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COLONIZATION AND LABORATORY MAINTENANCE OF *Anopheles albimanus* WIEDEMANN IN VENEZUELA(1)

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SUMMARY

Anopheles albimanus is one of the main vectors of malaria in Central America and the Caribbean, based on its importance, there are previous reports of the successful colonization of this species in Latin America countries. Mosquitoes were collected in the Aragua State of Venezuela colonized in the laboratory, using a simple and efficient maintenance method. Based on life table calculations under well established laboratory conditions, the Survival Rate Probability was constant and always close to 1 in immature stages, the Reproductive Net Rate (Ro) was 3.83, the generation time (Tc) was 24.5 days and the Intrinsic Growth Rate (rm) was 0.0558. This is the first report of the colonization of *A. albimanus* in Venezuela.

KEYWORDS: Colonization; *Anopheles albimanus*; Venezuela; Laboratory.

INTRODUCTION

Different mosquito colonies have been established worldwide in order to study the biological, ethological and ecological variables involved in dynamics of disease transmission, and to evaluate control measures against the parasites and their vectors (DAVIS, 1994). Some colonies of *Anopheles* are maintained by induced copulation and therefore are not fully adapted to laboratory conditions (ARRUDA et al., 1982, BURALLI & BERGO, 1988; KLEIN et al., 1990). However, some species and strains adapt fully, undergoing copulation, reproduction and colony replacement in the insectary; this is the case for *Anopheles gambiae, An. sacharovi, An. arabiensis, An. dirus, An. stephensi and An. freeborn* (KASAP & KASAP, 1983; VAUGHAN et al., 1994). Under such conditions, these species can also be evaluated for the susceptibility to *Plasmodium* infection.

Anopheles albimanus is an important vector of malaria in Central America (FLEMING, 1986) and was previously so in Venezuela (GABALDON, 1949). This species has therefore been studied and colonized in Central America by different researchers. ROZEBOON (1936) published the first report of laboratory breeding of *An. albimanus* while mass rearing methods were published by FORD & GREEN (1972), DAME et al. (1974) and BAILEY et al. (1980b), the latter providing a reliable technique for the rapid colonization of this species. In South America, *An. albimanus* has been colonized by CARRILLO et al. (1981) in Colombia and by GABALDON and ULLOA in Venezuela (ULLOA, personal communication). In 1993 we initiated the colonization of this *An. albimanus* and herein we present for the first time in Venezuela, its successful maintenance in the laboratory.

MATERIALS AND METHODS

Mosquito collections

Female mosquitoes were collected both from human baits or from the walls of a stable in the town of Magdaleno (10° 11'N, 67° 37'W) at the southeastern Valencia lake shore of the Aragua state. Mosquitoes were collected between 18:00 and 22:00 hours, for 4 days each month over a total of twelve months. Females were placed in 250 ml screen-topped glasses and maintained in a hermetic and humid icebox.

Female *An. albimanus* mosquitoes were fed in the laboratory on hamsters and domestic pigeons, and maintained in plastic cages of 15x27x30 cm. For oviposition a damp white tissue paper was laid in the bottom of the container. These containers were kept in an insectary of 3.5x3.0x2.7m, with natural light, maintaining a monthly average temperature of 27 ± 2°C and relative humidity of 80.5 ± 7.5 %. Eggs were placed in small (26x16x3 cm) plastic pans of water for incubation and hatching.

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First instar larvae were transferred to light plastic containers (33x21x6cm), approximately 200 larvae in each, with commercial mineral water from a deep well. Larvae were fed upon a dry food mixture of a fine powder of bovine liver, fish food, beer yeast and wheat germ. The powder was added to the water two or three times a day. Batches of 400 pupae were transferred with a dropper to plastic containers within the cages, in order to allow adult emergence. Adults were fed ad *libitum* on 20% (v/v) honey solution. To maintain humidity, cages were placed on a shelf covered with a wet tissue. Females were presented with a host for blood feeding from three days after eclosion.

Population studies

Fecundity and fertility were evaluated based on a modification of the BAILEY'S technique (1980a). Engorged females were placed individually in small plastic cups (60 ml), covered with gauze and provided with wet filter paper and a cotton pad. Fecundity, was estimated based on the average numbers of eggs laid by each female (ROCHSTEIN, 1975). Fertility was estimated from the numbers of eggs hatched per female. Population parameters for immature stages were calculated from age-specific life-tables (RABINOVICH, 1980; SERVICE, 1993), from a population of 400 larvae, divided in four cohorts of 100 individuals. Reproductive Potential, Reproductive Net Rate (Ro = Σlxmx) and Generation Time (Tc = Σxlxmx/Σlxmx) were estimated, based on the Survival Time (lx) of a cohort of 120 females using the equation mx = NHF/NHV (NHF= No. of fertile females, NHV = No. of live females at age x), where NHF was substituted by the mean of the eclosion divided by two and previously estimated as a constant 65,6/2=32.8. The Growth Intrinsic Rate (rm.) was based on these estimates and according to SOUTHWARD (SERVICE, 1993).

RESULTS

A very high mortality in the early collections was observed, such that the colony was initiated only from later collections From the 573 females *Anopheles albimanus* collected in the field, an average of 2818 individuals were obtained by generation (Table 1). The relative mortality per instar is uniform as shown in the life table (Table 2), and the population exhibits a Type I survival curve (Fig. 1) where the survival probability up to the end of the stage, is constant and close to 1. The sum instar longevity (17.4 days) exceeds the time total real time (23.4 days) taken for the immature phase, since the different stages are overlapped. The reproductive potential of this colony was estimated, demonstrating a Reproductive Net Rate (Ro) of 3.83, a Generation Time (Tc) of 24.5 days and an Intrinsic Growth Rate (rm) of 0.00558.

The fecundity (Fig. 2) fluctuates around a mean of 89.8 ± 5.5 eggs/female, after an unsteady decrease related to the parental generation. The fertility shows a dramatic descent in F1 and thereafter is stabilized around an eclosion mean of 66.4 ± 7.8 and a proportion of 0.69 eggs eclosioned per female.

DISCUSSION

The results demonstrate the complete adaptation of *Anopheles albimanus to* laboratory conditions. The pronounced diminution in fertility of the first generation, might be a consequence of the non-adaptation of the early generations. However, fertility values suggest that copulation occurred in some of these females, as observed by CARRILLO et al. (1981). ROZEBOON (1936) pointed out that *An. albimanus* copulates easily in small cages if humidity is sufficiently high, and in our case it was necessary to maintain

TABLE 1
Mortality larval and reproductive values of *A. albimanus* reared in the laboratory

Generation	Larval mortality	Estimated No. of emerging adults	Mean of eggs per female	Proportion of No. of eggs eclosioned	No. of larval in the next generation
P		573	122.3	0.95	13,550
F1	93.2	927	81.7	0.1	556
F2	33.1	372	80.6	0.38	3,845
F3	31.2	2.65	96.5	0.78	5,200
F4	50.2	2,588	97.4	0.76	5,300
F5	50.5	2,625	81.9	0.84	7,000
F6	37.2	4,399	80	0.88	7,000
F7	32.9	4,699	99.3	0.69	5,600
F8	50.9	2,752	96.3	0.58	7,750
F9	37.7	4,830	92.4	0.67	6,700
F10	31	4,593	91.7	0.62	6,400

X=2818

TABLE 2

Relative mortality per instar of *An. albimanus* which were reared in the laboratory

Instar	Age in days	Age in days at beginning of instar	No. of entering instar	Deaths in instar	Relative proportion dying in instar	Proportion dying daily in instar
I	2.7	0	100	1	0.01	0
II	2.3	2.7	99	3	0.03	0.01
III	6	5	96	1	0.01	0
IV	5.3	10.3	95	2	0.02	0
Pupa	7.1	17.4	93	9	0.1	0.01
Adult	14.7		84			

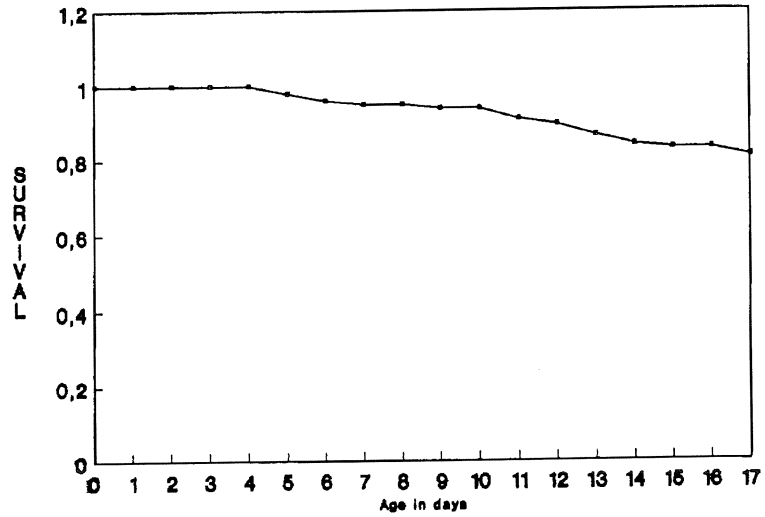

Fig. 1 - Survivorship curve for immature stages of *An. albimanus* maintained in the laboratory

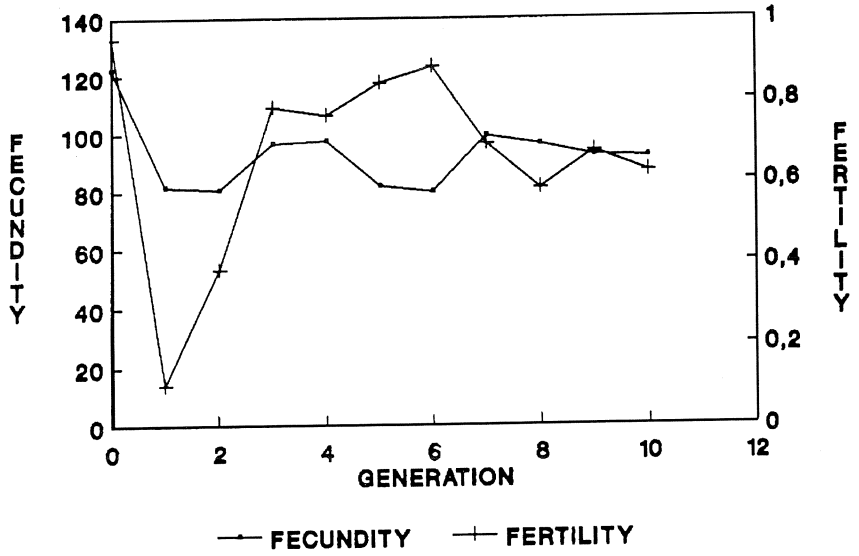

Fig. 2 - Mean values of fecundity and fertility of *An. albimanus* by generation

humidity above 80%. The reproductive potential of the Aragua colony is different from the reported by WEIDHASS et al. (1974) and BAILEY et al. (1979), who obtained an Intrinsic Growth rate increased rate for this species of 0.4-4.8x from the field and 34.8x from the laboratory, respectively. This is the first report of the colonization of *An. albimanus in* Venezuela and we provide a description of materials and simple methods used to initiate and maintain the colony.

RESUMO

Colonização e manutenção no laboratório de *Anopheles albimanus* Wiedemann na Venezuela

Anopheles albimanus é dos principais vetores da malária na América Central e Caribe, sendo a primeira vez que conseguimos sua colonização na Venezuela. Os mosquitos foram capturados no Estado Aragua e colonizados no laboratório através de um método simples e eficiente. Estimaram-se parâmetros populacionais usando tabelas de vida em condições laboratoriais bem controladas, observando-se probabilidade de sobrevivência constante próxima a 1 para os estadios imaturos. O potencial reprodutivo está representado por uma taxa reprodutiva (Ro) de 3,83, período entre gerações de 24,5 dias e uma taxa intrínseca de crescimento (rm) de 0,05.

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