ENVIRONMENTAL FACTORS ASSOCIATED WITH LARVAL HABITATS OF MALARIA VECTORS IN NORTHERN KYUNGGI PROVINCE, REPUBLIC OF KOREA¹

DAVID M. CLABORN,²³ PAUL B. HSHIEH,² DONALD R. ROBERTS,² TERRY A. KLEIN,⁴ BRIAN C. ZEICHNER⁵ AND RICHARD G. ANDRE²

ABSTRACT. The larval habitats of malaria vectors near the Demilitarized Zone of the Republic of Korea (ROK) were sampled from June through September 2000 to determine larval abundance and to identify environmental factors associated with high larval density. Six primary habitats were identified: rice fields, irrigation ditches, drainage ditches, stream pools, irrigation pools, and marshes. Most habitats harbored similar densities of larvae until August and September, when population densities in rice fields declined and those in irrigation pools increased. The primary vector in the ROK, *Anopheles sinensis*, occurred in water with a wide range of values for environmental factors, including pH, total dissolved solids, percent of surface covered with floating vegetation, and nitrate and phosphate concentrations. No environmental factor or combination of factors were found that were predictive of high larval densities. This study suggests that larval *Anopheles* are capable of developing in a wide range of stagnant, freshwater habitats in northern Kyunggi Province, ROK.

KEY WORDS Anopheles sinensis, Anopheles lesteri, Anopheles yatsushiroensis, habitat, Republic of Korea, water quality, *Plasmodium vivax*

INTRODUCTION

After an absence of more than 10 years, malaria (Plasmodium vivax) has reemerged in the Republic of Korea (ROK) (Feighner et al. 1998). The number of detected cases grew from 1 in 1993 to 3,719 in 1999. Of all cases diagnosed in the ROK through 1999, 83 (2.23%) cases occurred in American military personnel or Korean augmentees to the U.S. Army. As of December 2000, 16 confirmed cases had occurred in American personnel stationed in the ROK for that year alone (Preventive Medicine Directorate, 18th MEDCOM, personal communication). To date, the focus of the disease has been in Paju County just south of the Korean Demilitarized Zone (DMZ) that separates North and South Korea. Large populations of military personnel, both American and Korean, are stationed in this area.

The strain of *P. vivax* transmitted in the ROK is well adapted to a temperate climate and demonstrates both short and long incubation periods. Long incubation periods of greater than 6 months increase the risk of returning soldiers reintroducing malaria to the continental United States, making malaria control in Korea even more important (Walter Reed Army Institute of Research 1999). One of the best-documented instances of local transmission in the USA occurred after the visit of an American soldier to a Girl Scout camp in California where vector *Anopheles* were abundant (Brunetti et al. 1954).

Until recently, malaria prevention policies for American soldiers in high-risk areas of the ROK relied exclusively on vector control and personal protective measures (PPM). However, Strickman et al. (2001) reported acceptable vector and disease control in areas treated with residual insecticides and command emphasis on personal protection. The U.S. malaria policy was modified in 1999 by placing more than 6,000 American soldiers stationed north of the Imjim River on chloroquine/ primaquine chemoprophylaxis. Although these drugs are still effective against the strain of *P. vivax* in the ROK, some concerns exist about relying on this method as the primary means of malaria control. For instance, soldiers stationed in areas south of the Imjim River often train for several days in camps considered to be high-risk areas north of the river. During 1999, soldiers training in high-risk areas were placed on chemoprophylaxis before the exercise until the end of the malaria season in October. During the 2000 malaria season, only soldiers residing north of the Imjim River were placed on chemoprophylaxis. Soldiers entering these areas only for training depended solely on the use of PPM, including permethrin-treated uniforms, proper wearing of the uniform (pants tucked into the boots and shirt sleeves rolled down), and topical repellents.

In addition to PPM, an area-based control method might provide better malaria control in those areas with transient military populations. Strickman et al. (1999) suggested that larviciding in mosquito habitats around American installations could be a useful control method if such efforts were coordi-

¹ This manuscript reports original research and does not necessarily reflect the policy of the Department of Defense or the U.S. Navy.

² Uniformed Services University of Health Sciences, 4301 Jones Bridge, Bethesda, MD 20814-4799.

³ Present address: Navy Disease Vector Ecology and Control Center, Jacksonville, FL 32212-0043.

⁴ Preventive Services Directorate, 18th Medical Command, Unit 15281, APO AP 96205-0054.

⁵ U.S. Army Center for Health Promotion and Preventive Medicine, Entomological Sciences Program, APG, MD 21010-5403.

nated with Korean civilian authorities. One advantage of larviciding would be that protection could be conferred on all soldiers in the high-risk areas, regardless of their length of exposure. However, the cost of such a larviciding program is unknown and would require significant information on the bionomics of vector larvae in the malaria endemic areas. In addition to the coordination of a larviciding program with the surrounding Korean community, cost would be a major consideration. Cost comparisons with other control methods, especially chemoprophylaxis, would be essential.

The cost of larviciding is a function of the size of the larval habitats and the cost of treatment per unit area. However, the following 3 questions about the local vectors must be answered before the size and location of larval habitats requiring treatments can be determined. What larval habitats occur in the high-risk area? What species of mosquito larvae occur in the local habitats? Which mosquito species are proven malaria vectors? This paper describes efforts to answer these questions and to develop a quick and reliable means of surveying habitats in Korean malaria foci.

Successful larval control requires the ability to identify larval habitats and to distinguish between sites with high and low vector populations in a timely manner (Wood et al. 1991). Limited mosquito control assets in the ROK require prioritization of the areas in need of pesticide applications; this can be achieved with larval surveillance. One approach to surveillance is to identify key environmental factors that predict the presence of vector populations, then to use these factors as markers to predict the presence of significant larval densities either in an indirect survey or in a remote sensing system. Many such environmental factors have been identified for a variety of vectors. For example, throughout much of its extensive range, Anopheles pseudopunctipennis Theobald was associated with green filamentous algae and aquatic vegetation in sunlit freshwater stream pools (Manguin et al. 1996b). In southern Mexico, high populations of Anopheles albimanus Wiedemann were linked to flooded, unmanaged pastures at an elevation below 25 m (Rodriguez et al. 1996). Positive associations between the density of An. pseudopunctipennis and the presence of filamentous algae or the plant Heteranthera spp. occurred during the dry season (Savage et al. 1990). In Venezuela, Anopheles aquasalis Curry was collected most often in brackish mangrove swamps and larval populations varied with the water salinity, whereas Anopheles oswaldoi Peryassu was associated with permanent freshwater habitats (Grillet 2000). In Belize, the habitats of Anopheles darlingi Root were characterized as being river margins with patches of floating debris (Manguin et al. 1996a). Vector populations also have been associated with pH and percent tree cover (Rejmankova et al. 1998).

Researchers have developed models to predict

the presence or abundance of vector populations based on environmental factors by using logistic regression (Savage et al. 1990), multivariate regression (Rodriguez et al. 1996, Grillet 2000, Moncayo et al. 2000), and global information system techniques (Roberts and Rodriguez 1994, Roberts et al. 1999). Models of this type have the potential to improve the efficiency of malaria vector control and surveillance programs in the ROK, but few if any have been developed for the indigenous malaria vectors.

Malaria vectors in the Republic of Korea

In a review of Korea's arthropods of public health importance, Chow (1973) mentioned only 2 potential malaria vectors: Anopheles sinensis (Wiedemann) and Anopheles yatsushiroensis Miyazaki. Of these 2 species, Chow considered the former to be the most important vector. This conclusion generally has been considered correct by subsequent authors, but questions about the taxonomy of An. sinensis and others in the Hyrcanus Group have caused significant confusion. In particular, the difficulty of distinguishing An. sinensis from Anopheles lesteri Baisas and Hu is of some importance because the vector potential of the latter species is unknown. Tanaka and others (1979) considered An. lesteri, rather than An. sinensis, to be the probable primary vector of malaria in Japan, but little work has been performed on this species on the Korean peninsula.

In a survey of malaria vectors in Kyonbuk, Korea (Joo and Kang 1992), An. sinensis was the only species considered, even though it generally is considered to be strongly zoophilic. Ree and others (1973) reported only 3 Anopheles species caught in light traps (An. sinensis, Anopheles sineroides Yamada, and An. yatsushiroensis). Anopheles sinensis comprised 95% of the anophelines and 18% of the total mosquito population. Strickman and others (1999) caught the same 3 species in light traps in the northern ROK, but also caught 1 specimen identified as An. lesteri. In that study, larval surveillance indicated densities of 0.02 to 3.1 larvae/ dip in rice fields surrounding a military training area with a probable history of malaria transmission. A later collection (Strickman et al. 2001) caught many more adult An. lesteri and An. vatsushiroensis, none of which were positive by enzymelinked immunosorbent assay for P. vivax circumsporozoite antigen. In fact, only 2 out of 2,376 An. sinensis tested for malaria infection were positive. Therefore, although An. sinensis is not considered to be an efficient vector, its relative abundance and the detection of *P. vivax* antigen only in this species implicate this mosquito as the primary vector in the ROK.

This study investigated the distribution of larvae of *Anopheles* among various larval habitats and attempted to identify environmental factors that could

Teared to adults (Julie-September 2000).					
Species (no. specimens)					
reared to adult stage	Habitats where species was found				
Anopheles sinensis (442)	Rice field, stream pool, drain- age ditch, irrigation ditch, marsh				
An. lesteri (42)	Rice field, stream pool, irriga- tion ditch				
An. yatsushiroensis (3)	Irrigation pond, drainage ditch				
Aedes vexans (277)	Rice field, stream pool, drain- age ditch, irrigation ditch, irrigation pond, marsh				
Culex vagans (41)	Rice field, stream pool, drain- age ditch				
Cx. pipiens (79)	Rice field, drainage ditch, irri- gation ditch, irrigation pond, marsh				
Cx. orientalis (36)	Rice field, stream pool, drain- age ditch				
Cx. tritaeniorhynchus (30)	Rice field, drainage ditch, irri- gation ditch				
Cx. mimeticus (7)	Rice field, stream pool				
Cx. hayashi (3)	Irrigation pond				
Cx. bitaeniorhynchus (10)	Irrigation ditch, stream pool				
Cx. rubensis (1)	Marsh				

Table 1. Mosquito species taken from larval habitats in northern Kyunggi Province, Republic of Korea, and reared to adults (June–September 2000).

be used to efficiently locate the sites producing the greatest number of malaria vectors.

MATERIALS AND METHODS

The study was performed from June through September 2000 in the northwestern part of Kyunggi Province, ROK, about 60 km north of Seoul, and near the DMZ in the vicinity of Munsan, Paju County. This area was characterized by a temperate climate with 4 distinct seasons. Average temperatures ranged from -7 to 2°C in the winter and up to 20–29°C in the summer. Monsoons usually affect the area, with the heaviest average rainfall by season occurring in the summer months (mean = 258mm/month). The summer of 2000 was unusually dry; however, typhoon-associated rainfall was significant and provided enough precipitation to flood many of the study sites, especially the streamside pools. Sample sites were concentrated in the areas surrounding the Camp (CP) Casey complex and CP Greaves (both U.S. military bases). The CP Casey complex consisted of 3 military facilities that included the contiguous CPs Castle and Hovey. A large, permanent stream that was prone to flooding transected the complex. The towns of Tongduchon and Tokori surrounded the CP Casey complex on 3 sides, but rice cultivation continued at the interfaces between the camps and villages.

The CP Greaves area was smaller and located in

a rural environment. A high bluff overlooking the Imjim River bordered the southern perimeter of CP Greaves. Rice farming was extensive on both sides of the river and continued from the camp into the DMZ. Cattle and goat herds were located near the eastern extension of CP Greaves.

The area around CP Greaves was characterized by low, wooded hills separated by shallow valleys. The valleys usually were planted in rice, but also supported crops of peppers, corn, onion, ginseng, and cabbage. Camp Casey was surrounded by steeper, rockier hills that may have exceeded 300 m in height. Other than the 1 large stream transecting CP Casey, few natural streams occurred in the study areas. Numerous irrigation ditches and ponds were interspersed between the rice paddies, providing a variety of potential larval habitats.

Six habitats were identified for sampling purposes: rice fields, irrigation ditches, drainage ditches, marshes, stream pools, and irrigation pools. The rice fields were typically small (less than 1,300 m²) and were bordered by dikes ranging in height from 0.3 to 2.0 m. Early in the season, the dikes were typically mowed and devoid of tall vegetation; however, the degree to which the borders were maintained varied, and some became overgrown later in the season. The rice fields were transplanted with seedlings in May through early June. Irrigation or flooding of the rice fields was inconsistent so that some were flooded whereas others at the same stage of plant development were not. Irrigation ditches ranged in size from small troughs 0.3 m deep to large canals up to 2 m wide and 1 m deep. Drainage ditches also varied in size and were frequently lined with concrete or stone. Stream pools were observed only at CP Casey and were all upstream of a wastewater treatment plant. Pools ranged in depth from a few centimeters up to 1 m. and in surface area from less than 1 m² to about 100 m². They often supported heavy algal mats and had sand or mud bottoms; large stones or concrete sides surrounded most of the pools. Irrigation ponds were located near the rice fields and provided water for irrigation. The ponds were usually about 1.5 m deep (range: 800-2,100 mm) with steep sides. Early in the season, few larvae were found in these sites, but by August, large populations of larvae occurred among the algae growing along the edge. Marshes were not common so only 2 were sampled during this study. The marshes supported a wide range of vegetation and were always flooded. The 2 marshes were included only for a general survey of the species present and were not included in statistical analyses.

Data collection: Initially, 60 study sites were selected and located with a Garmin III[®] handheld global positioning satellite (GPS) unit (Garmin International Inc., Olathe, KS). The GPS positions were collected for entry into a geographic information systems database to determine the size of larval habitats in high-risk areas. Thirty sites were

Habitat	June	July	August	September
Rice paddies	0.1a	0.4a	0.3a	0.0a
	(42, 0.9, 0–5.3)	(42, 0.9, 0–4.7)	(49, 0.4, 0–1.9)	(41, 0.6, 0–3.0)
Ditches	0.1a	0.4a	0.1a	0.1a
	(10, 0.3, 0-0.9)	(11, 1.8, 0-6.0)	(17, 0.3, 0-0.9)	(14, 1.4, 0–4.7)
Pools and ponds	0.0b	0.7a	0.4a	5.0b
	(8, 0.0, 0–0.1)	(12, 4.3, 0–14.7)	(14, 1.4, 0–3.0)	(11, 13.1, 0–39.3)

Table 2. Median number of larvae per dip (*n*, standard deviation, and range are given in parentheses) of Anopheles spp. collected in larval habitats of northern Kyunggi Province, Republic of Korea (June–September 2000).¹

¹ Median values within a column followed by the same letter are not significantly different (Kruskal–Wallis; P > 0.05). Irrigation ditches and drainage ditches were combined into 1 category for purposes of statistical analysis. Stream pools and irrigation ponds also were combined.

located around each of the 2 camps, Greaves and Casey. As the season progressed, other sites became available and were added to the study for a total of 93 sites: 50 rice paddies, 13 stream pools, 12 irrigation ditches, 11 drainage ditches, 5 irrigation pools, and 2 marshes.

Larval sampling was performed around the perimeter of each study site with a standard 400-ml plastic dipper. The circumference of each site was measured then divided by 30 so that 30 dips could be equally spaced around the site. Initially, all anopheline larvae and a sample of culicine larvae were removed with pipettes and stored in plastic bags for transport to the central laboratory at CP Casey. When Anopheles spp. exceeded 50/dip, approximately 10% of the larvae were taken for rearing. The plastic bags were filled with water from the study site then stored in an insulated chest for transport to the laboratory for rearing. All collections were performed between 0700 and 1200 h. With the exception of those added later in the study, each habitat was sampled once each month from June through September. After each additional site was added, it was sampled monthly through September. A value of 0 was recorded for sites that were dry, flooded with running water, or otherwise incapable of supporting larval populations, as well as for suitable sites with no larvae in 30 dips.

In June and July, water analysis was conducted on 1 of the 30 dips from each site. Nitrate and phosphate concentrations were determined with a pocket colorimeter (Hach Co., Loveland, CO); total dissolved solids and pH were both determined with Hach[®] Pocket Pal testers Model 46700-12.

Percent of water surface covered by duckweed

(*Lemna* spp.) or algae was estimated visually. Three observers made independent estimates of the duckweed or algal cover and the mean of the 3 estimates was recorded.

Rearing procedures: Because the species of Korean Anopheles are impossible to distinguish by larval characteristics, the larvae were reared to the adult stage for specific identification. Mosquito rearing was performed in an air-conditioned watertesting laboratory at CP Casey. A plastic enclosure was placed over a countertop and the interior was warmed continuously with two 100-W incandescent light bulbs. Larvae were exposed to the light continuously. The larvae were transferred to 16-oz plastic cups filled with tap water that had been allowed to sit overnight to reduce chlorine residuals. Larvae were fed finely ground Tetramin® Tropical Flakes fish food (Tetra GmbH, Melle, Germany). Each 4th-stage larva was moved to an individual plastic rearing vial. The 4th-stage larva and pupal exuvium were placed in 80% ethanol in a 0.25dram glass vial with a rubber stopper for taxonomic study. The adults were preserved according to procedures of Belkin and others (1965), then identified to species according to Lee (1998). Voucher specimens of adults and associated exuviae were deposited at the U.S. National Museum of Natural History, Smithsonian Institution, Washington, DC.

Statistical analyses: Mean and median densities of Anopheles (number of larvae/30 dips) were determined with the UNIVARIATE procedure in SAS (SAS Institute 2000). Medians were compared with the Kruskal–Wallis test. Data with similar coefficients of variation were compared with Tukey's honestly significant difference test. Similar analyses

Table 3. Percentage of area covered with algae in 3 larval habitat types of northern Kyunggi Province, Republic of
Korea (June-September 2000).

Habitat	n	Mean	Median	Standard deviation	Coefficient of variation	Range
Rice fields	50	7.6	Oa	20.1	2.6	0-80
Ditches ²	20	18.9	0a	26.9	1.4	0-75
Pools ²	15	19.4	Oa	32.3	1.7	0-100

¹ Medians followed by the same letter are not significantly different (Kruskal–Wallis test; P = 0.05).

² Irrigation ditches and drainage ditches were combined into 1 category for statistical analysis. Stream pools and irrigation ponds also were combined.

Habitat n Mean Median' deviation of variation Ra							
Rice fields	50	40.2	30a	38.4	1.0	0–100	
Ditches ² Pools and ponds ²	18 15	6.0 2.0	Ob Ob	14.1 0.7	2.4 0.4	0-50 0-10	

 Table 4.
 Percentage of area covered with Lemna spp. in 3 habitat types of northern Kyunggi Province, Republic of Korea (June–September 2000).

¹ Medians followed by the same letter are not significantly different (Kruskal–Wallis test; P > 0.05).

² Irrigation ditches and drainage ditches were combined into 1 category for statistical analysis. Stream pools and irrigation ponds also were combined.

were used to compare environmental parameters by habitat, with both types of ditches combined into 1 category and both types of pools similarly combined.

To develop a predictive model for sites with high densities of larval *Anopheles*, each environmental parameter was dichotomized at the median and also at the 75th percentile in 2 different models to allow analysis by logistic regression. The LOGISTIC procedure (SAS Institute 2000) was used to perform stepwise logistic regression modeling of the probability of a site being in the top quartile of larval density as a function of water quality and amount of surface covered by vegetation.

RESULTS

Of the 14 putative species collected during the study period, only 3 were anophelines (Table 1). These mosquitoes were all identified as adult specimens by using morphological characteristics described in Tanaka et al. (1979) and Lee (1998). Adult characteristics used to separate An. sinensis and An. lesteri may not be reliable, so the relative numbers of these 2 species must be considered with some caution. Nevertheless, the majority of Anopheles reared to adulthood (81%) appeared to be An. sinensis, with An. lesteri representing only 18.8%. Only 3 specimens of An. yatsushiroensis were reared to the adult stage during the study, representing less than 1% of the Anopheles. Other species that were identified in significant numbers were Aedes vexans (Theobald) and Culex pipiens Linnaeus.

Anopheles sinensis was collected in all 6 of the habitats sampled (Table 1). Anopheles lesteri was only obtained from rice fields, stream pools, and

irrigation ditches, but this finding may be due to the small number of specimens reared to the adult stage. Densities of larval *Anopheles* for most habitats were similar in June and July (Table 2). A trend toward greater larval densities in ponds and stream pools started in July and this trend eventually led to significantly greater densities in these habitats in August and September when compared to ditches and rice fields. The median number of larvae per dip in rice fields was never great, although as many as 50 larvae were collected in individual dips. Generally, distributions of larvae were highly clustered within rice fields.

In September, many rice fields were drained in preparation for harvesting, resulting in the elimination of larval habitats; however, very large larval densities developed in the pools, especially irrigation lagoons contiguous to the drained rice fields. The pools supported the highest larval densities observed during the 4-month study.

Tables 3-8 describe environmental factors for each of the habitats. The factors overlapped in their ranges and no statistically significant differences were noted between habitats, with the exception of percent surface covered by floating duckweed (Lemna spp.). Rice fields had significantly greater coverage with this plant, although percent coverage was not predictive of larval densities when using logistic regression techniques (P = 0.45). The similarity in water-quality parameters is perhaps not surprising given that several of the habitats are interconnected with a complex irrigation system that moves water from streams, rivers, and wells to irrigation ditches to rice fields then back again. In some fields, this movement was continuous and resulted in a slow movement of water through the paddies. Other fields were more stagnant and prone

Table 5. Mean nitrate concentration (mg/liter) in 3 habitat types of northern Kyunggi Province, Republic of Korea(June-September 2000).

Habitat	n	Mean ¹	Standard deviation	Coefficient of variation	Range
Rice fields	37	0.9a	0.8	0.9	0-2.8
Ditches ²	16	1.4a	1.1	0.8	0-4.0
Pools and ponds ²	12	1.2a	0.7	0.6	0.4-2.6

¹ Means followed by the same letter are not significantly different (Tukey's honestly significant difference; P > 0.05).

² Irrigation ditches and drainage ditches were combined into 1 category for statistical analysis. Stream pools and irrigation ponds also were combined.

Table 6.	Phosphate concentration (mg/liter) in 3 habitat types of northern Kyunggi Province, Republic of Korea
	(June–September 2000).

Habitat	n	Mean ¹	Standard deviation	Coefficient of variation	Range
Rice fields	44	0.4a	0.5	1.25	0.1–2.4
Ditches ²	18	0.7a	0.6	0.9	0.1 - 2.1
Pools and ponds ²	14	0.3a	0.3	1.0	0.0–1.3

Means followed by the same letter are not significantly different (Tukey's honestly significant difference; P > 0.05).

² Irrigation ditches and drainage ditches were combined into 1 category for statistical analysis. Stream pools and irrigation ponds also were combined.

to intermittent drying. Environmental factors also were similar for infested and noninfested sites (i.e., fields with and without detectable larval populations). The range of each factor is reported for infested and noninfested fields in Table 9. The results indicate that anopheline larvae in the ROK are present in habitats with significant variations in water quality and plant coverage.

Stepwise logistic regression analysis revealed no factors that were predictive of high larval densities (P > 0.05). The most likely candidate for predicting highly infested fields was percent duckweed coverage adjusted for nitrate and phosphate concentrations in July (P = 0.06), but the overall model was nonsignificant (P = 0.24).

The percent of each habitat in which at least 1 larva was detected varied monthly. For rice fields, peak percent infestation (77%) was observed in July, but fell to 20% in September when most of the paddies were drained. In contrast, none of the irrigation pools yielded anopheline larvae in June, but by September, all of them supported significant larval populations. Larval populations in stream pools and drainage ditches were variable, because they were prone to heavy flooding from typhoonassociated rains.

DISCUSSION

Larval Anopheles in the ROK were present in a wide range of the environmental factors investigated during this study. Because of the ability of the larvae to survive in such a variety of conditions and habitats, none of the water-quality or vegetation indices recorded in this study were reliable predictors of high larval density. This conclusion is consistent with previous research by Ikomoto and Sakaki (1979), who found a lack of consistent correlation between population density of *An. sinensis* and the pH and temperature of rice field water. However, a positive correlation was found between the number of immatures and the NH₄-N concentration in that study that was not reflected in the nitrate concentration analysis of our work.

In our study, larvae were collected from a variety of habitats and, at least early in the growing season, showed limited identifiable preference for habitat type. The larvae seemed capable of exploiting most stagnant or slowly moving water habitats in the study area. However, nearly 26% of the sites sampled in July did not support detectable larval populations; the reason for the lack of larval populations in these habitats is unknown but may be related to chemical fertilizers and pesticides applied to the paddies at irregular intervals. Another possibility is that the number of potential oviposition sites exceeded the number of gravid females.

Assuming that most of the *Anopheles* in the area were *An. sinensis*, this malaria vector is capable of development in areas with or without floating vegetation and in water with a broad range of pH, dissolved solids, and nitrate and phosphate concentrations. However, other environmental factors not investigated during this study may serve as limiting factors. It is important to note that the sampling method used during this study was designed for study of large habitats that could be identified and analyzed remotely. A sampling technique that analyzes at the level of a microhabitat (i.e., algal mats vs. overhanging vegetation) might be more likely to identify environmental parameters that are predictive of vector abundance at a different scale.

The species collected during this study occurred in different proportions than they did in a contemporary adult trapping study in the same study site

Table 7. Values of pH in 3 habitat types of northern Kyunggi Province, Republic of Korea (June-September 2000).

Habitat	n	Mean ¹	Standard deviation	Coefficient of variation	Range
Rice fields	38	7.5a	1.1	0.1	5.7–9.6
Ditches ²	16	7.4a	1.1	0.1	6.3-10.0
Pools and ponds ²	14	8.0a	1.2	0.2	6.2–9.6

¹ Means followed by the same letter are not significantly different (Tukey's honestly significant difference; P > 0.05). ² Irrigation ditches and drainage ditches were combined into 1 category for statistical analysis. Stream pools and irrigation ponds also

were combined.

Habitat	n	Mean ¹	Standard deviation	Coefficient of variation	Range
Rice fields	38	159a	80.5	0.5	0-375
Ditches ²	16	200a	78.8	0.4	116-438
Pools and ponds ²	14	178a	55.8	0.3	106-285

Table 8. Total dissolved solids (ppm) in 3 habitat types of northern Kyunggi Province, Republic of Korea (June-September 2000).

¹ Means followed by the same letter are not significantly different (Tukey's honestly significant difference; P > 0.05).

² Irrigation ditches and drainage ditches were combined into 1 category for statistical analysis. Stream pools and irrigation ponds also were combined.

(Burkett et al. 2001). In particular, very few An. yatsushiroensis were obtained during this larval study, whereas this species comprised nearly 48% of Anopheles captured with a Shannon trap during the same time in the same location. Several factors may contribute to the inconsistency between the larval and adult studies. Differential mortality in the rearing process may have occurred that resulted in the death of most of the larval An. yatsushiroensis. Mortality was significant throughout the study. with less than 20% of the larvae attaining adulthood. Alternatively, the distribution of this species in rice paddies may be such that perimeter dipping does not reflect the proportion of species present in the rice field habitat. Finally, another habitat may exist that is exploited preferentially by An. yatsushiroensis. Of these 3 possibilities, the 3rd seems most likely. The adult trapping performed by Burkett and others was done near CP Greaves, an area around which land mines remain from the Korean War. These mines prevented larval sampling in some areas, especially in wooded, hilly areas, and along the banks of the Imjim River, Habitats within these areas or habitats within the river may serve as the preferred larval habitats of An. yatsushiroensis. Tanaka et al. (1979) reported that this species is common in rice fields but that it is more common in hilly or mountainous areas.

The densities of anopheline larvae were similar for most habitats in June. In September, when the

Table 9. Mean (range) of environmental variables in study sites with and without larval *Anopheles*, Kyunggi Province, Republic of Korea (June–September 2000). No significant differences were noted between infested and noninfested habitats (Kruskal–Wallis; P = 0.05).

Environmental	Larval presence				
factor	Yes	No			
% algal coverage	11.3 (0-80)	13.8 (0-100)			
% Lemna spp. cov-					
erage	31.7 (0-100)	17.3 (0-100)			
Nitrates (mg/liter)	0.9(0-2.8)	1.4(0.1-4.0)			
Phosphates (mg/		· · · ·			
liter)	0.4(0-2.4)	0.4(0.05-2.05)			
pH	7.6 (5.9–9.9)	7.6 (5.7–10)			
Dissolved solids	. ,	· /			
(ppm)	164.0 (0-375)	182.0 (103-438)			

rice paddies were drained, larval densities in the irrigation pools greatly increased. In fact, the greatest larval densities during the entire 4 months occurred in September in the stream pools and irrigation lagoons. A larval control program directed toward pools and ditches late in the season would eliminate the greatest concentrations of the vector population, although the relative size of the rice field habitat would still make this habitat a major contributor to overall vector numbers.

Several aspects of the biology of malaria vectors in the ROK must be addressed in future studies. First, the larval habitat of An. yatsushiroensis should be identified. This species may play some role as a malaria vector and thus might be important in the epidemiology and control of this disease. Second, other environmental factors should be investigated as possible predictors of the presence and abundance of larval Anopheles. One factor that seems to have a substantial impact on larval density that was not addressed in this study is the pattern of flooding and draining in the rice fields. Many fields were drained in the middle of the growing season, perhaps accidentally. More attention should be directed to the effect of flooding and draining on species composition and abundance within the rice field habitat. Third, the taxonomy of Anopheles in the ROK must be clarified before specific work on the biology and control of members of this genus can be completed. In particular, the ability to distinguish between An. lesteri and An. sinensis by using adult morphological characteristics is questionable. At present, numbers of specimens for these species from this study are tentative until further systematic work to validate species identification is completed.

This paper reports the 1st part of a larger study that used remote sensing to determine the size and quality of larval habitats of *Anopheles* within the flight range around 2 U.S. military bases in the ROK. This information will be used to estimate the cost of a larviciding program and to compare that estimate to the cost of providing chemoprophylaxis. It should be noted that the U.S. government is not considering any unilateral larviciding efforts, and would implement such a malaria control program only with the support and concurrence of ROK governmental authorities and local landowners.

ACKNOWLEDGMENTS

We are extremely grateful to the soldiers of the 702nd Preventive Medicine Section; the 2nd Infantry Division; the 5th, 38th, and 154th Medical Detachments; the 168th Medical Battalion (Area Support); and the 18th Medical Command for their valuable assistance in conducting larval surveillance during the field phase of this study. Special thanks go to McKinley Rainey, William Herman, Kenneth McPherson, and Alex Ornstein for their support in providing personnel during this study. Thanks also are due to Hung-chol Kim and Kwanwoo Yi, who provided valuable technical assistance in specimen identification. The manuscript was reviewed by Richard Thomas, Tomoko Hooper, Susan Langreth, and Art Lee. Funding was provided by the Department of Defense, Global Emerging Infections System, U.S. Army Medical Research and Materiel Command's Military Infectious Disease Research Program, and National Aeronautics and Space Administration (grant NAG5-8532). This manuscript is in partial fulfillment of the requirements for the Doctorate of Public Health at the **Uniformed Services University of Health Sciences** by D.M.C.

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