BIONOMICS OF *ANOPHELES MINIMUS* AND ITS ROLE IN MALARIA TRANSMISSION IN THAILAND

S. Ratanatham, E.S. Upatham, C. Prasittisuk*, W. Rojanasunan, N. Theerasilp, A. Tremongkol and V. Viyanant

Department of Biology, Faculty of Science, Mahidol University, Bangkok, Thailand; and *Malaria Division, Department of Communicable Diseases Control, Ministry of Public Health, Bangkok, Thailand.

INTRODUCTION

Anopheles (Cellia) minimus Theobald, one of the main malaria vectors in Thailand, is prevalent in foothills with slow running streams in most parts of the country except in the south. In the first paper of this field research programme, Upatham *et al.*, (1988) reported on the bionomics of *An. maculatus* complex and their role in malaria transmission in Thailand. In this paper, we report on the bionomics of *An. minimus* and its role in malaria transmission in Pakchong district, Nakhon Ratchasima province, Thailand.

MATERIALS AND METHODS

Mosquito catching sites: Ban Moh Mai Khaen, located in village no. 5, Mou Si canton, Pakchong district, Nakhon Ratchasima province, upper central Thailand, was selected as mosquito catching sites, from January 1984 to June 1985. The area is mountainous and covered with forest. The village composed of 212 houses with 590 inhabitants. The main occupation of villagers is maize farming and forest reservation park labourers. This area has a long history of malaria since the beginning of the malaria eradication programme. Residual indoor spraying with DDT at 2 g/m^2 has been used since 1960.

Entomological techniques: Mosquito catchings on human and animal baits were conducted monthly between January 1984 and June 1985, from 1800 to 0600 hours with 10 min rest for each hour. Four humans, 2 indoors and 2 outdoors, and one cattle were used as baits to collect mosquitoes for 6 consecutive nights of each month studied.

The captured female anopheline mosquitoes were kept in mosquito cups marked with identification sites and hours of catching, and were held in a cooler at ambient temperature. In the following morning, they were identified and dissected to examine the tracheoles of ovaries to determine their age composition (Detinova, 1962). In parous specimens, the guts and glands were removed and examined for oocysts and sporozoites, respectively.

Based on the parous figures and field observation, the gonotrophic cycles of *An. minimus* were designated 2 days for rainy and summer seasons (March – November) and 3 days for cool season (December − February). Hence, the probability of daily survival was calculated using the formula $p = \sqrt[G]{P}$ (where p = probability of daily survival, G = gonotrophic cycle, and P = proportion of parity) (WHO, 1975). Subsequently, the life expectancy was determined using the formula $1/(1-\ln p)$ (Garrett−Jones and Grab, 1964).

Meteorological records: Temperature and relative humidity were continuously recorded by a standard automatic thermohygrograph placed in a screen. Daily precipitation and hourly wind velocity were recorded during the nightly mosquito bite collections.

RESULTS

Fig. 1 shows monthly densities of *An. minimus*, together with rainfall, air temperature and relative humidity at catching sites in Pakchong study area, from January 1984 to June 1985. The densities of *An. minimus* varied from month to month, with major peaks between September and November. In summer season (February to May), the density was low, with an average of less than 10 mosquitoes per night in both 1984 and 1985. In wet season (June to October), the density increased rapidly in July and reached its highest peak in October. In dry and cool season (November to January), the density was high in November and decreased so rapidly in December and January that the mosquitoes became very scarce in the following summer season. Statistical analysis in Table 1 shows significant positive correlations of the densities of *An. minimus* with monthly rainfall and relative humidity, but shows significant negative correlations with air temperature and wind velocity.

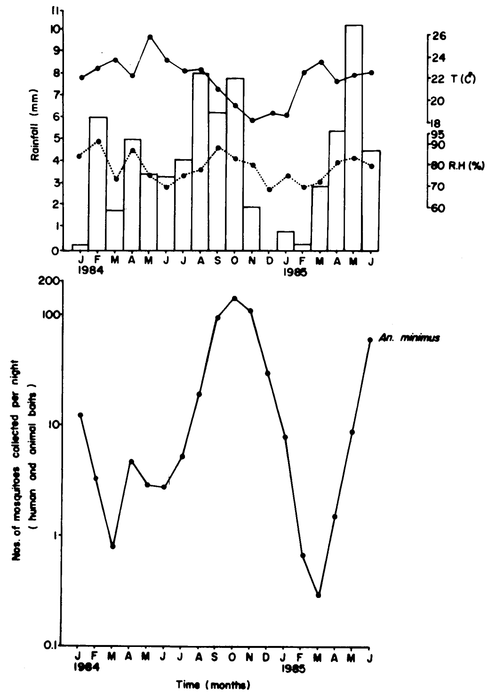

Fig. 1–Total numbers of *An. minimus* caught monthly on human and animal baits, together with rainfall, air temperature and relative humidity at catching sites in Pakchong study area.

Table 2 shows man-vector contact and host preference of *An. minimus* at Pakchong study area. The results indicated that *An. minimus* preferred to bite animal rather than human, that they tended to bite human more outdoors than indoors and thus indicating a tendency towards zoophilic and exophilic habits. The results also revealed that the ratio of *An. minimus* feeding on human and cattle was 1.0 : 2.6 (864 : 2,269). Their outdoor contact with human was higher than indoor (618 vs 249). The ratios of mosquitoes biting human indoor to human outdoor and to animal were 1.0 : 2.6 : 9.1 (249 : 618 : 2,269).

Biting cycles of *An. minimus* on human and cattle interpreted in percentages of total collections in summer (February to May), wet (June to October), and cool seasons

Table 1

Pearson correlation coefficients of the densities of *An. minimus* collected monthly with meteorological factors at Pakchong study area, from January 1984 to June 1985.

Meteorological factors	Correlation coefficients (r)	P-value	N (days)
Rainfall (daily)	−0.1234	0.102	108
Rainfall (monthly)	0.3271	0.007	108
Wind velocity	−0.5718	0.000	84
Air temperature	−0.4214	0.000	108
Relative humidity	0.2649	0.003	108

Table 2

Man-vector contact and host preference of *An. minimus* at Pakchong study area, from January 1984 to June 1985.

Catch (month)	Human (per man/night)		Animal	Total (per night)
	Indoor	Outdoor	(per bait/night)	
1984				
Jan	10* (0.8)**	20 (1.7)	49 (8.2)	79 (13.2)
Feb	4 (0.3)	5 (0.4)	12 (2.0)	21 (3.5)
Mar	1 (0.1)	1 (0.1)	3 (0.5)	5 (0.8)
Apr	2 (0.2)	8 (0.7)	20 (3.3)	30 (5.0)
May	1 (0.1)	1 (0.1)	15 (2.5)	17 (2.8)
June	3 (0.3)	1 (0.1)	12 (2.0)	16 (2.7)
July	2 (0.2)	8 (0.7)	22 (3.7)	32 (5.3)
Aug	11 (0.9)	47 (3.9)	102 (17.0)	160 (26.7)
Sept	40 (3.3)	107 (8.9)	409 (68.2)	556 (92.7)
Oct	68 (5.7)	189 (15.8)	606 (101.0)	863 (143.8)
Nov	14 (1.2)	45 (3.8)	622 (103.7)	681 (113.5)
Dec	8 (0.7)	36 (3.0)	135 (22.5)	179 (29.8)
1985				
Jan	2 (0.2)	7 (0.6)	39 (6.5)	48 (8.0)
Feb	0	0	4 (0.7)	4 (0.7)
Mar	1 (0.1)	0	1 (0.2)	2 (0.3)
Apr	0	0	9 (1.5)	9 (1.5)
May	6 (0.5)	15 (1.3)	35 (5.8)	56 (9.3)
June	76 (6.3)	128 (10.7)	174 (29.0)	378 (63.0)
Total	249 (1.2)	618 (2.9)	2,269 (21.0)	3,136 (29.0)

* = number of specimens collected for 6 nights,

** = number of specimens collected per man night or per bait night.

(November to January), from 18 00 to 06 00, are shown in Fig. 2. The biting cycles of *An. minimus* on human and animal differed considerably. On human, the mosquitoes were early biters. In summer and wet seasons, the biting peaks were between 21 00 and 22 00, with an additional peak at 05 00 during wet season. In cool season, the biting peak was at 18 00. On animal, the mosquitoes bit throughout the night, with no major peak in either wet or cool season, but with two peaks during the first and third quarter of the night in summer season.

Fig. 3 shows the indoor and outdoor biting cycles of *An. minimus* on human interpreted in percentages of total collections in summer, wet, and cool seasons, from 18 00 to 06 00.

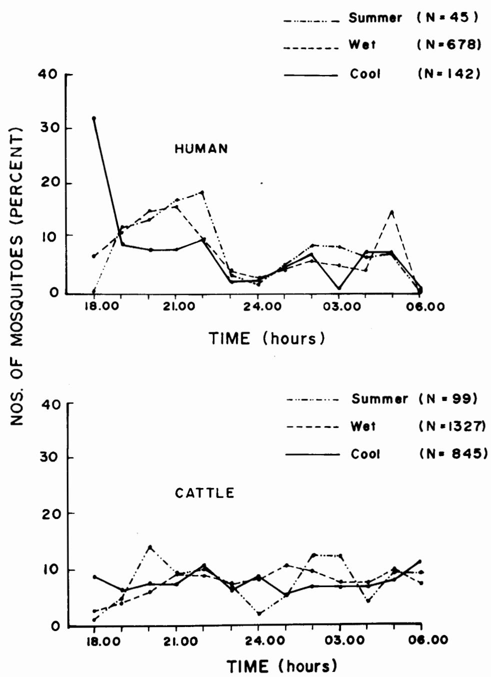

Fig. 2–Biting cycles of *An. minimus* on human and cattle interpreted in percentages of total collections in summer (February to May), wet (June to October), and cool seasons (November to January), from 1800 to 0600 h at catching sites in Pakchong study area.

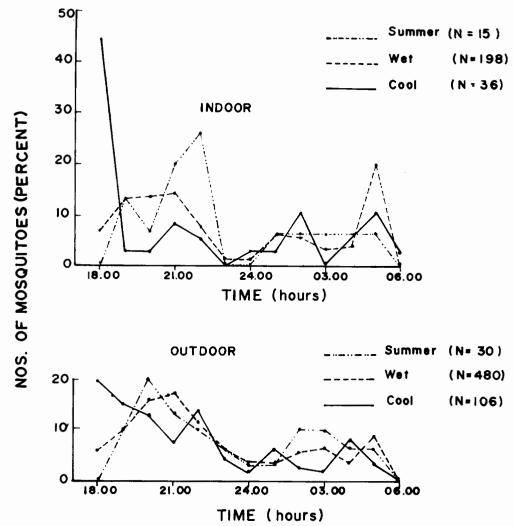

Fig. 3–Indoor and outdoor biting cycles of *An. minimus* on human interpreted in percentages of total collections in summer, wet, and cool seasons, from 18 00 to 06 00 h at catching sites in Pakchong study area.

The indoor biting patterns fluctuated throughout the night in all seasons, with biting peaks at 22 00, 21 00 and 05 00, and 18 00, respectively, for summer, wet, and cool seasons. The outdoor biting patterns were similar for all seasons, with intensive bites during the early hours of the night. The outdoor biting peaks were 20 00, 21 00, and 18 00, respectively, for summer, wet, and cool seasons.

The proportion of parity and life expectancy of *An. minimus* are shown in Table 3. The life expectancy ranged from 1.8 to 11.5 days. Attempts were made to compare life expectancy of *An. minimus* of different seasons using only monthly data which rendered more than 10 mosquitoes dissected for parous rate. The results indicated that the values of the life expectancy in summer, wet and cool seasons were 3.0 to 5.8, 2.7 to 7.0, and 6.1 to 11.5 days, respectively.

Parasitological data: Out of 1,518 guts of

Table 3

Proportion of parity and life expectancy (age composition) of *An. minimus* at Pakchong study area, from January 1984 to June 1985.

Time (month)	Proportion of parity	Life expectancy days
1984		
January	0.76 (72) *	10.9
February	0.37 (19)	3.0
March	0.40 (5)	2.2
April	0.62 (29)	4.2
May	0.71 (17)	5.8
June	0.75 (16)	7.0
July	0.55 (31)	3.3
August	0.48 (156)	2.7
September	0.73 (523)	6.4
October	0.59 (690)	3.8
November	0.72 (639)	6.1
December	0.72 (173)	9.1
1985		
January	0.77 (47)	11.5
February	1.00 (3)	–
March	0.50 (2)	2.9
April	0.33 (9)	1.8
May	0.65 (51)	4.6
June	0.61 (326)	4.1

* Age composition in parenthesis.

An. minimus dissected, 0.4% oocyst rate was recorded, and out of 1,560 salivary glands examined, there was no positive sporozoite detected. This low percentage of mosquito infection correlated with the few *Plasmodium falciparum* positive cases of blood survey carried out by the Anti-Malaria Programme in the study area.

DISCUSSION

At Pakchong study area where there was a cleared forested foothill, *An. minimus* was prevalent most of the year, with a major peak between September and November, and a small irregular peak between February and April 1984. The variations of density patterns of the summer months in 1984 and 1985 were probably due to the climatological differences.

Ismail *et al.*, (1974, 1975 and 1978) reported that *An. minimus* was prevalent in some foothill areas with two population peaks, at the beginning and the end of the rainy season. The authors found that there was only one major peak between September and November, during which high precipitation was recorded, and that the mosquito densities were still high at the beginning of the cool season.

The results of the present study showed that *An. minimus* bit both human and animal, with the animal biting densities significantly higher throughout the seasons of occurrence. In addition, the *An. minimus* mosquitoes bit human outdoor approximately 2.5 times higher than indoor. Ismail *et al.*, (1978) reported the outdoor biting as 5.8, 5.0 and 4.4 times higher than indoor biting in 1972, 1973, and 1974, respectively, at village no. 5, Salongphan canton, Muaklek district, Saraburi province. The results of the biting cycle on human showed the at *An. minimus* were early night biters, especially in the cool season, and were early morning biters in the wet season. Naturally, this situation increases the chances of man-vector contacts since those time periods are coincident with various activities of the people around their houses.

The longer duration of life expectancy of *An. minimus* in the cool season than other seasons, coupling with their early night biting habit point to the possibility that the mosquitoes possess a greater ability to transmit malarial parasites to human in the cool season than in other seasons. Rutledge and Gould (1969) reported that both *P. falciparum* and *P. vivax* infected subjects tended to be more infective during the cool season (November to January), and thus reflecting the seasonal trend in gametocytaemia.

The results of parasitological data showed very low rates of oocysts and no sporozoites from dissecting guts and salivary glands of the mosquitoes, respectively, during the 18–month study periods. This situation was coincident with the low natural malaria epidemiology owing to the Anti-Malaria Programme in the area. However, the exophagic behaviour of *An. minimus* and their early morning biting habit in the wet season may increase the chance of people becoming infected with malaria during the outbreak season.

An. minimus are one of the main malaria vectors in the forested hilly areas in most parts of Thailand (Ismail *et al.*, 1978). In Pakchong study area, *An. minimus* possessed characteristics of being a good malaria vector because they were prevalent in high numbers during the wet, and early dry and cool seasons. In addition, their life expectancy was longer in the dry and cool season than in other seasons. *An. dirus* are also known as another main malaria vector in the forested hilly areas. But, their prevalence was very low in number, only 0.5% (115), which supported the fact that the ecological conditions became unfavourable for the breeding of *An. dirus* and more favourable for *An. minimus*. Consequently, the *An. minimus* vector became more prevalent, and the *An. dirus* vector disappeared.

SUMMARY

The bionomics of *Anopheles minimus*, one of the main malaria vectors in Thailand, were conducted in Pakchong district, Nakhon Ratchasima province, from January 1984 to June 1985. The prevalence of *An. minimus* was influenced by monthly rainfall, relative humidity, temperature and wind velocity, with a major peak of density from September to November. *An. minimus* preferred to feed on animal rather than on human, tended to bite human more outdoors than indoors, and thus exhibiting zoophilic and exophilic behaviour. The biting activity of the mosquitoes on animal exhibited high densities throughout the night in all seasons, whereas on human they tended to be an early evening biter in the dry cool season, and early morning biter in the wet season, and thus increasing the chance of man-vector contact.

The life expectancy of *An. minimus* varied from month to month, ranging from 2.7 to 11.5 days, with the longest longevity during the dry cool season. The natural malaria infection rate of this species was very low. Out of 1,518 dissected guts, only 0.4% were found infected with oocysts. There were no sporozoites detected in the 1,560 dissected salivary glands.

ACKNOWLEDGEMENTS

The authors thank Dr. S. Phinichpongse, former Director of Malaria Division, Department of Communicable Disease Control, Ministry of Public Health, Bangkok, Thailand for his support and the entomological field staff to carry out this investigation. Special thanks to Mr. Weerapol Photichitti, Regional Entomologist and Mr. Sarayut Chaiaei, team leader, and his staff of the Entomological Section in Pakchong, Mr. Wanchad Sumanochitraporn, Mr. Sompob Boontim and Ms. Suchittra Jariyasesth who contributed a great deal to the success of this investigation. Appreciation also to Dr. Rauf Ali for his assistance in statistical analyses.

This investigation was supported by the United States National Academy of Sciences, Board on Science and Technology for International development Research Grants Program, Number MVR-TH-583-29.

REFERENCES

GARRETT-JONES, C. and GRAB, B., (1964). The assessment of insecticidal impact on the malaria mosquito's vectorial capacity, from data on the proportion of parous females. *Bull. WHO., 31* : 71.

ISMAIL, I.A.H., NOTANANDA, V. and SCHEPENS, J., (1974). Studies on malaria and responses of *Anopheles balabacensis balabacensis* and *Anopheles minimus* to DDT residual spraying in Thailand. I. Pre-spraying observations. *Acta Trop., 31* : 129.

ISMAIL, I.A.H., NOTANANDA, V. and SCHEPENS, J., (1975). Studies on malaria and responses of *Anopheles balabacensis balabacensis* and *Anopheles minimus* to DDT residual spraying in Thailand. II. Post-spraying observations. *Acta Trop., 32* : 206.

ISMAIL, I.A.H., PHINICHPONGSE, S. and BOONRASRI, P., (1978). Responses of *Anopheles minimus* to DDT residual spraying in a cleared forested foothill area in central Thailand. *Acta Trop., 35* : 69.

RUTLEDGE, L.G. and GOULD, D.J., (1969). Factors affecting the infection of anophelines with human malaria in Thailand. *Trans. Roy. Soc. Trop. Med. Hyg., 63* : 613.

WHO., (1975). Manual on practical entomology in malaria. II. Methods and techniques. *WHO Offset Publ. No. 13* : 121.