related with the ecological modifications caused by the Tucurui dam: Turlock, Kwatta-like, ORO, SLE, Maguari/Xingu, and Triniti viruses. Each was chosen in the following discussion for some eco-epidemiologic peculiarity.

Turlock (TUR: Bunyaviridae: Bunyavirus, group Turlock)

TUR virus is typically a *Culex*— bird transmitted agent. In Brazilian Amazonia, it has been isolated sporadically from at least 11 different bird species (Hervé *et al* 1986) and from mosquitoes. In the dam area, pre and during flooding period has each furnished 2 strains. Although the virus has not been isolated again from the area — as is also the case for the Altamira region since 1974 — it is probable that the enormous proliferation of *Culex* spp during flooding favoured a raising of enzootic circulation, which was reflected in the positive serology in the sera of 6 birds, bled during the first half of 1986 (during the total period of study, 13 birds of 12 species had been positive).

Kwatta-like (Rhabdoviridae, group Kwatta)

This virus is ecologically comparable to TUR, with a *Culex* — bird cycle (Hervé *et al* 1986). It was present in the area before flooding (1 strain) and showed an enhanced circulation during flooding, 4 strains being isolated from *Culex* (Culex) sp. However, unlike TUR virus, it was also present in the Altamira area at the same time (3 strains isolated in 1982-83), indicating that favourable eco-climatic conditions were possibly independent of changes caused by the dam.

Oropouche (ORO: Bunyaviridae: Bunyavirus: group Simbu)

This virus, not prevalent in the Tucurui area's human serological surveys (cf supra), at first did not appear to us to have been favoured by the dam impoundment, nor the populations of its usual ceratopogonid midge vector. However, 1 strain has been isolated recently (April 1988: possibly an imported case, from Araguaina, Gois, see below) and 10 seroconversions occurred in the human population between the beginning of 1986 and the middle of 1988. Further, the continuous circulation of ORO virus among the wild vertebrates is attested by relatively high annual rates of positive animals, mainly birds and monkeys during the flooding period. The annual prevalence rates of positive birds (during the flooding phase, the bird sera were all negative) were between 2% and 7.1%, with 2 peaks in 1983 and 1986. The monkey sera collected during the flooding period showed a positivity of 8.5% (too few monkeys were tested during the other periods).

Besides the confirmation of the effective role of birds (22 different species), monkeys (5 species) and sloths (1 species) in the sylvatic cycle of this arbovirus (cycle hypothesized by Hervé *et al* 1986 and previous authors), these findings are compatible with the creation of ecological conditions that favoured the transmission of ORO virus. But recent studies (Vasconcelos *et al* 1989) have brought some support for another interpretation. In fact, such an enhancement of transmission may also have been due to generally favourable ecological conditions that developed simultaneously in a much wider area. The latter is supported by a) rise in antibody prevalence in human sera collected during 1987 both in the reference region and the dam area, and b) the demonstration of a large epidemic which occurred some 300 km southeast of Tucurui from December, 1987 to March, 1988. Unfortunately, ceratopogonid midges collected from the dam area during 1988 have not yet been sorted.

Saint Louis Encephalitis (SLE: Flaviviridae: Flavivirus)

SLE virus is predominantly a *Culex* — bird circulating arbovirus. However, it has been isloated from monkeys and other canopy-dwelling mammals (sloths, marsupials and bats). Until recently, it was considered as a canopy-transmitted agent, but isolations from a great variety of ground-living mosquitoes and from an armadillo (*Dasypus novemcinctus*) have suggested the existence of other cycles at lower levels (Hervé *et al* 1986; unpubl data).

In the reference region, SLE has probably been circulating from 1974 to 1986, with 2 epizootic periods in July - November 1974 and February - August 1983. It probably remained in the area at a low (enzootic) level of transmission, as was suggested by the isolation of two strains, in May 1984 and April 1986, respectively.

In the dam area, SLE virus was isolated in 1982, in 1984 before flooding and during a 2-month period after outset of flooding, and one more time 1 year after, in 1985. Human seroconversions have been detected in November 1984 (3), April and May 1985 (2). A positive reaction against this arbovirus has been detected in the serum of one marsupial Didelphis marsupialis, captured in 1985, thus confirming the possibility of this very common, versatile mammal being one of the amplification hosts at ground-level. Annual rates of antibodies in birds (10 species) were low (0.4% - 0.8%) but attained 3.4% in 1984.

Although the flooding of the dam favoured a great proliferation of culicine vectors (especially *Cx declarator*), and 6 strains were isolated during this period (versus 3 from Altamira), the present data has not revealed a clear-cut difference between the transmission patterns in the 2 regions. One interesting finding was a very high prevalence of flavivirus antibodies (35.7% of 255 sera, 1985) in two sympatric species of large terrestrial tortoises (*Chelonoidis carbonaria* and *C denticulata*), the populations of which were concentrated on islands. Unpublished experimental infection of tortoises by SLE virus have shown that both specific and group reacting antibodies are appearing simultaneously (although the latter at lower rates), allowing us to suspect SLE virus as the natural causative agent of these serological findings. However, the exact significance of this has not yet been elucidated, especially the inter-tortoise vector. An as yet unidentified diurnal *Culex (Melanoconion)* mosquito has been observed to be avidly attracted by these reptiles, but this apsect deserves more studies.

Maguari/Xingu (two jointly studied Bunyaviridae: Bunyavirus: group Bunyamwera)

Strains, possibly of one or both of these 2 arbovirus types (the necessary plaque reduction neutralization tests have not yet been completed; see Calisher *et al* 1987 for other types and subtypes in the complex), have shown a possible shift of mosquito hosts. In reference to other areas where these arboviruses have been encountered, the natural hosts are diurnal mosquito species such as *Ae scapularis*, *Ae serratus*, *Ae fulvus*, *Psorophora ferox*, and various Sabethini.

In the Tucurui dam area, there have been 4 isolates between 1986-87 (nearly 2 years after flooding completion), from the crepuscular-nocturnal *An nuneztovari* and *An triannulatus*. However, the hypotheses of host shift needs to be tested by a precise identification of the strains — for example, a third distinct type may have appeared — and by looking for the presence of specific antibodies in wild vertebrates.

Triniti (TNT: ungrouped Togaviridae)

The sylvatic cycle of TNT virus was not characterised until recently. It is transmitted by diurnal sabethine (genera *Sabethes*, *Wyeomyia*, *Trichoprosopon*) and/or diurnal anopheline (*An nimbus*) mosquitoes between diurnal rodents (Hervé *et al* 1986). It has been isolated from these vectors in 1983, from the Altamira region (2 strains) and in Tucurui during the pre-flooding period (3 strains). One year after the flooding of the dam area (November - December 1985), 4 strains were unexpectedly isolated from *An triannulatus*. A shift — and delay — similar to those already noted for the Maguari/Xingu viruses may also well have occured with TNT virus. Nevertheless, it is not clear if such a shift in vector species was accompanied by a shift in vertebrate hosts.

Arboviruses with Transmission Patterns Apparently not Modified

Eight arboviruses did not show clear-cut differences between their transmission patterns in the reference region and the ecologically modified dam area.

Yellow Fever and Mayaro (YF: Flaviviridae: Flavivirus)

YF virus is the most important cause of human deaths by arbovirus in Brazil, despite the well-proven efficacity of the vaccine. In fact, it appears logistically impossible to vaccinate a sufficient proportion of the people living far from the cities, to prevent man-to-man rural and/or sylvatic outbreaks which seem associated with fatal cases (Travassos da Rosa *et al* 1987).

In the Altamira region, human cases have been confirmed, either by virology, histopathology, and/or serological conversion, nearly each year from 1977 to 1984, and in 1987-88. In the Tucurui dam area, one strain was obtained from a (*Chiropotes satanas*) a little before the outset of flooding and 2 human cases (1 fatal) were confirmed in 1987. With regard to human serology, the routine tests have not allowed separation between natural contaminations and antibodies acquired from vaccination. However its actual sylvatic circulation was indicated by an 9.1% rate of antibodies in the monkey sampling done in 1984, and one positive reaction in the sera of a *Cebus apella* monkey, bled in 1985.

It is probable that the *Haemagogus* — monkey (with sporadic contacts with man) cycle of this virus has not been modified by dam construction and thus, that vaccination of all people entering such an area must be maintained.

The same can be said for Mayaro (MAY: Togaviridae: *Alphavirus*) virus, which is ecologically extremely similar. Isolated from man in Tucurui in November, 1984 (along with seroconversions in October (1), December (2), 1984, April, 1985 (1), April, 1986 (1), and February, 1989 (1)), and during the seventies in Altamira, it has shown a high and fairly constant prevalence of antibodies in human sera collected in the 2 areas. In Tucurui, during 1984, 48.2% of the sampled monkeys had antibodies to it. Thereafter, 4 monkeys and 2 birds (*Arremon tactiturnus* and *Tyrannus melancholicus*) were positive in 1985. Two other birds, *Formicivora grisea* and *Cercomacra tyrannica* were positive in 1983 and 1987, respectively. This arbovirus, whose sylvatic cycle seems to be identical to that of YF (Hervé *et al* 1986), would be a good "indicator" of the ecological potentiality of any area for the sylvatic circulation of YF virus in the absence of human cases or when vaccine-acquired antibodies against YF are obscuring the sylvatic pattern.

Itaporanga (Bunyaviridae: group Phlebotomus)

This virus has been isolated from *Culex* sp mosquitoes, collected in Altamira and Tucurui, in May and June, 1985, respectively. From Tucurui, 1 monkey (*Saguinus midas*: 1982), 3 marsupials (*Didelphis marsupialis*: 1984, 1985), 3 rodents (*Proechimys* sp *Dasyprocta primnolopha*: 1982; *Agouti paca*: 1984), 1 bat (1986) and 3 birds (*Micrastur gilvicollis*: 1986; *Thamnomanes caesius*: 1986; 1988) had

TABLE 3

Probably new arbovirus types in the Altamira (ALT) and the Tucurui (TUC) hydroelectric dam regions, Para, Brazilian Amazonia, 1974–1988; NC = not classified; NG = not grouped.

Virus name	*Date of coll.*	*Host*	*Prototype*	*Genus*	*Antigenic group*
		ALTAMIRA, PARA, 1974–1988			
Acatinga	09/06/88	Phlebotominae sp.	AR 482250	*Orbivirus*	Changuinola
Altamira	07/30/74	"	AR 264277	"	"
Assurinis	09/03/88	"	AR 482249	"	"
Bacajai	"	"	AR 482267	"	"
Gorotire	09/06/88	"	AR 482251	"	"
Uxituba	04/12/86	"	AR 452652	"	"
Ambe	09/06/82	Phlebotominae sp. (males)	AR 407981	*Phlebovirus*	Phebotomus
Joa	03/29/79	Phlebotominae sp.	AR 371637	"	"
Tapara	02/04/83	Phlebotominae sp. (males)	AR 413570	"	"
Cajazieras	04/08/86	Chiroptera sp.	AN 447884	NC	NG
Codajas	05/04/84	*Haemagogus janthinomys*	AR 423137	NC	NG
Irii	02/18/82	Phlebotominae sp.	AR 408005	NC	NG
Pirixa	05/02/84	*Lonchophila thomasi*	AN 422840	NC	NG
		TUCURUI, PARA, 1984–1988			
Aracai	02/20/84	Phlebotominae sp.	AR 425269	*Orbivirus*	Changuinola
Aratau	08/27/84	*Lutzomyia carrerai*	AR 428812	"	"
Aruana	08/27/84	" "	AR 428815	"	"
Canoal	10/01/84	*Lu ubiquitalis/Lu shawi*	AR 433317	"	"
Coari	09/30/84	*Lu carrerai/Lu infraspinosa*	AR 433343	"	"
Jandia	07/20/85	*Lu ubiquitalis/Lu shawi*	AR 440489	"	"
Jatuarana	07/20/85	" "	AR 440497	"	"
Mucura	05/30/85	*Anopheles triannulatus*	AR 455230	"	"
Pacaja	07/20/85	*Lu ubiquitalis/Lu shawi*	AR 440503	"	"
Parakana	10/31/84	*Culex declarator*	AR 463992	"	"
Paranati	"	*Culex coronator*	AR 439007	"	"
Pependana	07/20/85	*Lu ubiquitalis/Lu shawi*	AR 440504	"	"
Pindobai	02/21/86	Phlebotominae sp.	AR 482675	"	"
Piratuba	12/14/85	"	AR 478781	"	"
Surubim	07/20/85	*Lu ubiquitalis/Lu shawi*	AR 440507	"	"
Timbozal	07/25/85	*Lu carrerai/Lu infrasponosa*	AR 440541	"	"
Tocantins	05/20/86	Phlebotominae sp.	AR 486776	"	"
Tracambe	06/03/85	*Anopheles nuneztovari*	AR 455156	"	"
Tuere	05/20/86	Phlebotominae sp.	AR 484704	"	"
Jacunda	10/31/84	*Myoprocta* sp.	AN 428329	*Phlebovirus*	Phlebotomus
Morumbi	04/12/88	*Homo sapiens*	H 475236	"	"
Arumateua	30/10/84	*An (Nyssorhynchus) sp.*	AR 437811	*Bunyavirus*	Anopheles A
Caraipe	08/26/84	" "	AR 428793	"	"
Tucurui	02/24/84	" "	AR 422535	"	"
Jatobal	05/04/84	*Nasua nasua*	AN 423380	"	Simbu
Papura	04/15/85	*Psychodopygus davisi*	AR 450572	NC	NG
Trocara	02/24/84	*Aedes serratus*	AR 422431	NC	NG
Uriurana	12/16/85	Phlebotominae sp.	AR 479776	NC	NG

antibodies for this arbovirus. Thus, its enzootic circulation may not have been modified by the dam presence.

Ilheus (Flaviviridae: Flavivirus)

Ilheus, a widely distributed and pathogenic arbovirus (Pinheiro *et al* 1986), does not seem to have been influenced by the dam presence. Although it has not been isolated in the 2 regions of study since May, 1984, and March, 1984, respectively, human seroconversions, detected in September, 1985 (1), February, 1986 (1), April, 1986, (1), February, 1987 (5), and August, 1987 (1), attested to its continuous presence, and enzootic circulation in the dam area. In Altamira, specific antibodies were detected in the sera of bats (1 in 1985 and 3 in 1986). In Tucurui, various birds (1982; 1988), 1 monkey (Alouatta belzebul: 1984), 2 rodents (Dasyprocta prymnolopha: 1982; Myoprocta sp: 1984) and 1 marsupial (Marmosa cinerea: 1987) were positive.

Icoaraci (Bunyaviridae: Phlebovirus)

Icoaraci virus was not isolated from either vertebrate or from arthropods, after August, 1983, and November, 1983 in Altamira and Tucurui, respectively. However, its continuous enzootic circulation in the dam region was demonstrated by the detection of specific antibodies in the sera of 2 monkeys *Alouatta nigerrima* and *Saimiri sciureus*: 1983), 1 marsupial (Marmosa cinerea: 1983) and 7 rodents (*Proechimys* sp 1982-83, 1985). In the reference region, rodents of the same genus were found positive in 1983, 1984 and 1986.

Tucunduba (Bunyaviridae: Bunyavirus: group Bunyamwera)

This arbovirus is transmitted mostly by sabethine mosquitoes and some diurnal species of other tribes. It has been active in the Altamira region from 1978 to 1981 (8 strains), and in the dam area before (1 strain, 1983) and after (1 strain, 1987) the flooding period. It is noteworthy that the later strain was not isolated from the known mosquito hosts but from *An triannulatus*.

As there has been no epizootic episode, a dead-end infection seems a more acceptable hypothesis than any vector-shift to explain this aberrant isolation. Whatever the case, the dam does not seem to have affected the transmission of this arbovirus.

Eastern Equine Encephalitis and Western Equine Encephalitis

The Eastern Equine Encephalitis (EEE: Togaviridae: Alpahvirus) virus, which is largely distributed in the Amazonian region (Pinheiro *et al* 1986) and with a complex sylvatic cycle (Hervé *et al* 1986), has not been isolated from the Altamira region since October, 1974, and from the dam area since July, 1983, but have probably been present in the latter area as was suggested by a seroconversion detected in November, 1985 in a human serum and positive serological reactions detected in the sera of a monkey (*Saimiri sciureus*: 1984) and a bird (*Thamnophilus aethiops*: 1985). The presence of the Western Equine Encephalitis (WEE: Togaviridae: Alphavirus) virus in the dam area was suggested by the detection of specific antibodies in the sera of numerous birds (at least 14 species), collected each year from 1982 to 1988.

The dam impoundment does not seem to have affected the transmission patterns of these two arboviruses.

New Ecological Data About Little Known Arboviruses

Isolations of 5 rarely encountered or little known arboviruses from material collected in the dam area, will allow us to confirm previously suspected hosts or cycles. One of these is Bush Bush (BSB: *Bunyaviridae*: Bunyavirus: group Capim) virus, isolated in 1959 in Trinidad Island W.I., far from the Brazilian Amazonian region, between 1960 and 1975 from a man in Santarem, from *Culex* sp mosquitoes (1 strain), and *Ameiva ameiva* lizard (1 strain: not cited in Hervé *et al* 1986), and sentinel mice (18 strains) near Belém city. One strain of this virus was obtained in Tucurui, from the wild terrestrial rodent *Proechimys* gr *guyannensis*, 9 years after the former isolations.

Aruac (Rhabdoviridae: unclassified: ungrouped) virus, described from Trinidad, is similar and was isolated twice from *Culex* spp, near Belém and in the dam area, Tucurui, 13 years later.

Munguba virus (Bunyaviridae, *Phlebovirus*, Phlebotomus group; Travassos da Rosa *et al* 1983) have been isolated once from phlebotomine sandflies collected during the post-flooding period in Tucurui (1986). The only other isolation was the prototype, 6 years before, from phlebotomine sandflies collected on the northern side of Amazon river, some 500 km from Tucurui.

Santarem virus, an ungrouped Bunyaviridae, isolated first from an Oryzomys rodent species in 1973, and later in 1984 from phlebotomine sandflies, has been isolated recently in 1987 from sandflies in the Tucurui area.

The virus Acado-like (Reoviridae: Orbivirus: group Corriparta), has been isolated from *Cx declarator* collected near Belém city, in 1984, and once from *Cx coronator* collected the same year (some days before outset of flooding) in the dam area.

Emergence of New Types of Arboviruses

During the period of study, 40 new types of arboviruses were isolated (Table 3). Twenty-four (60%) of them were classified in the Changuinola group of the genus Orbivirus (Reoviridae) (Travassos da Rosa *et al* 1984 and unpubl. data). Five (12.5%), 3 (7.5%), and 1 (2.5%) were classified in the Phlebotomus, Anopheles A, and Simbu groups of Bunyaviridae, respectively. The remainders (7 or 17.5%) are yet neither classified nor grouped. None was isolated in more than one region and except for the *Phlebovirus*, the numbers of new types in each group were superior in the dam area than in the reference one.

At this time, serological data for these new arboviruses are very scarce, and only the following arbovirus-vertebrate associations may be inferred: Jacunda virus/rodents, Jatobal virus/carnivorous - primates, Trocara virus/rodents.

DISCUSSION AND CONCLUDING REMARKS

In this first attempt to classify the different arboviruses according to their various ecological reactions to the construction of a big dam, we have encountered one main difficulty: the possible grouping of the same virus in more than one category. This is the consequence of a great variety and/or complexity of the cycles of these arboviruses, combined with a lack of ecological information.

Some numbers will express the richness of the region: a total of 74 different types, at least 53 (71%) of them were isolated from the Tucurui area — one third of the total known types in Brazilian Amazonia — versus 37 (50%) from Altamira, only 300 km distant from the former. At this date and with comparable sampling efforts, the two areas have furnished only 16 (21.6%) viral types in common. These results merit the following remarks: 1) the diversity of arboviruses in the studied regions is high, 2) the endemicity rate of the arboviruses is high, and 3) the initial hypotheses made about the ecological similarity of the two studied regions may need to be revised.

Amazonian arboviruses were classified by Hervé *et al* (1986) in various types of cycles, based on the usual hosts (vertebrates and arthropods), and eventually incorporating human contaminations. The presently reported studies have not modified these schemes, but to the contrary, have showed that they may be useful for the forecasting of possible impacts.

For example, a great proliferation of mosquitoes such as *Anopheles (Nyssorhynchus)* spp has favoured the circulation of the arbovirus Guaroa and 6 types pertaining to the Anopheles A group. Temporary conditions, which prevailed during the flooding period, were favourable to a great increase in *Culex (Culex)* spp density, thus enhancing the enzootic circulation of various arboviruses like Turlock, Kwatta-like, and SLE. *Aedeomyia squamipennis*, multiplying with a certain delay after the flooding was completed, supported an outbreak of the bird-associated Gamboa virus. This agent, formerly not isolated in Brazilian Amazonia, was possibly introduced by a migrating bird.

Another ecological factor, which has possibly played a role in the temporary amplification of arbovirus circulation, and in the appearance of viruses so far undetected, may be the concentration (natural or through the saving operation) of stressed wild animals on the isles and the banks of the lake. New types of biotypes (swamps) may also have attracted some new species for the region.

Arbovirus studies in relation with the construction of hydroelectric dams are scarce, especially in the neotropical region. Two projects have implicated arbovirological studies: the Bayano dam in Panama (Galindo *et al* 1983) and the Brokopondo dam in Surinam (Dutch Guiana) (Leentvaar, 1967). In the Amazonian River Basin, the present study is the first of that kind, therefore it may be ecologically unreliable to compare it with other studies done in fairly distinct geographical and/or climatic environments. The only detailed information of a similar nature is that of the Bayano Project and the following discussion emphasizes the most salient differences and similarities.

The effects of flooding seem to have been very different in some cases:

— neither the VEE virus or its closest Amazonian relative, Mucambo virus, have been isolated in Tucurui (versus numerous strains in Panama); the only indication of the presence of the latter virus in Tucurui was a human seroconversion detected in May, 1985.;

— no virus of the Chagres (Bunyaviridae: *Phlebovirus*; group Phlebotomus) complex has been isolated in Tucurui (versus numerous strains in Panama);

— EEE virus was possibly favoured by the Bayano dam impoundment, but it nevertheless circulated at a low level. As in the Tucurui area, its real impact remains to be appraised.

On the other hand, the effects of dam impoundment were comparable in the following instances:

— SLE virus showed an epizootic-like episode during the flooding periods of both projects;

— the epizootic of Gamboa viruses have followed a similar pattern in the both places, characterised by an 8 month delay between the outset of flooding and the first isolation from the mosquito host; however, the duration of the epizootic (not including the 2 pre-flooding strains isolated from birds) was significantly shorter in Tucurui than in the Bayano lake region (14 versus probably more than 18 months); another difference between the 2 series of results is that no strain has been isolated from male *Ad squamipennis* mosquitoes collected in Tucurui, thus making the possibility of transovarial transmission improbable in the latter area;

— the enzootic transmission of YF and MAY viruses does not seem to have been modified in any respect, in either of the 2 regions;

— after an initial outbreak, some arboviruses, like the Utinga/Utive/Pintupo complex (Bunyaviridae: group Simbu: Seymour *et al* 1983), seem to have "disappeared" from the Bayano area, but doubts remain about the significance of such a lack of isolation;

— a number of new (or unclassified) types were isolated in the groups Changuinola, Phlebotomus and others, in either cases.

The reported findings are of a preliminary nature but they show the necessity of lomgitudinal and long-term studies for a better understanding of the extremely diverse and complex cycles that characterise the Amazonian arboviruses.

An important factor that emerged from the studies is the necessity for a reference or control area for comparative purposes and that it cannot be just the dam area before flooding (for the present study, the reference area has been chosen a posteriori). Moreover, it ought to be chosen not so far from the dam as it was in the present case.

For future studies, it would be interesting: a) to define the ecological characteristics of the new equilibrium which, we think, may now have been established in the region of the Tucurui dam and b) to adopt a more provisional attitude (hypothesis testing) during field work in regions of planned dam impoundments.

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