

1 **Appendix**

2 **Method details**

3 *16s-rRNA gene amplicon sequencing of water samples from natural oviposition sites*

4 In La Lopé, after aliquoting a subset of water samples into formaldehyde solution, we kept the
5 water in -20 °C in the field until returning to CIRMF, Franceville, Gabon. We centrifuged the samples
6 after thawing using a Beckman TJ-6 centrifuge (Beckman Coulter, USA) at 6000 rpm (maximum speed)
7 for 30 minutes to collect microbial cells. The liquid was removed immediately after centrifuge, and we
8 extracted DNA using the QIAGEN Blood and Tissue kit (QIAGEN, USA) following the manufactural
9 protocol. The DNA was stored at -20 °C until brought back to the lab at Yale University. In Rabai, due to
10 lack of access to centrifuge, we instead used a filtering approach to collect microbiome from water
11 samples. Specifically, upon bringing the water samples back to the field station, we pushed around 50 mL
12 water of each sample using a sterile syringe through a Millipore Sterivex filter unit (SVGPL10RC, EMD
13 Millipore, USA) with 0.22 µm pore size to collect all microbial cells. We then sealed the filtering unit and
14 frozen them at -20 °C until bringing them back to Yale University. Under aseptic conditions, we peeled
15 the members from the filter unites and extracted DNA from them using the DNeasy PowerWater Kit
16 (QIAGEN, USA) according to the manufactural protocol.

17 We followed the same protocol as in Kozich et al. (2013) to prepare two sequencing libraries for
18 samples collected in La Lopé and Rabai, respectively. The protocol amplified the V4 region of the
19 bacterial 16s-rRNA gene with dual indexes, which allows us to multiplex multiple samples in one
20 sequencing. The primers were prepared in Dr. Andrew Goodman's lab at Yale University as Glden et al.
21 (2017). PCR was conducted using the Q5 High-Fidelity DNA Polymerase (New England Biolabs Inc.,
22 USA) with a similar PCR cycle in Kozich et al. (2013). The amplification products were cleaned using
23 SPRI beads (AMPure XP, Beckman Coulter, USA), and we determined the DNA concentrations by Qubit
24 (Qubit Fluorometric and dsDNA HS Assay Kit, Thermal Fisher, USA). We then mixed the samples with

25 equal quantity. To evaluate the sequencing and analysis performance, we added four positive controls into
26 the mixed sequencing library, including two samples of genomic DNA from Microbial Mock Community
27 B (HM-276D and HM-277D, BEI Resources, NIAID, NIH as part of the Human Microbiome Project)
28 (Nelson, Morrison, Benjamino, Grim, & Graf, 2014), one genomic DNA from ZymoBIOMICS (D6305)
29 and one mixed community cells from ZymoBIOMICS (D6300). The final library was then examined on
30 Bioanalyzer (Thermo Fisher, USA) to confirm the amplicon size and sent to the Yale Center for Genome
31 Analysis for sequencing using Illumina MiSeq (Illumina, USA).

32

33 *Characterizing the volatile chemical profiles of oviposition sites in Rabai, Kenya*

34 We collected 8-15 mL water samples from a subset of oviposition sites (Table 1) in glass vials
35 previously washed and baked at 400 °C. An air pump (Casella Apex Pro, UK) was used to generate an
36 airflow at 0.2 L/min to extract the volatiles from the water in the vials for 24 hours. The chemicals in the
37 airflow leaving the vials were captured using Volatile Collection Trap (Volatile Collection Trap LLC,
38 USA) with PoraPak-Q and later eluted in 200 µL Hexane (Sigma-Aldrich, USA). 1-Bromoheptane
39 (B67570, Sigma-Aldrich, USA) were mixed in the Hexane as an internal standard with a concentration of
40 100 ng/µL. We started volatile extraction in the evening of each collection day, which is less than 12
41 hours from the water collection. An empty vial was used every day as a positive control to characterize
42 the background chemical profile in the airflow. The solution after elution was kept at -20 °C until shipped
43 back to Yale. We analyzed the chemical profile using gas chromatography-mass spectrometry (GC-MS,
44 Agilent 7890A/5975C, Agilent Technologies, Inc. USA) at Yale West Campus Analytical Core.

45 We analyzed the GC-MS results using MSD ChemStation F.01.03.2357 (Agilent Technologies,
46 Inc. USA). We first identified any compounds that exist only in the samples or have a substantially higher
47 quantity in the samples compared to the positive control. The compounds were then identified using the
48 National Institute of Standards and Technology (NIST) reference library (v 2.2, Scientific Instrument

49 Services, USA). We quantified the concentrations of each compound using the area underneath the
50 corresponding peak(s) and used the 1-Bromoheptane peak to translate area to absolute concentration. We
51 removed compounds found in only one oviposition sites which could result from identification errors. We
52 also excluded compounds that do not exist in nature (<http://www.thegoodscentscompany.com>), which
53 suggested contamination or misidentification. After filtering the compounds, 11 oviposition sites without
54 any of the remaining chemical compounds were also removed.

55

56 *Mosquito colonies*

57 *Ae. aegypti* collected from natural oviposition sites were kept alive in the field to establish
58 colonies. In La Lopé, due to the low number of *Ae. aegypti* collected, we pooled all individuals from each
59 habitat to establish a single forest colony and a single village colony. We also distributed several bamboo
60 traps and performed human landing catches in La Lopé forest to supplement the larval collection. In
61 Rabai, all colony-forming *Ae. aegypti* came from natural breeding sites. We established two forest
62 colonies (one from the deep area in the forest and another from the edge of the forest adjacent to the
63 Chang'ombe village) and four domestic colonies collected from the domestic oviposition sites in the four
64 villages, respectively. In both La Lopé and Rabai, we blood-fed the females in the field multiple times,
65 which is necessary for egg development and collected the eggs (the second generation) on seed
66 germination papers (SD7606, Anchor Paper Company, USA). After producing eggs, *Ae. aegypti* were
67 preserved in ethanol in the field for genetic analysis.

68 The eggs of the two La Lopé colonies and the four Rabai domestic colonies were brought back to
69 Yale at the end of the fieldwork. The two Rabai forest colonies were first brought back to the McBride
70 Lab at Princeton University and maintained as described in Rose et al. (2020) with code name K66 and
71 K67. A copy of the third generation of these two colonies was later sent to Yale while the original copy
72 was continued at Princeton University. We kept the mosquitoes in the insectary with 27 °C constant

73 temperature, 50%-70% relative humidity, and a 12h/12h light/dark cycle. Eggs were hatched with
74 deionized water supplied with fish food (TetraMarine Saltwater Granules, Tetra, German), and pupae
75 were transferred into insect rearing cages (BugDorm-1) that are roughly 30 x 30 x30 cm. We provided
76 10% sugar water to adults constantly and fed them multiple times with sheep blood (DSB050, Hemostat,
77 USA) at least five days after they emerged. Three days after feeding, we provided four cups (two black
78 cups and two white cups) lined with seed papers to collect eggs from gravid females. The eggs were dried
79 slowly in the insectary and kept for up to six months.

80 To control for the possible laboratory adaptations of these colonies to our specific insectary and
81 rearing regime, in addition to the eight colonies in our lab (named “Powell” strains), we also acquired
82 four more colonies from Rabai that had been reared in the McBride lab at Princeton University until the
83 fifth generation (named “McBride” strains). They were maintained as described in Rose et al. (2020).
84 These four colonies consist of the two Rabai forest colonies (K66 and K67) as well as two Rabai
85 peridomestic colonies that were independently collected from the Chang’ombe village (K65) and the
86 Mbarekani village (K63), respectively. In total, we examined two colonies from La Lopé and ten colonies
87 from Rabai in our oviposition assays.

88

89 *Laboratory oviposition assay*

90 Each batch of the oviposition assays included two to four colonies, with at least one forest colony
91 and one village colony, and we synchronized all colonies from hatching until the end of the experiment.
92 The mosquitoes used for the laboratory oviposition choice assays were reared in the similar protocol as
93 the main colonies described in the main texts, with a few changes to reduce between-experiment
94 variations introduced by the rearing process. Firstly, after hatching, the first-instar larvae were transferred
95 into new larval trays with the density of one larva per 5mL water and fed with a fixed amount of larval
96 food. Secondly, we kept around 360 adults, including both sexes in a 17.5 cm cube cage (BugDorm-

97 4M1515) instead of the larger cage for colony maintenance. Lastly, we fed females with sheep blood only
98 once about 5-8 days after more than 90% of pupae emerge into adults. Females were allowed to feed for
99 one hour. We removed all males and females that did not appear engorged immediately after feeding.

100 Experiments started roughly 72 hours after blood-feeding. We used a binary-choice design except
101 for experiments examining bacterial density (described in detail in the following paragraphs). Five gravid
102 females were transferred into a 15 x 15 x 15 cm customized cage with messes covering both lateral sides
103 and the top side (Figure S1). We used more than one animal per cage as preliminary trials with single
104 females per cage rendered very low rates of response. We placed two or five black cups (one-oz plastic
105 food container cups) containing 12 mL solution that differ in the variable of interest in the cage. The cups
106 were lined with seed-germination papers for collecting eggs. Positions of the choices were selected
107 randomly among the two or five cup positions (Figure S1). The experimental cages were kept in an
108 environmental chamber (model PG031, Darwin Chambers Company, USA) with 27 °C, 70% humidity,
109 10 lux light intensity and a 12h/12h light/dark cycle, which is more accurately regulated than the insectary
110 room. Locations of the cages in the incubator were determined randomly. We allowed the females to lay
111 eggs for 24 hours and counted the number of eggs in each cup at the end of the experiments.

112

113 **Reference**

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124 **Tables**

125 **Table S1.** Methods to measure physical variables of mosquito oviposition sites in La Lopé, Gabon

Variable	Measurement method	Location*
Diameter 1	Measured as the longest diameter of the water surface. For tires lying horizontal, this variable is measured as the diameter of the outer circle.	Both
Diameter 2	Measured as the longest diameter of the water surface that is perpendicular to diameter 1. For tires lying horizontal, this variable is measured as the diameter of the inner circle.	Both
Circumference	Calculated from diameter 1 and diameter 2 for containers with regular-shaped openings (e.g., round), or estimated from photos.	Both
Surface area	Calculated from diameter 1 and diameter 2 for containers with regular-shaped openings (e.g., round), or estimated from photos.	Both
Container depth	Measured as the distance from the bottom of the container to the lowest point of the entrance of the container	Rabai
Water depth	Measured as the distance from the bottom of the container to the water surface	Rabai
Volume	The exact volume was either calculated from surface area, and depth if the shape of the water body is roughly cylinder (e.g., bamboo traps, buckets, boxes, and cans), or estimated from the volume of water collected into the bottle when the site was emptied. When the measurement of exact volume was not possible, the volume was estimated by researchers in the field.	Both
Height	Measured as the distance between the lowest point of the entrance of the sites to the ground next to the site. For rock pools and other breeding sites that sit on the ground, the height is zero.	Both
Temperature difference	Ambient temperature was measured by HOBO UX100-011 Temperature and Relative Humidity Loggers (Onset, MA, USA). A logger was placed beside the oviposition site until the read stabilize. Another logger calibrated to the first one was placed in the field station to record the diurnal fluctuation of ambient temperature, which was then subtracted from the temperature measured by the oviposition sites to calculate the temperature difference.	Both

Humidity difference	Relative humidity was measured by HOBO UX100-011 Temperature and Relative Humidity Loggers (Onset, MA, USA). A logger was placed beside the oviposition site until the read stabilize. Another logger calibrated to the first one was placed in the field station to record the diurnal fluctuation of relative humidity, which was then subtracted from the humidity measured by the oviposition sites to calculate the humidity difference.	Both
Canopy coverage	Measured by a spherical densiometer (Convex Model A, Forestry supply # 43887). The densiometer was held as close to the entrance of the breeding site as possible. Canopy coverage was estimated facing north, west, south, and east. The mean value of the four directions was used for future analysis. The direction was determined by a compass in the forest.	Both
pH ⁺	La Lopé: Measured by a WTW-3110 pH-meter (Xylem, USA) from water samples collected in 50 mL sterile conical tubes less than 24 hours from collection. The water samples are kept in an icebox or at 4 degrees before measuring and recovered to room temperature when measuring. Rabai: Measured by a Hach Pocket Pro+ Multi 2 Tester (Hatch, USA).	Both
Conductivity ⁺	La Lopé: Measured by a WTW-3310 conductivity-meter (Xylem, USA) from water samples collected in 50 mL sterile conical tubes less than 24 hours from collection. The water samples are kept in an icebox or at 4 degrees before measuring and recovered to room temperature when measuring. Rabai: Measured by a Hach Pocket Pro+ Multi 2 Tester (Hatch, USA).	Both
Salinity	Measured on-site by a Hach Pocket Pro+ Multi 2 Tester (Hatch, USA)	Rabai
Total dissolved solids (TDS)	Measured on-site by a Hach Pocket Pro+ Multi 2 Tester (Hatch, USA)	Rabai
Water temperature	Measured on-site by a Hach Pocket Pro+ Multi 2 Tester (Hatch, USA)	Rabai

126 * This column indicates whether this variable was measured in both La Lopé and Rabai (“Both”), or only
127 in Rabai (“Rabai”).

128 ⁺ pH and conductivity were measured slightly differently in La Lopé and Rabai.

129

130

131 **Table S2.** Details of laboratory oviposition assays

Variable	Methods to prepare oviposition choices	The choice resembling forest oviposition sites	The choice resembling village oviposition sites	Mosquito colonies
Water samples collected in the field	About 10 mL water samples were collected from randomly selected 18 oviposition sites in Rabai forest and 18 in Rabai villages. The water samples were frozen in -20 °C. The forest and village water samples were randomly paired to create 18 pairs and used to in an oviposition assay with nine cages of forest mosquitoes and nine cages of domestic mosquitoes.	Water samples collected directly from Rabai forest oviposition sites, including sites present and absent of <i>Ae. aegypti</i> .	Water samples collected directly from Rabai domestic oviposition sites, including sites present and absent of <i>Ae. aegypti</i> .	Kwa Bendegwa domestic colony, Powell strain; Rabai forest deep colony, Powell strain
pH	We adjusted the pH of 1x Phosphate-buffered saline (PBS) solution using hydrochloric acid (HCl) and sodium hydroxide (NaOH) to the desired value.	pH = 7.18 at the beginning of the experiment, which roughly equals the median pH of forest oviposition sites in Rabai present with <i>Ae. aegypti</i> (pH = 7.2). The pH at the end of the experiment is 7.1.	pH = 8.71 at the beginning of the experiment, which is slightly higher than the median pH of domestic oviposition sites in Rabai present with <i>Ae. aegypti</i> (pH = 8.4). The pH at the end of the experiment is 7.8.	Kwa Bendegwa domestic colony, Powell strain; Rabai forest deep colony, Powell strain
Shading	We placed the experimental cage in the center of a 17.8 x 17.8 x 17.8 cm cardboard box, which allows light coming into the cage only from the top. The top side of the box was divided evenly into two halves, each covering one of the two cups in the cage. The two halves were modified to represent the shading of the forest and village oviposition sites in Rabai. To maximize the difference in shading, we placed the two oviposition cups against the opposite walls of the cage instead of 7.6 cm away as in other experiments.	The "forest" half of the top side of the cardboard box has 30 holes of 0.8 cm diameter wide, which in total counts for ~ 8% of the surface area. This condition mimics the median canopy coverage in Rabai forest oviposition sites present with <i>Ae. aegypti</i> (92%).	The "domestic" half of the top side of the cardboard box has no holes mimics the complete canopy coverage in most Rabai domestic oviposition sites present with <i>Ae. aegypti</i> .	Kwa Bendegwa domestic colony, Powell strain; Rabai forest deep colony, Powell strain

Combination of pH, salinity, and shading	We adjusted the pH of distilled water using hydrochloric acid (HCl) and sodium hydroxide (NaOH) and adjusted the conductivity using sodium chloride (NaCl). Different shading conditions were created, as described above.	pH = 7.11, conductivity = 901 μ L, shading = 92%	pH = 8.30, conductivity = 503 μ L, shading = 100%	Kwa Bendegwa domestic colony, Powell strain; Rabai forest deep colony, Powell strain
Larval density	We hatched eggs of a Rabai forest strain and a Rabai village domestic strain simultaneously. The second-instar larvae of each strain were transferred to two new trays with 800 mL distilled water at different larval densities that represent the conditions of forest vs. domestic oviposition sites. We added 50 pellets of fish food per tray on the same day of larvae transferring and removed all larvae after three days. The water holding larvae of the two strains at the same larval density were mixed in equal quantity and used as the choices in the oviposition assay.	Water holding larvae for 3 days at the density of 50 larvae / 800 mL (1 larva / 16 mL). This larval density roughly matches the median larval density of all mosquito species in Rabai forest oviposition sites present with <i>Ae. aegypti</i> .	Water holding larvae for 3 days at a density of 1 larva / 800 mL. This larval density roughly matches the median larval density of Rabai domestic oviposition sites present with <i>Ae. aegypti</i> .	Kwa Bendegwa domestic colony, Powell strain; Rabai forest deep colony, Powell strain

Bacterial community composition *	<p>During fieldwork, we preserved water samples from a subset of oviposition sites that were present with <i>Ae. aegypti</i> using 80% glycerol. These samples include 10 from La Lopé village, 5 from La Lopé forest, 10 from Rabai forest, and 10 from Rabai village. The glycerol preservation allows the bacteria to stay alive with minimal changes over time. We kept the preservations in -80 °C.</p> <p>Glycerol preservations from each habitat in each location were mixed with equal quantity to create in total four mixed stocks. We inoculated the forest and village mixed stock in 10 mL Lysogeny broth (LB). The bacterial cultures were shaken at 200 rpm in 37 °C for 24 hours. The cell densities of the two cultures were measured by OD600 light absorption. We diluted the bacterial cultures to 1.25×10^9 cells/mL in 10 mL LB media, and then added 500 mL sterilized water. The final cell density is 2.5×10^7 cells/mL. The diluted bacterial solutions made from the forest vs. village bacterial stocks were used as the two oviposition choices.</p>	The diluted bacterial culture generated using bacterial glycerol samples collected from forest oviposition sites present with <i>Ae. aegypti</i> .	The diluted bacterial culture generated using bacterial glycerol samples collected from village oviposition sites present with <i>Ae. aegypti</i> .	All colonies
Bacterial density *	We generated bacterial cultures following the same protocol as described above but inoculated the same LB media with both the forest and the village bacterial stocks. After the 24-hour growth, the bacterial culture was diluted using fresh LB to four different cell densities: 1.25×10^9 , 2.5×10^8 , 5×10^7 , and 1×10^7 cells/mL, and then added to 500 mL sterilized water. The final cell densities are 2.5×10^7 , 5×10^6 , 1×10^6 , and 2×10^5 cells/mL. A fifth choice is 10 mL LB in 500 mL sterilized water with no bacteria.	The median bacterial density in Rabai forest oviposition sites present with <i>Ae. aegypti</i> is 1.5×10^6 cells/mL.	The median bacterial density in Rabai forest oviposition sites present with <i>Ae. aegypti</i> is 3.0×10^5 cells/mL.	All colonies

132 * We used the two La Lopé bacterial stocks in oviposition assays with the La Lopé mosquito colonies, and Rabai bacterial stocks in oviposition
133 assays with the Rabai mosquitoes.

134 **Table S3.** Single variable comparisons across oviposition site groups, habitats, and *Ae. aegypti* present vs. absent sites in La Lopé

Variable	All oviposition site groups*	Forest vs. Peridomestic ⁺	<i>Ae. aegypti</i> absent vs. present ⁺
Longest diameter	$\chi^2 = 1.54$, df = 3, p = 0.673	W = 1127, p = 0.231	W = 619.5, p = 0.777
Second diameter	$\chi^2 = 2.79$, df = 3, p = 0.425	W = 842, p = 0.257	W = 786.5, p = 0.163
Circumference	$\chi^2 = 0.52$, df = 3, p = 0.915	W = 995.5, p = 0.905	W = 698, p = 0.617
Surface area	$\chi^2 = 0.98$, df = 3, p = 0.806	W = 909.5, p = 0.563	W = 733, p = 0.394
Height of container opening	$\chi^2 = 23.59$, df = 3, p < 0.001[#]	W = 540.5, p < 0.001	W = 885.5, p = 0.003
Water volume	$\chi^2 = 6.33$, df = 3, p = 0.096	W = 727.5, p = 0.038	W = 839, p = 0.054
Temperature difference	$\chi^2 = 15.64$, df = 3, p = 0.001	W = 501.5, p < 0.001	W = 739.5, p = 0.359
Humidity difference	$\chi^2 = 19.56$, df = 3, p < 0.001	W = 1515, p < 0.001	W = 480, p = 0.091
Canopy coverage	$\chi^2 = 10.2$, df = 3, p = 0.017	W = 1334, p = 0.004	W = 515, p = 0.181
pH	$\chi^2 = 1.48$, df = 3, p = 0.687	W = 912.5, p = 0.58	W = 741.5, p = 0.348
Conductivity	$\chi^2 = 2.34$, df = 3, p = 0.506	W = 1089, p = 0.376	W = 611, p = 0.713
Microbial density (log transformed)	$\chi^2 = 1.33$, df = 3, p = 0.722	W = 138, p = 0.412	W = 170, p = 0.889
Shannon index at ASV level	$\chi^2 = 3.68$, df = 3, p = 0.298	W = 487, p = 0.108	W = 508, p = 0.218
Shannon index at Species level	$\chi^2 = 8.71$, df = 3, p = 0.033	W = 387, p = 0.005	W = 501, p = 0.257
Shannon index at Genus level	$\chi^2 = 8.54$, df = 3, p = 0.036	W = 390, p = 0.006	W = 493, p = 0.307
Shannon index at Family level	$\chi^2 = 7.37$, df = 3, p = 0.061	W = 411, p = 0.012	W = 491, p = 0.321

135 * Kruskal-Wallis rank sum test

136 ⁺ Wilcoxon rank sum test

137 [#] Statistically significant results were marked in bold

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140 **Table S4.** Pairwise single variable comparisons between oviposition site groups in La Lopé

Variable	Forest <i>Ae. aegypti</i> absent vs. Forest <i>Ae. aegypti</i> present*	Forest <i>Ae. aegypti</i> absent vs. Peridomestic <i>Ae. aegypti</i> absent*	Forest <i>Ae. aegypti</i> absent vs. Peridomestic <i>Ae. aegypti</i> present*	Forest <i>Ae. aegypti</i> present vs. Peridomestic <i>Ae. aegypti</i> absent*	Forest <i>Ae. aegypti</i> present vs. Peridomestic <i>Ae. aegypti</i> present*	Peridomestic <i>Ae. aegypti</i> absent vs. Peridomestic <i>Ae. aegypti</i> present*
Longest diameter	W = 131.5 p = 1	W = 666.5 p = 1	W = 352.5 p = 1	W = 70 p = 1	W = 38 p = 1	W = 142.5 p = 1
Second diameter	W = 112 p = 1	W = 552 p = 1	W = 207 p = 0.392	W = 56.5 p = 1	W = 26.5 p = 1	W = 127 p = 1
Circumference	W = 125 p = 1	W = 610.5 p = 1	W = 286 p = 1	W = 69.5 p = 1	W = 29.5 p = 1	W = 136.5 p = 1
Surface area	W = 119.5 p = 1	W = 565.5 p = 1	W = 252 p = 1	W = 62 p = 1	W = 30 p = 1	W = 133.5 p = 1
Height of container opening	W = 111 p = 1	W = 366.5 p = 0.005 #	W = 126 p < 0.001	W = 36 p = 0.802	W = 12 p = 0.233	W = 89.5 p = 0.164
Water volume	W = 104 p = 1	W = 490.5 p = 1	W = 161 p = 0.047	W = 56.5 p = 1	W = 19.5 p = 1	W = 128.5 p = 1
Temperature difference	W = 127 p = 1	W = 296.5 p = 0.005	W = 175 p = 0.097	W = 16 p = 0.071	W = 14 p = 0.452	W = 150.5 p = 1
Humidity difference	W = 121 p = 1	W = 863 p = 0.004	W = 497 p = 0.007	W = 96 p = 0.239	W = 59 p = 0.041	W = 174 p = 1
Canopy coverage	W = 98.5 p = 1	W = 730 p = 0.4	W = 458 p = 0.062	W = 90 p = 0.522	W = 56 p = 0.138	W = 194.5 p = 1
pH	W = 123.5 p = 1	W = 578 p = 1	W = 250.5 p = 1	W = 62 p = 1	W = 22 p = 1	W = 122.5 p = 1
Conductivity	W = 160 p = 1	W = 680 p = 1	W = 325 p = 1	W = 56 p = 1	W = 28 p = 1	W = 136 p = 1
Density of <i>Ae. aegypti</i>	- +	- +	- +	- +	W = 33.5 p = 0.961	- +

Density of all mosquitoes	W = 121.5 p = 0.723	- +	- +	- +	- +	- +
Microbial density (log transformed)	W = 33 p = 1	W = 76 P = 1	W = 57 P = 1	W = 35 P = 1	W = 24 P = 1	W = 55 P = 1
Shannon index at ASV level	W = 78 p = 1	W = 314 P = 1	W = 105 P = 0.524	W = 52 P = 1	W = 16 P = 1	W = 86 P = 1
Shannon index at Species level	W = 92 p = 1	W = 259 P = 0.270	W = 81 P = 0.087	W = 36 P = 1	W = 11 P = 0.595	W = 87 P = 1
Shannon index at Genus level	W = 94 p = 1	W = 262 P = 0.305	W = 84 P = 0.113	W = 34 P = 1	W = 10 P = 0.452	W = 88 P = 1
Shannon index at Family level	W = 94 p = 1	W = 279 P = 0.576	W = 86 P = 0.134	W = 35 P = 1	W = 11 P = 0.595	W = 89 P = 1

141 * Wilcoxon rank sum tests with multiple comparison correction using the *holm* method

142 # Statistically significant results were marked in bold

143 + Data not available for this comparison

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146 **Table S5.** Single variable comparisons across oviposition site groups, habitats, and *Ae. aegypti* present vs. absent sites in Rabai

Variable	All oviposition site groups*	Habitats*	Forest vs. Peridomestic**	Forest vs. Domestic**	Peridomestic vs. Domestic**	<i>Ae. aegypti</i> present vs. <i>Ae. aegypti</i> absent**
Longest diameter	$\chi^2 = 44$ df = 3 p < 0.001 #	$\chi^2 = 42.74$ df = 2 p < 0.001	W = 48.5 p = 0.003	W = 1 p < 0.001	W = 82 p = 1	W = 867 p < 0.001
Second diameter	$\chi^2 = 47.32$ df = 3 p < 0.001	$\chi^2 = 45.51$ df = 2 p < 0.001	W = 29 p < 0.001	W = 4.5 p < 0.001	W = 64.5 p = 0.416	W = 909 p < 0.001
Circumference	$\chi^2 = 45.98$ df = 3 p < 0.001	$\chi^2 = 44.37$ df = 2 p < 0.001	W = 37.5 p = 0.001	W = 0 p < 0.001	W = 82 p = 1	W = 884.5 p < 0.001
Surface area	$\chi^2 = 46.5$ df = 3 p < 0.001	$\chi^2 = 44.69$ df = 2 p < 0.001	W = 34 p = 0.001	W = 1 p < 0.001	W = 81 p = 1	W = 890.5 p < 0.001
Water volume	$\chi^2 = 42.89$ df = 3 p < 0.001	$\chi^2 = 42.87$ df = 2 p < 0.001	W = 57.5 p = 0.008	W = 0 p < 0.001	W = 61 p = 0.31	W = 837.5 p < 0.001
Container depth	$\chi^2 = 36.61$ df = 3 p < 0.001	$\chi^2 = 37.55$ df = 2 p < 0.001	W = 85.5 p = 0.077	W = 25 p < 0.001	W = 40.5 p = 0.035	W = 780.5 p = 0.001
Height of container openings	$\chi^2 = 9.53$ df = 3 p = 0.023	$\chi^2 = 10.14$ df = 2 p = 0.006	W = 275 p = 0.008	W = 480 p = 0.767	W = 41.5 p = 0.039	W = 372.5 p = 0.061
Water depth	$\chi^2 = 3.66$ df = 3 p = 0.301	$\chi^2 = 3.63$ df = 2 p = 0.163	W = 181 p = 1	W = 295 p = 0.24	W = 68 p = 0.551	W = 563 p = 0.559

Temperature difference	$\chi^2 = 1.77$ df = 3 p = 0.622	$\chi^2 = 2.82$ df = 2 p = 0.244	W = 115.5 p = 0.485	W = 420.5 p = 1	W = 139.5 p = 0.244	W = 535.5 p = 0.82
Humidity difference	$\chi^2 = 0.83$ df = 3 p = 0.842	$\chi^2 = 1.48$ df = 2 p = 0.478	W = 201.5 p = 1	W = 390.5 p = 1	W = 69 p = 0.597	W = 564 p = 0.551
Canopy coverage	$\chi^2 = 48.6$ df = 3 p < 0.001	$\chi^2 = 49.8$ df = 2 p < 0.001	W = 282 p = 0.004	W = 2.5 p < 0.001	W = 0 p < 0.001	W = 735 p = 0.004
pH	$\chi^2 = 28.1$ df = 3 p < 0.001	$\chi^2 = 27.84$ df = 2 p < 0.001	W = 24.5 p < 0.001	W = 121 p < 0.001	W = 93.5 p = 1	W = 812.5 p < 0.001
Conductivity	$\chi^2 = 6.27$ df = 3 p = 0.099	$\chi^2 = 4.06$ df = 2 p = 0.131	W = 221 p = 0.405	W = 513 p = 0.295	W = 87 p = 1	W = 489.5 p = 0.721
Salinity	$\chi^2 = 6.34$ df = 3 p = 0.096	$\chi^2 = 3.96$ df = 2 p = 0.138	W = 218.5 p = 0.461	W = 514.5 p = 0.28	W = 89 p = 1	W = 495 p = 0.775
Total dissolved solids	$\chi^2 = 6.66$ df = 3 p = 0.083	$\chi^2 = 4.32$ df = 2 p = 0.115	W = 223 p = 0.363	W = 516 p = 0.267	W = 87 p = 1	W = 489 p = 0.717
Water temperature	$\chi^2 = 7.25$ df = 3 p = 0.064	$\chi^2 = 4.73$ df = 2 p = 0.094	W = 203 p = 0.955	W = 546 p = 0.089	W = 96 p = 1	W = 313.5 p = 0.008
Density of <i>Ae. aegypti</i>	- +	$\chi^2 = 20.961$ df = 2 p < 0.001	- +	- +	- +	- +
Density of all mosquitoes	$\chi^2 = 37.25$ df = 3 p < 0.001	$\chi^2 = 37.24$ df = 2 p < 0.001	W = 228 p = 0.091	W = 794 p < 0.001	W = 160 p = 0.013	W = 755.5 p = 0.002

Microbial density (log transformed)	F = 8.43 df = 3 p < 0.001 ^	F = 13.47 df = 2 p < 0.001 ^	diff = -0.141 p = 0.959 ^	diff = 1.825 p < 0.001 ^	diff = 1.966 p = 0.001 ^	t = -1.914 p = 0.070 ^^
Shannon index at ASV level	$\chi^2 = 1.90$ df = 3 p = 0.593	$\chi^2 = 2.610$ df = 2 p = 0.271	W = 105 p = 0.275	W = 369 p = 1	W = 119 p = 1	W = 496 p = 0.787
Shannon index at Species level	$\chi^2 = 3.44$ df = 3 p = 0.329	$\chi^2 = 1.25$ df = 2 p = 0.536	W = 160 p = 1	W = 471 p = 0.967	W = 119 p = 1	W = 622 p = 0.179
Shannon index at Genus level	$\chi^2 = 4.91$ df = 3 p = 0.178	$\chi^2 = 2.73$ df = 2 p = 0.256	W = 176 p = 1	W = 509 p = 0.336	W = 122 p = 1	W = 650 p = 0.087
Shannon index at Family level	$\chi^2 = 5.69$ df = 3 p = 0.128	$\chi^2 = 3.72$ df = 2 p = 0.156	W = 199 p = 1	W = 522 p = 0.218	W = 121 p = 1	W = 671 p = 0.046

147 * Kruskal-Wallis rank sum test

148 ** Wilcoxon rank sum tests with multiple comparison correction using the *holm* method

149 + Data not available for this comparison

150 # Statistically significant results were marked in bold

151 ^ One-way Analysis of variance (ANOVA) with Tukey post hoc multiple comparisons of means

152 ^^ Student's t-test

153

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155 **Table S6.** Pairwise single variable comparisons between oviposition site groups in Rabai

Variable	Forest <i>Ae. aegypti</i> absent vs. Forest <i>Ae. aegypti</i> present*	Forest <i>Ae. aegypti</i> absent vs. Peridomestic <i>Ae. aegypti</i> present*	Forest <i>Ae. aegypti</i> absent vs. Domestic <i>Ae. aegypti</i> present*	Forest <i>Ae. aegypti</i> present vs. Peridomestic <i>Ae. aegypti</i> present*	Forest <i>Ae. aegypti</i> present vs. Domestic <i>Ae. aegypti</i> present*	Peridomestic <i>Ae. aegypti</i> present vs. Domestic <i>Ae. aegypti</i> present*
Longest diameter	W = 99.5 p = 0.262	W = 23.5 p = 0.016 #	W = 0 p < 0.001	W = 25 p = 0.153	W = 1 p < 0.001	W = 60 p = 1
Second diameter	W = 82 p = 0.061	W = 10 p = 0.002	W = 1 p < 0.001	W = 19 p = 0.051	W = 3.5 p < 0.001	W = 51.5 p = 0.546
Circumference	W = 91 p = 0.136	W = 14.5 p = 0.004	W = 0 p < 0.001	W = 23 p = 0.11	W = 0 p < 0.001	W = 60 p = 1
Surface area	W = 85.5 p = 0.087	W = 14 p = 0.003	W = 0 p < 0.001	W = 20 p = 0.065	W = 1 p < 0.001	W = 59 p = 1
Water volume	W = 132.5 p = 1	W = 29 p = 0.036	W = 0 p < 0.001	W = 28.5 p = 0.271	W = 0 p < 0.001	W = 47 p = 0.338
Container depth	W = 163.5 p = 1	W = 48.5 p = 0.403	W = 24.5 p < 0.001	W = 32.5 p = 0.485	W = 0.5 p < 0.001	W = 40.5 p = 0.164
Height of container openings	W = 203 p = 1	W = 146 p = 0.042	W = 311 p = 0.646	W = 92.5 p = 0.233	W = 169 p = 1	W = 41.5 p = 0.184
Water depth	W = 175 p = 1	W = 96 p = 1	W = 185.5 p = 1	W = 61 p = 1	W = 109.5 p = 0.528	W = 61 p = 1
Temperature difference	W = 151 p = 1	W = 66 p = 1	W = 237.5 p = 1	W = 49.5 p = 1	W = 183 p = 1	W = 117.5 p = 1
Humidity difference	W = 152 p = 1	W = 96.5 p = 1	W = 220.5 p = 1	W = 70 p = 1	W = 170 p = 1	W = 69 p = 1
Canopy coverage	W = 155.5 p = 1	W = 143 p = 0.063	W = 1.5 p < 0.001	W = 102 p = 0.044	W = 1 p < 0.001	W = 0 p < 0.001

pH	W = 129.5 p = 1	W = 12 p = 0.001	W = 60 p < 0.001	W = 7.5 p = 0.005	W = 61 p = 0.008	W = 86.5 p = 1
Conductivity	W = 119.5 p = 0.983	W = 113 p = 1	W = 282 p = 1	W = 90 p = 0.333	W = 231 p = 0.25	W = 69 p = 1
Salinity	W = 116.5 p = 0.824	W = 109 p = 1	W = 283.5 p = 1	W = 91.5 p = 0.27	W = 231 p = 0.256	W = 71 p = 1
Total dissolved solids	W = 118 p = 0.902	W = 113 p = 1	W = 284 p = 1	W = 92 p = 0.24	W = 232 p = 0.231	W = 69 p = 1
Water temperature	W = 215.5 p = 0.726	W = 118 p = 0.997	W = 352 p = 0.06	W = 71.5 p = 1	W = 194 p = 1	W = 77 p = 1
Density of <i>Ae. aegypti</i>	-	-	-	W = 69 p = 1	W = 19 p < 0.001	W = 140 p = 0.040
Density of all mosquitoes	W = 161.5 p = 0.926	W = 114 p = 0.711	W = 467 p < 0.001	W = 78 p = 0.711	W = 327 p < 0.001	W = 148 p = 0.015
Microbial density (log-transformed)	diff = -0.139 p = 0.994 ^	diff = - 0.158 p = 0.994 ^	diff = - 1.745 p = 0.005 ^	diff = 0.019 p = 0.999 ^	diff = -1.884 p < 0.001 ^	diff = -1.903 p = 0.006 ^
Shannon index at ASV level	W = 178 p = 1	W = 58 p = 1	W = 226 p = 1	W = 43 p = 1	W = 143 p = 1	W = 102 p = 1
Shannon index at Species level	W = 214 p = 0.809	W = 90 p = 1	W = 301 p = 1	W = 42 p = 1	W = 170 p = 1	W = 109 p = 1
Shannon index at Genus level	W = 212 p = 0.912	W = 98 p = 1	W = 326 p = 0.296	W = 46 p = 1	W = 183 p = 1	W = 113 p = 1
Shannon index at Family level	W = 207 p = 1	W = 115 p = 1	W = 336 p = 0.163	W = 52 p = 1	W = 186 p = 1	W = 113 p = 1

156 * Wilcoxon rank sum tests with multiple comparison correction using the *holm* method

157 # Statistically significant results were marked in bold

158 ^ Tukey post hoc multiple comparisons of means after one-way Analysis of variance (ANOVA)

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161 **Table S7.** Bacterial families with significantly different abundance between forest and village peridomestic oviposition sites in La Lopé

Class: Order	Family	Frequency in forest samples	Frequency in village samples	Proportion in forest samples	Proportion in village samples	log2 fold change *	p value *
Betaproteobacteria: Unknown	Unknown	1	2	0.015	0.002	-1.592	0.031
Flavobacteriia: Flavobacteriales	Flavobacteriaceae	2	2	0.216	0	-1.671	0.038
Betaproteobacteria: Burkholderiales	Unknown	11	0	0.035	0	-1.950	0.019
Unknown: Unknown	Unknown	4	0	0.035	0	-2.182	< 0.001
Betaproteobacteria: Methylophilales	Methylophilaceae	7	1	0.017	0	-2.277	0.043
Spartobacteria: Unknown	Unknown	7	0	0.001	0	-2.335	0.002
Armatimonadetes_gp5: Unknown	Unknown	4	0	0.001	0	-3.045	< 0.001
Betaproteobacteria: Burkholderiales	Burkholderiaceae	27	4	0.069	0.027	-3.212	0.003
Cyanobacteria: Family_XIII	GpXIII	1	0	0.002	0	-3.261	0.049
Gammaproteobacteria: Methylococcales	Methylococcaceae	1	0	0.014	0	-3.345	0.022
Gammaproteobacteria: Aeromonadales	Aeromonadaceae	9	0	0.002	0	-3.389	0.004
Chlamydia: Chlamydiales	Parachlamydiaceae	1	0	0.025	0	-3.530	< 0.001
Deltaproteobacteria: Bdellovibrionales	Bacteriovoracaceae	5	0	0.019	0	-3.562	0.002
Chlamydia: Chlamydiales	Simkaniaceae	4	0	0.007	0	-3.672	0.004
Cyanobacteria: Unknown	Unknown	1	0	0.016	0	-3.709	0.005

Cyanobacteria: Family_IX	GpIX	5	0	0.002	0	-4.646	0.002
Holophagae: Holophagales	Holophagaceae	8	0	0.002	0	-4.737	< 0.001
Actinobacteria: Actinomycetales	Microbacteriaceae	14	1	0.068	0.001	-4.976	< 0.001
Cyanobacteria: Family_XI	GpXI	1	0	0.232	0	-5.853	0.004
Bacilli: Bacillales	Bacillales_Incertae_Sedis_XII	0	4	0	0.048	23.199	< 0.001
Alphaproteobacteria: Rhodospirillales	Reyranella	0	1	0	0.221	5.351	< 0.001
Actinobacteria: Actinomycetales	Nocardiaceae	0	3	0	0.011	4.502	< 0.001
Verrucomicrobiae: Verrucomicrobiales	Verrucomicrobiaceae	0	2	0	0.024	3.585	< 0.001
Oligoflexia: Oligoflexales	Oligoflexaceae	1	9	0.001	0.002	3.459	0.004
Acidobacteria_Gp4: Aridibacter	Unknown	1	4	0	0.005	3.442	0.019
Acidobacteria_Gp3: Gp3	Unknown	0	8	0	0.003	3.255	< 0.001
Actinobacteria: Actinomycetales	Geodermatophilaceae	0	6	0	0.002	3.231	0.026
Alphaproteobacteria: Rhizobiales	Xanthobacteraceae	0	7	0	0.028	2.776	0.005
Alphaproteobacteria: Caulobacterales	Caulobacteraceae	10	20	0.002	0.007	2.268	< 0.001
Gammaproteobacteria: Pseudomonadales	Pseudomonadaceae	0	1	0	0.085	2.188	0.037
Betaproteobacteria: Burkholderiales	Oxalobacteraceae	0	19	0	0.071	2.041	0.007
Alphaproteobacteria: Rhizobiales	Methylobacteriaceae	10	21	0.001	0.002	2.002	0.001
Sphingobacteria: Sphingobacteriales	Sphingobacteriaceae	0	2	0	0.030	1.944	0.019

Alphaproteobacteria: Rhodospirillales	Acetobacteraceae	0	7	0	0.022	1.163	0.049
Epsilonproteobacteria: Campylobacterales	Campylobacteraceae	0	1	0	0.056	-4.509	0.019

162 * Calculate by R package *DESeq2*

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164

165 **Table S8.** Bacterial families with significantly different abundance between forest and village (domestic, peridomestic) oviposition sites in Rabai

Class: Order	Family	Frequency in forest samples	Frequency in village samples	Proportion in forest samples	Proportion in village samples	log2 fold change *	p value *
Deltaproteobacteria: Desulfuromonadales	Geobacteraceae	1	0	0.248	0	6.559	< 0.001
Betaproteobacteria: Rhodocyclales	Rhodocyclaceae	2	0	0.180	0	5.855	< 0.001
Bacilli: Bacillales	Paenibacillaceae_1	12	0	0.003	0	5.512	< 0.001
Methanobacteria: Methanobacteriales	Methanobacteriaceae	4	0	0.001	0	5.378	< 0.001
Deltaproteobacteria: Desulfobacterales	Desulfobulbaceae	2	0	0.002	0	5.187	0.001
Clostridia: Clostridiales	Peptococcaceae_1	1	0	0.008	0	4.987	< 0.001
Alphaproteobacteria: Rhizobiales	Methylocystaceae	3	0	0.026	0	4.970	< 0.001
Clostridia: Unknown	Unknown	2	0	0.001	0	4.546	0.001
Actinobacteria: Coriobacteriales	Coriobacteriaceae	3	0	0.002	0	4.161	0.019
Actinobacteria: Actinomycetales	Cellulomonadaceae	7	0	0.015	0	4.136	0.004
Clostridia: Clostridiales	Unknown	5	0	0	0	4.080	< 0.001
Deltaproteobacteria: Desulfovibrionales	Desulfovibrionaceae	4	0	0	0	4.017	0.007
Bacilli: Lactobacillales	Streptococcaceae	7	3	0.021	0.003	3.689	0.044
Gammaproteobacteria: Pseudomonadales	Pseudomonadaceae	15	7	0.034	0.005	3.637	< 0.001
Clostridia: Clostridiales	Ruminococcaceae	4	0	0.001	0	3.470	< 0.001

Clostridia: Clostridiales	Clostridiales_Incertae_Sedis_XIII	2	0	0	0	3.401	0.020
Clostridia: Clostridiales	Heliobacteriaceae	1	0	0.001	0	3.248	0.002
Bacteroidia: Bacteroidales	Unknown	1	0	0.001	0	3.173	0.039
Gammaproteobacteria: Enterobacteriales	Enterobacteriaceae	10	2	0.043	0.001	3.095	0.003
Actinobacteria: Actinomycetales	Thermomonosporaceae	1	0	0.001	0	2.946	0.035
Bacilli: Bacillales	Unknown	30	16	0.008	0.002	2.717	0.001
Actinobacteria: Actinomycetales	Streptomycetaceae	20	1	0.007	0	2.501	0.002
Negativicutes: Selenomonadales	Veillonellaceae	3	0	0.001	0	2.412	0.008
Bacilli: Bacillales	Planococcaceae	17	7	0.020	0.002	2.347	0.023
Gammaproteobacteria: Xanthomonadales	Xanthomonadaceae	19	16	0.025	0.002	2.121	0.003
Clostridia: Clostridiales	Clostridiaceae_1	7	1	0.002	0	1.758	0.020
Unknown: Unknown	Unknown	1	0	0.011	0	-1.526	0.033
Bacteroidia: Bacteroidales	Porphyromonadaceae	0	2	0	0.011	2.979	0.010
Planctomycetia: Planctomycetales	Planctomycetaceae	0	2	0	0.010	-1.584	0.030
Verrucomicrobiae: Verrucomicrobiales	Verrucomicrobiaceae	2	2	0	0.005	-1.844	0.032
Alphaproteobacteria: Rhizobiales	Methylobacteriaceae	3	6	0	0.012	-2.102	0.030
Deltaproteobacteria: Bdellovibrionales	Bdellovibrionaceae	0	2	0	0.003	-2.132	0.023
Actinobacteria: Actinomycetales	Micrococcaceae	3	10	0	0.002	-2.320	0.008

Alphaproteobacteria: Rhodospirillales	Acetobacteraceae	0	4	0	0.018	-2.554	0.001
Alphaproteobacteria: Rhodobacterales	Rhodobacteraceae	0	12	0	0.027	-2.666	< 0.001
Betaproteobacteria: Unknown	Unknown	0	6	0	0.007	-2.993	0.030
Cytophagia: Cytophagales	Unknown	0	3	0	0.001	-3.327	0.023
Gammaproteobacteria: Legionellales	Legionellaceae	0	1	0	0.014	-3.568	< 0.001
Cytophagia: Cytophagales	Cytophagaceae	0	6	0	0.027	-3.677	< 0.001
Unknown: Unknown	Unknown	0	12	0	0.005	-3.708	< 0.001
Flavobacteriia: Flavobacteriales	Cryomorphaceae	0	3	0	0.007	-3.833	0.008
Chloroplast: Chloroplast	Bacillariophyta	0	5	0	0.002	-5.268	0.005
Bacilli: Bacillales	Bacillales_Incertae_Sedis_XII	2	20	0.001	0.022	-5.770	< 0.001
Cytophagia: Cytophagales	Cyclobacteriaceae	0	10	0	0.006	-5.853	0.001
Betaproteobacteria: Methylophilales	Methylophilaceae	0	10	0	0.006	-6.034	< 0.001
Deinococci: Deinococcales	Deinococcaceae	0	18	0	0.001	-6.269	< 0.001
Alphaproteobacteria: Rhodospirillales	Reyranella	0	9	0	0.004	-6.701	< 0.001

* Calculate by R package *DESeq2*

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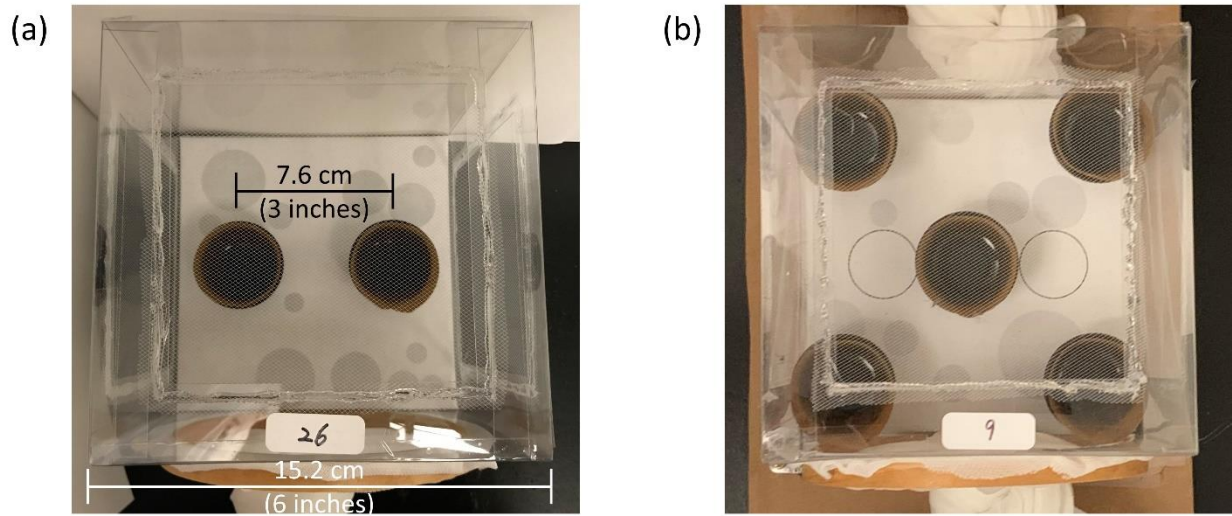
169 **Table S9.** Results of statistical models for laboratory oviposition assays

Variables	Colonies	Models *	Model comparison results
Water samples from the field	Kwa Bendegwa domestic & Rabai forest deep	Full model: egg counts in two cups ~ Colony Null model: egg counts in two cups ~ 1	Full model AIC: 226.69 Null model AIC: 225.76 $\chi^2 = 1.08$, df = 1, p = 0.300
pH			Full model AIC: 162.55 Null model AIC: 161.07 $\chi^2 = 0.52$, df = 1, p = 0.472
Shading			Full model AIC: 340.11 Null model AIC: 38.68 $\chi^2 = 0.58$, df = 1, p = 0.447
Combination of pH, conductivity, and shading			Full model AIC: 271.42 Null model AIC: 269.93 $\chi^2 = 0.52$, df = 1, p = 0.472
Larval density	Kwa Bendegwa domestic & Rabai forest deep	Full model: egg counts in two cups ~ Colony + (1 Experiment ID) + Null model: egg counts in two cups ~ 1 + (1 Experiment ID)	Full model AIC: 566.40 Null model AIC: 567.39 $\chi^2 = 2.98$, df = 1, p = 0.084
Bacterial community	La Lopé forest & La Lopé village	Full model: egg counts in two cups ~ Colony Null model: egg counts in two cups ~ 1	Full model AIC: 156.60 Null model AIC: 155.19 $\chi^2 = 0.59$, df = 1, p = 0.44
	All Rabai colonies	Full model: egg counts in two cups ~ Habitat + (1 Colony) + (1 Experiment ID) ++ Null model: egg counts in two cups ~ 1 + (1 Colony) + (1 Experiment ID)	Full model AIC: 816.87 Null model AIC: 814.46 $\chi^2 = 0.55$, df = 1, p = 0.451
		Full model: egg counts in two cups ~ Colony + (1 Experiment ID) Null model: egg counts in two cups ~ 1 + (1 Experiment ID)	Full model AIC: 820.15 Null model AIC: 810.46 $\chi^2 = 8.32$, df = 9, p = 0.503
Bacterial density	La Lopé forest &	Full model: egg count of each cup ~ Colony x Oviposition choice +	Full model AIC: 1039.6 Null model AIC: 1033.9

La Lopé village	(1 Cage ID) Null model: egg count of each cup ~ Colony + Oviposition choice + (1 Cage ID)	$\chi^2 = 2.29, df = 4, p = 0.683$
All Rabai colonies	Full model: egg count of each cup ~ Habitat x Oviposition choice + (1 Colony) + (1 Experiment ID) + (1 Cage ID) ⁺⁺⁺ Null model: egg count of each cup ~ Habitat + Oviposition choice + (1 Colony) + (1 Experiment ID) + (1 Cage ID)	Full model AIC: 4386 Null model AIC: 4374 $\chi^2 = 3.98, df = 8, p = 0.858$
	Full model: egg count of each cup ~ Colony x Oviposition choice + (1 Experiment ID) + (1 Cage ID) Null model: egg count of each cup ~ Colony + Oviposition choice + (1 Experiment ID) + (1 Cage ID)	Full model AIC: 4416.8 Null model AIC: 4370.7 $\chi^2 = 25.9, df = 36, p = 0.893$

170 * Negative-binomial models for testing bacterial density and beta-binomial models for the rest of the oviposition assays
171 + For oviposition assays that were conducted in multiple experiment cycles, experiment ID was included as a random effect
172 ++ Colonies were included as random factors in statistical models examining the effects of habitats
173 +++ Cage ID were included in oviposition assays to control for the paired structure of the five egg counts in each cage

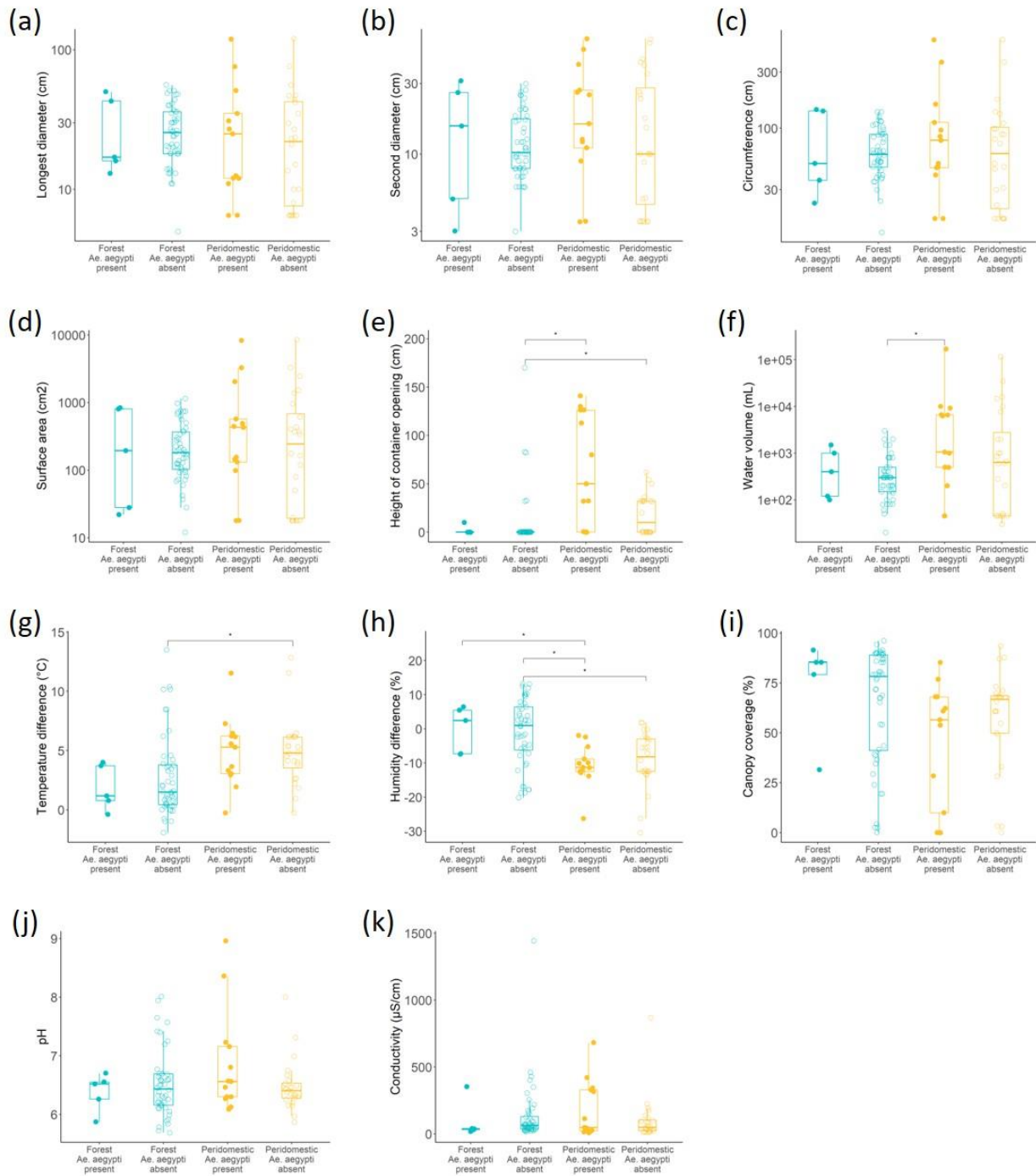
174 **Figures**



175

176 **Figure S1.** Set up of the laboratory oviposition assays with (a) two choices for testing most variables and
177 (b) five choices for testing bacterial density. The cage was built from a 15.2 x 15.2 x 15.2 cm transparent
178 plastic box with fine meshes covering the top and the two lateral sides. A cloth sleeve was attached to the
179 opening on the front side (bottom in the photos) through which the mosquitoes were introduced. The cage
180 has a white bottom with randomly generated gray circles that aims to provide visual stimuli for the
181 mosquitoes to navigate in the cage. The black cups, each lined with a piece of 4 x 13 cm seed germination
182 paper, were 7.6 cm away from each other in the two-choice set-up (a). In the five-choice experiments (b),
183 the five cups located at the four corners of the cage and the center.

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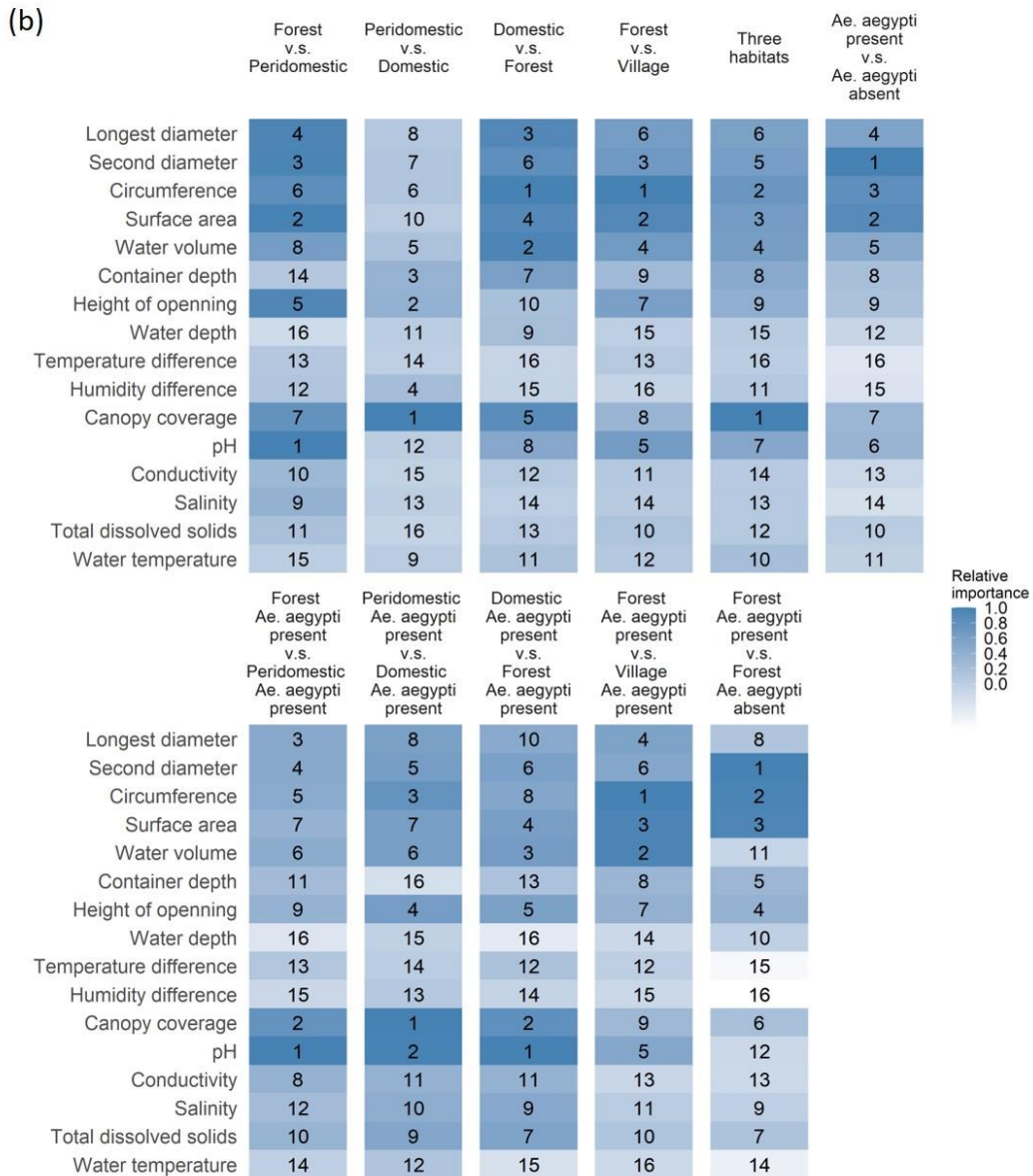
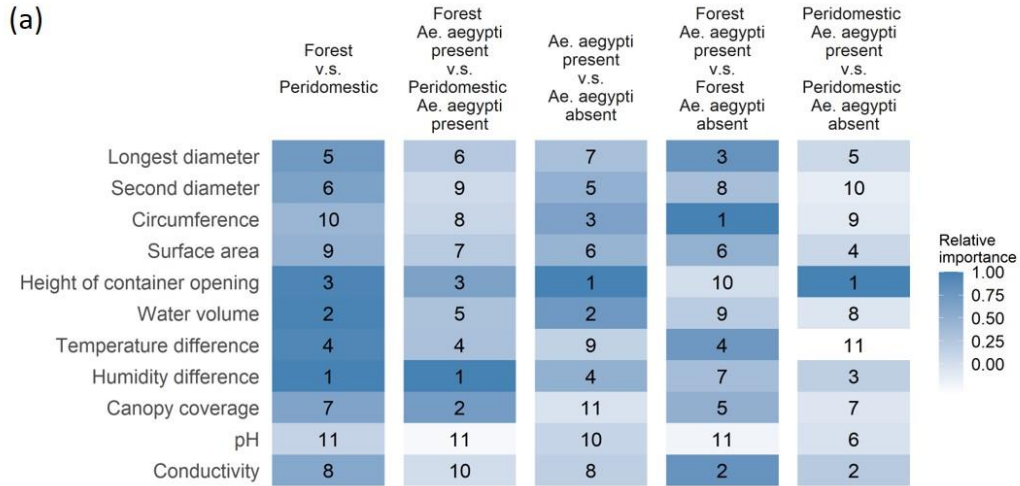


185

186 **Figure S2.** Comparison of individual physical variables between oviposition site groups in La Lopé. Each
 187 point represents a single oviposition site, and the boxplots show the minimum, 25% quartile, median,
 188 75% quartile, and maximum of all values. The colors and shapes are as in Figure 2. Differences between

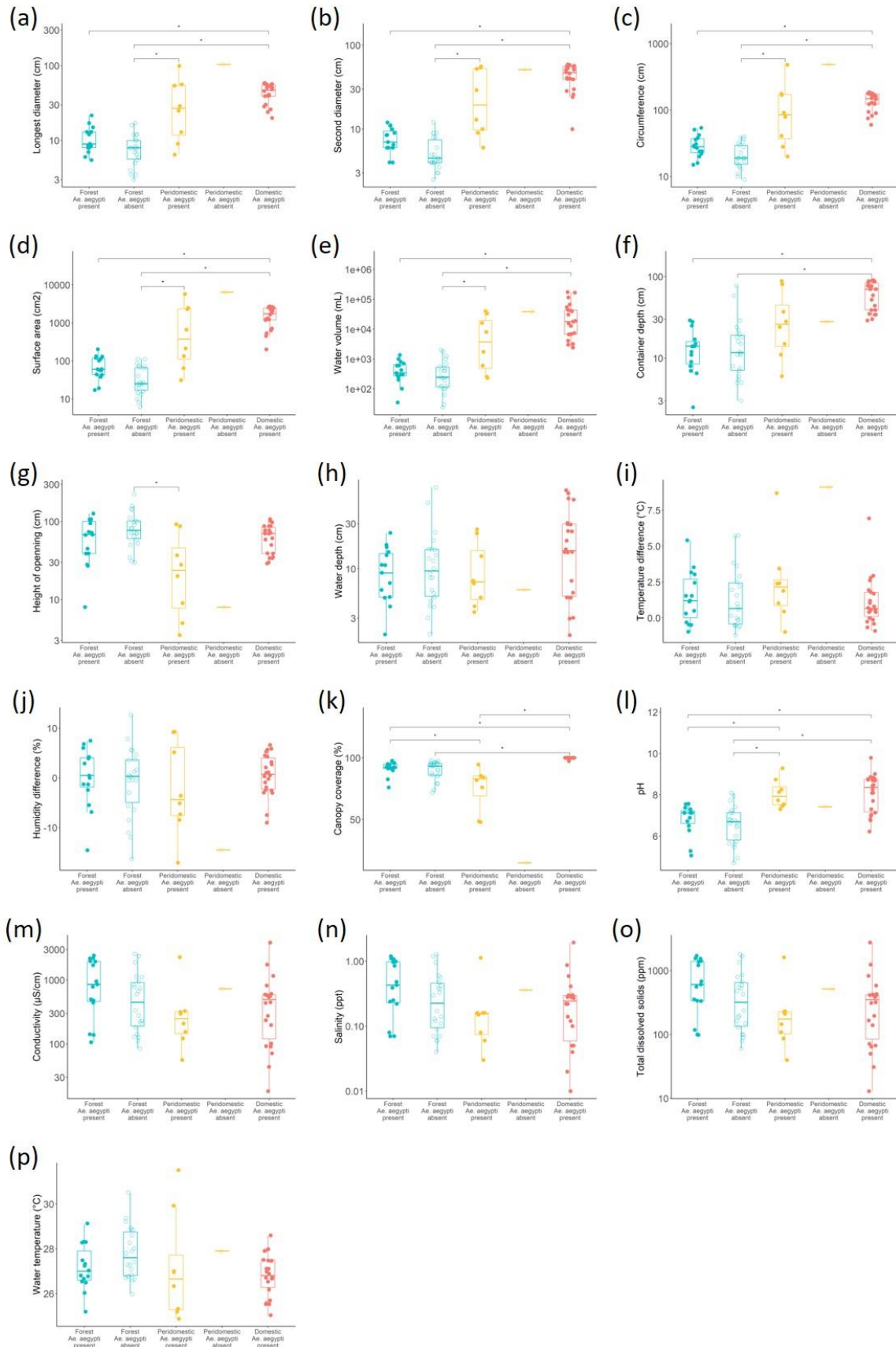
189 groups were tested using pairwise Wilcoxon rank sum test with *Holm* multiple comparison corrections (*:
190 $p < 0.05$, Table S3 and S4). Only significant comparisons are labeled.

191



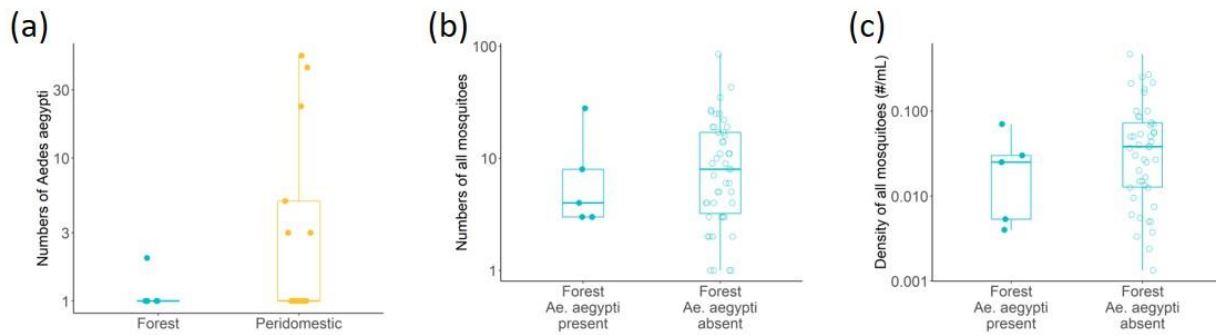
193 **Figure S3.** The importance of each physical variables in differentiating different comparisons between
194 habitats or oviposition site groups in (a) La Lopé and (b) Rabai. Each row represents one variable, and
195 each column describes the comparison, which is labeled above the figures. The color of each cell
196 quantifies the relative importance, which is estimated by random forest in R. The absolute importance
197 measures of all variables in each comparison were scaled to proportions of the variable that shows the
198 highest importance measure. The numbers in each cell indicate the rank of the variables.

199



201 **Figure S4.** Comparison of individual physical variables between oviposition site groups in Rabai. Each
202 point represents a single oviposition site, and the boxplots show the minimum, 25% quartile, median,
203 75% quartile, and maximum of all values. The colors and shapes are as in Figure 2. Differences between
204 groups were tested using pairwise Wilcoxon rank sum test with *Holm* multiple comparison correction (*:
205 $p < 0.05$, Table S5 and S6). Only significant comparisons are labeled.

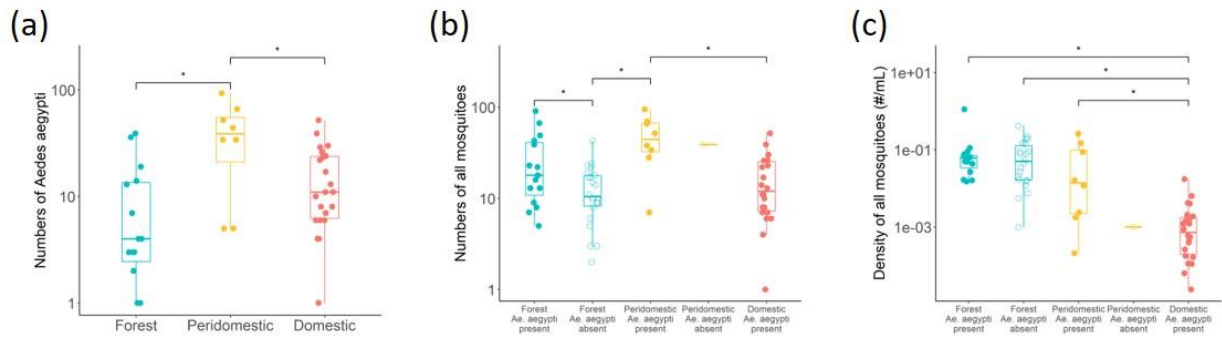
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208 **Figure S5.** Comparison of (a) number of *Ae. aegypti*, (b) number of mosquitoes of all species, and (c)
209 density of mosquitoes of all species in La Lopé. Mosquito species other than *Ae. aegypti* were only
210 screened in the forest habitat, so in (c), we only compared forest oviposition sites present vs. absent of *Ae.*
211 *aegypti*. Each point represents a single oviposition site, and the boxplots show the minimum, 25%
212 quartile, median, 75% quartile, and maximum of all values. The colors and shapes are as in Figure 2.
213 Differences of mosquito numbers between groups (a and b) were examined by negative-binomial models
214 tests (*: $p < 0.05$), while differences of mosquito density (c) between groups were tested using pairwise
215 Wilcoxon rank sum. Only significant comparisons are labeled.

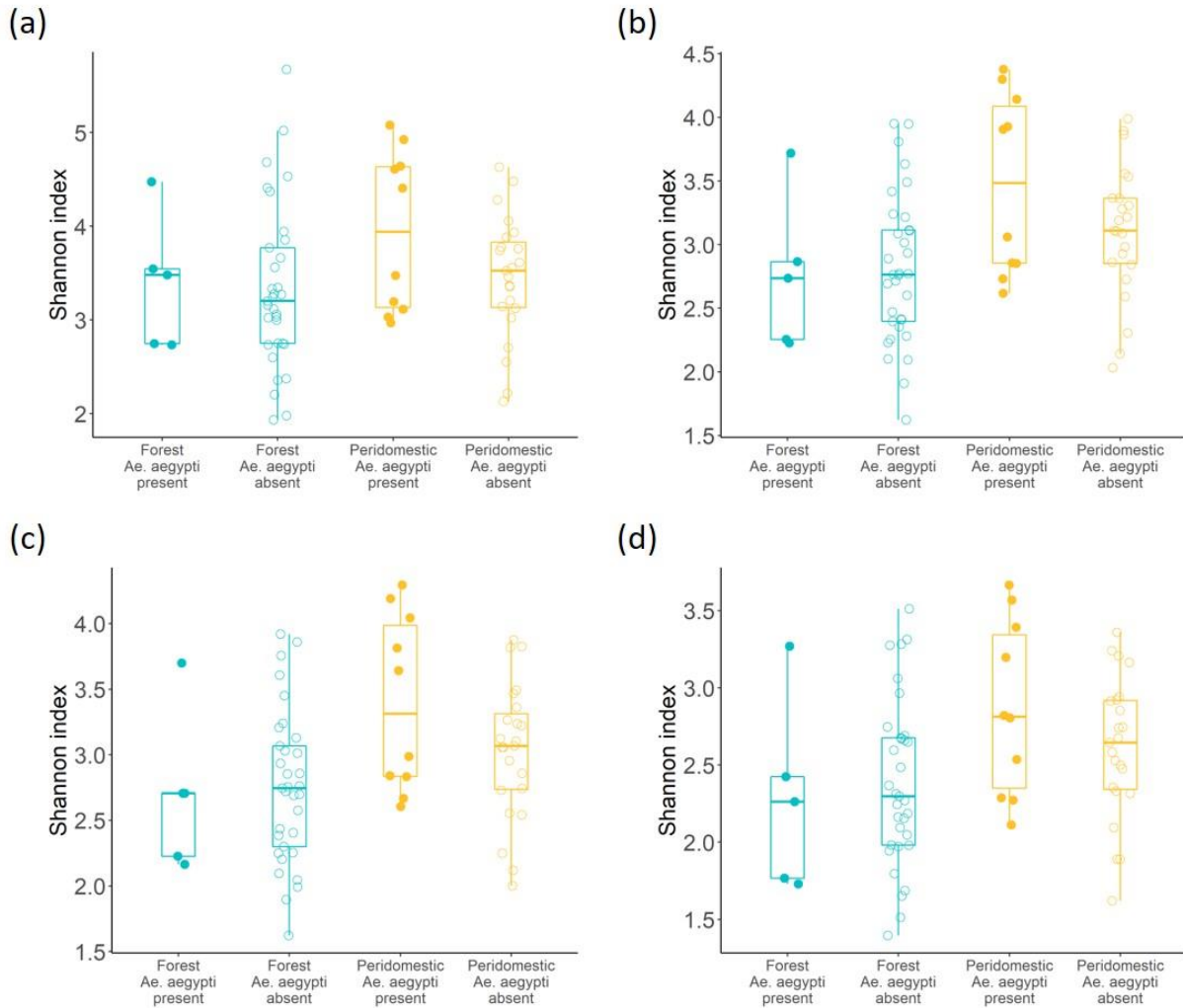
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217

218 **Figure S6.** Comparison of (a) number of *Ae. aegypti*, (b) number of mosquitoes of all species, and (c)
 219 density of mosquitoes of all species in Rabai oviposition site between habitats or oviposition site groups.
 220 Each point represents a single oviposition site, and the boxplots show the minimum, 25% quartile,
 221 median, 75% quartile, and maximum of all values. The colors and shapes are as in Figure 2. Differences
 222 of mosquito numbers between groups (a and b) were examined by negative-binomial models, while
 223 differences of mosquito density (c) between groups were tested using pairwise Wilcoxon rank sum test
 224 with *Holm* multiple comparison corrections (*: $p < 0.05$, Table S5 and S6). Only significant comparisons
 225 are labeled.

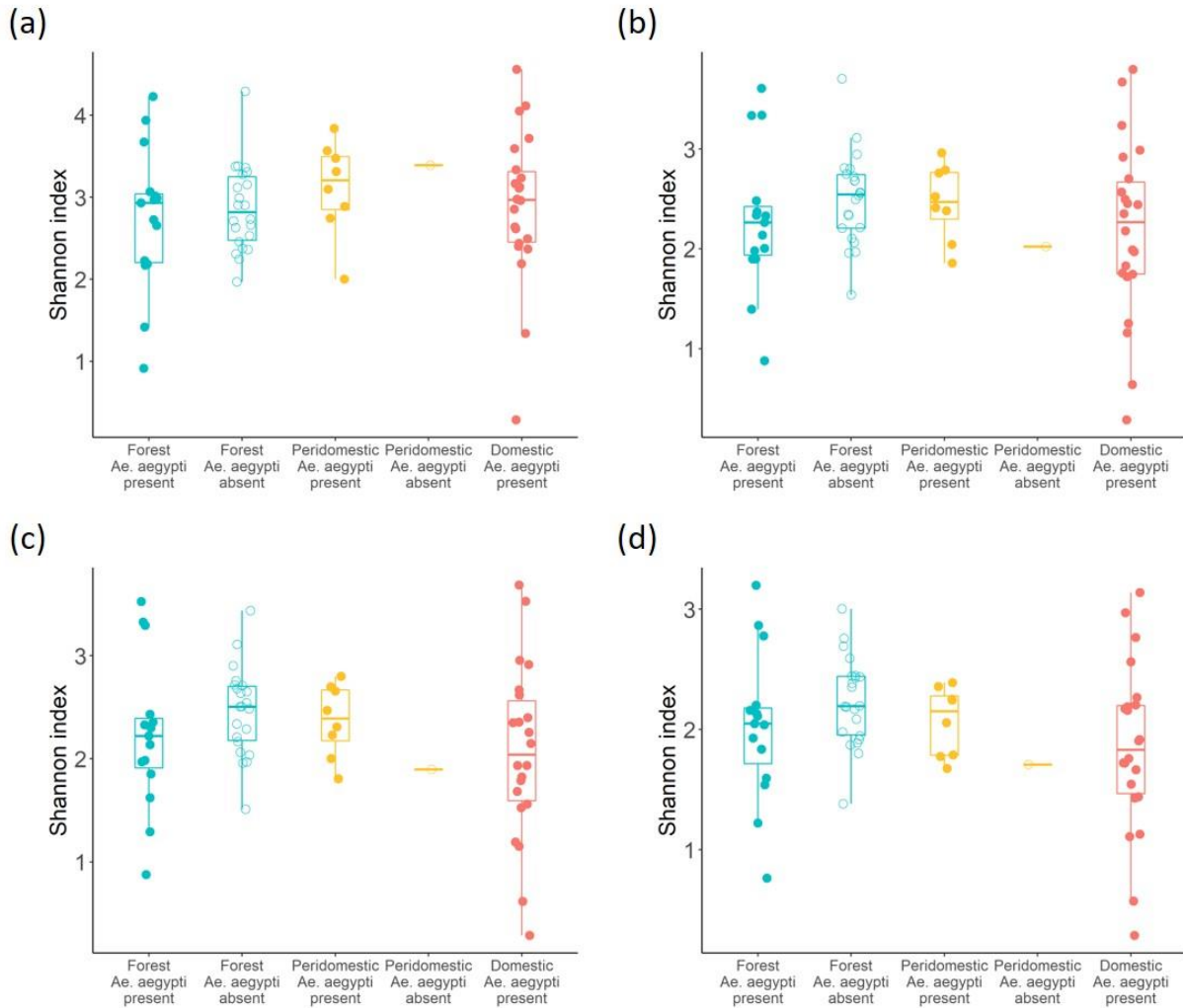
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228 **Figure S7.** Comparison of the Shannon index of the bacterial community in La Lopé oviposition sites at
 229 different taxonomic levels: (a) ASVs, (b) Species, (c) Genus, and (d) Family. Each point represents a
 230 single oviposition site, and the boxplots show the minimum, 25% quartile, median, 75% quartile, and
 231 maximum of all values. The colors and shapes are as in Figure 2. Differences between groups were tested
 232 using pairwise Wilcoxon rank sum test with *Holm* multiple comparison corrections. No significant
 233 difference was found in any tests (Table S3 and S4).

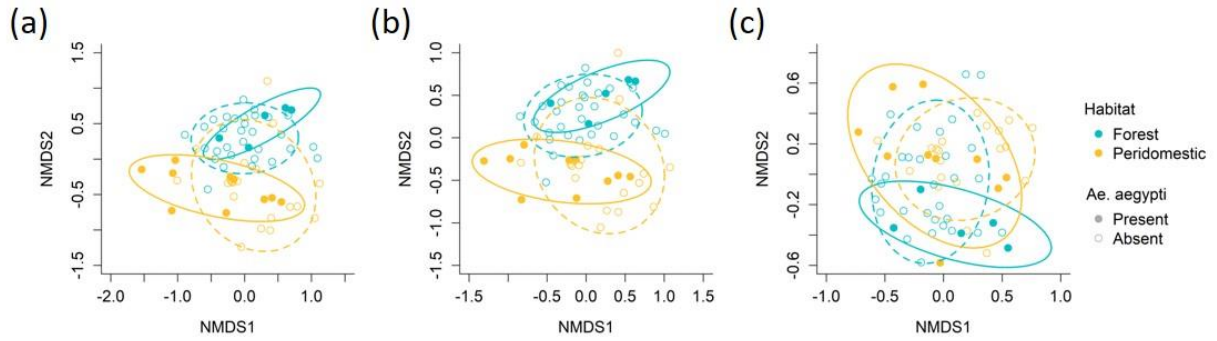
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235

236 **Figure S8.** Comparison of the Shannon index of the bacterial community in Rabai oviposition sites at
 237 different taxonomic levels: (a) ASVs, (b) Species, (c) Genus, and (d) Family. Each point represents a
 238 single oviposition site, and the boxplots show the minimum, 25% quartile, median, 75% quartile, and
 239 maximum of all values. The colors and shapes are as in Figure 2. Differences between groups were tested
 240 using pairwise Wilcoxon rank sum test with *Holm* multiple comparison corrections. No significant
 241 difference was found in any tests (Table S5 and S6).

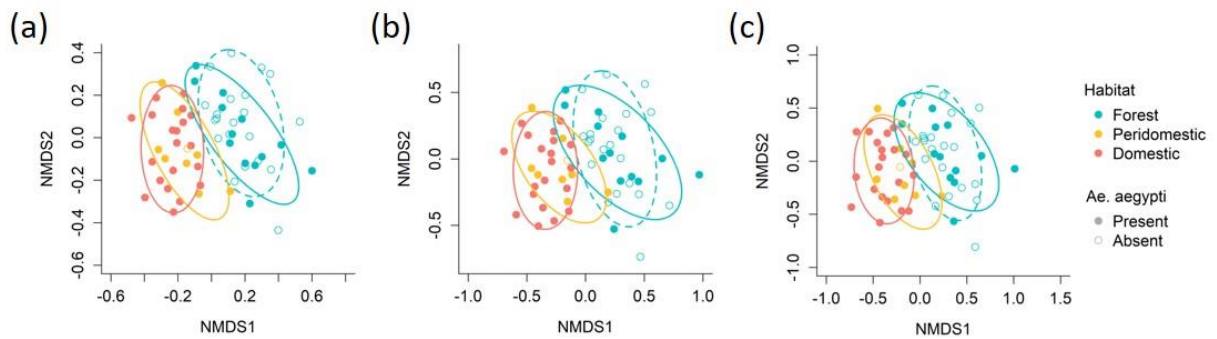
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243

244 **Figure S9.** NMDS analysis of bacterial community compositions in La Lopé oviposition site at (a)
 245 Species, (b) Genus, and (c) Family level. Each point represents an oviposition site. The color and shape of
 246 points, as well as the ellipses, are the same as in Figure 2.

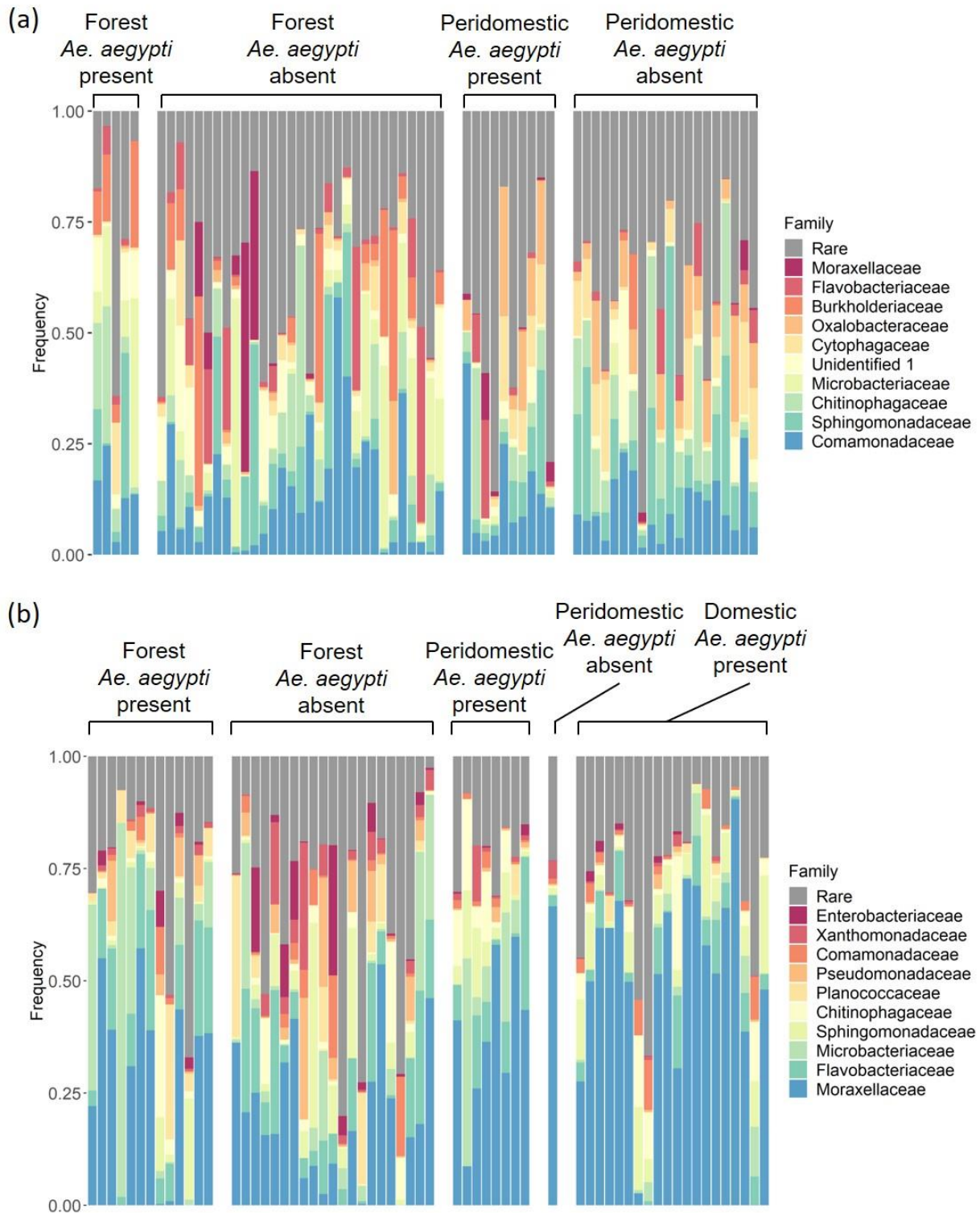
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249 **Figure S10.** NMDS analysis of bacterial community compositions in Rabai oviposition site at (a)
 250 Species, (b) Genus, and (c) Family level. Each point represents an oviposition site. The color and shape of
 251 points, as well as the ellipses, are the same as in Figure 2.

252



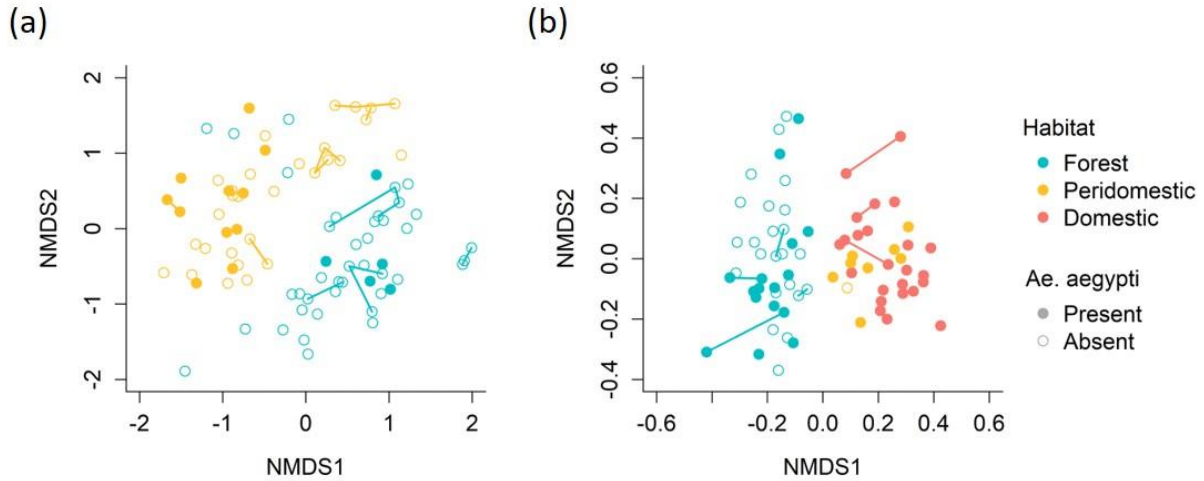
253

254 **Figure S11.** Frequency of the top ten bacterial Families in oviposition site in (a) La Lopé and (b) Rabai

255 oviposition sites. Each bar represents an oviposition site, and the length of each color represents the

256 proportion of the corresponding Family in the site. Other bacterial Families are grouped in the “Rare”
257 category, which shows as gray in the bar plots. Samples are grouped by oviposition site groups, which are
258 labeled above the bar plots.

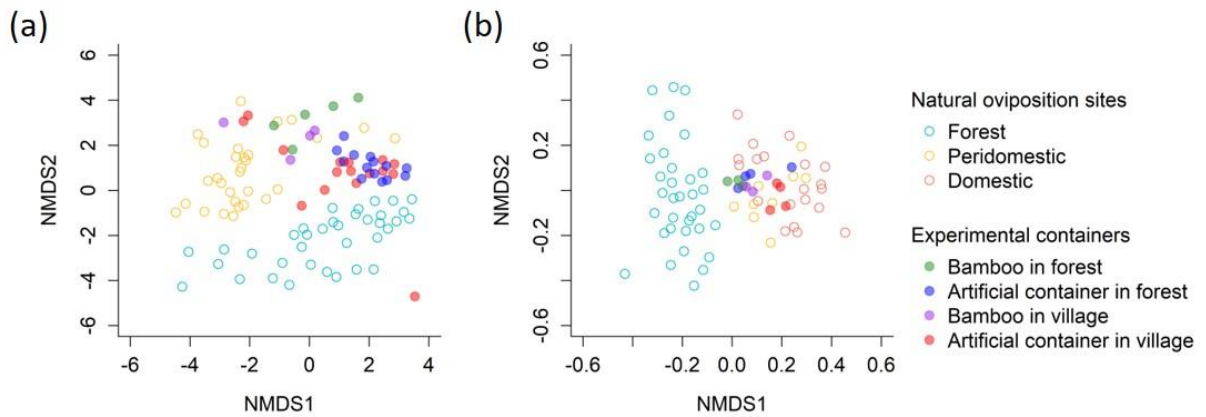
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261 **Figure S12.** Temporal variation of bacterial community compositions at the ASV level in oviposition
262 sites in (a) La Lopé and (b) Rabai. Each point represents an oviposition site. The color and shape of
263 points, as well as the ellipses, are the same as in Figure 2. Bacterial samples collected from the same
264 oviposition sites at different times are linked with segments.

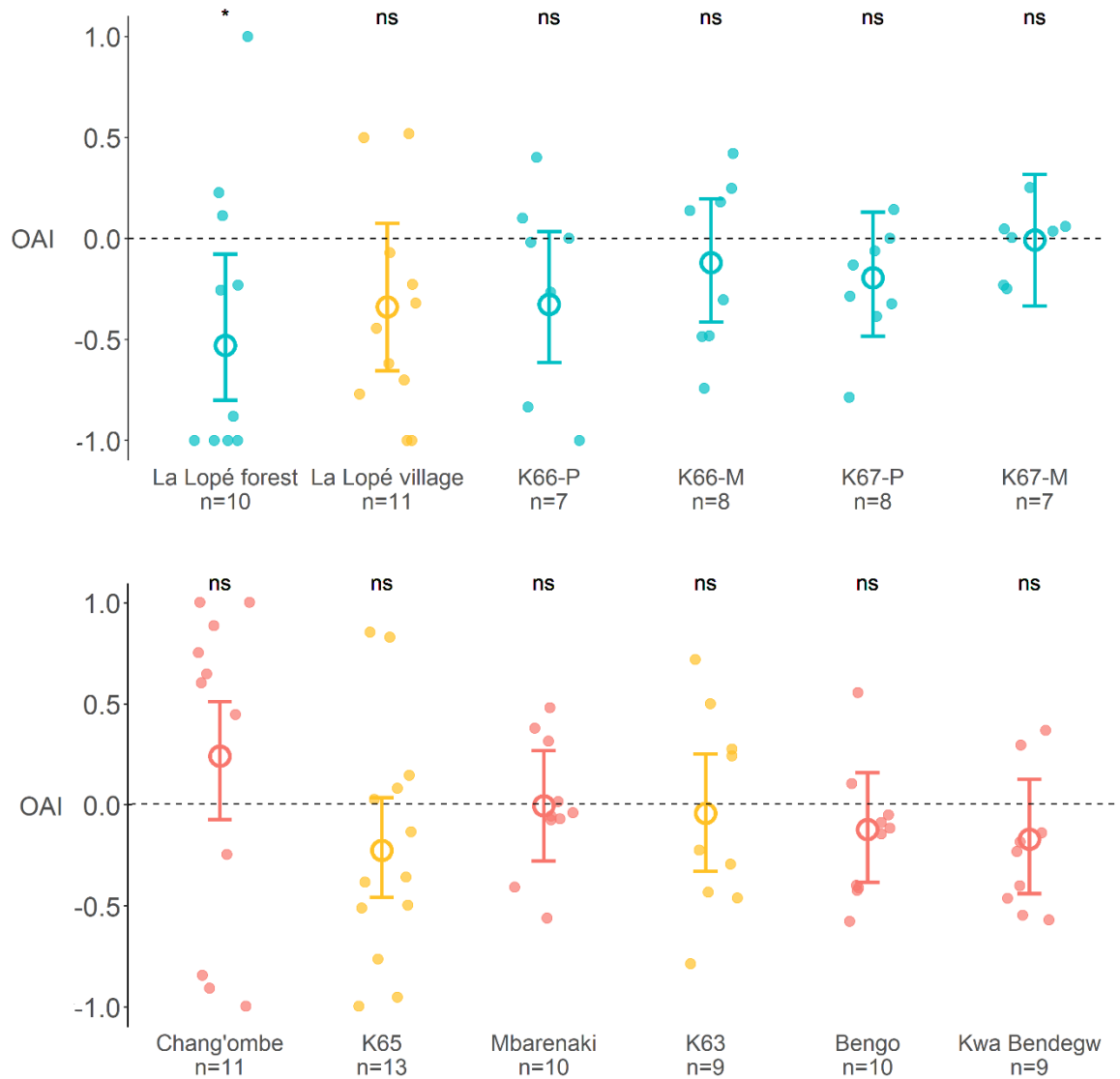
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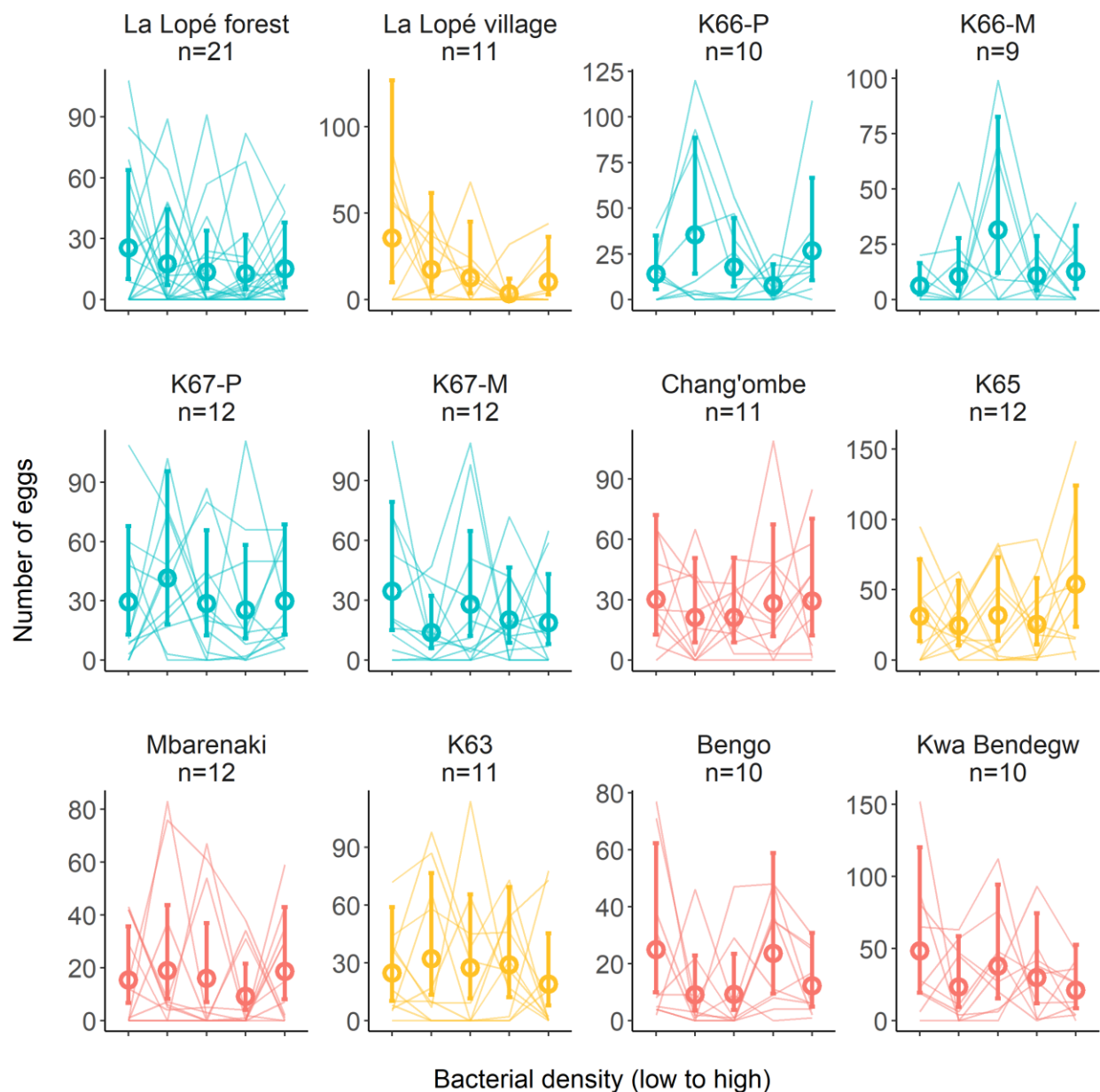
267 **Figure S13.** Bacterial community compositions of the containers used in the field oviposition choice
 268 experiments in (a) La Lopé and (b) Rabai. The NMDS was performed at the ASV level. Each point
 269 represents an oviposition site. Hollow points represent natural oviposition sites, and solid points represent
 270 experimental containers. The two types of experimental containers in the two habitats were differentiated
 271 by different colors.

272



273

274 **Figure S14.** Results of laboratory oviposition assay on forest vs. village bacteria culture summarized by
 275 colonies. The details of the two bacterial cultures were described in Table S2. Each point represents one
 276 cage with five gravid females, and the color indicates the habitat where the colony originated. Higher OAI
 277 indicates preference for forest bacteria cultures. K66-P and K66-M represents the two copies of the same
 278 forest colony maintained at Yale and Princeton University. A beta-binomial model was used to test
 279 differential preference among colonies, which results in no significant colony effects. The model also
 280 estimated the mean OAIs and the 95% confidence intervals of all colonies, indicated by the hollow circles
 281 and the error bars. The asterisks and 'ns' above each colony indicates whether the 95% CI excludes zero.



283

284 **Figure S15.** Results of laboratory oviposition assay on bacterial density summarized by colonies. Five
 285 cups were provided in each cage with increasing bacterial density at 0, 2×10^5 , 1×10^6 , 5×10^6 , 2.5×10^7
 286 cells/mL (details in Table S2), which correspond to the five columns in each subplot. Each line connects
 287 the five egg counts in one cage. Colors represent the habitats from where the colonies came. A negative-
 288 binomial model was used to fit the results of each oviposition assay. The model estimates the mean

289 number of eggs in each bacterial density and a 95% confidence interval, which are shown by the hollow
290 cycles and the error bars, respectively.

291

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