## Epidemio-entomological survey of Plasmodium vivax in Korea\*

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#### Introduction

Malaria due to Plasmodium vivax is one of the most important and widely spread protozoan diseases in Korea, Until World War II, malaria was popularly known as "Haru-geori" or "Chare-Bae", and recognized as one of the unavoidable diseases in summer months. During the period from 1940 to 1955, the movement of population and unsanitary conditions caused by the War increased the prevalence of Korean vivax malaria infections to such an extent that it became a major public health problem of nationwide significance. After Korean War, the epidemiological, clinical, and therapeutic studies on the Korean vivax malaria have been carried out extensively by many investigators, recognizing the urgency of the problem. From malaria pre-eradication survey on Korean vivax malaria, ROK-WHO malaria project (1960) reported that a total of 22,005 children, 1-14 year of age, were found to have an average spleen rate of 1.1% in a range of 0.0-3.8%.

According to parasitological survey findings, of 16, 498 blood samples, 17 cases of *vivax* malaria were microscopically confirmed. Ree et al.(1967) studied on natural infection of *P. vivax* in Anopheline mosquitoes in Korea, and reported that of 7,517 dissections of *A. sinensis*, only one was found to be naturally infected with the sporozoite.

After the beginning of the New Community project

in the third "Five-year Economic Development Plan" in 1972, the Korean Government established a plan to expand the land by cultivating hilly areas, thus practicing land reclamation, and also accompanied the establishment of an irrigation system due to dam construction, reformation of land, improvement of agricultural technique, and intense use of agricultural chemicals. However, increased rice production inevitably resulted in the expansion of mosquito larval habitats, and introduced important changes in the agroecosystems which determined the distribution and abundance of mosquitoes(Surtees, 1970; Mogi, 1984; Baik and Joo, 1991). Furthermore, rice fields also produced important vectors of human malaria and other viral diseases. Recently Joo and Wada (1985) conducted the seasonal prevalence and population density ofvector mosquitoes in Kyungpook Province, and Ahn et al.(1983) reported on 51 cases of malaria imported in Korea during 1970-1982, of which 37 cases were detected in the home country.

Every year only a few cases of malaria in Korea have been noted, although the infective cases of malaria are said to have disappeared in Korea after 1980, it was considered to be due to lack of attention given to the problem of mosquito borne diseases.

Kyungpook Province is in the south-eastern part of Korea, and some endemic foci of malaria in northern counties of the Province were found during a malaria survey in 1960-1962.

The present study attempts to estimate the

<sup>\*</sup> The results of this study were presented at the Spring meeting of the Korean Society for Parasitology(1991).

population dynamics of vector mosquitoes in Kyungpook Province from 1987 to 1991, and is to observe the present status of Korean *vivax* malaria infections among residents.

## Geographical conditions of surveyed areas

Kyungpook Province is situated in the southeast part of the Korean peninsula, having an area of 19,700 square kilometers. Three areas in the Province were selected as vector mosquito survey stations(Fig. 1).

Keimyung University training farm in Kyungsan county was the main study area. It is located about 15 kilometers E. S. E. of Taegu city, situated on a low hill studded with copse, orchards, rice fields and small swamps. The farm covers an area of approximately 250,000 square meters and consists of pasture, animal shelters, vinyl houses, and human dwellings. Large numbers of cattle, pig, fowls, dogs, sheep and deer are raised on this farm but there are no wild animals which could be important as hosts of vector mosquitoes. Another area selected for this survey station was the Agricultural research farm in Kyungpook Provinical office of Rural development. It is located in Dongho village, a northern district of Taegu city. The total area of rice field in this survey station was about 150,000 square meters. The north and west corners of the rice field contain the house-dwellings and the east boderline is limited in the foothills of a low mountain range.

The rice field irrigation begins in May and ends in late August or early September: during this period the rice fields form the main breeding places for the vector mosquitoes. The third area selected for this survey station was Shincheon-ri in Anjung-myun, Yeongpung county. It is a rural village of about 300 residents and situated along the bank of Namweon stream, one of the large tributaries of the Naktong river. It is about 170 kilometers by road from Taegu city and lies in the hilly and mountainous area of the northern part of Kyungpook Province at 37 degree north latitude. The average elevation is about 170 meters above the sea level. Most of the housedwellings lie beside the road, which runs north and

south through the village, and a few houses are scattered about in the surrounding rice fidlds.

Meteorological Data: Meteorological data for the period of the present survey was provided by the Taegu branch of the Korea Meteorological Agency. These data are shown in Table 1.

The study areas are under the influence of a typical continental climate of an eastern coast affected by both high atmospheric pressure from the cold continent and a low one from the Pacific Ocean in the summer season. Therefore, seasonal fluctuation of air temperature and precipitation, which is of fundamental importance to understand the dynamics of mosquito populations, is very great.

#### Materials and Methods

Light trap operation: In order to observe the seasonal prevalence of the vector mosquito populations, light trap collections were performed as follows: A light trap was fixed 1.5m above the ground at trapping spots, the piggery A, the cow-shed B, and the house-dwelling C, and operated from dusk to dawn on one-night per week schedules. Mosquito collected at each station were counted by species.

Indices of mosquito abundance: In order to compare the annual abundance of *A. sinensis*, mean per cent index(MPI) which was proposed by Maeda et al.(1978), was used.

Human baited trap: In order to determine the relative numbers and species of mosquito which were attracted by human beings, human baited trap were performed as follows: A man was allowed to lie on the floor of a tent 2.6×2.0m and 1.5m in height. An open window 2.0×1.5m permitted entry of mosquitoes. All mosquitoes biting or attempting to bite were collected between 19:00 and 06:00 hours on one night in July and August in 1990.

Dissection of mosquito: All the mosquitoes biting or attempting to bite were collected either on the skin with a sucking tube or with on insect net, and killed with ethyl ether. They were transferred into a glass tube and kept in an ice box until they were individually identified and dissected in the la-

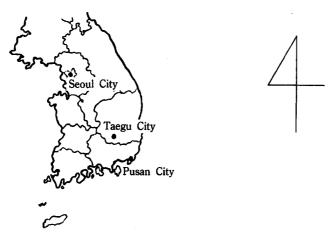




Fig. 1. Surveyed areas(●) in Kyungpook Province, Korea.

boratory. Dissection of the mosquito specimens were made usually the next morning. After each mosquito was identified and numbers recorded, it was transferred on a slide glass with a drop of 0.6% saline solution. They were examined for the determination of the ovarian age using the methods reported by Detinova(1962). Records were made for each mosquito whether it was nulliparous or parous, and if parous the number of follicular relics was determined. After this all other body parts, especially salivary glands, were examined for malarial infections.

Collection of resting mosquitoes: In order to determine the resting places of mosquitoes in day-time, oral aspirators, and hand nets, about 40 Cm in diameter, made of fine mosquito netting were used to catch adult mosquitoes resting in human and animal shelters. All the mosquito specimens were individually examined for species under a binocular dissecting microscope and counted.

Collection of larvae: In order to estimate the species and density of mosquito larvae and pupae, 30 fixed rice fields were dipped from April to October at one week intervals in 1989-1991. The dipper was 15 Cm in diameter and 5 Cm in depth with a wooden handle of 60 Cm in length. At the outset a collector stood at a point on the side of rice field, and took a dip on the water surface, which was thought to be most favorable for the breeding of the larvae and pupae within the reach of the dipper.

In each rice field, the dipping was made ten times which was thought to be necessary to determine the distribution pattern of the numbers of mosquito larvae in a rice field(Wada et al. 1971). The total number of mosquito larvae in the study areas were estimated according to the methods described by Wada and Mogi(1974).

Insecticide susceptibility test: In order to estimate insecticide resistance on A. sinensis, blood-fed females of A. sinensis were collected from human and animal shelters with an insect net or with a sucking tube, and transferred into the cage.

The mosquitoes were allowed to oviposit in an insectary at  $30\pm1$  C and 70-80% relative humidity with 16 hours of illumination per day. The approxi-

mately 300 first instar were reared in enamel pans measuring 50×40Cm filled to 2Cm depth of water and fed on crushed powders of laboratory mouse pellets and adults were fed on 5% sugar solution.

Insecticide resistance was modified from those described by Yasutomi et al.(1986). Toxicities of the organophosphorus were determined with fourth instar.  $LC_{50}$  values were calculated from the average of two replicates. From these data the regression of the probit mortality on log dosage was computed and the  $LC_{50}$  were obtained.

Parasitological survey: Thick and thin blood films were made for the determination of the incidence and densities of species of malaria parasites among residents aged from 1 to 70 living in northern parts of Kyungpook Province. Blood films were fixed in methanol and treated with 4% Giemsa stain for 30 minutes. Thick film fields were examined microscopically(1,000x) for the presence of malarial parasites. Parasite densities were recorded as the number of parasite per 200 white blood cell.

#### Results

The five years' observation of the earlist dates in which *A. sinensis* begin to be collected by light traps and air temperature and humidity at that time in Kyungsan county, Kyungpook Province, Korea is presented in Table 2.

A. sinensis were first collected in the light traps from 1987 to 1991 in April, between the 2nd and 20th days, but on March 15 in 1989. At that time the air temperature ranged from 2.4-24.3C and humidity from 45-61 per cent. The average number of A. sinensis per trap-night was from 0.3 to 1.0.

Table 3 lists the dates of peak population of A. sinensis collected in the surveyed area and the meteorological data at that time. The highest population density of A. sinensis was observed during the period from early July to mid-July in every year.

The air temperature was between 18.6 and 34.8 C, and the relative humidity from 59 to 85 per cent. The maximum number of *A. sinensis* in 1987 was 1,247 per trap-night. In 1988 the number decrea-

Table 1. Monthly maximum and minimum temperature, relative humidity, and total precipitation reported by regional Meteorological Center in Taegu, Korea during period from 1987 to 1991

		1987	1988	1989	1990	1991
Jan.	T*:	-10.7-11.7	- 9.2-11.6	- 9.4-12.5	-11.5-10.8	- 8.6-12.0
	H**:	14 - 58	19-54	23 - 66	39 - 56	36 - 94
	P***:	44.4	12.6	110.7	22.3	20.6
Feb.	T:	- 9.9-19.6	-9.9-14.1	- 5.9-17.1	- 2.3-22.1	-12.4-11.0
	H:	20 - 58	19 - 56	22 - 64	38 - 58	37 - 93
	P:	43.9	2.9	90.5	45.3	44.4
Mar.	T:	- 4.3-20.3	- 5.0-19.5	- 3.9-22.1	- 3.8-12.9	- 2.6-24.7
	H:	24 - 59	21 - 58	19-60	52 - 70	32 - 91
	P:	51.4	45.2	100.4	85.5	79.3
Apr.	T:	0.9 - 27.9	3.7 - 30.4	3.5 - 28.2	2.7 - 27.3	- 2.7-27.3
	H:	19 - 56	19 - 53	19 - 56	38 - 58	33-81
	P:	42.7	49.6	34.1	90.5	93.2
May	T:	7.2 - 30.8	7.7 - 32.0	9.5 - 32.7	8.9 - 30.8	6.7 - 34.1
	H:	19 - 63	17 - 60	24 - 64	44 - 65	29-83
	P:	62.3	64.1	46.3	143.1	21.9
Jun.	T:	10.6 - 36.3	14.4 – 35.1	12.6 - 32.9	10.0 - 33.0	14.8-33.8
	H:	21 - 66	26 - 71	25 - 70	53 - 70	52-86
	P:	138.7	85.9	103.3	200.5	104.7
Jul.	T:	18.0 - 34.8	15.4 – 38.0	15.2 – 35.5	10.0 - 35.3	19.0 – 35.0
	H:	33 - 77	43-79	23 - 79	66 - 75	63-89
	P:	275.7	215.1	306.6	251.3	425.3
Aug.	T:	18.7 - 32.5	16.1 - 36.5	18.5 – 35.4	13.1 - 38.5	17.5 – 33.0
	H:	49 - 82	42 - 76	35 - 72	57 <del>- 7</del> 0	63 - 86
	P:	327.2	74.5	149.8	125,9	282.5
Sep.	T:	8.9-31.5	11.7 - 32.3	11.8 – 30.1	11.5 - 33.2	13.2 - 32.0
	H:	24 - 71	36 - 74	28 - 77	54 - 74	59-89
	P:	11.0	52.7	196.4	197.0	154.1
Oct.	T:	2.5 - 29.9	2.1 - 28.0	1.6 - 25.9	4.2 - 26.6	4.8 - 26.9
	H:	21 - 64	22 - 61	19-66	53 - 69	47 - 72
	P:	44.6	3.9	18.8	13.9	1.3
Nov.	T:	- 5.0-22.2	- 1.7-21.7	- 2.1-21.3	- 1.3-23.7	- 3.1-20.7
	H:	15-63	19 - 53	16-64	39-56	16-59
	P:	51.8	3.4	61.9	43.5	13.4
Dec.	T:	- 8.8-20.1	-8.0-17.4	- 5.1-19.3	-9.6-19.0	- 6.8-17.0
	H:	17-56	23 - 59	10-64	42 - 81	17 - 64
	P:	0.4	7.1	28.6	2.1	55.5

<sup>\*</sup> T: temperature(C)

\*\* H: relative humidity(%)

\*\*\* P: precipitation(mm)

sed to 1,109.3, and in 1989 the number abruptly increased to 3,495, in 1990 the number decreased again to 518.3.

Table 4 shows the dates *A. sinensis* were not collected in the surveyed area according to the years studied. *A. sinensis* was not observed during the period from late October to mid-November. The air temperature at that time ranged from 2.6-24.6 C and humidity from 39 to 70 per cent.

The seasonal prevalence of *A. sinensis* collected by light traps are summarized in Table 5. In general, *A. sinensis* was collected in 8 months, from April to November in every year studied, but 9 months in 1989. In 1988, the average number of female *A. sinensis* per trap-night in April was 0.1, it increased to 0.7 in May, 128.8 in June, and reached the maximum number, 537.6 in July. In August, the average number decreased to 74.3, in September 60.6, in October 2.4, and in November 0.1. The general patterns of seasonal prevalence of *A. sinensis* in the other years are similar to those for 1988, but the pattern

of monthly change in 1989 is dissimilar to those for 1988. In 1989 *A. sinensis* first appeared in mid-March, it subsequently increased and reached a maximum of 1,331.4 in July. In August the number abruptly decreased to 48.6, and in September the number increased again to 121.1. There was a gradual decrease from October, with a very small number of them until mid-November in the traps.

The results of relative abundance and MPI calculation for *A. sinensis* in successive years after 1987 in correlation with the incidence of Korean *vivax* malaria are shown in Table 6 and illustrated by Fig. 2. It was found that the total number of *A. sinensis* progressively increased during the initial three years from 1987 to 1989. A marked decrease in MPI was obtained in 1990 and 1991.

Table 7 shows the results of the number of total and engorged female *A. sinensis* by light traps collected at three locations during 5 years from 1987 to 1991. The overall rate of engorgement, as calculated by dividing the number engorged with the total,

Table 2. Five years' obser	rvations of the earliest da	ates Anopheles sinensis	begin to appear in	Kyungsan county,
Kyungpook Provi	ince, Korea, together witl	h meteorological data		

Year	Earliest date when mosquito appeared		Temperature (Range °C)	Humidity (%)	Averge No./ trap-night	
1987	April	2	3.3-16.1	55	0.3(1/3)*	
1988	April 2	20	5.9 - 13.0	61	0.3(1/3)	
1989	March 1	15	4.8 - 16.1	46	0.3(1/3)	
1990	April 1	1	7.7 - 24.3	52	1.0(3/3)	
1991	April	3	2.4 - 19.6	45	0.3(1/3)	

<sup>\*</sup> Number in parentheses means the total number of female mosquitoes per traps.

Table 3. Dates of peak population of Anopheles sinensis and the meteorological data at that time

Yeat	Dates of peak population	Temperature (Range $^{\circ}$ C)	Humidity (%)	Average No./ trap-night
1987	July 9	21.7-34.8	59	1,247.0( 3,741/3)*
1988	July 6	20.5 - 24.6	70	1,109.3( 3,328.3)
1989	July 5	18.6 - 28.4	69	3,495.0(10,485/3)
1990	July 12	24.3 - 31.2	76	518.3( 1,555/3)
1991	July 18	20.2 - 23.0	85	730.7( 2,192/3)

<sup>\*</sup> Number in parentheses means the total number of female mosquito per trap-night.

Table 4. Dates of disappearance of *Anopheles sinensis* and the meteorological data

Year	Date of disappe of mosqui		Temperature (Range °C)	Humidity (%)
1987	November	19	2.6-13.0	39
1988	November	9	5.6 - 21.7	60
1989	November	15	4.7 - 14.2	64
1990	October	21	9.1 - 24.6	70
1991	October	31	9.1 – 19.3	64

which reflect the efficiency of blood-sucking activity on *A. sinensis* in 1990 were in the order of 62.7 per cent on cowstall, 43.7 per cent on piggery, and 1.3 per cent on human dwelling. The general patterns of engorgement rates in the other years are similar to those for 1990, but in 1989 are dissimilar.

The monthly fluctuation in the blood-sucking rate of A. sinensis is listed in Table 8. The blood-suc-

king rate varied greatly by different month in every year. The rate was 10.5-44.8 per cent in June, 8.3-58.7 percent in July, 12.9-42.5 in August, and 9.2-48.3 per cent in September, respectively, although the rate decreased on October, it was kept on the level of 5.0 per cent.

The data shown in Table 9 represents the biting rhythm of *A. sinensis* by light trap collections in a pigsty and on human baits through the night at interval of one hour. The biting activity was continued throughout the night.

In light trap collection in a pigsty, *A. sinensis* attempted to feed from 19:00-20:00 onward, and the peak numbers of the mosquito showed two peaks, one between 20:00-21:00 hour and another between 03: 00-04:00 hour in July and 04:00-05:00 hour in August. In human baited trap the peak hour of biting differed each month, for example between 24:00-01:00 hour in July 22:00-23:00 hour in August, when the tempe-

Table 5. Seasonal prevalence of *Anopheles sinensis* by the average number collected each traps during 5 years

V			Ave	erage num	ber of fem	nale mosqui	to per trap-nigh	nt	
Year	March	April	May	June	July	August	September	October	November
1987	0	0.1	0	108.2	542,6	50.8	18.1	0.4	0.1
1988	0	0.1	0.7	128.8	537.6	74.3	60.6	2.4	0.1
1989	0.2	0.7	2.5	401.0	1331.4	48.6	121.1	4.2	0.1
1990	0	0.6	1.9	89.4	271.9	180.8	84.3	0.2	0
1991	0	0.3	1.3	151.3	372.1	44.1	73.0	6.3	0

Table 6. Relative abundance of Anopheles sinensis population in successive years after 1987 in correlation with incidence of malaria

Vace	At the 3-station suburban		. Ma	Malaria	
Year —	Total No. collected	MPI*	NO. cases**	Incidence(%)	
1987	10.242	100.0	3	0.007	
1988	11,383	125.1	8	0.019	
1989	21,241	175.2	1	0.002	
1990	7,248	71.2	-	_	
1991	6,591	67.8	_	_	

<sup>\*</sup> MPI means Mean per cent index.

<sup>\*\*</sup> This cases are based on the reports from Cities and Provinces under Communicable Disease Control Laws.

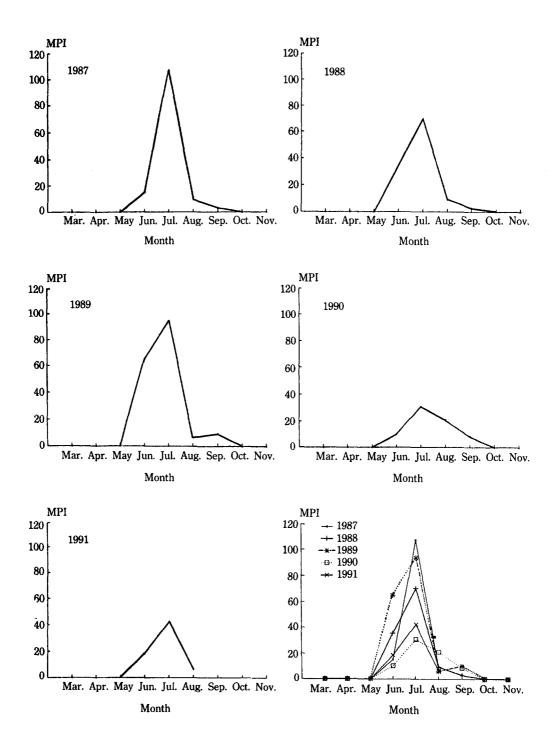


Fig. 2. Annual prevalence of *Anopheles sinensis* as shown in MPI caculated from the data of mosquito collection at 3 stations.

rature was between 26.9 and 29.3 C and the humidity 67-76 per cent in the field.

The age compositions of *A. sinensis* collected by human baited trap are compared with that of a pigsty by pig bait and listed in Table 10. The frequency distribution of the *A. sinensis* by the number of follicular relics counted after their dissection, and parous rates of both biting collections were relatively low, being found in 29.9 per cent in human bait and in 51.3 per cent in pig bait, respectively.

The positive rate for the sporozoite of *P. vivax* in the mosquito specimens reported in 1967 was compared with the results in 1990 and tabulated in

**Table 7.** Comparison of total and engorged number of *Anopheles sinensis* collected by light traps at three locations during five years, 1987-1991

Year	Location	No. collected	No. engorged(%)
1987	A**	3,492	1,034(29.6)*
	В	5,206	2,016(38.7)
	С	1,544	90(5.8)
	Subtotal	10,242	3,140(30.7)
1988	Α	4,632	535(11.6)
	В	4,274	1,540(26.0)
	С	2,477	41( 1.7)
	Subtotal	11,383	2,116(18.6)
1989	Α	4,258	1,384(32.5)
	В	15,286	940(6.1)
	С	1,697	55( 3.2)
	Subtotal	21,241	2,379(11.2)
1990	A	2,847	1,243(43.7)
	В	3,355	2,103(62.7)
	С	1,046	14( 1.3)
	Subtotal	7,248	3,360(46.4)
1991	Α	3,276	1,544(47.1)
	В	3,121	2,004(64.2)
	С	1,163	9(0.8)
	Subtotal	7,560	3,557(47.1)

Number in parentheses means the percentage of engorged females.

Table 11.

In 1967 the sporozoite of *P. vivax* were found in one specimen of *A. sinensis*, whereas no case was found in all of the specimens collected in 1990.

The age structure of immature stages of A. sinensis and the seasonal prevalence of total number of larvae plus pupae in the study area are shown in Table 12. The highest larval density in cultivated fields was  $21,226\times10^3$  on July 6th, 1990. After late September, such densities show a marked decrease and the larvae and/or pupae of A. sinensis were rarely found until rice plants were harvested. Factually, the general pattern of the seasonal prevalence in the study area was determined largely by the prevalence in cultivated fields. The total number reached its yearly peak during the period from late June to mid-July.

Table 13 shows the insecticide resistance levels of larval *A. sinensis* for one generation in the laboratory from collections in Kyungsan county and compared with those of susceptible laboratory strains.

The larval *A. sinensis* were highly resistant to malathion and diazinon, the LC<sub>50</sub> values being 14.50 ppm for malathion and 12.35 ppm for diazinon, but only low resistance was indicated in larval *A. sinensis*, LC<sub>50</sub> value being 0.085 ppm for fenthion.

The  $LC_{50}$  values to malathion and fenitrothion were about 4.12 and 3.31 times as high as those of susceptible strain.

The prevalence of *P. vivax* among the residents by sex and age groups are summarized in Table 14. A total of 1,546 residents were examined, no case was found to be infected with *P. vivax*.

## Discussion

Since Ross' description of the malaria-mosquito theory in 1897, the epidemiological, clinical, therapeutic, and entomological studies on the subject of malaria and its vector mosquitoes have been conducted by many investigators of Korea and other countries. The fact that patients with malaria are present among the residents in Korea have been known for a long time, and a disease called "Harugeori" (every otherday fever attack) or "Chare-bae" (enlarged sp-

<sup>\*\*</sup> Location A: Piggery, B: Cow-stall, C: House dwelling

Table 8. Monthly fluctuation of total and engorged number of Anopheles sinensis collected by light trap(1987-1991)

3.6 (1)	1	987	19	988	1	989	19	990	19	991
Month	No. colle- cted	No. engor- ged(%)								
Mar.	0	0	0	0	2	0	0	0	0	0
Apr.	1	0	1	0	7	0	5	2 (40.0)	3	0
May	0	0	11	0	38	7 (18.4)	23	3 (13.0)	19	2 (10.5)
Jun.	1,298	136 (10.5)	3,090	336 (10.9)	4,812	878 (18.2)	1,073	316 (29.5)	1,815	813 (44.8)
Jul.	8,112	2,886 (35.6)	6,451	1,510 (23.4)	14,645	1,198 ( 8.2)	3,268	1,917 (58.7)	4,093	2,297 (56.1)
Aug.	609	98 (16.1)	892	115 (12.9)	596	140 (23.5)	1,870	795 (42.5)	661	148 (22.4)
Sep.	217	20 ( 9.2)	909	155 (17.1)	1,090	154 (14.1)	1,012	489 (48.3)	876	290 (33.1)
Oct.	4	0	29	0	50	2 (4.0)	2	0	93	7 (7.5)
Nov.	1	6	0	0	1	0	0	0	0	0

Table 9. The results of overnight Anophels sinensis collection by light trap in a pigsty and on human baits(1990)

T.T	July 27-	28	August 1	3-14	A	ugust 30	0-31
Hour	Light trap	Man	Light trap	Man	Light	trap	Man
19:00-20:00	10( 4)*	0	13( 3)	0	22(	2)	0
20:00-21:00	50(7)	0	136(28)	0	123(	75)	1
21:00-22:00	40(10)	3(1)	38(11)	3	29(	12)	1
22:00-23:00	17(8)	0	19(8)	6(2)	13(	4)	2
23:00-24:00	8(2)	1	15(4)	2	4(	1)	1
24:00-25:00	10(2)	10	10(3)	1	8(	3)	3
01:00-02:00	14(6)	3	8(1)	3	4		3
02:00-03:00	11(2)	0	8(1)	3	2		2
03:00-04:00	18(5)	5	9(1)	5	3		0
04:00-05:00	7(1)	3	13(1)	3	12(	5)	0
05:00-06:00	0	0	9(3)	0	8(	3)	0
Total	185(47)	25(1)	278(63)	25(2)	228(	105)	16
Temperature(℃)	33.0	0-26.1	3	0-80		30.8	3-25.0
Humidity(%)	4:	3-89	3	0-80		52	2-88

<sup>\*</sup> Number in parentheses means number of engorged female mosquitoes.

Table 10. Age composition of *Anopheles sinensis* collected in 1990 as determined by the number of follicular relics

Bait	No. dissected	Freque th		stribut of reli		percent
		N*	1	2	3	– parous (%)
Human Pig	67 413	47 201	16 145	4 59	8	29.9 51.3

<sup>\*</sup> N means nulliparous.

leen) in the past is now accepted to be nothing but malaria. In the studies on malaria in Korea, little work was done until World War II, although malaria has been recognized as one of the unavoidable diseases in the summer months in Korea and A. sinensis was thought to be the main vector.

In 1959, the Ministry of Health initiated a prevention and eradication program against malaria in Korea with the technical support of the World Health Organization. During 1961 a total of 13,000 reports with corresponding blood smears were examined at the Central Malaria Eradication Service of the Ministry of Health and Social Affairs, of these 5,206 posi-

tive *P. vivax* cases were confirmed(Paik and Tsai, 1963). Throughout 1962 to the end of October, a total of 6,369 blood smears were examined, and 2,575 positive cases were confirmed.

A study of Paik and Tsai(1963) reported that the number of positive P. vivax cases during the period from 1961 to 1962 found in the northeastern area including the four Provinces of Kyungpook, Kangweon, Kyunggi and Chungpook comprised 97.3 per cent of all positives. They also commented that almost all localities in the southwestern(plain) area were non-active and retrogressive in malaria occurrence, where malaria transmission was low. In contrast the northeastern (mountainous) area had a rather large number of residual foci where malaria transmission was high. Whang(1963) carried out a blood survey on narcotic addicts in asylums in Korea from 1959 to 1962, and reported that of 3,991 smears collected, 13 cases were found to be positive for P. vivax, 23 positive for P. falciparum, and 1 positive for P. malariae. He also commented that P. malariae and P. falciparum in Korea were not natural infections but resulted through contaminated syringes co-

Table 11. Comparison of demonstration for sporozoite of P. vivax in vector mosquitoes in 1967 and 1990

	<b>5</b>	Ree et	al.(1967)	Presen	t survey	
Locality	Bait	No. dissected	No. positive(%)	No. dissected	No. positive(%)	
Yeongju	Cow	1,514	0	145	0	
3	Human	73	0	92	0	
	Subtotal	1,587	0	237	0	
Andong	Cow	689	0	_	_	
	Human	12	0	-	_	
	Subtotal	701	0	-		
Yeoju	Cow	434	0	-	_	
	Human	687	1(0.15)	-	_	
	Subtotal	1,121	1(0.09)	-	_	
Kyungsan	Pig	_	_	413	0	
	Human		_	67	0	
	Subtotal	-	_	480	0	
	Total	3,409	1(0.03)	717	0	

Table 12. Age structure of immature stages of Anopheles sinensis in the study area(1989-1991)

	Total No. in the study area at the median age of each stage(x $10^3$ )					
% Date	L1	L2	L3	L4	Pupa	
1989						
Jun. 30	745	541	286	64	45	
Jul. 7	952	1,408	379	39	58	
Jul. 14	2,337	916	329	62	27	
Jul. 21	3,890	1,021	596	115	46	
Jul. 28	149	251	98	125	10	
Aug. 18	85	51	21	83	7	
Aug. 25	17	59	17	25	0	
Sep. 1	120	144	66	36	9	
Sep. 8	18	63	59	89	0	
Total	8,313	4,454	1,851	638	202	
1990						
Jun. 15	0	24	330	118	18	
Jun. 22	39	136	613	183	56	
Jun 29	1,055	2;299	1,070	190	105	
		7,387				
Jul. 6 Jul. 13	5,748	•	6,580	1,227	284	
	1,498	413	247	164	128	
Jul. 19	120	230	268	76 50	25	
Jul. 27	190	251	106	50	15	
Aug. 2	63	88	31	27	15	
Aug. 10	9	68	26	4	6	
Aug. 19	128	18	30	21	12	
Aug. 23	43	16	16	11	7	
Aug. 30	86	136	131	71	7	
Sep. 6	203	74	41	43	13	
Sep. 20	12	32	95	25	6	
Total	9,253	11,213	9,606	2,221	697	
1991						
Jun. 15	443	195	99	53	38	
Jun. 21	35	257	585	253	43	
Jun. 29	988	1,129	781	274	25	
Jul. 4	3,995	2,094	657	96	0	
Jul. 10	325	517	74	7	0	
Jul. 20	489	457	223	37	0	
Jul. 27	1,061	430	168	77	4	
Aug. 2	74	208	238	22	0	
Aug. 7	2,796	480	170	55	0	
Aug. 14	1,144	527	104	38	10	
Aug. 21	515	493	756	164	42	
Aug. 30	982	2,667	360	99	226	
Sep. 6	358	382	294	123	92	
Sep. 13	3,604	1,232	773	398	175	
Sep. 20	1,228	474	306	287	41	
Sep. 27	531	199	159	75	39	
Total	18,568	11,741	5,747	2,058	735	

Table 13. Lc50 value and resistance ratio of larval Anopheles sinensis for tested insecticides

Towns of the town	LC <sub>50</sub>	Destatores		
Insecticides	Kyungsan	Nagasaki*	Resistance ratio	
O-P compounds				
Diazinon	12.35	3.16	3.19	
Malathion	14.50	3.52	4.12	
Fenthion	0.085	0.125	0.68	
Fenitrothion	21.50	6.50	3.31	
Pyrethroids				
Phenothrin	0.25	-	<del>-</del>	
Allethrin	0.94	_	_	

<sup>\*</sup> Data reported by Department of Medical Entomology, NIH, Japan.

Table 14. Prevalence of P. vivax among residents in Kyungpook Province, Korea(1991)

Age group (Y)	Male		Female		Total	
	No. examined	Percent positive	No. examined	Percent positive	No. examined	Percent positive
0- 9	16	0	14	0	30	0
10-19	19	0	13	0	77	0
20-29	39	0	38	0	77	0
30-39	144	0	115	0	259	0
40-49	158	0	141	0	450	0
50-59	213	0	237	0	450	0
60-	167	0	199	0	366	0
Total	774	0	772	0	1,546	0

mmonly used among the narcotic addicts. Similar results among narcotics patients in Korea have been obtained by Seo and Rim(1959).

Many factual works on malaria among Korean military personnel have been reported by many investigators since the participitation of Korean troops in the Vietnamese War(Kim et al., 1966; Kim, 1967; Lee et al., 1967; Choi, 1969; Kim, 1968; Seo et al., 1970, Soch, 1970). Kim(1967) reported 1,780 malaria cases of Korean troops in south Vietnam during 1 year from November 1,1965, and noted that the malaria mortality rate was 111 per thousand per year.

Seo et al.(1970) in a parasitological study of Ko-

rean Forces in South Vietnam reported that 224 cases or 95.3 per cent out of 235 malaria patients were found to be infected by ring form trophozoites of *P. falciparum* parasites, one case or 0.4 per cent of *P. vivax*, and the other 10 patients or 4.3 per cent were of mixed infection. After the Vietnamese War, because many military personnel with tropical malaria and tertian malaria returned to this country, the possibility of its spread in Korea was considered and discussed by a number of scientists. However, with effective treatment and control measures no further spread occurred. After the beginning of the New Village Movement in the third Five year Economic

Development Project, the epidemiological and ecological studies on malaria and its vector mosquitoes in Korea have made remarkable progress through the labors of medical parasitologists, entomologists, and Public Health officials. As presented in Table 15, the number and incidence rate reported during the period from 1973 to 1990 is based on the Yearbook of Public Health and Social Statistics, Korea.

Table 15. The reported number and incidence of malaria in Korea from 1973

Year	Number notified	Incidence(%)
1973	747*	2.2*
1974	611	1.8
1975	314	0.9
1976	32	0.09
1977	11	0.03
1978	-	_
1979	1	0.003
1980	_	_
1981	_	_
1982		_
1983	_	_
1984	_	_
1985	_	_
1986	<del></del>	-
1987	3	0.007
1988	8	0.019
1989	1	0.002
1990	_	

<sup>\*</sup> Number and incidence is based on the report from cities and Provinces under Communicable Disease Control Law.

Factually, this is no indication of the true incidence among the residents in Korea, because the reports from cities and Provinces are not sufficient to determine the true incidence of all malaria cases.

As shown in Table 15, the malaria cases reported in and after 1977 were very few and seemed not too significant as a public health problem.

The exact cause for the decrease of indiginous malaria in Korea in not known, but there are several factors possibly being due to control measures against malaria by means of free treatment of patients and drug prophylaxis, and to spraying of insecticides against mosquitoes, cutting down of grass around dwellings, etc. In recent years, the number of malaria cases reported in and after 1987 increased and it was estimated that they were mostly imported by Korean businessman or travellers from tropical regions such as Southeast Asia, Africa, America, or other countries. In practice, many people who recently went abroad for various purposes increased from year to year, and actually many cases of parasitic diseases were imported by people returning to this country from tropical countries. Thus, several reports of imported malaria were published by many investigators of Korea (Kim et al., 1966; Yoon et al., 1967; Kim, 1967; Kim, 1968; Ahn et al., 1983).

Ahn et al.(1983) reported on 51 cases of malaria imported in Korea during the period from 1970 to 1982, of which 37 cases were detected abroad or in the home country. They involved 4 P. vivax and 15 P. falciparum cases. The number of malaria cases according to the localities from where they returned is 12 cases from Africa such as Libia, Kenya, Cameroon, Nigeria, Liberia, etc., 11 cases from Saudi Arabia and Iran, etc., 9 cases from Southeast Asia such as Burma and Indonesia, etc., and 1 case from Australia. There were 2 cases of foreigners(India and Malaysia) among them. Similar results have been obtained by Soh et al.(1985) In the survey on Anopheline mosquitoes in Korea, with a consideration of their importance as malaria vector, Kobayashi (1928) examined many specimens of the adult and larval mosquitoes collected from various localities, and reported for the first time that the principal vector of malarial parasite was A. sinensis. A study of Yamada(1936) reported that among the blood meals of A. sinensis collected from animal sheds, 82.5 per cent fed on cows and none on man, however, among those collected in human dwellings and railway station in Seoul, 54.8 per cent and 80.3 per cent fed on man, respectively, and Yokoo(1944) in a study on the distribution and biology of mosquitoes reported that the seasonal prevalence of A. sinensis first appeared in late March and they trapped large num bers during the period from late June to midJuly.

He also stressed that A. sinensis seemed to serve as a vector of setariasis of domestic animals in Korea. Whang(1962) carried out the biological observations on Anopheline mosquitoes in Korea, with special reference to A. sinensis. It was found that Anopheline mosquitoes began to appear from April or May and disappeared in October each year. He also commented that Anopheline mosquitoes had a mainly zoophilic habit, and since its man-biting rate was rather low, it would cause a low infection rate of malaria. Similar results in the vector mosquitoes have been obtained by Nagai(1925), Kobayashi(1929), Yamada (1936), Yokoo(1944), Paik et al.(1965), and Hong (1970).

Seasonal fluctuation in numbers of *A. sinensis* have been reported previously (Paik et al., 1965; Hong, 1970; Joo and Wada, 1985), but this is the first report on a complete year's collection with one-night per week schedules. It seems that the month of highest average catch in night was July, when the temperature was between 18.6 and 34.8C and the humidity 59-76 per cent. The earlist date of appearance and disappearance in this survey was in mid-March or early and mid-April and in late October or mid-November each year. The main factors contributing to the earliest time of appearance and the change in the density of the *A. sinensis* each year were considered to be breeding sites such as rice fields and swamps, etc., rainfall, temperature and humidity, etc.

The seasonal prevalence of *A. sinensis* has been shown usually in the number of *A. sinensis* collected by a light trap, but the numbers were found to fluctuate day by day. Therefore, the total or average number seems to be unfit for comparison of the abundance of *A. sinensis*. On the basis of this point, the mean per cent index(MPI), being calculated from the totals of vector mosquito collections at stations in comparison with those in the standard year, were used for the comparison of the annual abundance of vector mosquitoes. The results shown in Table 5 and Table 6 indiciate that a marked decrease in MPI was obtained in 1990-1991, and accompanied with a decrease in the number of mosquitoes. Al-

though the main reasons for the decrease in population levels of vector mosquitoes are not readily apparent, it was considered to be due to the intermittent irrigation or early planting of rice plants, extensive uses of chemical insecticides and herbicides in farming, high temperature and small precipitation, livestock and natural enemies, and the reduction of rice fields by urbanization. Such consideration was also recognized by Baik and Joo(1991), Russell and Rao (1940), Surtees(1970) Chandler and Highton(1975), Chandler et al.(1975), and Mogi(1984).

In this survey, A. sinensis blood feeding success observed sometimes a reflection of mosquito abundance. A. sinensis abundance during 1989 was greater than in other years observed, and the same relationship held true for the number of engorged mosquitos collected during those years. However, monthly engorgement success was not always linked to overall mosquito collected abundance. In 1989, A. sinensis was most abundant in July, but engorgement of female A. sinensis had the highest rate in August.

The results of a monthly fluctuation of engorged number of *A. sinensis* during the period from 1987-1991 indicated that blood feeding was significantly associated with rainfall. Such consideration was also recognized by Olson et al.(1983), Russel(1986), Day and Curtis(1989), and Baik and Joo(1991).

In the biting rhythm of *A. sinensis* in this survey, this species appeared to be active throughout the whole night, but was more active during darkness, after sunset to mid-night. At that time the temperature was between 26.9 and 30.1 C and the humidity 67-76 per cent. In the present study the nocturnal periodicity activity of the female *A. sinensis* was not always similar by collection methods on the same night. Although the environmental factors such as temperature, light, humidity, etc., should be important in determining the attraction of mosquitoes, this data can not be explained fully only by the hourly changes of these factors when the collection was made. Further work along this line is needed.

Ree et al.(1967) carried out a study on the natural infection of *P. vivax* in *A. sinensis* in Korea, and reported for the first time that of 7,517 dissections

of A. sinensis, only one specimen was found to be naturally infected with the sporozoite in August, 1962. They also commented that the abundance of Anopheline mosquitoes did not always seem to coincide with the incidence of malaria.

As for experiments with Korean *P. vivax*, valuable information was published in Japan by Otsuru(1949) who succeeded in infecting 25.7 per cent of Japanese *A. sinensis* fed on a patient with this parasite. Similar experiments were performed by Ham(1970). He also reported that oocysts were found in 17 per cent of the *A. sinensis* fed on patients.

In the present survey, the population density of A. sinensis was relatively abundant but no infection for the occysts or sporozoites of P. vivax was found among 717 female A. sinensis dissected. Such a result seems to be affected by human and socioeconomic factors in relation to the ecology of vector mosquitoes, i.e., the gradual elevation of living standards of the residents, gradual awakening and behavior of the residents to protect themselves from mosquito biting using such preventives as mosquito nets and insecticide sprays. The relatively short period of mosquito season each year may also affect the transmission of Korean vivax malaria.

The immature stages of the Anopheline mosquitoes occur in a variety of habitats among which the most important is the rice fields in Korea. The ecology of this species in various rural and suburban areas in Korea and other parts of the world have been studied by many investigators. Some of their results indicate that the seasonal fluctuation in the larval population densities in paddy water is markedly different from field to field in the same area and/or from year to year in the same field.

In the present survey, the total number of larval A. sinensis in rice fields was highest in early July, when the fields were undergoing rice transplanting, and its number progressively decreased in early August by insecticide sprays. After mid-September, densities showed a marked decrease and both larvae and pupae of the vector mosquitoes were rarely found until rice plants were harvested.

A similar result of peak abundance of vector mos-

quitoes from rice fields in Kyungpook Province of Korea was recorded by Baik and Joo(1991) who also reviewed published observations from other parts of the world. In Korea, A. sinensis were always collected from parsley fields and open stagnant but relatively clean water, also in stream pools or sluggish streams near the villages in early April. A study of Whang(1962) reported that only female A. sinensis with sperms in the spermathocae hibernated in cellars and animal sheds, etc., because male mosquitoes were not found. Further work along this line is needed. He also commented that A. sinensis from various habitats moved progressively in a series of dispersal flights until whole areas was finally repopulated by mid-summer each year.

From their study on the toxicity of agricultural pesticide applications to several mosquito species in south Korean rice fields, Self et al.(1973) reported that the use of agricultural insecticides had reduced larval rice field populations throughout Korea. On the contrary, Chandler and Highton(1976) and Service(1977) reported an increase in larval densities and adult population, respectively following the application of insecticides to control rice pests in the Kisumu area of Kenya. In both cases these increases were attributed to the suppression of predator populations(Snow, 1983). There is evidence that the widespread use of insecticides in agriculture has been responsible for the selection of resistant strains of vector mosquitoes in various parts of the world.

In the studies on insecticide resistance of vector mosquitoes in Korea, Lee(1969) reported for the first time that *A. sinensis* as a malaria vector was found to be resistant to dieldrin and susceptible to DDT, but *C. tritaeniorhynchus* was highly resistant to both dieldrin and DDT. Ree et al.(1979) studied the control effects of agricultural pesticides against mosquito populations of rice paddy breeding species in Korea, and reported that the mortality rates of *A. sinensis* on the second days of the treatment were 12 per cent to BPMC, 38 per cent to diazinon, and 10 per cent to fenitrothion. They also commented that this species had developed a high degree of resistance to some insecticides, and that organophosphorus

and carbamate insecticides seriously affected the population of the aquatic organisms in the rice fields besides mosquitoes.

In recent years, from their studies on epidemioentomological survey of Japanese encephalitis in Korea, Baik and Joo(1991) reported that *C. tritaeniorhy*nchus had developed high resistance to most of the insecticides as compared with the results of susceptible strains reported by Yasutomi and Takahashi (1987). Similar results of insecticide resistance of *A.* sinensis and *C. tritaeniorhynchus* were reported by Shim and Kim(1980 and 1981), Yasutomi et al.(1986), and Baik and Joo(1987) who also reviewed published observations from other parts of the world.

The results of the present study indicate that the resistance ratio to organophosphorus compounds in Kyungsan colonies of *A. sinensis*, relative to susceptible colonies, were nearly 4 times. Resistance being higher of *A. sinensis* suggests that this is probably related to some difference in the opportunity of contact of the insecticides and fungicides. Such considerations were also recognized by Self et al.(1973), Mogi(1978), and Yasutomi and Takahashi(1987).

### **Summary**

The seasonal prevalence and population dynamics of *Anopheles sinensis* in relation to the epidemic of *Plasmodium vivax*, and the ecology of this vector mosquito in Kyungpook Province were studied during the period of 5 years from 1987 to 1991, and the incidence of *Plasmodium vivax* among the residents in northern parts of the Province were also studied.

Anopheles sinensis first collected in April between the 2nd and 20th days, but in March from the 15th day in 1989, and trapped in large number during the period from early July between the 5th and 12th. There was a gradual decrease from mid-August, with a very small number of them collected until mid-November, each year. The average number of Anopheles sinensis in July was 542.6 per trap-night in 1987, but in 1989 increased abruptly to 1,331.4, it then decreased to considerably lower levels, with an average of 271.9 in 1990 and 372.1 in 1991.

In the trend of nocturnal activity of Anopheles sinensis, with oncoming darkness, this vector mosquito becomes very active, gradually decreasing in activity toward mid-night, however, then slightly increasing toward dawn.

The immature stage of *Anopheles sinensis* first found in the rice fields contributed to peak adult densities in early July. The highest larval densities of *Anopheles sinensis* in the study area was  $21,226 \times 10^3$  in early July 1990.

The larval Anopheles sinensis showed high resistance level and resistance ratios against 3 kinds of organophosphorous compounds, diazinon, malathion, and fenitrothion, but low resistance against fenthion.

A total of 1,546 residents were examined, no case was found to be infected with *Plasmodium vivax*. Key-words: *Anopheles sinensis*, seasonal prevalence, population dynamics, *Plasmodium vivax*.

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# 韓國에 있어서 三日熱 마라리아의 疫學的, 媒介動物學的 調査

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## (國文抄錄)

慶北地域에 있어서 三日熱 마라리아 妹介모기, 중국얼룩날개모기(Anopheles sinensis)의 季節的 出現消長과 群集變動이 三日熱 마라리아 發生과 流行에 미치는 影響을 糾明하기 위해 1987年부터 5年間 慶山郡 1個所에 誘蛟燈으로 1週日에 한번씩 成蟲을, 1989年부터 3年間 慶北 農村 振興院 附屬農場에서 모기幼蟲을 採集調査함과 아울려 流行地域으로 알려진 慶北 北部地域 住民들에서의 마라리아 感染狀을 調査하였다.

1987年부터 1990年까지 誘蛟燈에 처음으로 중국얼룩날개모기가 採集되는 날자는 年度別로 큰 差異를 나타내었으며, 3月中旬에서 4月中旬사이에 採集되었고, 가장 높은 群集密度를 보인것은 7月初旬에서 中旬이었으며, 그 後 점점 減少하여 11月初旬과 中旬부터는 전혀 採集되지 않았다. 採集된 모기중 吸血한 個體率은採集場所別로 差異가 있었으며 그 率은 0.5-67.9% 였다. 중국얼국날개모기의 採集되는 數는 1987年이후 增加하여 1989年 가장 많은 數가 採集되었으며, 그 後 급격히 減少하여 1990年에는 가장 적은 數가 採集되었다. 夜間活動性은 22時에서 23時사이에 가장 旺盛하였으며, 그 後 점차 減少하다가 새벽 4-5時에 다시 약간 增加하였다. 중국얼룩날개모기幼蟲은 6月中旬에 논에서 처음 發見되었고 그 密度는 7月初旬 및 中旬에 가장 높았으며 1㎡당 平均數는 1990年 7月 初旬에는 141.5마리였다. 9月中旬이후 부터는 그 密度가 현저히 減少하였다. 중국얼굴날개모기幼蟲은 3種의 有機燒劑에 對하여 높은 抵抗性을 나타내었다. 慶北 北部地域 住民 1,546名에 있어서 三日熱 마라리아 感染狀을 調査하였던 바 마라리아 感染者는 發見할 수 없었다.