These data demonstrate that the fire ant has a magnetic sense and that a change in magnetic-field direction disrupts the initial homing ability of workers. Thus, fire ants can now be included with the increasing variety of organisms having a demonstrated ability to perceive the earth's magnetic field for orientation or navigation. The sensory mechanisms by which the geomagnetic field is transduced into neural signals are still unclear, but an internal ferromagnetic material, magnetite, has been found in many of these organisms [15]. We propose that magnetite is also likely to be involved in fire ant geomagnetic sensing and plan to test for its presence.

Central place foragers such as ants must search widely, yet keep track of their location relative to their nest. Powerful selection for efficient foraging strategies in these animals has produced complex systems of navigation which use information from a variety of sources [1, 3]. It is well known that celestial information (the sun, the pattern of polarized light, the moon) and terrestrial landmarks can be important visual cues [16]. Beyond maze learning [17], and gravitational [9] and pheromonal orientation [5] already reported for fire ants, our studies add geomagnetic orientation to form a complex hierarchy of possible homing and orientation mechanisms that contribute to the success of this species. The demonstration of geomagnetic orientation in an ant species may be broadly applicable and provide an explanation of worker orientation within subterranean nests and the ability of some ants to forage successfully without obvious cues (visual or otherwise), a puzzle which has confounded researchers for years [18].

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1. Wehner, R.: Ann. Rev. Entomol. *29*, 277 (1984)
2. Wehner, R., Harkness, R.D., Schmid-Hempel, P., in: Information Processing in Animals, p. 1 (M. Lindauer, ed.). Stuttgart: G. Fischer 1983
3. Jaffé, K., Ramos, C., Lagalla, C., Parra, L.: Insectes Soc. *37*, 101 (1990)
4. Porter, S.D., Tschinkel, W.R.: Environ. Entomol. *16*, 802 (1987)
5. Wilson, E.O.: Anim. Behav. *10*, 148 (1962)
6. Vander Meer, R.K., in: Fire Ants and Leaf-cutting Ants: Biology and Management, p. 201 (C.S. Lofgren, R.K. Vander Meer, eds.). Boulder, CO: Westview 1986
7. Wilson, E.O.: Anim. Behav. *10*, 134 (1962)
8. Kidd, K.A., Apperson, C.S.: J. Agric. Entomol. *1*, 212 (1984)
9. Marak, G.E., Wolken, J. J.: Nature *205*, 1328 (1965); Howard, D.F., Tschinkel, W.R.: Behaviour *56*, 157 (1976); Vander Meer, R.K.: unpublished
10. Vinson, S.B., MacKay, W.P., in: Applied Myrmecology: A World Perspective, p. 496 (R.K. Vander Meer, K. Jaffé, A. Cedeño, eds.). Boulder, CO: Westview 1990
11. Blakemore, R.P.: Science *190*, 377 (1975)
12. Mather, J.G., Baker R.R.: Nature *291*, 152 (1981)
13. Rosengren, R., Fortelius, W.: Insectes Soc. *33*, 306 (1986)
14. Hölldobler, B.: Behav. Ecol. Sociobiol. *1*, 3 (1976)
15. Kirschvink, J.L., in: Biomagnetism: An Interdisciplinary Approach, p. 501 (S. Williamson, ed.). New York: Plenum 1983
16. Rosengren, R.: Acta Zool. Fenn. *150*, 1 (1977)
17. Stratton, L.O., Coleman, W.P.: J. Comp. Physiol. Psychol. *83*, 7 (1973)
18. Cosens, D., Toussaint, N.: Anim. Behav. *33*, 541 (1985)

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Inhibition of Male Calling by Heterospecific Signals

Artifact of Chorusing or Abstinence During Suppression of Female Phonotaxis?

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Various animals in which sexually advertising males signal acoustically exhibit the curious phenomenon of refraining from calling in the presence of certain heterospecific song [1]. This interaction has been reported from orthopteran insects, anurans, birds, and mammals. Typically, the response is unilateral and short-term: the inhibited species adjusts the timing of its calls such that they are broadcast during brief gaps in the inhibitor's song. In several instances the affected species exhibits a long-term response as well in which its diel periodicity is shifted and the singing periods of the inhibitors are largely avoided [2–4]. Heterospecific inhibitory responses have been proposed to be either artifacts of intraspecific male-male chorusing interactions or abstinence from calling at a time when females would not be effectively or "correctly" influenced by it [1, 5]. My recent studies on female phonotaxis in the neotropical katydid *Neoconocephalus spiza* (Orthoptera: Tettigoniidae) provide results consistent with the latter hypothesis but do not necessarily negate the former one. Consideration of the nature of female choice in *N. spiza* suggests that limitations in female perception ultimately drive male inhibition.

Four species of *Neoconocephalus* katydids are common in the lowlands of central Panama [6], and males of each call during evening hours throughout much of the year. *N. spiza* males are inhibited from singing by the calls of any of the other three (*N. affinis, N. punctipes, N. triops*), and in areas where populations of inhibitors are high, *N. spiza* shifts from nocturnal to diurnal calling [3]. While microhabitat differences between the species exist, their songs are sufficiently loud (> 80 dB SPL at 1 m; 0 dB re 20 μPa) that, given the low threshold for inhibition (≈ 40 dB), the calling of *N. spiza* males is commonly stifled in the

field. The songs of all four species are similar in frequency range (9–16 kHz), but their temporal patterns differ. *N. spiza* produce short "chirps" (17–74 ms in length) at 1.82–3.60 s^{-1}, whereas the other three species produce "whines" or "buzzes" that continue uninterruptedly from several s to 30 min or longer.

To investigate potential causes of heterospecific inhibition in *Neoconocephalus* katydids, I studied the phonotactic orientation of female *N. spiza* toward conspecific calls either broadcast alone or in the presence of *N. affinis* calls. Female nymphs were collected in the vicinity of Gamboa, Panama, and reared to maturity under natural photoperiodic and temperature conditions in a screened room isolated from calling males. I tested the responses of these females, ≥ 20 days past the adult molt, to *Neoconocephalus* songs in an acoustically insulated arena (diameter 4 m) in a darkened laboratory during natural scotophase. The arena was illuminated from above by diffused red light to facilitate observation.

In each trial I released an individual female in the arena center and, after an initial 5-min "accustoming" period, presented *Neoconocephalus* calls from two loudspeakers on opposite sides. Loudspeakers #1 and #2 broadcast *N. spiza* and *N. affinis* song, respectively. After 10 min, calls from loudspeaker #2 ceased, while those from loudspeaker #1 (*N. spiza* chirps) continued alone for another 10 min. I monitored the movement and behavior of the female throughout the 25-min trial. Females were tested once in this experiment, and the 20-min playback was begun only for individuals that remained within a 20-cm radius of the arena center during the accustoming period.

Playback stimuli were made from genuine calls recorded on DAT tape. *N. spiza* chirps (rate ≈2.5 s^{-1}) and a continuous *N. affinis* buzz thus recorded were copied onto channels 1 and 2, respectively, of a computer signal editing system. These signals were continuously "looped" and transferred back to a 20-min stereo DAT tape segment, the stimulus tape; channel 2 was turned off at the 10-min juncture. For phonotaxis trials the stimulus tape was played on a DAT recorder and broadcast via two high-frequency loudspeakers. I adjusted the amplitudes of these broadcasts such that each was 57 dB at the arena center; this relatively low SPL reduced the possibility of saturating the insect's hearing [7]. The initial recordings of the calls and the phonotaxis trials were all conducted at ambient temperatures of 25–27 °C.

Sixteen of the tested females remained at the arena center during the accustoming period, and ten of these insects exhibited phonotaxis during the playback. All ten insects remained stationary until *N. affinis* song ceased but then quickly walked or jumped to within 40 cm of loudspeaker #1 and remained there for at least 30 s (Fig. 1). The latencies for phonotaxis in these ten trials were substantially longer than latencies obtained when these same females were exposed solely to *N. spiza* chirps from the beginning of a playback session (Fig. 1). Therefore, failure to orient during the initial 10 min of the playback trials could not be attributed merely to a prolonged latency in female phonotaxis.

Suppression of orientation toward male chirps in female *N. spiza* exposed to heterospecific calls was not predicted at first. The two stimuli presented were 180 ° apart with respect to the female, and, based on the neurophysiological capabilities of other ensiferan orthopterans [8], separate processing of the calls was presumed (such capabilities were not found, however, in a caeliferan orthopteran [9]). Unlike the one previous test of this phenomenon (in a hylid frog) [10], females did not merely prefer conspecific calls over a composite of conspecific + heterospecific ones in a two-

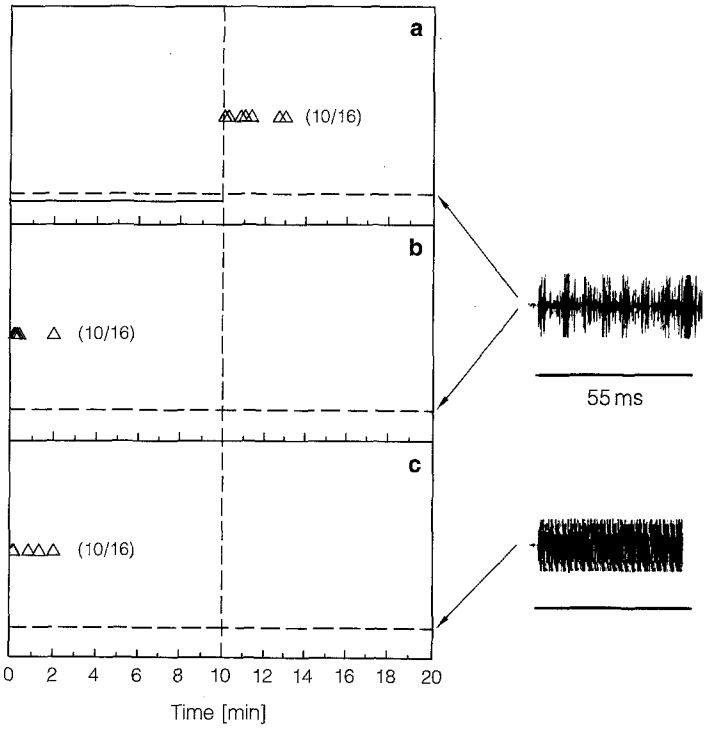

Fig. 1. Latencies of phonotaxis of *N. spiza* females presented with a) *N. spiza* chirps and *N. affinis* song from two respective loudspeakers, b) *N. spiza* chirps alone from one loudspeaker, c) *N. spiza* modified (330-s^{-1} pulse rate) chirps alone from one loudspeaker. *Dotted* and *solid horizontal lines* in each graph indicate playback durations of *N. spiza* and *N. affinis* calls, respectively, and oscillograms to the right indicate envelopes and pulse rates of *N. spiza* chirps. *Triangles* represent times when phonotaxis of individual females to *N. spiza* chirps began. In each test the *N. spiza* chirp rate was ≈2.5 s^{-1}, chirp length was ≈55 ms, and amplitude was ≈57 dB (at the female). Modified chirps used in (c) were made by digitally editing the genuine chirps used in (a) and (b). Females were tested singly, and the same individuals were used in all three experiments. Parenthetic values are proportions of tested females that exhibited phonotaxis. Phonotaxis latencies of given individuals are significantly longer in (a) than in (b) ($P < 0.01$; sign test), but values in (b) and (c) do not differ significantly ($P > 0.20$)

choice test. Rather, *N. spiza* females failed to orient at all during heterospecific calling.

An additional reason for surprise at finding complete suppression of orientation was that acoustic stimuli eliciting phonotaxis in female *N. spiza* were not delimited by a restrictive set of parameters. *N. spiza* chirps consist of 2–10 "pulses" (sound made by a complete cycle of forewing movement), but the specific pulse rate ($\approx$150 s^{-1}) within the chirp is unnecessary for phonotaxis. A modified 55-ms chirp with a 330-s^{-1} pulse rate (modification was accomplished via the computer signal editing system with which I placed pulses so close together that there were no intervals between them) and a natural chirp elicited comparable responses (Fig. 1). Furthermore, females responded (at a given temperature) to a range of chirp rates from 2 to 6 s^{-1} and to chirps (with the specific pulse rate) of lengths from 55 to 150 ms (Fig. 2). Therefore, the obscuring of critical signal parameters by a continuous call in the background seemed improbable.

A study of male synchronous chorusing, however, showed that females given a choice of two stimuli were very selective with respect to the relative length and timing of chirps [11]. In particular, they exhibited an overriding preference for the leading chirp in a closely synchronized sequence. This preference was effected by stimulation from the sudden onset of sound, a feature retained by the leading chirp.

The attractiveness of leading chirps may originate in a "sensory bias" [12] and/or Fisherian or direct sexual selection [13] in which *N. spiza* female preference coevolved with a male trait of producing mostly leading chirps during signal interactions. In either case the ability of females to hear, assess, and orient toward chirps would be expected to be severely limited during heterospecific calling. That is, chirps masked by continuous sound might simply provide insufficient stimulation because of the obscuring of sudden onsets of sound, or females forced to evaluate several males against a background of continuous sound might desist to avoid making an "incorrect" choice even if absolute stimulation is sufficient. Thus, heterospecific inhibition of calling in *N. spiza* would be coupled – either genetically [14–16] or via adaptive intersexual coevolution [17] – with suppression of female phonotaxis.

Despite the suppression of female phonotaxis, likely the outcome of limitations in female hearing or orientation as indicated above, inhibition of male calling by heterospecific song could also be an artifact of chorusing. Signal interactions in male *N. spiza* are governed by an "inhibitory-resetting" mechanism in which individuals are mutually inhibited from calling by each other's chirps but are then "reset" such that the chirp period following inhibition is shortened [11]. This mechanism entails inhibition of calling throughout the duration of any continuous acoustic stimulus. Inhibitory-resetting appears to have been selected for because a male adhering to it increases its chance of producing leading chirps [11]. Therefore, inhibition of calling by heterospecific song could represent, in part, an incidental consequence of signal interactions. Nonetheless, one might question, given that naturally occurring chirps are uniformly short, why a mechanism wherein a male is immediately reset by the onset of another's call is not employed. Such resetting would be equally effective in yielding leading chirps but would not generate silence for many hours per night. This point either argues against the hypothesis that inhibition is a by-product of signal interactions or suggests the existence of constraints on the evolution of an ideally designed neural circuitry.

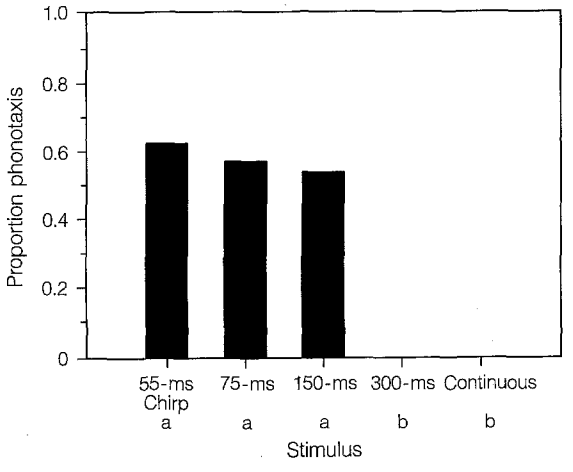

Fig. 2. Phonotactic responses of *N. spiza* females to male chirps of four different lengths and a continuous stimulus containing the same pulse structure as a chirp. Chirp length was varied by digitally editing a single genuine 55-ms chirp. Stimulus presentation was the same as in Fig. 1b and c. Up to five different stimuli were presented to an individual in separate tests during a given night; the sequences of tests for the various individuals were arranged in a Latin square design. Bar height indicates the proportion of tested females exhibiting phonotaxis (see text) to the stimulus; stimuli followed by different superscript letters have significantly different proportions ($P < 0.05$; McNemar test)

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1. Schatral, A., in: The Tettigoniidae: Biology, Systematics and Evolution, p. 152 (W.J. Bailey, D.C.F. Rentz, eds.). Berlin: Springer 1990
2. MacKinnon, J.: Behaviour *22*, 3 (1974)
3. Greenfield, M.D.: Anim. Behav. *36*, 684 (1988)
4. Römer, H., Bailey, W.J., Dadour, I.: J. Comp. Physiol. A *164*, 609 (1989)
5. Greenfield, M.D., in: The Tettigoniidae: Biology, Systematics and Evolution, p. 71 (W.J. Bailey, D.C.F. Rentz, eds.). Berlin: Springer 1990
6. Walker, T.J., Greenfield, M.D.: Trans. Am. Entomol. Soc. *109*, 357 (1983)
7. Wendler, G.: Naturwissenschaften *76*, 128 (1989)
8. Pollack, G.S.: J. Comp. Physiol. A *158*, 549 (1986)

9. Helversen, O. von: ibid. *154*, 837 (1984)
10. Schwartz, J.J., Wells, K.D.: Herpetologica *39*, 121 (1983)
11. Greenfield, M.D., Roizen, I.: Nature *364*, 618 (1993)
12. Ryan, M.J., Fox, J.H., Wilcyznski, W., Rand, A.S.: ibid. *343*, 66 (1990)
13. Kirkpatrick, M., Ryan, M.J.: ibid. *350*, 33 (1991)
14. Doherty, J., Hoy, R.: Quart. Rev. Biol. *60*, 457 (1985)
15. Boake, C.R.B.: Trends Ecol. Evol. *6*, 225 (1989)
16. Butlin, R.K., Ritchie, M.G.: Biol. J. Linn. Soc. *37*, 237 (1989)
17. Bauer, M., Helversen, O. von: J. Comp. Physiol. A *161*, 95 (1987)

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Roles of Nasonov and Queen Pheromones in Attraction of Honeybee Swarms

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Pheromones are crucial mediators of intraspecific communication in animals [1], especially in social insects where a variety of social interactions occur among many individuals. Honeybees (*Apis mellifera*) reproduce by swarming, a pheromonally mediated process in which thousands of individuals leave the parental nest and fly to a new nest site. Honeybee Nasonov pheromone consists of seven terpenes [2], of which five, when combined, elicit pheromonal activity equal to the natural pheromone in assays for clustering and orienting bees [2, 3]. This worker-produced pheromone is released by scout bees as they scent mark good potential nest cavities and signals the location of the nest cavity entrance. Nasonov pheromone also is released when a lost queen is discovered, when a nest entrance lost by natural calamity or beekeeper manipulation is found, and at low odorant sugar syrup feeders or water sources [3, 4]. Queen mandibular gland secretion is a blend of two dozen identified, plus many unidentified, compounds [5, 6]. Natural queen pheromone, or a blend of five pheromone components in their natural ratios, attracts queenless swarms, causes a retinue of workers to form around an artificial queen, and inhibits queen rearing [6, 7]. In preliminary experiments, queen pheromone did not affect attractancy of artificial cavities to swarms [8]. Our results revealed the independent role of long-range and short-range pheromones in the honeybee swarming process by establishing artificial nest cavity stations for feral bee swarms. Long-range attraction was investigated in a crossover experiment in which honeybee swarms were provided a choice of an untreated cavity, a queen pheromone-containing cavity, and either two cavities containing Nasonov pheromone or two cavities containing both Nasonov and queen pheromone (Fig 1, Exp. I). The results of this experiment revealed that neither the untreated nor the queen pheromone-scented cavities were attractive to swarms, whereas Nasonov pheromone-scented cavities were highly attractive. The presence of queen pheromone in addition to Nasonov did not improve swarm attraction (Fig. 2, Exp. IA). Visual observations confirmed these results: scout bees were almost never observed investigating untreated or queen pheromone-containing cavities, but were intensively investigating any cavity containing Nasonov pheromone.

Short-range attractiveness of both Nasonov and queen pheromones was analyzed in a second experiment by altering the design such that at all 20 cavity stations, nest site-seeking scouts could choose among cavities having lures containing no pheromone, queen pheromone, Nasonov pheromone, or the two pheromones together (Fig. 1, Exp. II). Swarms chose the cavities containing Nasonov plus queen pheromones in 17 of 19 cases. Untreated and queen pheromone cavities were not selected (Fig. 2, Exp. II). These results indicate that queen pheromone alone lacks either long-range or short-range attractiveness to scouts and moving swarms; but in the presence of Nasonov pheromone, it exhibits strong short-range attractive properties. Confirmation that queen pheromone attraction in the presence of Nasonov pheromone occurs only at short range was revealed by the results of the three pairs of cavity stations in Experiment IB that were separated by only 100 m. Although these station pairs were relatively close to each other, there was no preferential attraction of swarms to the Nasonov plus queen pheromone cavities (Fig. 2, Exp. IB), demonstrating that the short-range effects of queen pheromone operated at distances less than 100 m.

Our results are the first documented example of separate pheromones produced by individuals of two castes operating independently and at times synergistically to control swarm movement. This interaction between worker and queen pheromones provides a new dimension to our understanding of how social insects integrate worker and queen functions to colony-level coordination of complex social behavior. Scout honeybees are highly attracted to Nasonov pheromones – but not to queen pheromone – and use Nasonov pheromone to mark cavity entrances. They recruit other scouts, which use the Nasonov pheromone for long- and short-range orientation to the cavity [3, 11]. During flight to a cavity, naive worker bees and the queen might follow an aerial trail of Nasonov pheromone [11] generated by scout bees that fly back and forth through the moving swarm [11–13].