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EFFECTS OF MAGNETIC FIELDS ON MOSQUITOES

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ABSTRACT. Phylogenetically diverse organisms, including some insects, are able to detect and respond to magnetic fields comparable to the Earth's magnetic field. Because of their tremendous importance to public health, mosquitoes were tested for the presence of remanent ferromagnetic material indicative of a biological compass and also tested for behavioral responses to magnetic fields. Using a superconducting quantum interferometry device, we found that significant remanence was probably due to attraction of ferromagnetic dust onto the surface of live or dead mosquitoes. Most mosquitoes placed in a 1.0-gauss, uniform magnetic field moved until they were oriented parallel to the field. Two of 3 species of mosquitoes tested took fewer blood meals in a rotating magnetic field than in the Earth's normal magnetic field.

KEY WORDS Magnetic fields, mosquitoes, *Anopheles, Aedes, Culex, Culiseta, Psorophora*

INTRODUCTION

The ability to detect magnetic fields and then respond to them behaviorally is present in a wide variety of organisms (Gould 1984, Kirschvink et al. 1985). Bacteria, fish, reptiles, birds, and mammals orient in response to magnetic fields comparable in strength to the Earth's natural fields. Among insects, 2 species of beetles (Arendse 1978), a butterfly (Jones and MacFadden 1982), and the honeybee (Gould et al. 1978, Towne and Gould 1985) change direction in response to the orientation of magnetic fields, have the capability of detecting magnetic fields, or alter specific behaviors according to the direction of the field to which they have been exposed.

Although magnetic fields can influence biochemical reactions directly (Cremer-Bartels et al. 1984), the most commonly observed effect involves organs that function like compasses by providing information on the orientation of the magnetic field surrounding the individual organism. Where the magnetic sensing organ has been identified, it usually includes crystalline magnetite (Fe_3O_4) particles, although magnetic iron sulfide compounds have been found in several species of bacteria (Farina et al. 1990, Mann et al. 1990). Because of spatial, daily, and seasonal variation in the Earth's magnetic field, organisms could potentially accomplish a number of purposes by detecting the fields (Jones and MacFadden 1982). Like a compass, organisms could sense direction, with or without the ability to distinguish polarity of the field. Organisms that travel relatively long distances might be able to use geographic differences in the magnetic field, which vary over the surface of the Earth in total strength (from 0.24 to 0.68 gauss) as well as in both vertical and horizontal directions (Skiles 1985). These geographical differences could potentially function as a global map with magnetic landmarks. Finally, diel and seasonal periodicity of the Earth's magnetic field, caused mainly by lunar tidal effects on currents in the ionosphere (Campbell 1989, Winch 1989), could contribute toward entrainment of periodic behavior.

The ability of mosquitoes to sense and respond to magnetic fields had never been examined before the current study. Considering the tremendous practical importance of mosquitoes as vectors of diseases such as malaria and dengue, as well as the discomfort caused directly by their bites, the potential use of magnetic fields to alter mosquito behavior is a topic worth examination.

MATERIALS AND METHODS

Two kinds of experiments were conducted in order to determine whether magnetic fields influence mosquito behavior. First, the magnetic properties of mosquitoes were measured in a superconducting quantum interferometry device (SQUID) to determine whether the mosquitoes contained ferromagnetic particles that could be associated with detection of magnetic fields (Fuller et al. 1985). The 2nd experiment involved observation of live mosquitoes in uniform magnetic fields generated by large Helmholtz coils (Grant and Phillips 1990).

Detection of magnetic remanence: The SQUID is a standard laboratory apparatus (Quantum Design Magnetic Property Measurement system Model MPMS2, San Diego, CA) that measures the tiny current generated in superconducting coils as a sample in helium gas passes through them in the presence of an externally applied magnetic field. The data signal is simple amperage, but software provided with the machine calculates electromagnetic units (emu) generated by the sample. Following extensive trials, the following procedure was used to determine ferromagnetism of samples:

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Table 1. Mosquito sources. Sources of mosquitoes used in superconducting quantum interferometry device (SQUID) and behavioral experiments.

Scientific name	Source	Location collected[1]
Aedes aegypti (L.)	Colony	WRAIR
Ae. albopictus (Skuse)	Colony	AFRIMS
Ae. taeniorhynchus (Wied.)	Colony	USAMRIID
Ae. triseriatus (Say)	Field	Beltsville, MD
Ae. vexans (Meigen)	Field	Champaign, IL
Anopheles dirus Peyton and Harrison	Colony	AFRIMS
An. freeborni Aitkin	Field (SQUID)	Escondido, CA
	Colony (Behavior)	NIH
An. gambiae Giles	Colony	NIH
An. punctipennis (Say)	Field	Escondido, CA
An. stephensi Liston	Colony	WRAIR
Culex apicalis Adams	Field	Escondido, CA
Cx. quinquefasciatus Say	Colony	INHS
Cx. thriambus Dyar	Field	Escondido, CA
Cx. tritaeniorhynchus Giles	Colony	AFRIMS
Culiseta particeps (Adams)	Field	Escondido, CA
Psorophora columbiae (Dyar and Knab)	Field	Bowie, MD
Toxorhynchites splendens (Wied.)	Colony	AFRIMS

[1] WRAIR, Walter Reed Army Institute of Research, Washington, DC; AFRIMS, Armed Forces Research Institute of Medical Sciences, Bangkok, Thailand; USAMRIID, U.S. Army Medical Research Institute for Infectious Diseases, Fort Detrick, MD; NIH, National Institutes of Health, Bethesda, MD; INHS, Illinois Natural History Survey, Champaign, IL.

Mosquitoes were packed in a gelatin capsule (NDC 0002-2411-02 No. 4, Eli Lilly Co., Indianapolis, IN) with a small hole at 1 end to allow equalization of pressure. The capsule was wrapped in a few layers of Teflon tape (TFE Pipe Joint Tape, MIL-T027730A, Hercules Chemical Co., Inc., New York, NY) so that it would fit snugly in a plastic drinking straw (19.7 cm, wrapped, Sweetheart Cup Co., Inc., Chicago, IL). The straw was slipped on a Teflon plug wrapped onto the brass alloy sample rod. A pair of holes punched in the straw between the Teflon plug and sample was necessary to prevent loss of the capsule when the sample space was purged of air. The sample was loaded into the SQUID, the field run up to +1,000 gauss, then to 0 gauss, then to −1,000 gauss, and finally back to 0 gauss. Fifteen measurements of emu were taken at each of the 0-gauss fields, using the mean of the last 10 measurements for further calculations. Reported standard deviation was the mean of the sample standard deviations from each of the series of 0-gauss measurements. Ferromagnetism was one half of the difference between the 2 measurements expressed as emu. In order to determine that 1,000 gauss adequately coerced ferromagnetic particles and also to assure that the observed phenomenon was ferromagnetism rather that paramagnetism or diamagnetism, some initial samples were measured at a series of external magnetic fields, generating a hysteresis loop.

Preliminary trials showed that contamination of samples with magnetic material was a major source of error in performing the SQUID work. To minimize this problem, samples were handled with nonmagnetic tools, larval mosquitoes were reared in plastic containers, and adult mosquitoes were held in paper cups with nylon screen. Particulate contamination from surfaces was avoided by covering work surfaces with fresh paper and handling samples with frequently changed latex gloves. Although most gelatin capsules were clean to begin with, swabbing with acetone eliminated background in those few capsules that apparently contained magnetic particles. Some samples were loaded in a laminar flow hood in a further attempt to reduce contamination by air-borne magnetic particles.

Honeybees (*Apis mellifera* L.) were measured in the SQUID as a control, because they are known to contain a magnetic sensing organ in the abdomen. Three individual bees were triple rinsed in acetone and loaded into gelatin capsules. One bee was cut into 2 pieces, separating the head and thorax from the abdomen. Each piece was placed into a separate capsule for measurement. The bees were obtained alive from a hive at the Illinois Natural History Survey, Champaign, IL, in October 1994.

Mosquitoes came from several sources (Table 1) and were handled in various ways. Some of the 1st groups of mosquitoes had been dried in plastic containers and then loaded into gelatin capsules in the open in the SQUID laboratory. Later, dried mosquitoes were loaded into the capsules in a laminar flow hood in order to reduce the chances of contamination with magnetic particles. All of the results reported are for adult mosquitoes that had not taken any blood since emergence. One measurement was made on live female *Culex quinquefasciatus* Say that were anaesthetized with acetone immediately before loading them into a capsule.

Effects of magnetic fields on mosquito behavior: Behavioral experiments were conducted in a cham-

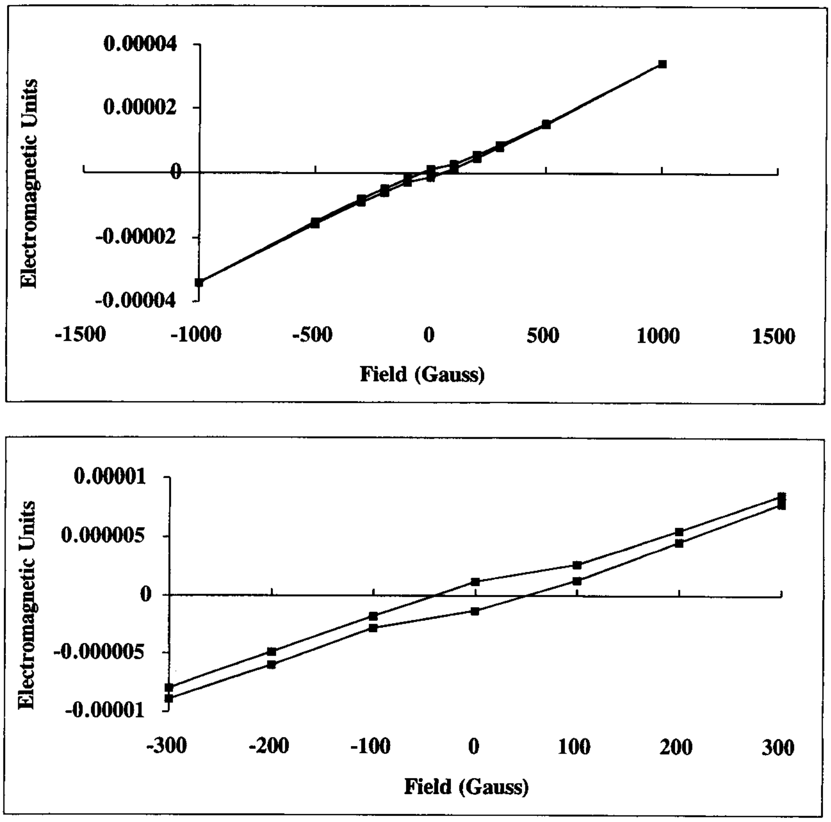

Fig. 1. Hysteresis loop of honeybee (*Apis mellifera*) abdomen with ferromagnetic moment of 9.41 × 10^{-7} electromagnetic units. Lower frame is enlargement to indicate gap between 0-gauss measurements.

ber constructed from 5.1-cm polyvinyl chloride pipe forming a cuboidal frame 1.83 m on a side. Six Helmholtz coils of 100 turns each were wrapped around the cube in 3 dimensions, placing the coils 91 cm apart and 46 cm from the edge of the cube. Each pair of coils in one dimension was powered by a magnet controller (High Power Precision bipolar Magnet Controller, Applied Magnetics Laboratory, Inc., Baltimore, MD), which provided current-regulated direct current up to 5 amperes. The entire chamber was covered in 2 layers of black plastic to provide complete darkness. Using this system, it was possible to create magnetic fields up to 1.9 gauss in any direction, with less than 10% variation in field strength and direction in a space of about 0.76 m^3 in the middle of the chamber. It also was possible to cancel the Earth's magnetic field, creating a null field. The current settings necessary to make a magnetic field of a given strength and orientation were determined by measuring the field with 3 gaussmeters (Gaussmeter GM1A with probe PM85, Applied Magnetics Laboratory, Inc.). The probes for the gaussmeters were placed in the center of the chamber, oriented at right angles to each other. Gaussmeters were calibrated in a 0-gauss chamber (Zero Gauss Chamber Type Z6-3, 1.9 cm diameter × 27.9 cm length, Applied Magnetics Laboratory, Inc.) before each use.

The 1st behavioral experiments tested the effect of a horizontal, 1-gauss field on orientation. Ten female mosquitoes were placed in a cylindrical paper carton covered with a layer of nylon screen and a layer of plastic film. The carton was divided into 4 equal quadrants with lines on the inside surface, creating a cross on the bottom when viewed from above. The carton was oriented so that the axis of the field bisected opposite sections. Mosquitoes were given the opportunity to respond to the magnetic field by inducing flight with taps of the carton against the table. Mosquitoes were then left undisturbed in the magnetic field for 5 min with no observer in the chamber and in total darkness. An observer reentered the chamber and counted the number of mosquitoes resting in each quadrant, being careful not to disturb the mosquitoes by illuminating the chamber with subdued light produced by a flashlight with 2 index cards over the lens. Experiments were performed with groups of 10 mosquitoes, testing each group 4 times. The chamber remained oriented in the same direction during the 4 tests, but the field was rotated clockwise horizontally by 90° each time. In this way, the possi-

Table 2. Remanence in bees. Ferromagnetism in worker honeybees (*Apis mellifera*).

Sample	Bee part	Ferromagnetism (emu × 10^{-7})[1] Mean	Sample SD
Bee No. 1	Head/thorax	1.52	0.32
	Abdomen	9.41	0.37
Bee No. 2	Entire	4.84	0.40
Bee No. 3	Entire	2.90	0.52

[1] emu, electromagnetic units.

bility of unknown, nonmagnetic cues was eliminated, because only the magnetic field changed direction. The experiment was repeated with 4 groups of mosquitoes during the day, which provided 16 separate counts. Two days of these experiments were performed for each of the following species, *Anopheles freeborni* Aitken, *An. gambiae* Giles, and *An. stephensi* Liston. Data from the orientation experiments were analyzed nonparametrically, calculating the 95% confidence limits for percentages (Steel and Torrie 1960) for the percentage of mosquitoes in sections parallel to the axis of the magnetic field compared to the percentage in sections perpendicular to the field.

The other set of behavioral experiments was designed to determine whether an unnatural magnetic field configuration could disrupt feeding. A Plexiglas® tube (15.2 cm in diameter, 91.4 cm in length) was used to confine mosquitoes to a linear path leading to a host. Groups of 10 mosquitoes 7–11 days old and starved 24–48 h were introduced through a hole in the middle of the tube and then allowed 10 min in total darkness to get a blood meal from a host at 1 end of the tube. Mosquitoes were removed from the tube after an experiment to determine whether or not each had taken a blood meal. To eliminate host odors that might influence orientation, the tube was cleaned between trials with distilled water followed by 95% ethanol and the tube was only handled with gloved hands. *Anopheles freeborni* were tested using a restrained mouse for bait, performing 7 sets of experiments under each of the following 3 conditions: the Earth's natural field, a horizontal field of 1 gauss rotating 90° each 15 sec (i.e., 1 rpm), and a null field. *Anopheles stephensi* was tested in 7 sets of experiments using a mouse as a blood source, comparing the Earth's field with a rotating 1-gauss field. This species was also tested using a human hand as a blood source, comparing the Earth's field to a rotating 1.9-gauss field. *Anopheles gambiae* was tested with a human hand as a blood source, comparing the Earth's field to a rotating 1.9-gauss field in 3 sets of tests. Results were analyzed using chi-square for 2 × 2 tables, the comparison consisting of number of mosquitoes bloodfed or not bloodfed under pairs of magnetic conditions. Analysis was performed on the combined data for all trials of each species.

Table 3. Remanence in normal mosquitoes. Ferromagnetism in mosquitoes processed in the open and tested in gelatin capsules.

Sample	Ferromagnetism (emu × 10^{-7})[1] Mean	Sample SD
Empty capsule before	1.45	0.22
Culex quinquefasciatus (30 females)	10.75	0.41
Empty capsule after	11.55	0.43
Empty capsule before	1.50	0.30
Aedes taeniorhynchus (11 females)	3.65	0.19
Empty capsule after	3.70	0.30
Mosquitoes replaced in capsule	6.20	0.37
Empty capsule before	0[2]	—
Aedes vexans (30 females)	5.05	0.29
Empty capsule after (cleaned)	0[2]	—
Mosquitoes replaced in capsule	12.90	0.42

[1] emu, electromagnetic units.

[2] Hysteris loop closed with no gap between 0-gauss readings.

RESULTS

Detection of magnetic remanence

Five of 8 empty gelatin capsules had no remanent ferromagnetic moment and the other 3 had moments less than 2.42 × 10^{-7} emu. Based on these results, any moment less than 2.42 × 10^{-7} emu was considered below background. Two of the 3 bees had a significant remanent ferromagnetic moment, with the moment concentrated in the abdominal portion of the bee (Table 2). The shape of the hysteresis loop (Fig. 1) showed that the observed moment was ferromagnetic in nature.

Groups of mosquitoes also had significant remanent magnetic moments (Table 3), averaging 3.6 × 10^{-8} emu per female *Cx. quinquefasciatus*, 3.3 × 10^{-8} emu per female *Aedes taeniorhynchus* (Weidemann), and 1.7 × 10^{-8} per female *Aedes vexans* (Meigen). The nature of the moment was identical to that of the bees, as illustrated by the hysteresis loop (Fig. 2). Surprisingly, the signal remained in the gelatin capsule after removal of the mosquitoes. Even more remarkably, the signal increased by a factor of 1.7–2.6 when the mosquitoes were reintroduced in the same capsule. Living *Cx. quinquefasciatus* females anesthetized with acetone produced similar results. Fifteen females in a capsule that originally had no moment produced a signal of 1.9 × 10^{-6} emu, but after removal of the mosquitoes, the capsule retained a moment of 1.7 × 10^{-6} emu. Efforts to protect the mosquitoes from ferromagnetic dust by loading them into capsules in a laminar flow hood reduced the observed remanent ferromagnetic moment below the threshold for 13 species of mosquito in 5 genera (Table 4). Only 1

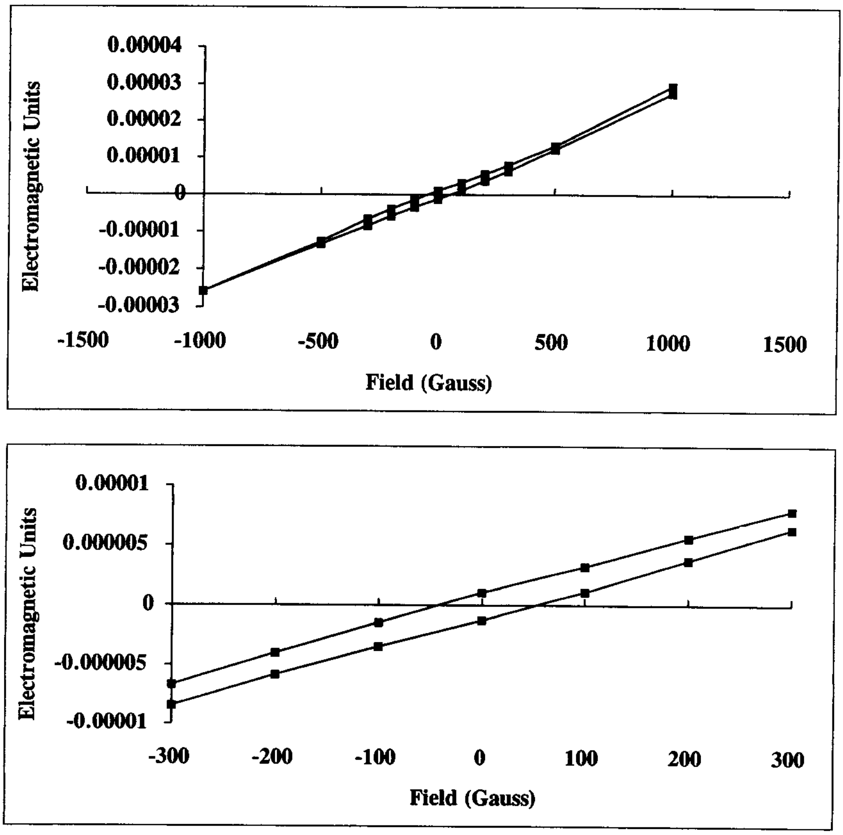

Fig. 2. Hysteresis loop of 30 female *Culex quinquefasciatus* mosquitoes with ferromagnetic moment of 10.75 × 10^{-7} electromagnetic units. Lower frame is enlargement to indicate gap between 0-gauss measurements.

Table 4. Remanence on dust-free mosquitoes. Ferromagnetism in mosquitoes processed in laminar flow hood.

Species	Number of specimens	Ferromagnetism (emu × 10^{-7})[1]	
		Mean	Sample SD
Aedes aegypti	30	0[2]	—
Ae. albopictus	10	2.25	1.20[3]
Ae. triseriatus	13	2.19	0.50
Ae. vexans	30	1.14	0.31
Anopheles dirus	30	1.42	0.46[3]
An. freeborni	22	0.96	0.35
An. punctipennis	4	1.58	0.27
An. stephensi	10	1.04	0.32
Culex apicalis	11	1.34	0.34
Cx. thriambus	18	2.24	0.21
Cx. tritaeniorhynchus	20	1.63	0.43
Culiseta particeps	3	1.82	0.27
Toxorhynchites splendens	10	1.38	0.21
Psorophora columbiae	10	4.25	0.33
Empty capsule after		0	—

[1] emu, electromagnetic units.

[2] Hysteresis loop closed with no gap between 0-gauss readings.

[3] Measured only once, error based on shape of curve.

species, *Psorophora columbiae* (Dyar and Knab), produced a significant signal of 4.2 × 10^{-8} emu/female.

Effects of magnetic fields on mosquito behavior

Behavioral experiments showed that females of *An. freeborni, An. gambiae,* and *An. stephensi* have a greater tendency to rest on the sides of a container parallel to a magnetic field than on the sides perpendicular to the field (Table 5). For all 3 species, approximately two thirds of the females (62–68%) came to rest in the 2 quadrants of a round container parallel to a uniform, horizontal 1.0-gauss field. Differences were statistically significant, as indicated by the lack of any overlap in 95% confidence limits of the percentages of mosquitoes in pairs of quadrants. Also, great consistency occurred between trials for individual quadrants, adding more confidence that the observed effect was real.

Results of experiments testing the proportion of mosquitoes that would blood feed under various magnetic conditions are presented in Fig. 3. Statistically significant reductions in feeding were observed for 2 of the species exposed to rotating mag-

Table 5. Orientation in magnetic field. Distribution of mosquitoes in relation to a horizontal, 1.0-gauss magnetic field.

Species	Day	Total no. (% ± 95% CL) mosquitoes	
		Parallel	Perpendicular
Anopheles freeborni	1	100 (62 ± 7.5)	60 (38 ± 7.5)
	2	107 (67 ± 7.3)	53 (33 ± 7.3)
An. gambiae	1	110 (69 ± 7.2)	50 (31 ± 7.2)
	2	106 (66 ± 7.3)	54 (34 ± 7.3)
An. stephensi	1	108 (67 ± 7.3)	53 (33 ± 7.3)
	2	109 (68 ± 7.2)	51 (32 ± 7.2)

netic fields. When *An. freeborni* were exposed to a rotating 1.0-gauss field a 39% reduction in feeding occurred compared to mosquitoes in the Earth's field ($n = 70$ mosquitoes, $\chi^2 = 5.77$, $P = 0.016$). When *An. stephensi* were exposed to either a 1.0-gauss or 1.9-gauss rotating field (combining the results from both experiments) a 27% reduction occurred in feeding ($n = 140$ mosquitoes, $\chi^2 = 4.31$, $P = 0.038$). Exposure of *An. gambiae* to rotating fields and of *An. freeborni* to null fields did not affect the rate of blood feeding.

DISCUSSION

Our procedures for using the SQUID were adequate to detect the magnetic remanence in honeybees. As observed by Gould et al. (1978), we saw variation between individual bees and concentration of the signal in the abdomen (Table 2). The maximum signal detected by Gould et al. (1978) was in freshly killed bees and reached 2.7×10^{-6} emu, compared to a maximum signal of 9.4×10^{-7} emu observed in our study.

Applying the same technique to measurement of mosquitoes, we found that some specimens had a significant magnetic remanence, but that the material causing this remanence was easily left behind in the sample chamber (Table 3). In 2 of 3 cases, the signal was greater when mosquitoes were replaced in the same sample chamber. On the other hand, a series of 13 species of mosquitoes in 5 genera did not have significant magnetic remanence when they were handled in a way to avoid external contamination (Table 4). Only a single species, *Ps. columbiae,* seemed to contain ferromagnetic material internally.

Analysis of these data suggests that most mosquitoes probably lack an internal magnetic organ analogous to that found in bees. However, the external surfaces of both dead and live mosquitoes seem to have a great affinity for ferromagnetic particles in the air. Such particles may be common in the air, as dust at low elevations has been found to contain significant quantities of ferromagnetic material. In a study of dust settling onto the ocean, Young et al. (1991) found that 10–15% of the dust was composed of iron with a mean diameter of 2–3.5 µm. Analyzing urban dust in France, Berry et al. (1980) measured 8% as iron oxide and 6% as amorphous aluminum or iron. The ubiquitous nature of iron dust is discussed as a confounding factor in laboratory tests (Jones and Wallace 1992, Kobayashi et al. 1995). Considering what may be the universal presence of ferromagnetic dust and the great affinity of external surfaces of mosquitoes for such dust, it seems possible that a covering of ferromagnetic dust could be a constant feature of many species of mosquitoes. Such a covering might be involved in biological mechanisms with which at least some species of mosquitoes detect magnetic fields.

Although the mechanism is uncertain, 3 species

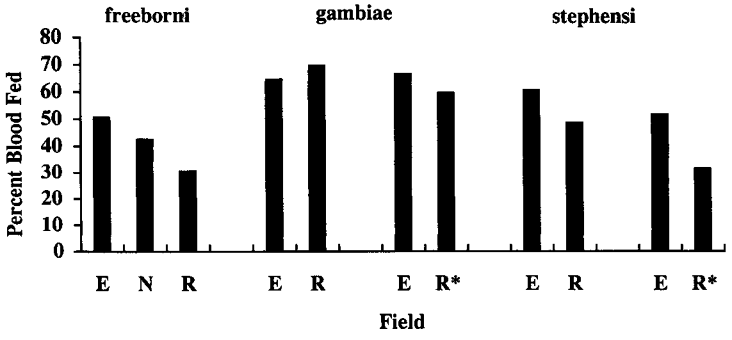

Fig. 3. Percentage of mosquitoes taking blood meals during exposure to Earth's magnetic field (E), null field (N), rotating horizontal 1.0-gauss field (R), or rotating 1.9-gauss field (R*).

of *Anopheles* mosquitoes were able to orient in a magnetic field, with approximately two thirds of them preferring to rest parallel to the field rather than perpendicular to it. This sort of axial orientation in a magnetic field without a preference for either pole is similar to orientation observed in bees (Towne and Gould 1985) and sandhoppers (Arendse 1978).

We can only speculate on the adaptive advantages of the ability of mosquitoes to orient in magnetic fields. One possibility is that orientation in the Earth's magnetic field supplements other means for maintaining a straight line of flight toward an attractive stimulus over a relatively short distance. Another possibility is that migratory species (e.g., *Ae. vexans*) use their sensitivity to magnetic fields as an aid in reaching specific areas suitable for oviposition or for host seeking. At the least, a magnetic sense would tend to keep the migratory species moving in a straight line during long-distance travel.

The disruption of blood feeding by *An. stephensi* and *An. freeborni* observed in this study were probably the result of the unnatural disturbance created by a moving magnetic field. Possibly, the mosquitoes were able to detect the magnetic fields but sensed a magnetic pattern that was impossible in nature, distracting them from feeding. Perhaps a stronger field or a different pattern of rotation would cause greater disruption of feeding.

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