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### Permalink

<https://escholarship.org/uc/item/21z7m3ds>

### Journal

American Journal of Tropical Medicine and Hygiene, 73(1)

### ISSN

0002-9637

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### Publication Date

2005-07-01

### DOI

10.4269/ajtmh.2005.73.157

Peer reviewed

## SPATIAL DISTRIBUTION OF ANOPHELINE LARVAL HABITATS IN WESTERN KENYAN HIGHLANDS: EFFECTS OF LAND COVER TYPES AND TOPOGRAPHY

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**Abstract** The distributions of anopheline larval habitats were aggregated in valley bottoms in Kenya in both the rainy and dry seasons, although the degree of aggregation was higher in the dry season than in the rainy season. Larvae of the *Anopheles gambiae* complex larvae were found more frequently in habitats in farmlands and pastures. However, *An. funestus* larvae were found more frequently in natural swamps and pastures. Canopy cover was the only variable significantly associated with the occurrence of the *An. gambiae* complex and *An. funestus*. The average canopy cover was significantly less in the habitats with the *An. gambiae* complex and *An. funestus* larvae than those without the anopheline larvae. Thus, land cover types and topographic features showed important effects on the distribution of anopheline larval habitats. These results suggest that clearing riparian forests would improve growing conditions of the *An. gambiae* complex and *An. funestus* larvae in Kenyan highlands.

### INTRODUCTION

Malaria, a major human health threat, occurs globally in tropical and sub-tropical regions. In the east African highlands, the threat is mounting as shown by more frequent malaria outbreaks in areas where malaria was previously rare.<sup>1–4</sup> Unlike their counterparts in malaria-endemic regions, recent studies suggested that the residents of highland areas generally lack immunity to *Plasmodium falciparum* and are particularly vulnerable to malaria infection.<sup>5,6</sup> Several hypotheses have been proposed to explain the increased malaria transmission in the highlands, including land-use changes, global climate changes, increased drug resistance, cessation of malaria control activities, and demographic changes.<sup>2,7–11</sup>

In recent years, Kenya has experienced very rapid human population growth. The population of this country has almost doubled since 1980.<sup>12</sup> Population growth in the western Kenya highlands is a particularly severe problem, due in part to a lack of family planning and to migration from other areas. As a consequence of dramatic human population increase, there have been unprecedented land-use changes in the highlands. For example, since 1965 the Malava Forest in the Kakamega district has been reduced from 600 to less than 100 hectares.<sup>13</sup> Most rain forests have been cleared for crop planting, cattle grazing, commercial logging, firewood collection, and housing construction.<sup>14</sup>

The drastic land-use alteration can promote vector-borne disease transmission in several ways.<sup>15,16</sup> First, more human-made aquatic habitats become available for the *Anopheles gambiae* complex, which is the primary malaria vector in Africa. For example, increased cattle grazing creates more open temporary habitats that may serve as mosquito breeding habitats.<sup>17,18</sup> Second, the physical and chemical properties of mosquito larval habitats may change. Third, the microclimate of mosquito larval habitats may change. Habitats well exposed to sunlight may have a higher temperature than those in shaded areas, and thus mosquito larval development rate may be increased. For example, a higher adult density of the *An. gambiae* complex was found in houses near cultivated swamps

than in those near natural swamps in a Ugandan highland site.<sup>9</sup> Natural swamps are primarily populated with tall grasses such as papyrus, whereas cultivated swamps are planted with agricultural crops such as maize.<sup>18</sup> Since cultivated swamps generally receive more exposure to sunlight than those in natural swamps, the ambient air temperature in the cultivated swamp area was significantly higher than that in natural swamps.<sup>9</sup> Moreover, mosquito larval predators may be more prevalent in natural swamps than in cultivated swamps.<sup>19</sup> These differences in aquatic habitat characteristics may have important effects on anopheline larval development and survivorship, and on the distribution of suitable larval habitats.

The aim of the present study is to examine the effects of land use/land cover types on the distribution of anopheline larval habitats in a western Kenya highland site. We determined the spatial distribution pattern of larval habitats and analyzed several physical and chemical components in the aquatic habitats that may be affected by land use and may be relevant to mosquito larval development. We also examined the effects of topography on the spatial distribution of larval habitats. The east African highland region contains numerous small valleys and basin-like depressions in a plateau.<sup>7</sup> Stagnant water bodies are more likely to form in valley bottoms and basin-like depressions than in the hills.<sup>18</sup> Thus topography may be an important factor for the distribution of aquatic habitats in the highlands. Knowledge of the effects of land use/land cover types and topography on mosquito larval habitat distribution is useful for predicting the distribution and abundance of malaria vectors and the impact of land-use practice on malaria transmission, and thus for designing novel strategies for malaria intervention.

### MATERIALS AND METHODS

**Study area.** We established a study area of approximately 4 km<sup>2</sup> along Yala River in the Kakamega district, western Kenya, where epidemics of malaria have claimed several hundred lives in recent years.<sup>20</sup> The elevation of the study area ranged from 1,400 to 1,580 meters above sea level. During the period between May 24 and August 31, 2002, the air temperature ranged from 11.4°C to 33.6°C, and the average air temperature was 19.3°C. In 2002, the long rainy season persisted

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TABLE 1  
Total surface area (m<sup>2</sup>) of each land cover type and occurrence of habitats found during the rainy season and dry season\*

	Farm	Forest	Pasture	Road	Swamp
Total surface area (m <sup>2</sup> ) (%)†	2,377,923 (59.6)	715,984 (17.9)	799,061 (20.0)	42,942 (1.1)	55,237 (1.4)
Rainy season					
Number of anopheline-positive sites (%)	108 (44.1) <sup>a</sup>	44 (34.4) <sup>b</sup>	74 (42.8) <sup>ac</sup>	23 (16.4) <sup>d</sup>	28 (34.6) <sup>bc</sup>
Number of anopheline-negative sites (%)	137 (55.9)	84 (65.6)	99 (57.2)	117 (83.6)	53 (65.4)
Total number of sites (%)	245 (100)	128 (100)	173 (100)	140 (100)	81 (100)
Dry season					
Number of anopheline-positive sites (%)†	16 (34.0) <sup>a</sup>	5 (29.4) <sup>ab</sup>	6 (35.3) <sup>a</sup>	1 (11.1) <sup>b</sup>	20 (20.0) <sup>b</sup>
Number of anopheline-negative sites (%)	31 (66.0)	12 (70.6)	11 (64.7)	8 (88.9)	80 (80.0)
Total number of habitats (%)	47 (100)	17 (100)	17 (100)	9 (100)	100 (100)

\* The letters following the proportion of anopheline-positive sites indicate the results of the Tukey-type multiple comparison tests. The values with the same letter were not statistically significant at the  $P = 0.05$  level after adjustment with the Bonferroni correction.

† The total does not include the surface area coverage of the river and streams within the study area, which was 75,477 m<sup>2</sup>. Therefore, the actual size of the study area was 4,066,624 (m<sup>2</sup>).

from April through early June, and the dry season started in July and ended in September. In early 2002, 707 houses and 2,217 inhabitants were counted within the study area. The inhabitants live in traditional stick and mud houses with thatch or iron sheet roofs. The area included small patches of indigenous forests along the river and streams. Deforestation and cultivation along the forest edges are ongoing. The major crop grown in this area was maize.

**Land cover types.** Prior to the larval habitat survey, we estimated the surface area (meters<sup>2</sup>) of each land cover type within the study area. Land cover types were classified into five categories: farmlands, forests, pastures, roads, and natural swamps. Farmlands were characterized by the presence of any agricultural crops and bare ground that had been prepared for planting crops. Pastures were grasslands used for grazing. An area with a mixture of grass and shrubs was also classified as pasture. Natural swamps were characterized by the presence of emergent aquatic plants. Forests and natural swamps were the categories with the least modification by anthropogenic activities. A multispectral one-meter ground resolution Ikonos image (Space Imaging, Atlanta, GA) of the study area was acquired on April 12, 2001. Each land cover type was visually digitized using the geographic information

system (GIS) software package ArcView 3.3 (Environmental Systems Research Institute, Redlands, CA). The digitized land covers were validated by ground truthing. The surface area of each land cover type was estimated by first projecting the land cover layer into Universal Transverse Mercator 1983 zone 36N and then using an Avenue Script that calculates the surface area in ArcView. All streams (including the Yala River) and roads in the study area were also mapped and digitized using the satellite image in the GIS and validated by ground truthing.

**Habitat characterization.** We first counted and categorized all aquatic habitats (excluding running water and water in containers in houses and in tree holes) with respect to land cover types in the long rainy season (May 17 to May 30, 2002) and dry season (August 13 to August 21, 2002). Land cover types, where aquatic habitats were located, were classified into the above five categories. The location and elevation of each habitat was recorded using the global positioning system in differential mode.<sup>21</sup> Occurrence of anopheline larvae was examined at each habitat using a standard dipper (size = of 350 mL). Water was dipped up to 20 times. When a habitat was too small to make 20 dips, water was dipped as many times as possible.

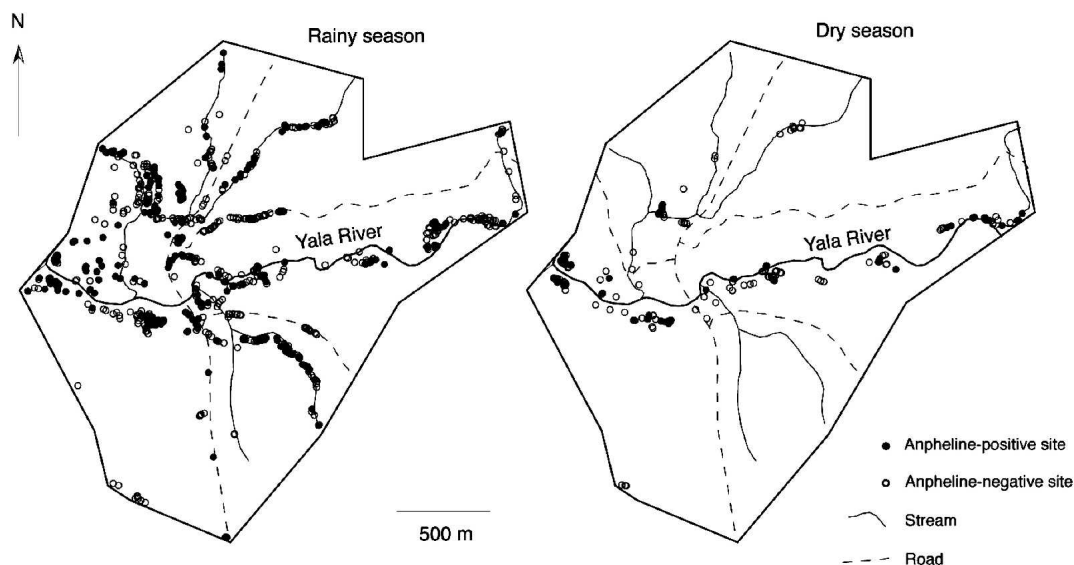


FIGURE 1. Distributions of anopheline larval habitats in the rainy season and dry season in the study area in Kakamega, western Kenya. m = meters.

We also characterized 130 and 50 randomly selected aquatic habitats (including both anopheline-positive and -negative habitats) with respect to physicochemical variables in the long rainy season and dry season, respectively. The selected habitats were characterized using the following variables: 1) canopy cover, 2) presence of filamentous algae, 3) turbidity, 4) habitat size (volume), 5) pH, and 6) concentrations of ammonium ( $\text{NH}_4^+$ ), nitrate ( $\text{NO}_3^-$ ), and orthophosphate ( $\text{PO}_4^{3-}$ ). Canopy cover was estimated with a spherical densiometer.<sup>22</sup> Surface water was sampled and analyzed for the above chemical variables and turbidity following the standard methods of the U.S. Environmental Protection Agency.<sup>23,24</sup> The volume (meters<sup>3</sup>) of an aquatic habitat of 10 evenly spaced grids was established by measuring its length, mean width, and mean depth, and calculated as the product of these three indices.

In the habitat characterization with the physicochemical variables, anopheline larvae were collected and preserved in 95% ethanol from each positive habitat, and third and fourth instar larvae were identified to the *An. gambiae* complex, the *An. funestus* complex, and the other anopheline species based on a morphologic key using a microscope.<sup>25</sup> Larvae of the *An. gambiae* complex and the *An. funestus* complex were further analyzed with the ribosomal DNA (rDNA) polymerase chain reaction (PCR) method.<sup>26,27</sup> First and second instar larvae were subjected to rDNA-PCR analysis for species identification directly.

**Data analysis.** We determined whether the distribution pattern of anopheline larval habitats was clustered in the study area in each season using point pattern spatial analysis (Ripley's K-function).<sup>28</sup> In this analysis, we only used the data from the anopheline-positive habitats. When it was clustered, we determined whether topography influenced the distribution pattern of anopheline larval habitats using two geographic variables, the distance of each habitat to the nearest stream and its elevation. The Kolmogorov-Smirnov one-sample test was used to determine whether anopheline larval habitats were confined toward valley bottoms in each season. The distance of each larval habitat to the nearest stream was estimated in the GIS. Similarly, we examined the relationships between elevation and the distribution of anopheline habitats. We further determined whether the occurrence (presence/absence) of anopheline larvae was significantly correlated with these geographic variables and season using logistic regression analysis.

Since anopheline breeding sites are generally reduced during the dry season,<sup>29,30</sup> we suspected that the reduction is reflected in the difference in habitat distribution patterns between the dry season and rainy season. We used bivariate point pattern analysis to determine whether the distribution of anopheline-positive habitats in the dry season was more confined than in the rainy season using the statistical software package Programita.<sup>28,31</sup> When it was significantly clustered, we used a *t*-test to compare the average distances from the nearest streams and the average elevations of anopheline-positive habitats between the seasons.

Since occurrence of the *An. funestus* complex larvae is generally limited to larger, semipermanent or permanent habitats with aquatic vegetation,<sup>30,32,33</sup> its habitat distribution pattern may be different from the *An. gambiae* complex. We determined whether the distribution of the *An. funestus* complex-positive habitats was significantly more clustered than that of

the *An. gambiae* complex using bivariate K-function analysis. When it was significantly clustered, we compared the average distances of larval habitats to the nearest streams and the average elevations of larval habitats between the two species complexes. In the analysis for the species complexes, the data from both seasons were included because the number of anopheline habitats during the dry season was not large enough to perform a separate analysis.

Associations between the occurrence of anopheline larval habitats and the land cover types were examined for each season using the chi-square test. The Tukey-type multiple comparison test was used to test differences in proportion of larval occurrence among the land cover types.<sup>34</sup> The association was also examined for each species complex. In the analysis for each species complex, the data from both seasons were included together and road was excluded because few positive sites were found within this land cover type. Logistic regression analysis was used to examine the association be-

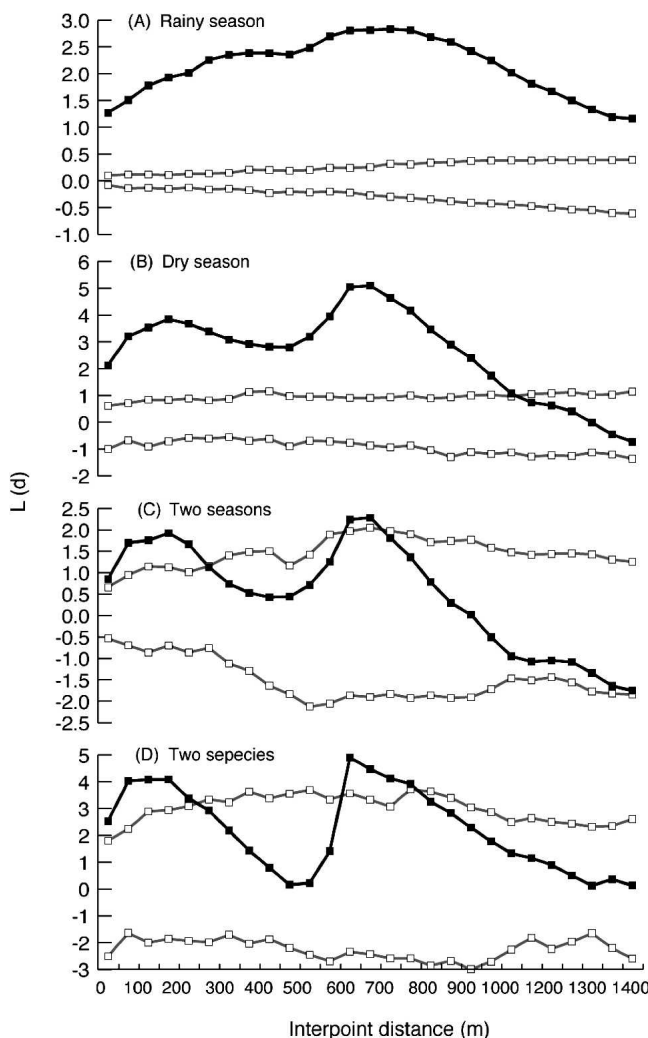


FIGURE 2. Results of K-function analysis on the spatial relationship between anopheline larval habitats and distances to the nearest streams in the rainy season (A) and dry season (B), and results of bivariate K-function analysis comparing the distribution patterns between the rainy and dry seasons (C) and between the *Anopheles gambiae* complex and *An. funestus* (D). The bold lines are the observed value of the test statistic  $L(d)$  at a given distance  $d$ , and the gray lines indicate the 95% confidence intervals.  $m$  = meters.

tween occurrence of each species complex and environmental variables. We further examined whether the characteristics of larval habitats (including both positive and negative sites) varied significantly among the land cover types using two-way analysis of variance for continuous variables and chi-square analysis for categorical variables. Tukey multiple comparison tests were used for post-hoc analyses. The statistical significance was 0.05 and adjusted with the Bonferroni correction if necessary. All continuous variables were log-transformed.

## RESULTS

**Species composition.** Among the 1,021 anopheline larvae collected, 43 (4.2%) were *An. implexus*, and 987 larvae belonged to the *An. gambiae* complex and the *An. funestus* complex that were subjected to further molecular identification. We found 585 (57.3%) *An. gambiae*, 73 (7.1%) *An. arabiensis*, and 122 (11.9%) *An. funestus* s.s. larvae. A total of 207 (20.3%) anopheline larvae were not identified due to PCR amplification failure. Because the proportion of *An. arabiensis* was small, we pooled *An. arabiensis* and *An. gambiae* in the following analyses.

**Relationships with topography.** During the rainy season survey, 767 aquatic habitats were identified in the study area,

and anopheline larvae were found at 277 habitats (36.1%; Table 1 and Figure 1). The number of aquatic habitats was reduced to 190, and 48 habitats (25.3%) were anopheline-positive during the dry season. Figure 2 shows measures of the observed  $L(d)$  and the 95% confidence interval plotted for values of interpoint distance ranging between 0.1 and 1.4 km. For clustering to be significant at the  $P < 0.05$  level, the observed  $L(d)$  must lie above or below the 95% confidence interval lines. We found that anopheline larval habitats were significantly clustered up to 1.4 km during the rainy season, and clustering peaked at 0.7–0.9 km (Figure 2A). During the dry season, larval habitat clustering occurred up to 1.0 km and peaked at 0.7 km (Figure 2B). Bivariate K-function analysis found that the magnitude of the K-function estimate of larval habitats for the dry season was significantly greater than that for the rainy season at distances up to 0.3 km and approximately 0.6 km (Figure 2C). That is, larval habitat during the dry season was significantly more clustered compared with the rainy season at the distances. Similarly, *An. funestus* larval habitats were significantly more clustered than the *An. gambiae* complex habitats in the rainy season (Figure 2D).

Anopheline larval habitats were generally clustered near the streams (Figures 1 and 3). In the rainy season survey, 84.4% of anopheline-positive habitats were located within

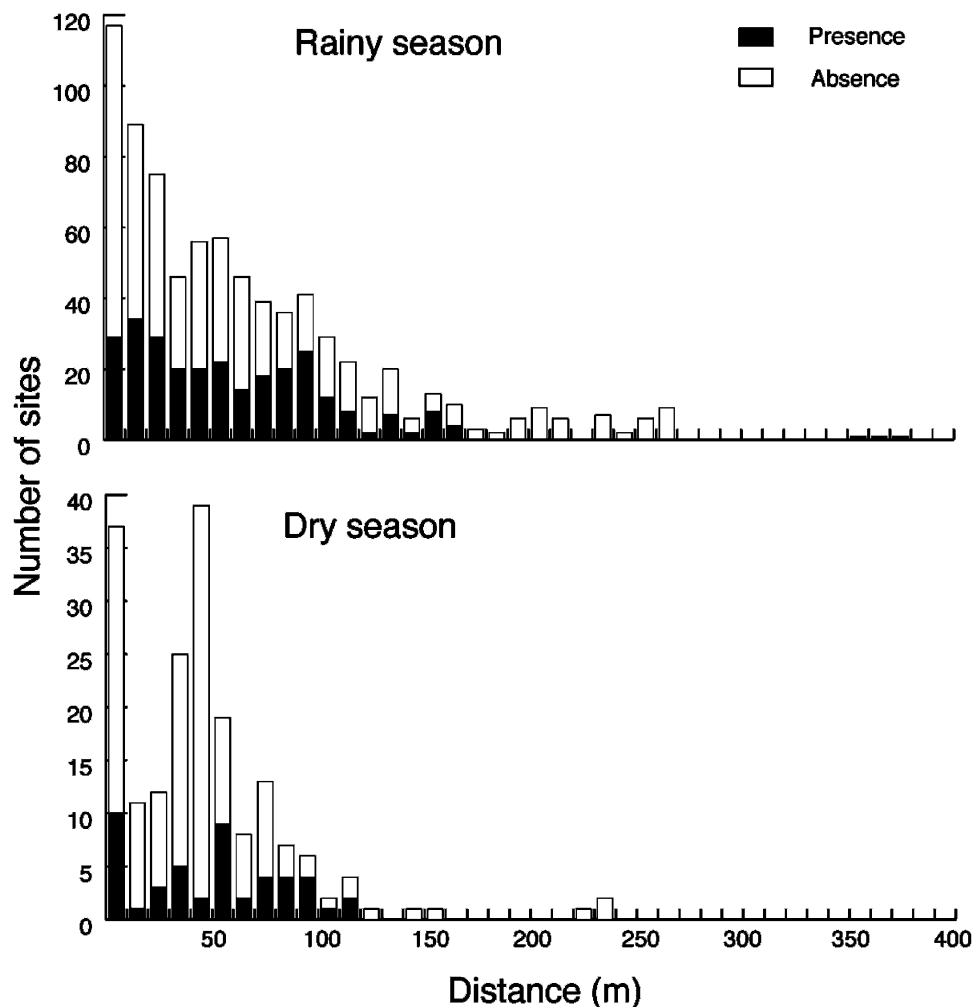


FIGURE 3. Distribution of anopheline larval habitats with respect to the distance to the nearest stream. m = meters.

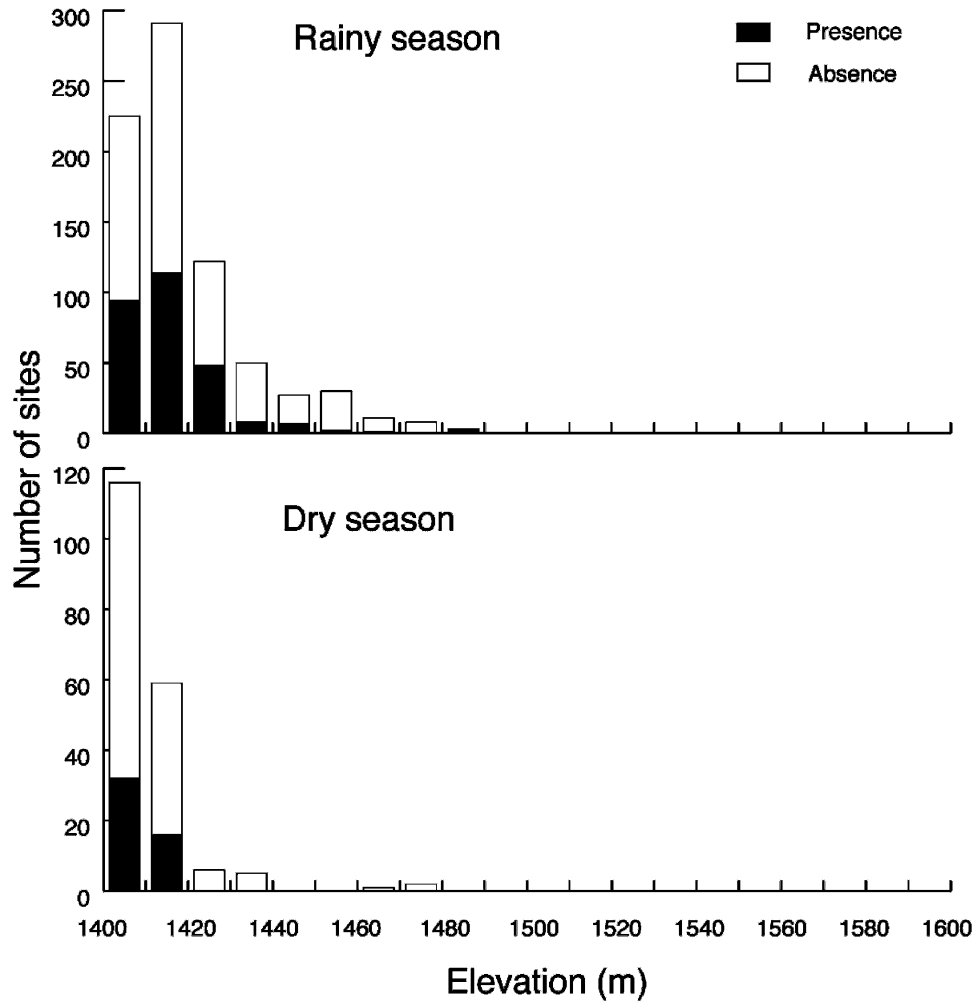


FIGURE 4. Distribution of anopheline larval habitats with respect to elevation. m = meters.

100 meters from streams, whereas 93.6% of positive habitats were distributed within 100 meters in the dry season (Kolmogorov-Smirnov one-sample test:  $Z = 7.63$ ,  $n = 277$ ,  $P < 0.001$  for the rainy season and  $Z = 2.66$ ,  $n = 47$ ,  $P < 0.001$  for the dry season). Larval habitats were also clustered in the lower area; 75.1% and 100% of anopheline-positive habitats were found in the valley bottom within 1,400–1,420-meters range of elevation during the rainy season and dry season, respectively (Figure 4). The average distance of anopheline larval habitats to the nearest stream in the dry season (mean = 54.8 meters, SE = 4.9) was significantly shorter than in the rainy season (mean = 67.0 meters, SE = 4.9,  $t = 4.35$ ,  $P = 0.038$ ). The average elevation of anopheline-positive habitats in the rainy season was significantly higher than in the dry season (rainy season: mean = 1,415.7 meters, SE = 0.68,  $n = 277$  and dry season: mean = 1,408.0 meters, SE = 1.63,  $n = 48$ ,  $t = 4.35$ , degrees of freedom [df] = 323,  $P < 0.001$ ). Logistic regression analysis showed that the occurrence of anopheline larvae was significantly correlated with elevation and season (Table 2).

**Relationships with land cover types.** Farmlands were the major land cover type, covering 59.6% of the study area; this was followed by pastures (20.0%) and forests (17.9%; Table 1). Swamps had little coverage (1.4%). Farm, forest, and pas-

ture were almost equally dominant in the area within 50 meters from streams (Figure 5); however, the proportion of farm to the other land cover types increased with an increase in distance from streams. More than one-third (35.9%) of the total forest cover was present in the area within 50 meters from streams. The aquatic habitats in forests had significantly higher canopy coverage than the habitats in farmlands, pastures, and swamps (Table 3). The proportion of habitats with filamentous algae was significantly higher in farmlands and

TABLE 2

Results of logistic regression analysis determining whether the occurrence of anopheline larvae was correlated with three independent variables, the distance of a habitat from the nearest stream, elevation, and season

	Presence	Absence	P
Distance (m)*	58.6 ± 3.3	60.8 ± 2.4	0.147
Elevation (m)*	1,414.5 ± 0.8	1,418.2 ± 0.6	< 0.001
Season†			< 0.001
Rainy	277 (36.1%)	490 (63.9%)	–
Dry	48 (25.3%)	142 (74.7%)	–
Total number of habitats	325	632	–

\* Values for distance and elevation are means ± SE.

† Values for season are numbers of habitats (%).

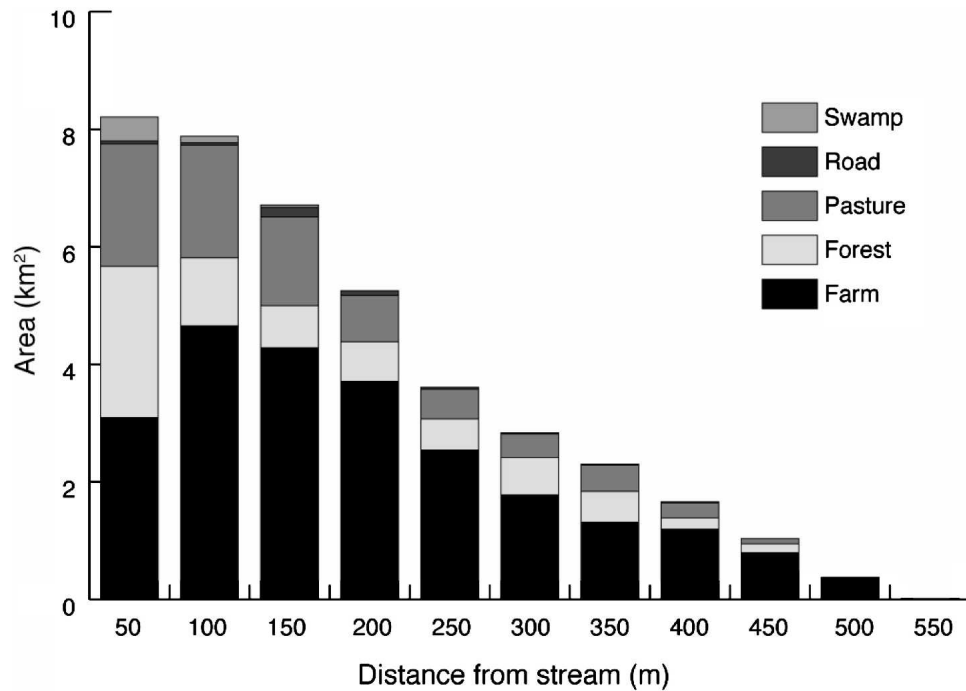


FIGURE 5. Relationships between area of each land cover type and distance from streams. m = meters.

swamps than in forests and pastures. However, the nitrogen and phosphate contents, pH level, and turbidity of aquatic habitats were not significantly varied among the land cover types.

Anopheline-positive habitats were found most in farmlands (39.0%), followed by pastures (26.7%), during the rainy season (Table 1). During the dry season, positive habitats occurred most in the swamps (41.7%), followed by farmlands (33.3%). The average size of habitats in swamps was significantly larger than in the other land cover types (Table 3). The association between the occurrence (presence/absence) of anopheline larvae and land cover types was statistically significant during the rainy season ( $\chi^2 = 36.69$ ,  $df = 4$ ,  $P <$

0.001), but not significant during the dry season ( $\chi^2 = 5.13$ ,  $df = 4$ ,  $P = 0.274$ ; Table 1). In particular, the proportion of positive habitats to negative habitats was highest in the farmlands and pastures in both the rainy season (range = 42.8–44.1%) and the dry season (range = 34.0–35.3%), but lowest in road ditches (16.4% and 11.1% for the rainy and dry seasons, respectively).

We found that the larvae of the *An. gambiae* complex were found more frequently in farmlands and pastures than in larval habitats in the forests ( $\chi^2 = 10.53$ ,  $df = 3$ ,  $P = 0.015$ ), while *An. funestus* larvae were found more frequently in swamps and pastures than in farmlands and forests ( $\chi^2 = 9.85$ ,  $df = 3$ ,  $P = 0.020$ ; Table 4). Among the eight physicochemical variables examined, canopy cover was the only factor significantly associated with the occurrence of the *An. gambiae* complex and *An. funestus* larvae (Table 5). The average canopy cover was significantly less in the *An. gambiae* complex-positive habitats and in the *An. funestus*-positive habitats than in the negative habitats.

TABLE 3  
Characteristics of aquatic habitats found in different land cover types\*

Variables	Farm	Forest	Pasture	Swamp
Canopy cover (%)	33.8 ± 4.1 <sup>a</sup>	87.5 ± 2.4 <sup>b</sup>	21.6 ± 3.9 <sup>a</sup>	18.4 ± 2.8 <sup>a</sup>
Habitat size (m <sup>3</sup> )	18.0 ± 7.9 <sup>a</sup>	25.4 ± 10.0 <sup>a</sup>	11.5 ± 4.0 <sup>a</sup>	70.5 ± 14.4 <sup>b</sup>
Habitat with algae (%)	31.8 <sup>a</sup>	10.5 <sup>b</sup>	6.8 <sup>b</sup>	25.9 <sup>a</sup>
NH <sub>4</sub> <sup>+</sup> (mg/L)	0.89 ± 0.16 <sup>a</sup>	2.00 ± 0.60 <sup>a</sup>	1.03 ± 0.18 <sup>a</sup>	1.99 ± 0.82 <sup>a</sup>
NO <sub>3</sub> <sup>-</sup> (mg/L)	1.11 ± 0.35 <sup>a</sup>	0.66 ± 0.40 <sup>a</sup>	0.62 ± 0.22 <sup>a</sup>	0.17 ± 0.11 <sup>a</sup>
PO <sub>4</sub> <sup>-3</sup> (mg/L)	0.38 ± 0.10 <sup>a</sup>	0.31 ± 0.00 <sup>a</sup>	0.18 ± 0.06 <sup>a</sup>	0.18 ± 0.06 <sup>a</sup>
pH	6.3 ± 0.1 <sup>a</sup>	6.1 ± 0.1 <sup>a</sup>	6.2 ± 0.1 <sup>a</sup>	6.1 ± 0.0 <sup>a</sup>
Turbidity (NTU)	98.5 ± 21.0 <sup>a</sup>	85.9 ± 12.7 <sup>a</sup>	58.5 ± 10.0 <sup>a</sup>	95.2 ± 11.9 <sup>a</sup>
Number of sites	44	38	44	54

\* Values are the mean ± SE except for the proportions (%) of habitats with filamentous algae. The letters following the numerical values indicate the results of Tukey-type multiple comparison tests. The values with the same letter were not statistically significant at the  $P = 0.05$  level after adjustment with the Bonferroni correction. NTU = nephelometric turbidity units.

## DISCUSSION

The present study demonstrated that topographic features and land cover types play important roles in the spatial distribution of anopheline larval habitats. Anopheline larval habitats were generally located near the valley bottom or streams in our study area, which was a typical highland site in western Kenya characterized by hill-valley topography. Larval habitat distribution became more confined toward the valley bottoms during the dry season. Topographic features affect the formation of aquatic habitats. For example, stagnant aquatic habitats are more prevalent in valley bottoms than on hills because it is more difficult for water to accumulate on hill slopes due to surface runoff, whereas in the valley

TABLE 4  
Occurrence of *Anopheles gambiae* complex and *An. funestus* larval habitats in different land cover types\*

	Farmland	Forest	Pasture	Swamp
<i>An. gambiae</i> complex (%)	19 (43.2%) <sup>a</sup>	7 (18.4%) <sup>b</sup>	21 (47.7%) <sup>a</sup>	17 (31.5%) <sup>a</sup>
<i>An. funestus</i> larvae (%)	2 (4.6%) <sup>a</sup>	1 (2.6%) <sup>a</sup>	11 (25.0%) <sup>b</sup>	10 (18.5%) <sup>b</sup>
Total number of habitats	44	38	44	54

\* The letters following the proportion of the *An. gambiae* complex–or *An. funestus*–positive sites indicate the results of the Tukey-type multiple comparison tests. The values with the same letter were not statistically significant at the  $P = 0.05$  level after adjustment with the Bonferroni correction.

bottom stagnant aquatic habitats are formed by means of surface runoff from uphill, and from springs and groundwater seepage. Since the groundwater level is lowered during the dry season, the distribution of stagnant aquatic habitats becomes more confined toward valley bottoms. In our study area, most parts of roads are in the mid-slope of hills where rainwater drains quickly. As a result, in our study area puddles associated with roads were short-lived; in contrast, anopheline larvae are often found in habitats associated with roads in the lower area of the Lake Victoria Basin.<sup>17,32</sup> The Lake Victoria Basin is generally more flat, and anopheline larval habitats are widely dispersed.<sup>29,30</sup>

The results from this study suggest that land cover types affect availability and suitability of aquatic habitats for anopheline larvae. Among the physicochemical variables of aquatic habitats that we examined, we found that canopy cover was the only factor significantly associated with the occurrence of the *An. gambiae* complex and *An. funestus* larvae. Unshaded aquatic habitats occurred more often in farmlands and pastures than forests. Therefore, the *An. gambiae* complex larval habitats were more frequently observed in farmlands and pastures. Larval habitats of the *An. gambiae* complex also commonly occur in temporary habitats in swamp margins in valley bottoms.<sup>18</sup> It is interesting to note that the number of anopheline larval habitats in farmlands and pastures was greatly reduced in the dry season, mostly due to low availability of aquatic habitats. Farmers stop drawing water to farmlands from streams through irrigation ditches during the dry season, and there is little rainfall. *Anopheles funestus* also prefer habitats in unforested areas, but they tend to occur more often in larger, semipermanent or permanent habitats with emergent plants.<sup>30,32,33</sup> The confined distribution of *An. funestus* larval habitats observed in the present study is probably due to limited distribution of long-standing aquatic habitats.

Frequent occurrence of the *An. gambiae* complex larvae in temporary and sunlit habitats may result from a combination of several factors. At least three plausible explanations for this phenomenon are suggested. First, the *An. gambiae* complex females preferentially select small, open habitats for oviposition.<sup>35</sup> Second, larval predation is less prevalent in temporary habitats than in large, permanent habitats.<sup>19,36–38</sup> Third, the *An. gambiae* complex is a typical r-strategist (a species that is opportunistic and reproduces rapidly when density-independent limiting factors are not present), exploiting the increased resources of warmer, open habitats that tend to produce more algae (the main food source for the *An. gambiae* complex) than do shaded habitats.<sup>39</sup> Since maize is a dominant crop in this study area, maize pollen, which is an excellent food source, may also enhance survivorship of the *An. gambiae* complex in open habitats in farmlands.<sup>40,41</sup> Moreover, warmer temperatures encountered in small and open habitats during daytime hours shorten larvae-to-pupae development time, and subsequently larval mortality due to desiccation is reduced.<sup>32</sup> The *An. gambiae* complex may have evolved to exploit these favorable conditions by selecting small and open habitats for oviposition.

Our results suggest that land use practices affect the growing conditions of anopheline larvae and habitat availability in the African highlands. Patches of forest still remain along streams in the Kenyan highlands such as our study area, and clearing these riparian forests will create more larval habitats for the malaria vectors. Assessed from the perspective of larval ecology, our results support the hypothesis that clearing forests leads to an increased incidence of malaria.<sup>15</sup> This study also demonstrated that mapping the distribution of anopheline larval habitats may be used as a foundation for larval control. Our finding on the aggregated distribution of larval habitats in the highland sites suggests that larval control can be targeted to aquatic habitats in farmlands and pastures

TABLE 5  
Characteristics of habitats with/without the *Anopheles gambiae* complex and *An. funestus* larvae, and results of logistic regression analysis determining which variables were correlated with the occurrence of the mosquitoes\*

Variables	<i>An. gambiae</i> complex			<i>An. funestus</i>		
	Presence	Absence	<i>P</i>	Presence	Absence	<i>P</i>
Canopy cover	26.4 ± 3.6	43.7 ± 3.4	0.002	20.7 ± 5.7	40.1 ± 2.8	0.021
Habitat size (m <sup>3</sup> )	25.6 ± 7.4	38.2 ± 7.5	0.525	28.9 ± 8.3	34.5 ± 6.3	0.07
Habitat with algae (%)	18.8	19.8	0.769	9.0	26.8	0.135
NH <sub>4</sub> <sup>+</sup> (mg/L)	1.09 ± 0.15	1.71 ± 0.43	0.808	0.81 ± 0.23	1.60 ± 0.32	0.243
NO <sub>3</sub> <sup>-</sup> (mg/L)	0.13 ± 0.04	0.15 ± 0.04	0.978	0.15 ± 0.07	0.14 ± 0.03	0.991
PO <sub>4</sub> <sup>-3</sup> (mg/L)	0.27 ± 0.06	0.26 ± 0.05	0.937	0.25 ± 0.11	0.27 ± 0.04	0.384
ph	6.3 ± 0.1	6.2 ± 0.0	0.075	6.2 ± 0.1	6.2 ± 0.0	0.527
Turbidity (NTU)	79.4 ± 12.2	88.2 ± 9.1	0.994	49.6 ± 7.2	90.5 ± 8.2	0.089
Number of sites	74	106		26	154	

\* Values are the mean ± SE except for the proportions (%) of habitats with filamentous algae. NTU = nephelometric turbidity units.



near streams or valley bottoms. Larval control may be more effective if implemented during the dry season when the distribution of larval habitats is more strictly confined toward valley bottoms.

Received July 24, 2004. Accepted for publication January 5, 2005.

Acknowledgments: We thank H. Atieli, B. Ndenga, M. Okonji, S. Ajuma, W. Miheso, and P. Lutiali for field assistance, U. Kitron and J. Clennon for assistance with statistical analyses, M. Ombok for assistance with the global positioning system in differential mode, L. Carson for critical comments, and T. Wiegand for providing the statistical software package Programita.

Financial support: This work was supported by National Institutes of Health grants R01 AI 50243 and D43 TW01505.

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