Bionomics of *Anopheles aquasalis* Curry 1932, in Guaraí, State of Rio de Janeiro, Southeastern Brazil-I. Seasonal Distribution and Parity Rates

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From a total of 12,721 anophelines collected in a lowland area in Guaraí, Rio de Janeiro, from November 1991 to October 1992, 99.7% (12,688) were Anopheles aquasalis*. This species occurred throughout the year, but in higher numbers from April to September, when rainfall was low or moderate. The proportion of parous females in June was significantly higher than the annual rate.* An. aquasalis *was weakly attracted by a light-trap, and no significant differences in abundance were detected between nights with and without moonlight.*

Key words: mosquito bionomics - *Anopheles aquasalis* - Culicidae - parity - Brazil

Anopheles (Nyssorhynchus) aquasalis Curry l932, is a coastal mosquito whose malaria vector status and behavior vary throughout its distribution in South America. Its occurrence is usually associated with the presence of brackish water which limits its distribution in Brazil to no more than 30 km from the coast. Exceptions were observed by Lucena (1946), Deane et al. (1948), Rachou et al.(1950) and Rachou (1958).

An. aquasalis has been a primary vector of malaria in coastal Brazil (Deane 1986). Preferred sources of blood and frequency indoors of *An. aquasalis* vary geographically, and its vector status may depend on density. The species is sometimes considered zoophilic, exophilic and a secondary malaria vector, as in some areas of the Amazon and in southeastern Brazil. However, *An. aquasalis* has been considered anthropophilic and endophilic and a primary vector in some localities in northeastern Brazil (Boyd 1926, Galvão et al. 1942, Coutinho & Ricciardi 1943, Ramos 1943, Lucena 1946, Coutinho 1947, Deane et al. 1948, Rachou et al. 1950, Ferreira l964).

This paper describes the local anopheline fauna and the monthly frequency of nulliparous and parous *An. aquasalis* females at Guaraí, a locality in the "Baixada Fluminense", the coastal lowlands in Rio de Janeiro, southeastern Brazil. *An. aquasalis* has been regarded as one of the malaria vectors in "Baixada Fluminense" together with *An. darlingi* and *An. albitarsis* since early in the century (Boyd 1926, Root 1926). Guaraí, was up to few years ago, considered as part of the municipality of Magé, one of the most malarious areas in "Baixada Fluminense" until the 1950's. The annual incidence of malaria in Magé decreased to non endemic levels in the following decade due to extensive drainage and indoor spraying of DDT put into practice since the 1940's (Deane 1988, Moraes 1990). Recently (1970-1995), *An. aquasalis* has been considered the only potential vector during minor malaria outbreaks reported in "Baixada Fluminense" (Santanna 1995, pers. comm.). In spite of the epidemiological importance of *An. aquasalis* in this area few observations have been carried out, on its bionomics except for descriptions of its resting-places and blood-meal sources (Flores-Mendoza & Deane 1993, Flores-Mendoza et al. 1996).

MATERIALS AND METHODS

The study was carried out in Guaraí, municipality of Guapimirim (until recently considered part of Magé), State of Rio de Janeiro, from November 1991 to December l992. The field work was performed on a dairy farm, named "Meia Noite" (22°41'S and 42°57'W), where the Guaraí River crosses the road BR-493, between the villages of Magé and Itambi (Fig.1). The sedimentary terrain is flat and crossed by channels of brackish water and several rivers and streams that flow from a distant mountain chain (the "Serra dos Orgãos") to Guanabara Bay.

The land is predominantly pasture with a few scattered trees and small patches of sugar cane and banana. There is frequently a breeze in Guaraí due to its proximity to the seashore and lack of high vegetation. The terrain is frequently flooded during the rainy season and by tides.

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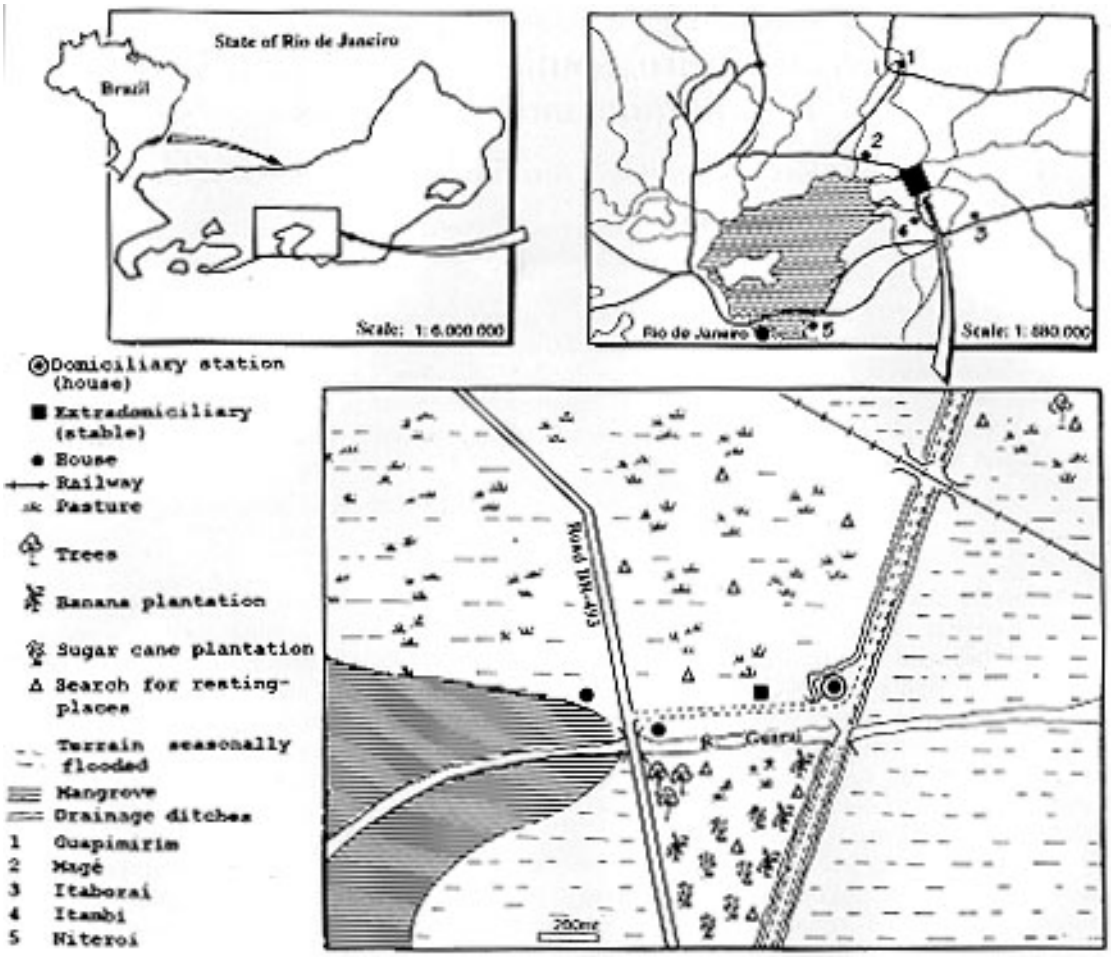

Fig. 1: study area in Guaraí, State of Rio de Janeiro, Brazil.

The few houses on "Meia Noite" farm are separated by large pastures, where the cattle browse during the day. At night, the cattle are confined to an open wooden stable where they stay from sunset to dawn.

Monthly rainfall averages and monthly minima, maxima and averages of relative humidity and temperature were obtained from Magé (Fig. 1), the nearest meteorological station. Monthly tide variations in Guanabara Bay were provided by the local Brazilian Navy Office.

Domiciliary station - Mosquito captures were performed inside and immediately outside of a house, which was nearly 500 m from the road BR-493, and 30 m from the Guaraí River and surrounded by narrow drainage ditches. The house was built simply with adobe and was inhabited by the same family (four persons) throughout the study. Its living-room, two bedrooms, kitchen, bathroom and small veranda were lined with tiles but had no ceiling. Windows had neither curtains nor screens. The house was frequently visited by neighbors to watch television from 6:00 to 9:00 p.m.

Extradomiciliary station - The cattle stable, 100 m from the domiciliary station, was selected for captures outside the house (Fig. 2).

Types of collections - At the domiciliary station, collections were conducted with two human baits, one indoors and another immediately outside. Indoor collections included specimens resting on the walls. Extradomiciliary collections were performed on a single horse, on one human bait, and from the wooden surfaces of the stable (extradomiciliary station). The catches on human bait and on the horse were carried out twice a month, from 7:00 to 9:00 p.m. from November 1991 to December 1992. Mosquitoes were aspirated also twice a month from the walls of the house from 6:55 to 7:00 p.m., 7:55 to 8:00 p.m. and from 9:00 to 9:05 p.m., and on the walls of the stable from 9:05 to 9:15 p.m. Correlations between monthly abundance of *An. aquasalis* females in night catches and rainfall were analyzed by Spearman's rank-order coefficient (Siegel 1956).

Adult mosquitoes were also collected resting in vegetation and by light trap. Potential natural

Fig. 2: "Meia noite" farm domiciliary and extradomiciliary stations in Guaraí, State of Rio de Janeiro, Brazil.

resting places were searched in open fields outside 500 m of the domiciliary station. This search was conducted from 9:00 a.m. to 1:00 p.m. two or three times a month. Mosquitoes were collected with a manual aspirator, an aerial insect net (30 cm ring diameter), and a battery powered portable suction apparatus slightly modified from Natal and Marucci (1984).

One light trap (Falcão 1981, slightly modified by Aguiar et al. 1985) was operated for 19 hr in May and 5 hr in June, 1992 at one edge of the stable, while human bait collections were being conducted at the domiciliary station.

Mosquitoes were transported to the laboratory in cardboard cages and morphologically identified to species, using taxonomic keys (Forattini 1962).

The ovaries of approximately 10% of females caught in each month were dissected to estimate parity by the tracheolar skeining method of Detinova (1962). Only females whose ovaries were in stages I or II of Christophers (1911) were considered for parity determinations. The number of dissected females varied monthly according with the density. Chi-squared tests were used to compare the monthly frequency of parous *An. aquasalis* females with the mean annual frequency.

Additionally, the influence of moonlight on the abundance of *An. aquasalis* was evaluated by comparing night collections during full and new moons performed in February, March, May, August and October. The two-tailed sign test (Siegel 1956) was used for data analysis.

RESULTS

The mean monthly relative humidity in Guaraí ranged from 85.8% in October to 91.8% in April (Fig. 3a).

Most rain fell in Guaraí between September and January, although a small peak in rainfall was recorded in July (Fig. 3b). The rainiest month was January (124.0mm^3) and the driest were May (18.0mm^3) and June (2.0mm^3). The temperature ranged from a minimum of 9°C in August to a maximum of 30.5°C in December, and the monthly average ranged from 15.2°C in October to 22.6°C in December.

The tide (Fig. 3b) ranged from 0-18 cm minimum to 133-155 cm maximum; the average in the last five years was around 75 cm.

Anopheline fauna - Only three anopheline species were collected. The predominant species was *An. aquasalis* with 12,688 specimens (99.74%), followed by *An. albitarsis* (0.04%), and *An. darlingi* (0.02%) (Table I).

Seasonal distribution (Table II) - *An. aquasalis* was collected throughout the year, but in higher numbers from April through August. Its abundance diminished after September, coinciding with an increase in rainfall (Fig. 3b). No significant correlation between rainfall and mean *An. aquasalis* density was detected (r_s = -0.3379, P > 0.05 - d.f. = 10), although the species was clearly more common during dry and moderately rainy periods and less abundant during the hot and rainy summer.

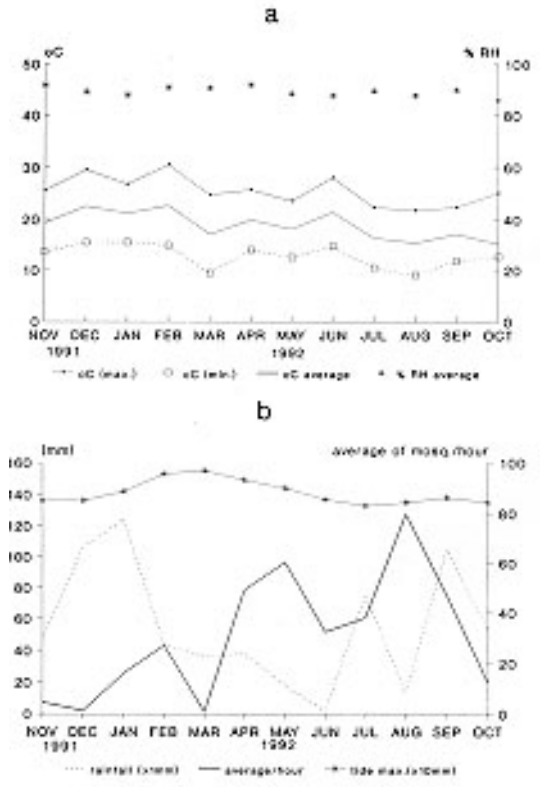

Fig. 3: a - Monthly variation of temperature (maximum, minimum and average), and relative humidity of the air. b - Monthly average (per hour) of *Anopheles aquasalis* females captured at the domiciliary (human bait indoors, outdoors and inner walls) and extradomiciliary stations (human and animal baits), and variation of rainfall and tide, in Guaraí, State of Rio de Janeiro, Brazil, November 1991 to October 1992.

The parous rate of *An. aquasalis* in June (Table III) was significantly higher than the annual rate (chi-squared = 91.29, $P < 0.05$). Other months were not significantly different from the annual rate ($P > 0.05$), except April when the proportion of nulliparous was significantly high (chi-squared = 4.344, $P < 0.05$)

The hourly averages of *An. aquasalis* females were of 34.46 ± 37.48 (SD) and 30.32 ± 28.45 (SD) during new and full moons respectively. There was no significant difference between the hourly abundances with and without moonlight ($P = 0.376$).

Only 56 *An. aquasalis* females and one male and two *An. albitarsis* males were collected in 24 hr of light trap operation in May and June 1992.

DISCUSSION

The two main seasons in southeastern Brazil are the hot and rainy summer occurring from September to January, and the dry fall and winter from February to September, when the lowest temperatures are usually recorded. In Guaraí, *An. aquasalis* was more abundant after the rainy season or during the moderately rainy period. Heavy rainfall may flush breeding sites, strand larvae and pupae, cause egg mortality and therefor, reduce the abundance of adults. By contrast, moderate rains sustain the breeding places and facilitate their connections to tidal waters. During the dry months of May, June and August, the tides still cause the Guaraí River to overflow, and surrounding breeding sites were maintained by brackish water.

Similar results were obtained by Galvão et al. (1942) studying a population of *An. aquasalis* from Pará in Amazonian Brazil, where this mosquito was more abundant between April and June when rainfall was moderate. Berti et al. (1993) found that *An. aquasalis* adults were not abundant in Guayana, Sucre State, Venezuela, during the heaviest rains. The opposite was observed by Senior-White (1952) in Trinidad, by Silvain and Pajot (1981) in Suriname, by Berti et al. (1993) in Santa Fé, Sucre State, Venezuela, by Lucena (1950) and Lourenço-de-Oliveira (1984), respectively, in northeastern and southeastern Brazil, where the abundance of *An. aquasalis* was highest in the rainy season.

The parous rate of *An. aquasalis* was relatively high in Guaraí, 46.8%, compared with the results of Panday (1973) in Suriname, where the parity rate of *An. aquasalis* was 46% at the end of July and beginning of August, but only 11% during the rest of the year. Berti et al. (1993) observed great differences in the incidence of parity of *An. aquasalis* in Santa Fé (82.5%) compared to nearly Guayana (37.4%), Venezuela.

The low yield of *An. aquasalis* in light-traps indicates that this anopheline is little attracted to this collection device.

No significant differences in abundance of *An. aquasalis* were detected between full and new moons in Guaraí, although, Senior-White (1951) reported that *An. aquasalis* activity was greater in the first 2 hr after sunset on moonlit than on moonless nights. On the other hand, Lourenço-de-Oliveira (1984) reported an average of 2.9 *An. aquasalis* per 10 hr of captures during nights with new moon, while collections at other moon phases were less abundant (1.6, crescent; 1.4 full and 1.0, waning). Collections of *An. nuneztovari*, a species beloging to the same Albimanus Section of *Nyssorhynchus* as *An. aquasalis*, were 1.86 times larger during moonlit nights in Venezuela than during those with no moon (Rubio-Palis 1992). Ribbands (1945) noted that *An. funestus* entered houses and took blood more often during moonlit nights.

The abundance of *An. aquasalis* with respect to moon phases as well as the variable parous rates need to be better evaluated throughout its territory with controls for variables, such as local landscape (vegetation, orography, availability of suitable resting-places), humidity and wind, generally disregarded by the authors.

TABLE I

Number of anophelines, females and males, collected per month, at the domiciliary (on human bait indoors and outdoors, and on the inner walls of a house) and extradomiciliary stations (on human and animal baits, on the internal wooden surfaces of the stable, and with light trap) and at natural resting places, in Guaraí, State of Rio de Janeiro, Brazil, from November 1991 to December 1992

		Anopheline species						
		An. aquasalis		*An. darlingi*		*An. albitarsis*		*An.*[a]
Year\Month		female	male	female	male	female	male	spp.
1991	November	60	-	-	-	-	-	1
	December	8	-	-	-	-	-	-
1992	January	323	1	-	-	-	-	2
	February	398	-	-	-	1	-	1
	March	235	-	2	-	-	-	1
	April	1,003	6	-	-	1	-	3
	May	1,114	16	-	-	-	-	2
	June	1,556	103	-	-	-	2	3
	July	944	186	-	-	-	-	3
	August	1,838	547	-	-	-	-	2
	September	2,213	652	-	-	-	-	4
	October	734	33	-	-	-	-	2
	November	417	200	-	-	1	-	1
	December	101	-	-	-	-	-	1
Total		10,944	1,744	2	-	3	2	26

a: non identifiable females

TABLE II

Monthly number of *Anopheles aquasalis* females captured at the domiciliary (on human baits indoors and outdoors, and on the inner walls) and extradomiciliary stations (on man and horse) in Guaraí, State of Rio de Janeiro, Brazil, from November 1991 to October 1992

Year/Month		Total collected	Hours spent with capture	Number of mosquito/ man-hour
1991	November	60	10.25	5.85
	December	8	6.16	1.21
1992	January	214	12.20	17.12
	February	349	12.25	28.49
	March	12	12.20	0.96
	April	674	12.25	55.02
	May	1,063	12.20	85.04
	June	459	12.33	27.23
	July	393	5.33	73.39
	August	1,029	12.25	84.00
	September	631	12.20	50.38
	October	196	10.20	18.67
Total		5,086	135.33	37.58

TABLE III

Monthly frequency of parous *Anopheles aquasalis* collected at the domiciliary and extradomiciliary stations in Guaraí, State of Rio de Janeiro, Brazil, from November 1991 to October 1992

		Number	Parous	
Year/Month		dissected	N°	%
1991	November	20	9	45.0
	December	4	3	75.0
1992	January	159	65	40.9
	February	188	79	42.0
	March	11	7	63.6
	April	271	99	36.5
	May	235	116	49.4
	June	166	136	81.9
	July	114	53	46.5
	August	266	113	42.5
	September	183	82	44.8
	October	58	22	37.9
Total		1,675	784	46.8

Our results show that *An. aquasalis* is the most abundant anopheline and the only potential malaria carrier in Guaraí. It is also the commonest potential vector in other coastal lowlands adjacent to Guanabara Bay where malaria outbreaks have been reported and *An. albitarsis* and *An. darlingi* have become increasingly rare in the past several decades (Santanna & Lourenço-de-Oliveira, pers. comm.). The risk of malaria outbreaks in "Baixada Fluminense", with the immigration of infected people from endemic areas of the Amazon, seems to be greater during the dry and moderate rainy seasons when *An. aquasalis* is more abundant especially in June when a high parity rate indicated extended survivourship.

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