Land Use and Land Cover Changes and Spatiotemporal Dynamics of Anopheline Larval Habitats during a Four-Year Period in a Highland Community of Africa

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Abstract. Spatial and temporal variations in the distribution of anopheline larval habitats and land use and land cover (LULC) changes can influence malaria transmission intensity. This information is important for understanding the environmental determinants of malaria transmission heterogeneity, and it is critical to the study of the effects of environmental changes on malaria transmission. In this study, we investigated the spatial and temporal variations in the distribution of anopheline larval habitats and LULC changes in western Kenya highlands over a 4-year period. *Anopheles gambiae* complex larvae were mainly confined to valley bottoms during both the dry and wet seasons. Although *An. gambiae* larvae were located in man-made habitats where riparian forests and natural swamps had been cleared, *Anopheles funestus* larvae were mainly found in permanent habitats in pastures. The association between land cover type and occurrence of anopheline larvae was statistically significant. The distribution of anopheline positive habitats varied significantly between months, during the survey. In 2004, the mean density of *An. gambiae* was significantly higher during the month of May, whereas the density of *An. funestus* peaked significantly in February. Over the study period, major LULC changes occurred mostly in the valley bottoms. Overall, farmland increased by 3.9%, whereas both pastures and natural swamps decreased by 8.9% and 20.9%, respectively. The area under forest cover was decreased by 5.8%. Land-use changes in the study area are favorable to *An. gambiae* larval development, thereby risking a more widespread distribution of malaria vector habitats and potentially increasing malaria transmission in western Kenya highlands.

INTRODUCTION

Malaria transmission intensity is spatially heterogeneous as a result of heterogeneities in vector abundance and capacity, human-vector contact rates, and other human host-related factors.¹ Adult vector abundance is positively associated with the availability and productivity of aquatic habitats, and the proximity of larval habitats to human dwellings has been found to be an important determinant of the risk of malaria transmission.² The highest malaria-risk areas are normally found within just a few hundred meters of the major larval habitats.^{3–5}

In the East African highlands there has been a recent increase in the regularity of malaria epidemics.⁶⁻⁸ Several mechanisms have been proposed to explain the emergence or re-emergence of epidemic malaria in the highlands, including land use change, demographic changes, climate variability, drug resistance, and cessation of mosquito and malaria control.9-13 These highland sites are fragile ecosystems and have come under pressure from rapid human population increases, and the associated increase in deforestation and farming. The unprecedented increase in the human population in the African highlands has induced dramatic changes in land use and land cover (LULC) and human settlement patterns. It has been suggested that these LULC changes have facilitated the transmission of malaria at several African highland sites.9-12 Recent studies in western Kenya highlands have shown that larval habitats in the deforested areas and cultivated swamps exhibited increased larval survivorship of Anopheles gambiae compared with larvae in the forested area.^{14,15} It is therefore likely that these modifications in land cover types will influence the spatial and temporal distribution of anopheline larval habitats and consequently affect malaria transmission.

*Address correspondence to Stephen Munga, Kenya Medical Research Institute, Centre for Global Health Research, Box 1578, Kisumu 40100. E-mail: munga_os@yahoo.com Although the temporal dynamics of anopheline larval habitats and adult mosquito abundance is significantly correlated with rainfall,¹⁶ the spatial distribution of larval habitats is often constrained by topography and water drainage, and modulated by LULC.^{5,14} For example, during a 2-year survey in Eritrea more than 90% of anopheline larvae were found in artificial aquatic habitats such as man-made stream edges, streambed pools, and drainage channels at communal water supply points.⁵ In western Kenya highlands, reduced canopy cover was significantly correlated with the increased occurrence, and shortened development times, of *An. gambiae* larvae.¹⁴ In urban Kisumu, western Kenya and urban Malindi, coastal Kenya, an estimated 30% of the area had experienced LULC changes during a period of 12 to 14 years (1980s and 1990s) based on Landsat satellite images of 28.5-m spatial resolution.¹⁷

In this study, we determined LULC changes over a period of 4 years in a 16 km² area in western Kenya highlands, using 1-m spatial resolution Ikonos images in combination with ground surveys. We then examined the spatial and temporal dynamics in the distribution of anopheline larval habitats to determine the impact of rainfall and LULC changes. This information is critical to our understanding of environmental determinants of malaria transmission heterogeneity at a micro-geographical scale. Furthermore, information on the spatial and temporal dynamics of immature stages of malaria vectors and the associated underlying factors may lead to the development of more cost-effective control strategies involving the targeted management of productive habitats.

MATERIALS AND METHODS

Study site. We established a study area (approximately $4 \times 4 \text{ km}^2$) at Iguhu village ($34^{\circ}45'E$ and $0^{\circ}10'N$) in Kakamega district, western Kenya. The elevation of the study site ranges from 1,420 to 1,580 m, and the area is transected by the Yala River.¹⁸ The average minimum and maximum monthly temperatures during 1970–2000 are 13.8 and $28^{\circ}C$ with the

hottest season in January–February and the coolest season in June–July. The average annual rainfall during this period was approximately 1,900 mm. Peak rainfall occurs between April and June followed by a shorter rainy season in October and November. The area has small patches of forests along the river and streams, which are remnants of a larger forest that has been cleared for cultivation and pasture.¹⁹ Steep and gently sloping hills and undulating topography characterizes the area.

Determination of LULC changes. This study examined the LULC changes in the study site between 2002 and 2006. A multispectral (blue, green, red, and infrared) Ikonos image with one meter ground resolution (taken on April 12, 2002) was used to classify the LULC types with a supervised classification method. The image was geometrically and radiometrically corrected to account for topographic distortions and atmospheric effects. A total of seven land-cover classes were used: farmland, pasture, forest, natural swamp, river/streams, shrubs, and road. Farmland was characterized either by the presence of an agricultural crop or bare ground that had been prepared for planting crops. Pasture was either grassland used for grazing or an area with a mixture of grass and shrubs. Natural swamp was characterized by the presence of emergent aquatic plants. Forest referred to areas with dense tree cover, normally with a closed canopy. Streams were classified as waterways less than one meter wide, whereas rivers were more than one meter wide. Each of the seven land-cover classes was determined based on their spectral signatures (expressed in terms of color or grayness), texture (the smoothness of the object), and structure (the spatial arrangement). Ground truthing was conducted in both dry and rainy seasons by direct field inspection of 185 points randomly selected by a script for random point generation in ArcGIS software (ESRI, Redlands, CA). In February-March 2006, the land use and land cover patterns in the study site were assessed through thorough ground investigations covering the entire study area. Ground truthing was done by surveying the entire study site while identifying the changes in land use. We used ground truthing because we could not find a cloud-free Ikonos image for April either in 2006 or 2007. In the places where land use had occurred, we marked the changes using polygons generated with a handheld global positioning system (GPS). These polygons were later downloaded to a computer using OziExplorer software (Des Newman, CA) and transferred to ArcView 3.3 (ESRI). Changes in LULC were determined by comparing the LULC types recorded in 2002 with the types observed on the ground. The changes were updated on the 2002 digital map in ArcGIS and the surface area of LULC change estimated.

Temporal and spatial dynamics of anopheline larval habitats. We conducted ground surveys on the spatial distribution of aquatic habitats and anopheline-positive habitats in the study area in the following months: November 2002; February and May 2003; February, May, August, November of 2004; and February 2005. We have previously reported the spatial distribution of anopheline larval habitats in May and August of 2002 in a 9 km² area within the study site.¹⁴ From April 2005 we implemented a vector control program in the study area, and the resulting larval and adult distribution will be reported elsewhere. In each sampling season, all aquatic habitats (excluding water containers in houses and tree holes) were thoroughly searched and their locations were recorded using a handheld Trimble GPS unit (Trimble Lavigation Limited, Sunnyvale, CA). The length and width of each habitat were measured. Land cover types and aquatic habitat locations were classified into the previous seven categories. Each aquatic habitat was examined for the presence of mosquito larvae using a 350 mL standard dipper. 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. Specimens that could not be identified in the field were taken to the laboratory for microscopic identification of species using morphological keys.^{20,21} In this study, we did not conduct polymerase chain reaction (PCR) analysis for members of *Anopheles gambiae* complex to the species level as our previous study at the same study site found that *A. gambiae s.s.* was the predominant species.¹⁴ In the larval distribution surveys from February 2004 onward, we counted the total number of *An. gambiae* and *Anopheles funestus* larvae in the dips, total water volume sampled, and total surface area of each habitat.

Statistical analysis. We determined the changes in land use and land cover over a period of 4 years using Arview 3.3. The changes in LULC were obtained by comparing the area of each land cover type in 2002 with that of February 2006. The surface area of each land cover type was estimated by first projecting the land-cover layer into WGS_84 Universal Transverse Mercator (UTM), zone 36 N, and then using an Avenue script that calculates the surface area in ArcView. We then estimated the distance of each breeding site from the nearest stream using geographic information systems (GIS). The distance of each breeding habitat to the nearest hydrological network (stream/Yala river) was estimated by changing the projection of both the geo-referenced habitats and the digitized hydrological network from GCS_WGS 1984 decimal degrees into UTM 36N projection using ArcView 3.3.

We tested for the randomness of the distribution of anopheline positive breeding sites, using the χ^2 test. We calculated the observed distribution of larval habitats within different intervals to the nearest streams/rivers, and the expected distribution of larval habitats based on the Poisson distribution. The χ^2 statistic was then computed. A significant departure from the Poisson distribution suggests a non-random distribution of larval habitats. Logistic regression analysis was used to test the association between the occurrence of anopheline larvae and land cover type. We also examined whether the density of *An. gambaie s.l.* and *An. funestus s.l.* larvae varied among seasons for the five samplings between February 2004 and February 2005 using one-way analysis of variance (ANOVA). When the differences were significant, we used Tukey-Kramer multiple comparison tests for post-hoc analyses.

RESULTS

LULC changes during the four-year period. At the beginning of the study period (2002) farmland was the major land use type making up 64.7% of the study area, followed by pasture (13.0%), forests (11.4%), shrubs (8.3%), swamp (0.8%), and road (0.9%). In February 2006, farmland area size had increased by 339,000 m², a 3.9% increase (Table 1), whereas forest, pasture, shrubs, and swamp area had reduced. The highest change in land use and land cover occurred in pasture, a reduction of 157,000 m², followed by forest (a reduction of 89,000 m²). Although the reduction of swamp area was only 22,000 m², it represented a 20.9% decrease. These LULC changes mainly occurred along the rivers/streams (Figure 1). Overall, the predominant land use change was the conversion of other land use and land covers into farmland (Table 1).

TABLE 1 Change of land use and land cover during a 4-year period (2002–2006) in the study site in Iguhu, Kakamega, western Kenya

Land cover	Area size in 2002* (%)	Area size in 2006* (%)	Changes in area size* (% as of original land use land cover type)
Farmland	8,733 (64.7)	9,072 (67.2)	+339 (+2.5%)
Forest	1,533 (11.4)	1,444 (10.7)	-89 (-0.66%)
Pasture	1,750 (13.0)	1,593 (11.8)	-157 (-1.2%)
River/streams	136 (1.0)	136 (1.0)	0(0%)
Road	119 (0.9)	119 (0.9)	0(0%)
Shrubs	1,117 (8.3)	1,046 (7.8)	-71 (-0.53%)
Swamp	105 (0.8)	83 (0.6)	-22 (-0.16%)
Total	13,493 (100)	13,493 (100)	0

*Area size is in 1,000 m².

Mosquito species composition. We sampled a total of 12,276 anopheline and 33,857 culicine larvae during the five surveys between February 2004 and February 2005. Among the anopheline larvae 9,977 (81.3%) were identified morphologically as *An. gambiae s.l.*; 2,049 (16.7%) as *An. funestus s.l.*; 219 (1.78%) as *Anopheles squamosus*; 21 (0.2%) as *Anopheles implexus*; and 10 (0.08%) as *Anopheles coustani* (Table 2). Higher *An. gambiae* abundance was observed during rainy seasons compared with dry seasons, whereas *An. funestus* larval abundance was quite stable (Figure 2). For example, a total of 5,027 *An. gambiae s.l.* larvae were collected in May 2004 (the long rainy season), about 4–8-fold higher than the dry seasons.

Temporal and spatial dynamics of anopheline larval habitats. The number of aquatic habitats was strongly associated with the amount of rainfall (Figure 3). The highest number of aquatic habitats (1,918) was recorded during May 2003, the long rainy season. Out of these aquatic habitats, 41% were positive for anopheline larvae (Table 3). The fewest habitats (469) were recorded during February 2005 (the dry season) and only 24% of these were positive for anopheline larvae. Overall, a total of 2,583 aquatic habitats were identified during wet and dry seasons (May and February) of 2003 compared with 2,090 aquatic habitats over the same seasons in 2004. The proportion of anopheline-positive habitats also varied

TABLE 2 Numbers and species of anopheline mosquito larvae collected during surveys between February 2004 and February 2005

Month and year	Anopheles gambiae s.l.	Anopheles funestus s.l.	Anopheles coustani	Anopheles implexus	Anopheles squamosus
February 2004	1,461	735	2	18	38
May 2004	5,027	378	4	3	48
August 2004	660	334	0	0	51
November 2004	2,415	352	0	0	74
February 2005	414	253	4	0	8
Total	9,977	2049	10	21	219

significantly among seasons ($\chi^2 = 162.6$, degrees of freedom [df] = 7, P < 0.001). For example, 42% of aquatic habitats were positive for anopheline larvae during wet and dry seasons in 2003, whereas only 32% of the habitats were positive in wet and dry seasons of 2004. One-way ANOVA found significant among-season differences in the density of *An. gambiae s.l.* larvae (F = 9.63, df = 4, 4626, P < 0.001) and of *An. funestus s.l.* larvae (F = 3.63, df = 4, 4626, P = 0.006) (Table 4).

Relationship between anopheline-larval occurrence and land use and land cover types. The association between land cover type and presence of anopheline larvae was statistically significant (P < 0.05) for all the months that the surveys were done except for August 2004 and February 2005 (Table 3). Overall, the highest proportions of anopheline positive habitats occurred in pastures (33%) and farmlands (32%), followed by swamp habitats (23%). Roads had the least number of anopheline positive habitats (15%), whereas habitats in forests had an 18% positive rate.

DISCUSSION

The western Kenya highlands have experienced a dramatic increase in human population because of immigration and high birth rate. It is estimated that the population of Kenya has doubled since the 1980s.²² The increase in population has led to unprecedented land-use changes in the highlands.



FIGURE 1. Land use and land cover changes in the study site in Iguhu, Kakamega, western Kenya between 2002 and 2006. Only areas that exhibited changes in land use and land cover (LULC) over the study period are presented. This figure appears in color at www.ajtmh.org.



FIGURE 2. Dynamics of the spatial distribution of anopheline larval habitats in the study area in Iguhu, Kakamega, western Kenya. Sampling was conducted in **A**, November 2002; **B**, February 2003; **C**, May 2003; **D**, February 2004; **E**, May 2004; **F**, August 2004; **G**, November 2004; and **H**, February 2005. This figure appears in color at www.ajtmh.org.

These land-use changes mainly occur in the form of deforestation and conversion of other land use types into farmlands to increase agricultural production. For example, since 1965 the Malava Forest in the Kakamega district has been reduced from 600 to less than 100 hectares.²³ Most rain forests have been cleared for crop planting, cattle grazing, commercial logging, firewood collection, and housing construction.¹⁹ In this analysis, we have shown that farmlands comprised the major land use type in the study area, followed by pasture. This study also shows that changes in LULC frequently occurred in the valley bottoms where large regions of natural swamps, forest, and pastures were converted into farmlands. The alteration of other LULCs into farmland and pastures represented 86.2% and 16.7% of the total changes that occurred during the study period.

Land use and land cover was significantly associated with the occurrence of anopheline larval habitats. *Anopheles gambiae* and *An. funestus* larvae frequently occurred in open and sunlit habitats in farmlands and pastures. Land cover type can influence the suitability and availability of anopheline larval habitats through its effects on temperature and food conditions.^{14,15} Habitats created by deforestation, cultivation of natural swamps, and cow hoof prints were found to be the most preferred breeding habitats in the study area. Previously, we found that mosquito larval survivorship was higher in such habitats.²⁴ Consistent to this finding we further showed that *An. gambiae* adults emerged exclusively from habitats in the open areas with an estimated productivity of 1.82 mosquitoes/ m²/week. The mosquito pupation rate in farmland habitats was significantly greater than in swamp and forest habitats and larval-to-pupal development times were significantly shorter.¹⁵ Land cover type may affect larval survivorship and adult productivity through its effects on water temperature and nutrients in the aquatic habitats, which may enhance pupation rates and shorten development times. Moreover, land cover type can also affect habitat characteristics, which are preferred by malaria vectors for oviposition. For example, we have previously demonstrated that gravid *An. gambiae* prefer to oviposit



FIGURE 3. Monthly variation in the amount of precipitation in Iguhu, Kakamega, western Kenya between 2002 and 2006. Rainfall variation is expressed as the percentage of deviation from the average during the study period (150.1 mm), i.e., (monthly rainfall - 150.1)/150.1 * 100.

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Land use type	Nov-02	Feb-03	May-03	Feb-04	May-04	Aug-04	Nov-04	Feb-05	Overall
Farm	0.42	0.49	0.45	0.28	0.39	0.11	0.22	0.21	0.32
Forest	0.22	0.38	0.09	0.13	0.14	0.14	0.15	0.20	0.18
Pasture	0.36	0.47	0.32	0.35	0.45	0.15	0.29	0.31	0.33
Road	0.00	0.33	0.13	0.00	0.29	0.08	0.36	0.00	0.15
Swamp	0.16	0.44	0.27	0.19	0.33	0.09	0.17	0.15	0.23
χ^{2*}	72.98	10.5	103.87	24.31	39.88	5.87	23.68	8.36	9.21
df†	4	4	4	4	4	4	4	4	4
Р	< 0.001	0.033	< 0.001	< 0.001	< 0.001	0.209	< 0.001	0.079	= 0.003

 TABLE 3

 Proportion of the larval habitats positive for anopheline larvae under different land cover types along with logistic regression analysis results

* χ^2 value indicates the results of logistic regression analysis for testing the association between occurrence of anopheline larvae in aquatic habitats and land use and land cover (LULC) types. † df = degrees of freedom.

in clear water found in open areas.²⁵ Consequently, larvae of *An. gambiae* complex were frequently observed in farmland and pasture habitats. *Anopheles gambiae* is an r-strategist, which exploits resources in open and warmer sunlit habitats,²⁰ and therefore has a large reproductive potential in areas predominated by farmlands and pastures.

Land use and land covers can have a profound impact on the vectorial capacity of anopheline mosquitoes. For example, LULC changes influence survivorship and biting frequency of An. gambiae.²⁶ Significant increases in the net reproductive rate of mosquitoes in the deforested area suggest that deforestation enhances mosquito reproductive fitness, increasing mosquito population growth potential in the western Kenya highlands. Deforestation was also found to shorten the sporogonic development time of Plasmodium falciparum in An. gambiae mosquitoes.²⁷ Houses in deforested areas had higher indoor temperatures, and the overall infection rate of mosquitoes housed in deforested areas was increased compared with that in forested areas. Vectorial capacity was estimated to be 77.7% higher in the deforested areas than in the forested areas of the same altitude,26 thereby emphasizing the trend of deforestation in western Kenya highlands leading to an increase in malaria epidemics. The association between infectious disease emergence and land-use changes is well established.²⁸ In the Amazon basin Vittor and others²⁹ concluded that deforestation and associated ecologic alterations are conducive to A. darlingi larval presence, and thereby increase malaria risk. In another study, Vittor and others³⁰ showed that A. darlingi displayed significantly increased human-biting activity in areas that have undergone deforestation and development associated with road development. While in the lowland areas of western Kenya, Mutuku and others³¹ found that larval habitats of Anopheles vectors of human malaria were associated with certain land cover types largely of human-modified and fragmented landscape consisting of agricultural and domestic land uses.

In this study, we showed consistent spatial clustering of larval habitats among dry and rainy seasons and among various years during the study period despite seasonal variability in the total number of aquatic habitats and proportion of anopheline positive habitats. Larval habitats were generally concentrated in the valley bottoms nearby the river or streams. This spatial clustering is likely caused by the hilly and gently sloping topography of the study area and that is characteristic of western Kenya highlands. Topographic features have previously been suggested to influence the formation of aquatic larval habitats and malaria transmission.^{14,32} The observed differences in the number of positive larval habitats between dry and rainy seasons likely reflect adult mosquito abundance and the availability of aquatic habitats, which are strongly influenced by rainfall. During the dry season, fewer larval habitats were available and they were confined to the valley bottoms, whereas aquatic habitats were more widely distributed during the rainy season.

Our findings on the impact of land use and land covers on the availability and suitability of anopheline larval habitats and habitat productivity have important implications on malaria management in the western Kenya highlands. First, the current trend of deforestation in the highlands can increase vectorial capacity of mosquitoes and the risk of epidemic malaria. Thus, forest preservation and new agricultural practices that maintain or increase agricultural productivity while reducing malaria vector survival are vital. Second, our results on the spatial and temporal variations in the distribution of anopheline larval habitats suggest that focal vector control operations could be targeted to habitats with higher larval densities during the dry season when anopheline positive habitats are strongly clustered spatially and are more limited in number. Larval habitats were mainly aggregated in the valley bottoms indicating that local inhabitants living closer to the valley bottoms are at a greater risk of human-vector contact.33 Treatment of such habitats using larvicides, possibly coupled with careful land management practices, and allowing riparian vegetation and swamp restoration may be a cost-effective mechanism for reducing malaria vector abundance and consequently reduce malaria transmission in western Kenya highlands.

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IABLE 4 Mean densities (mean + standard error) of Anopheles gambiae s.L and Anopheles funestus s.L larvae in aquatic habitats						
Month	February 2004	May 2004	August 2004	November 2004	February 2005	
A. gambiae s.l. A. funestus s.l.	$\begin{array}{c} 0.09 \pm 0.02^{a} \\ 0.04 \pm 0.10^{a} \end{array}$	$\begin{array}{c} 0.35 \pm 0.05^{b} \\ 0.03 \pm 0.00^{ab} \end{array}$	$\begin{array}{c} 0.08 \pm 0.03^{a} \\ 0.02 \pm 0.00^{b} \end{array}$	$\begin{array}{c} 0.19 \pm 0.04^{a} \\ 0.02 \pm 0.00^{b} \end{array}$	$0.05 \pm 0.01^{\circ}$ $0.02 \pm 0.01^{\circ}$	

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Numbers followed by the same letter are not significantly different at P < 0.05 by Tukey-Kramer multiple comparison tests for post-hoc analyses.

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REFERENCES

- Greenwood BM, 1989. The micro-epidemiology of malaria and its importance to malaria control. *Trans R Soc Trop Med Hyg 83:* 25–29.
- Awono-Ambene HP, Robert V, 1999. Survival and emergence of immature *Anopheles arabiensis* mosquitoes in market-gardener wells in Dakar, Senegal. *Parasite 6*: 179–184.
- Carter R, Mendis KN, Roberts D, 2000. Spatial targeting of interventions against malaria. *Bull World Health Organ* 78: 1402–1411.
- Edillo FE, Toure YT, Lanzaro GC, Dolo G, Taylor CE, 2002. Spatial and habitat distribution of *Anopheles gambiae* and *Anopheles arabiensis* (Diptera: Culicidae) in Banambani village, Mali. J Med Entomol 39: 70–77.
- Shililu J, Ghebremeskel T, Seulu F, Mengistu S, Fekadu H, Zerom M, Ghebregziabiher A, Sintasath D, Bretas G, Mbogo C, Githure J, Brantly E, Novak R, Beier JC, 2003. Larval habitat diversity and ecology of anopheline larvae in Eritrea. J Med Entomol 40: 921–929.
- Malakooti MA, Biomndo K, Shanks GD, 1998. Reemergence of epidemic malaria in the highlands of western Kenya. *Emerg Infect Dis 4*: 671–676.
- Lindblade KA, Walker ED, Onapa AW, Katungu J, Wilson ML, 1999. Highland malaria in Uganda: prospective analysis of an epidemic associated with El Nino. *Trans R Soc Trop Med Hyg* 93: 480–487.
- Shanks GD, Biomndo K, Hay SI, Snow RW, 2000. Changing patterns of clinical malaria since 1965 among a tea estate population located in the Kenyan highlands. *Trans R Soc Trop Med Hyg* 94: 253–255.
- Lindblade KA, Walker ED, Onapa AW, Katungu J, Wilson ML, 2000. Land use change alters malaria transmission parameters by modifying temperature in a highland area of Uganda. *Trop Med Int Health 5:* 263–274.
- Lindsay SW, Martens WJM, 1998. Malaria in the African highlands: past, present and future. *Bull World Health Organ 76:* 33–45.
- Mouchet J, Manguin S, Sircoulon J, Laventure S, Faye O, Onapa AW, Carnevale P, Julvez J, Fontenille D, 1998. Evolution of malaria in Africa for the past 40 years: impact of climatic and human factors. J Am Mosq Control Assoc 14: 121–130.
- Githeko AK, Lindsay SW, Confalonieri UE, Patz JA, 2000. Climate change and vector-borne diseases: a regional analysis. *Bull World Health Organ* 78: 1136–1147.
- Zhou G, Minakawa N, Githeko AK, Yan G, 2004. Spatial distribution patterns of malaria vectors and sample size determination in spatially heterogenous environments: a case study in the western Kenya highland. J Med Entomol 41: 1001–1009.
- 14. Minakawa N, Munga S, Atieli FK, Mushinzimana E, Zhou G, Githeko AK, Yan G, 2005. Spatial distribution of anopheline larval habitats in western Kenya highlands: effects of land cover types and topography. *Am J Trop Med Hyg* 73: 157–165.
- Munga S, Minakawa N, Zhou G, Mushinzimana E, Okeyo-Owuor JB, Githeko AK, Yan G, 2006. Association between land cover

and habitat productivity of malaria vectors in western Kenyan highlands. *Am J Trop Med Hyg* 74: 69–75.

- Hii JLK, Smith T, Mai A, Mellor S, Lewis D, Alexander N, Alpers MP, 1997. Spatial and temporal variation in abundance of *Anopheles* (Diptera: Culicidae) in a malaria endemic area in Papua New Guines. J Med Entomol 34: 193–205.
- Jacob B, Regens JL, Mbogo CM, Githeko AK, Keating J, Swalm CM, Gunter JT, Githure JI, Beier JC, 2003. Occurrence and distribution of *Anopheles* (Diptera: Culicidae) larval habitats on land cover change sites in urban Kisumu and urban Malindi, Kenya. J Med Entomol 40: 777–784.
- Zhou G, Minakawa N, Githeko AK, Yan G, 2004a. Association between climate variability and malaria epidemics in the East African highlands. *Proc Natl Acad Sci USA 101*: 2375–2380.
- Whitmore TC, 1997. Tropical forest disturbance, disappearance, and species loss. Laurence WL, Bierregaard Jr RO, eds. *Tropical Forest Remnants*. Chicago, IL: University of Chicago Press, 2–28.
- Gillies MT, De Meillon B, 1968. The Anopheline of Africa South of the Sahara. Johannesburg: The South African Institute for Medical Research.
- 21. Gillies MT, Coetzee M, 1987. A Supplement to the Anopheline of Africa South of Sahara. Johannesburg: The South African Institute for Medical research.
- 22. Nations U, 2003. *World Population Prospects: The 2002 Revision*. New York: United Nations.
- Brooks TM, Pimm SL, Oyugi JO, 1999. Time lag between deforestation and bird extinction in tropical forest fragments. *Conserv Biol* 13: 1140–1150.
- Munga S, Minakawa N, Zhou G, Githeko AK, Yan G, 2007. Survivorship of immature stages of *Anopheles gambiae s.l.* (Diptera: Culicidae) in natural habitats in western Kenya highlands. *J Med Entomol* 44: 758–764.
- Munga S, Minakawa N, Zhou G, Okeyo-Owuor JB, Githeko AK, Yan G, 2005. Oviposition site preference and egg hatchability of *Anopheles gambiae*: effects of land cover types. *J Med Entomol* 42: 993–997.
- 26. Afrane Y, Zhou G, Lawson BW, Githeko AK, Yan G, 2006. Effects of microclimatic changes caused by deforestation on the survivorship and reproductive fitness of Anopheles gambiae in western Kenya highlands. Am J Trop Med Hyg 74: 772–778.
- Afrane Y, Little TJ, Lawson BW, Githeko AK, Yan G, 2008. Deforestation and vectorial capacity of *Anopheles gambiae* Giles mosquitoes in malaria transmission, Kenya. *Emerg Infect Dis 14*: 1533–1538.
- 28. Patz J, Daszak P, Tabor G, Aguirre A, Pearl M, Epstein J, Wolfe N, Kilpatrick A, Foufopoulos J, Molyneux D, Bradley D, 2004. Unhealthy landscapes: policy recommendations on land use change and infectious disease emergence. *Environ Health Perspect 112*: 1092–1098.
- 29. Vittor AY, Pan W, Gilman RH, Tielsch J, Glass G, Shields T, Sánchez-Lozano W, Pinedo VV, Salas-Cobos E, Flores S, Patz JA, 2009. Linking deforestation to malaria in the Amazon: characterization of the creeding habitat of the principal malaria vector, *Anopheles darling. Am J Trop Med Hyg 81:* 5–12.
- 30. Vittor AY, Gilman RH, Tielsch J, Glass G, Shields T, Sánchez-Lozano W, Pinedo-Cancino V, Patz JA, 2006. The effect of deforestation on the human biting rate *Anopheles darling*, the primary vector of falciparum malaria in Peruvian Amazon. *Am J Trop Med Hyg* 74: 3–11.
- 31. Mutuku FM, Bayo NM, Hightower AW, Vulule JM, Gimnig JE, Mueke JM, Amimo FA and Walker ED, 2009. A supervised land cover classification of a western Kenya lowland endemic for human malaria: associations of land cover with larval *Anopheles* habitats. *Int J Health Geogr 8*: 19.
- 32. Githeko AK, Ayisi JM, Odada PK, Atieli FK, Ndenga BA, Githure JI, Yan G, 2006. Topography and malaria transmission heterogeneity in western Kenya highlands: prospects for focal vector control. *Malar J 5*: 107.
- Munyekenye OG, Githeko AK, Zhou G, Mushinzimana E, Minakawa N, Yan G, 2005. *Plasmodium falciparum* spatial analysis, western Kenya highlands. *Emerg Infect Dis* 11: 1571–1577.