included in the illustration were also approximately Gaussian [as opposed to exponential, multimodal, and so forth (*1*)].

If the simple model of motoneuron behavior described earlier is accurate, we should be able to predict these observed interspike-interval histograms from measurements on the synaptic noise and on the average properties of the spike-generating mechanism by simulating motoneuron behavior with the digital computer. Simulation is preferable to the analytic approach employed earlier (*4*) because idealizing assumptions inapplicable to some neurons are thus avoided.

To make this simulation, a linearly rising "membrane potential" was generated in the computer, and synaptic noise was added to this membrane potential ramp. When the membrane potential plus noise first reached the "firing level," the length of time since the previous crossing of the firing level was stored, and the membrane potential was reset to begin the process anew. In this way, a sequence of "interspike intervals" could be generated, and their statistics could be compared with those of the observed neuronal behavior.

Simulating a specific neuron's behavior requires that parameters be selected for the deterministic spike-generation process and for the synaptic noise. The values for the ramp slope, starting point, and firing level were measured from records of membrane potential. "Synaptic noise" for the simulation was derived from one of two sources. In one instance the output of a commercial white-noise generator was filtered to produce noise with the same statistical structure (Gaussian probability density and exponential autocorrelation function) as that observed in the neuron. In the other cases, tape-recorded samples of synaptic noise from the quiescent cell were used. Whatever the source of the noise voltage, it was sampled at 5 khz and added to the "membrane potential" ramp generated within the computer. It must be emphasized that all parameters of the simulation were estimated from intracellular recordings from the particular neuron whose behavior was being examined and that a neuron's own synaptic noise was used in the simulation; no parameters were obtained from the interspike-interval histograms we wished to predict.

Predicted interspike interval histograms for two motoneurons are shown in their cumulated form in Fig. 2. The obviously good agreement between predicted and observed histograms was confirmed by Kolmogorov-Smirnov goodness-of-fit tests (*5*); for all motoneurons included in this report, more often than one time in five, two observed histograms generated by the motoneuron itself would differ as much as the observed and predicted histograms.

The sensitivity of this method to inaccurate assumptions is illustrated by one of the cells (C202-6) included in Fig. 2. Although the firing level was constant in some cells, it was observed to increase linearly with time from the preceding spike in others. If this systematic rise in the level was ignored—that is, if a constant firing level was adopted in the simulation—the predicted interspike-interval histogram had a much smaller standard deviation than that observed for the motoneuron (Fig. 2B). By arbitrarily increasing the amplitude of the synaptic noise in the simulation by about 100 percent, we could make the predicted and observed standard deviations of the interspike-interval histograms match; however, their shapes were different, the predicted histogram being negatively skewed. When the increase in firing level (approximately 50 μv/msec) observed in the motoneuron was included in the simulation, the synaptic noise seen in the cells gave an accurate prediction of the observed interspike-interval histograms (circles, Fig. 2, A and B).

Although synaptic noise appears well-established as the dominant source of interspike-interval variability in at least some cells, the question of whether additional noise sources make important contributions in other cells naturally arises. This is a difficult question to answer, however, since any failure to predict accurately the observed interspike-interval histogram does not imply that sources of variability other than synaptic noise were the cause of failure. For example, if the firing level depends in some complicated way on the entire history of the membrane potential path, that is, if voltage fluctuations that do not cross the firing level still have some effect on the state of the neuron, then our predictions will fail because the model of the (deterministic) spike-generating mechanism is inadequate and not because something other than synaptic noise is a major source of noise. To identify a second major noise source, then, it must be shown that including its observed properties in the stimulation leads to accurate prediction of the interspike interval histogram. In one cell, we found the slope of the membrane potential ramp to vary randomly from interval to interval. Since including this additional observed behavior in the simulation leads to accurate prediction of the interspike-interval histogram, we have discovered at least one instance where a source other than synaptic noise (presumably variability arising in the spike-generating mechanism) is important. It is interesting to note that we have not yet found neurons in which threshold variability has appeared to contribute significantly to the interspike-interval variability.

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Honey Bees: Do They Use the Direction Information Contained in Their Dance Maneuver?

Abstract. *Direction communication experiments, when controlled against unilateral visitation of trained foragers at the experimental site, exhibited no evidence supporting the precision of communication suggested in the dance "language" hypothesis. Results compare well with those which might be expected on the basis of the geometric arrangement of the feeding dishes.*

A previous investigation demonstrated the importance of conditioned responses in the exploitation of food sources by honey bees (*1*). Experienced foragers were experimentally recruited to feeding sites, by means of odor stimuli, without their having gained information from the dance maneuver. That study further indicated that recruitment by the conditioned responses of experienced bees can account for a colony's efficient exploitation of food sources. Consideration of these results eventually led to a questioning of the validity of the dance "language" hypothesis.

Original evidence supporting the theory that naive bees utilize abstract dance information to determine the direction of food sources is largely derived from the "fan experiments" of von Frisch and co-workers (*2*). Those experiments were designed to ascertain the precision with which recruit bees use the direction information contained in the dance maneuver to locate a food source. In such a fan experiment, empty scented dishes were placed in slightly different directions 200 m from the hive. A group of marked bees regularly foraged scented sucrose solution only at an experimental dish placed in the central direction of the fan arrangement of control dishes, but at a greater distance (250 m) from the hive. During the course of the experiment, investigators tallied the number of unmarked bees approaching each dish.

In those earlier fan experiments the majority of recruited bees arrived at the experimental station or at a station, or stations, located in this same direction. The dance maneuver executed in the hive by the marked bees foraging at the experimental station contains precise direction information [within ± 8° for a distance of 200 m (*3*)]. Recruited bees presumably interpreted and used this dance information as they traveled to the prospective food dish.

A close examination of the design of such experiments reveals a lack of essential controls. For example, trained bees continually visited the experimental dish, while no bees routinely visited the control dishes. Therefore, the results of those experiments do not eliminate the possibility that recruit bees are attracted to (or near) the station having regular visitation by trained foragers. Kalmus (*4*) demonstrated the importance of feeding bees in attracting recruits to a site. Another possibility not eliminated by the design of the earlier experiments was that recruits were attracted to the geometric center of all feeding sites (*5*).

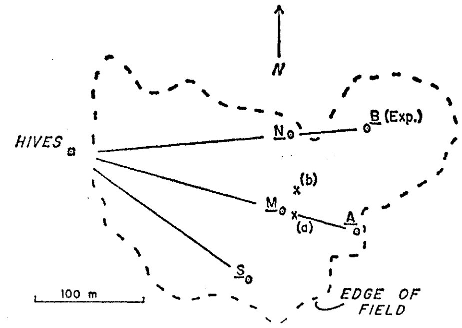

Fig. 1. Map of the experimental area. The experimental (dark-colored bees) and control hives (light-colored bees) were adjacent to one another at the edge of a football practice field (outlined by the broken line). The experimental site in the first series of experiments was *A*; in the second series, *B*. The three sites, north (*N*), middle (*M*), and south (*S*) served as controls and had dishes of sugar solution to which recruits could come. Points *a* and *b* indicate the geometric center of each set of four feeding sites for each experimental series.

Greater uniformity among the various stations (including controls against the effects of unilateral bee visitation at the experimental site) should not influence the pattern of recruitment if the recruited bees depend on the directional information of the dance to locate food sources as maintained by the dance "language" hypothesis [see von Frisch (*6, 7*) for detailed description of this hypothesis]. This present study contains fan experiments which incorporate such essential controls.

Two different bee colonies were used in these experiments. The experimental hive [different from the experimental hive used in a companion study (*5*)] contained at least 20,000 dark bees (*Apis mellifera*, Italian strain obtained from C. G. Wenner apiaries in northern California). The control hive contained about 15,000 light-colored bees. This strain, with a cordovan gene (*cd*) for light body-color, has been rendered isogenic with the Italian strain by repeated back-crossing and was furnished by H. H. Laidlaw of the University of California, Davis.

These adjacent hives were painted different colors and placed with entrances facing different directions to permit homecoming bees to orient readily to their own hive. Two other checks insured that drifting of foragers did not influence experimental results. Spot checks of bees entering and leaving each hive revealed that only rarely did an unmarked individual enter the wrong hive, in which case it was killed (by investigators). In addition, one or the other of the hives was closed periodically (at least once per day) while checks were made of all feeding stations. Neither dark nor light foragers arrived at any station when their respective hives were closed.

Two series of similarly designed experiments were conducted. In each series the three control feeding stations were positioned in an arc (70 m apart at 200 m from the hives), and the experimental station was 270 m from the hives. In the first series the experimental station was positioned behind the middle station (at location *A* of Fig. 1). In the second series the experimental station was located behind the north station (at location *B* of Fig. 1). Both control and experimental stations were located on a large level field of mowed green grass (football practice fields). The hives sat at one edge of this field.

Fifteen individually marked dark bees from the experimental hive were trained (*8*) to visit the experimental station; and light bees (also marked) from the control hive were trained to visit each of the feeding stations—20 to each of the control stations and five to the experimental. Subsequent references to the trained population at control and experimental feeding dishes will refer to the above numerical distributions of trained foragers, unless one of the hives was purposely closed. Trained bees were fed peppermint-scented 1.5*M* sucrose solution (five drops of peppermint oil per 500 ml of solution) at their respective stations and were replaced with new bees as needed throughout the experimental program. Each group of marked bees foraged only at the station to which trained. All unmarked visitors (or trained bees visiting incorrect stations) were killed.

Each of the two experimental series consisted of three types of experiments. In the first, or control experiment (Table 1, experiment 1), the control hive was closed, and only the 15 dark foragers regularly visited the experimental station. At time zero, all feeding dishes were provided with scented sucrose solution. For a 30-minute period all new (unmarked) recruits arriving at all stations were caught and placed in a container of alcohol by the person attending each dish and were counted later.

In both control experiments, 78 percent of the captured recruits arrived at stations located in the experimental direction, regardless of whether the experimental station stood behind the middle or the north station (Table 1). These control experiments were repeated with similar results. Moreover,

Table 1. Distribution of recruited bees in each of six experiments (three different types of experiments with the experimental site located either at *A* or at *B* in Fig. 1). In the first experiment of each series, dark bees visited only the experimental site. In the second experiment of each series, light bees visited all four sites while dark bees still visited only the experimental site. In the last experiment, an equal number of dark bees visited each of the four sites after the control hive had been removed. The numbers in parentheses indicate the distribution of bees recruited from the control hive. Asterisks indicate the control station behind which the experimental station was located.

Experiment	Recruits arriving at each station (%)				Total recruits (No.)	Wind		Date	Time
	Experimental	North	Middle	South		Direction (degrees)	Velocity (knots)		
				Experimental series 1					
1A	47	10	31*	11	99	160	4	11 Aug.	1030
2A	19	12	48*	21	150	240	11	11 Aug.	1400
	(10)	(6)	(53)	(31)	(245)				
3A	7	8	70*	15	61	210	9	18 Aug.	1210
				Experimental series 2					
1B	56	22*	14	8	36	140	8	16 Aug.	0950
2B	19	21*	52	8	125	140	8	16 Aug.	1040
	(10)	(18)	(57)	(16)	(351)				
3B	23	15*	57	5	74	240	6	18 Aug.	1020

these results are entirely consistent with those expected on the basis of the dance "language" hypothesis.

Following a control experiment, the control hive was opened, and its trained foragers frequented each of the four feeding stations while the trained dark bees continued to forage at the experimental site. As always, unmarked foragers were killed during this time. After this was accomplished, the second experiment began, and new recruits arriving at each station were collected during a 40-minute period. In the course of this experiment, the dish at the experimental station was replaced every 10 minutes with a clean one. The dirty dish from this station, in turn, alternately replaced one of the dishes at those two control stations not in a direct line with the experimental site. Switching dishes in this manner reduced the possible accumulation of specific hive odor (from the dark bees) at the experimental station and distributed it to other sites (*9, 10, 11*).

In these experiments (Table 1, experiments 2A and 2B), when feeding stations were more uniform with respect to trained foraging populations at each dish, between 50 and 60 percent of the total number of captured dark recruits arrived at stations located in the central direction, irrespective of where the experimental station was (at location *A* or *B* in Fig. 1). Furthermore, the identity between the distribution of light and dark recruits in each of the experiments is a result not consistent with the dance communication theory. The experiments were repeated twice and yielded results similar to those shown in Table 1.

Experiment 3 of each series involved removing the control hive and training 60 dark bees from the experimental hive to the four feeding stations, 15 to each station. The bees in each of these groups were not allowed to visit stations other than the one to which they had been trained. During a 40-minute period, while these bees continually foraged at the stations, all new recruits arriving at each station were collected as before (Table 1, experiments 3A and 3B).

The resulting nonuniformity of distributions of recruits suggests a bias favoring the central station in each experimental arrangement. This same bias is also apparent in the distributions obtained in experiments 2A and 2B. Three repetitions of experiment 3 again yielded similar results.

An additional experiment conjunctive to the previously described control experiments (1A and 1B) yielded information on the attractiveness of all four stations relative to that of the experimental station alone. During the 30 minutes preceding experiment 1A (Table 1), with the control hive closed and only the experimental station on the field, 25 new recruits arrived at that site. In the following 30 minutes (experiment 1A) a total of 99 unmarked bees landed at the four stations, 47 of which arrived at the experimental site.

The next morning, during similar weather conditions, a control experiment (of the 1B type—not in Table 1) was performed before the companion study. In the control a total of 85 recruits arrived at the four stations; 28 of these arrived at the experimental site. In the subsequent 30-minute companion study, when only the experimental station was on the field, 46 new recruits arrived at that site. Apparently, increasing the number of stations increased the number of recruits caught, which suggests that many more recruited bees search likely areas than ultimately locate a site regularly visited by hive mates (only 15 dark foragers visited the experimental site in each of the above experiments).

The control experiments (1A and 1B) essentially repeat the earlier fan experiments of von Frisch (*2*) with similar results; namely, the majority of recruits (78 percent) arrived at dishes in the direction presumably indicated by dancing bees. During experiment 2, however, when equal numbers of trained bees were permitted to visit all feeding stations, the pattern of recruit distribution differed from that obtained in the control experiments.

Comparison of one control to an experimental period (1B and 2B in Table 1) illustrates such a shift in recruitment distribution. Even though these periods were separated by only 20 minutes during similar environmental conditions and although they involved the same group of trained dark foragers at the experimental station, the furnishing of an equal number of regular visitors at each station yielded a markedly different distribution of dark recruits. Now, neither light nor dark recruits appeared to have responded to that precise direction information they could have received from dance maneuvers.

Identity between recruitment distributions of light and dark bees in the second experiment of each series and the consistent pattern in the percentage of recruitment evident between stations conceivably arise from the use of similar orientation cues. Apparently, providing equal visitation by trained bees at both control and experimental stations, although rendering them more identical in attractiveness, leaves them unequally distributed in space. Under the circumstances of these experiments, the middle station always lay closer to the geometric center of all sites (center of moments) than did any other station. In Fig. 1, points *a* and *b* locate the center of moments in each series of experiments—altering the location of the experimental site from *A* to *B* in that figure does not appreciably shift the center of moments. It remains near the middle site.

Just as the center of moments remains near the middle control station, so also were most recruits collected at the middle site when all stations became more similar in attractiveness. In either experiment 2A or 2B, the recruit distributions more closely correspond with the spatial distribution of dishes than with the results from the first (control) experiment of each series. [Statistical tests have not been employed in this or in other comparisons for various reasons; see note *9* in a related study (*5*).]

The results from the third experiment in each series (Table 1, 3A and 3B), where an equal number of marked dark bees foraged at all four sites (and in which the stations would be even more equal in attractiveness), correlate even better with the linear distance of each site from the geometric center of all sites. The close correspondence between the spatial arrangement of all stations and the distribution of recruited bees persists with little modification by wind conditions, possible interference between bees from the two different hives (*12*), and, perhaps even more important, slight differences in locality odors.

The results of the original fan experiments were interpreted by von Frisch as a demonstration of the precision with which honey bees use direction information. He concluded that the majority of searching bees, after leaving the hive, move within an angle deviating not more than 15° to the left or right of the experimental direction (*13*). While repetitions of such experiments yield data which seem completely consistent with the original results (experiments 1A and 1B), results obtained after the incorporation of essential controls into the experiments indicate that, if recruits use the direction information of the dance, they fail to do so with the precision suggested by the original experiments. Thus, although the waggle-dance maneuver contains relatively precise direction information, it would appear that local cues (including locality odor, food odor, hive odor, and bee odor at the site) dictate the location or locations at which recruits settle to feed.

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Honey Bees: Do They Use the Distance Information Contained in Their Dance Maneuver?

Abstract. *Regular visitors at one site (experimental) in a linear series of sites normally recruit inexperienced hive mates to or near that site. If bees from a second hive were allowed to forage at both control sites, however, recruits from the experimental hive, while orienting to these sites, exhibited no evidence of having used any distance information they might have received before leaving their parent hive.*

Important evidence for the theory that bees communicate and use abstract information concerning the distance of a food source comes from the step-experiments (*Stufenversuchen*) of von Frisch and co-workers (*1*). A linear series of stations were placed in one direction from a hive with a known number of marked visitors frequenting only one, the experimental. Repeated round trips by these marked bees, interspersed with waggle dances in the hive, preceded recruitment of new bees to or near the experimental site (*2*, *3*). Since the dance maneuver executed by successful bees upon their return to the hive contains information concerning the distance of the food site and since recruited bees generally arrived at the proper distance or close to it, recruited bees were presumed to have used the distance information contained in the dance before finally orienting to the specific odor of the food site.

The evidence gathered in support of distance communication in the dance "language" hypothesis might be an artifact of the experimental design rather than an indication that bees use distance information contained in the dance maneuver (see *2*, *4*). If not, and if recruited bees use the distance information furnished by regular foragers (and only secondarily use scent of food and of location upon their arrival at the approximate distance), then making both control and experimental sites more uniform in terms of bee visitation should not influence the overall distribution of recruits. I have studied the distribution of recruits both before and after bees foraged at control sites as well as at the experimental site.

All experiments in this series were conducted at the golf course of the Santa Barbara campus of the University of California, on an area largely devoid of living vegetation at that time of the year. One or two different hives provided the bees for each experiment, depending on the particular conditions required. One hive containing dark bees (*Apis mellifera*, Italian strain) was always used as the experimental hive to test the distance-communication hypothesis, whereas another hive of light bees [Italian strain, cordovan gene (*cd*) for body color] served as the control hive when needed (*5*). At the time of the experiments, the hives contained at least 30,000 and 10,000 bees, respectively.

The darker bees from the experimental hive were trained (*6*) to forage at only one of four possible sites in each of the experiments. The other three sites served as controls for this hive (Fig. 1). These sites were linearly arranged in an east-southeast direction from the hive so that the prevailing wind (from the south or southwest) was generally at right angles to the line of flight of bees from the experimental hive during each experiment. Placement of the sites 100 m apart insured that most recruits from the experimental hive would be exposed to relatively accurate information concerning the distance of the experimental site (*2*, *7*).