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An Analysis of Variability in the Feeding Motor Program of the Honey Bee; the Role of Learning in Releasing a Modal Action Pattern

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Received: November 30, 1987
Accepted: November 10, 1988 (G. Barlow)

Abstract

Sequences of behavior are highly predictable (stereotyped) during some segments and less predictable during transitions between those segments. Statistical characterization of behavior must involve observation of the behavior under different stimulus conditions, which includes how stimuli associated with behavioral releasers come to trigger behavior during learning. For example, the feeding motor program (FMP) of the honey bee Apis mellifera (Hymenoptera: Apidae) during proboscis extension can be divided into three response phases (REHDER 1987). We conditioned honey bees in an olfactory conditioning paradigm with one or several rewarded trials during which an odorant was paired with the sugar-water unconditioned stimulus (US); the latter elicits proboscis extension and feeding in properly motivated bees. By recording electromyogram activity from one of the muscles that move the proboscis during feeding, we quantified the bees' responses during an unrewarded test with either the conditioned odorant, a different (novel) odorant, or the sugar-water US. Various parameters of the response phases of the FMP varied in a consistent manner across these experimental treatments, with certain stimuli eliciting stronger, more consistent responses. The different response phases followed one another in time with some variability and statistical uncertainty. For example, the length of an individual licking movement with the glossa was relatively invariant, and may indicate that this parameter can be used to differentiate the FMP into more basic, independent units. Our work shows how learned information may release action patterns in ways slightly different from traditional sign-stimuli releasers.

Introduction

Ethological analyses attempt to describe the flow of an organism's behavior through time (BARLOW 1977). Ethologists routinely describe sequences of muscular actions by the changing relationships among different body parts (SCHLEIDT et
al. 1984). Behavior patterns are sequences of such defined motor acts. The sequences can be highly predictable during segments, yet can also be punctuated by periods of uncertainty on the part of the observer (a researcher or a conspecific) as to what sequence of acts the organism will perform next. An organism is said to have made a decision when there is a reduction in this uncertainty, that is, a predictable sequence of acts follows the period during which the organism's future behavior cannot be reliably predicted (Dawkins & Dawkins 1974).

The observation of highly predictable sequences of behavior led early ethologists to the idea of a fixed action pattern of behavior that is released by specific sign stimuli in the organism's environment (Barlow 1977). However, upon closer analysis many behavior patterns that were thought to be relatively invariant are in fact variable, especially when quantified under differing stimulus conditions. To draw attention to the higher degree of variability in the expression of a behavior pattern, Barlow (1977) used the term modal action pattern (MAP), which is a "recognizable spatiotemporal pattern of movement that can therefore be named and characterized statistically". Statistical characterization of MAP variability would necessarily involve observation under widely different stimulus conditions.

Variation in the control of feeding movements, for example, may reflect variation in the types of resources animals must harvest in order to survive. Through the employment of an appropriate feeding motor program (FMP) animals may be able to harvest a particular resource with less energy or time than through employment of another motor program in its FMP repertoire. Variation in the FMP may arise from stimulation with a variety of stimuli, among which are: (1) input from receptors that reflexively release the FMP, and (2) conditioned stimuli that are reliably associated with the resource. Case (2) involves some form of learning, where expression of the FMP is altered through an appetitive or aversive feeding experience (Gelperin 1983; Mptös & Cohan 1986a, b). Learned associations that predict resources are important for many organisms (Gould & Marler 1984). Therefore, an understanding of how learned stimulus associations release MAPs is crucial to understanding how MAPs are organized.

Honey bees (Apis mellifera) learn to manipulate complex floral structures with their proboscides in order to obtain nectar (Faegri & Van Der Pijl 1979); therefore, the FMP driving proboscis extension and subsequent glossal movements may be more variable than suggested by cursory visual inspection of the behavior. Upon application of sugar-water to one antenna or tarsus, a honey bee extends its proboscis (Kuwabara 1957; Menzel et al. 1974). The ensuing response can be divided into three temporal phases corresponding to various movements of the proboscis (Rehder 1987); extension, rhythmic 'licking' action by the glossa, and retraction. Furthermore, when sugar-water is immediately preceded by an olfactory stimulus, proboscis extension can later be elicited by the odorant alone because of the learned association (Kuwabara 1957; Menzel et al. 1974). Learned associations between floral odorants and nectar are important for controlling a honey bee's appetitive response under natural foraging conditions (von Frisch 1967).
In order to characterize the honey bee’s FMP statistically, we recorded electromyograms (EMG) from a muscle that is responsible for proboscis extension and control of movements of the glossa that coincide with uptake of nectar (Rehder 1987). Here we describe the variability in muscular contractions involved in response to types of stimulation limited to cases (1) and (2) above. Thereby we define aspects of the honey bee’s FMP that describe the decision structure (Dawkins & Dawkins 1974) of the bee’s behavior and show how a learned stimulus association influences expression of a MAP.

**Materials and Methods**

Worker honey bee foragers were collected as they exited their hives on foraging trips between 16.00 and 18.00 h on the day prior to testing. The hives were out-of-doors when tests were conducted between April and September, or in-doors in a flight room during the winter. After placing bees individually into unsealed glass vials, they were cooled to 3 °C, soon after which they could no longer move. Each bee was then harnessed in a small piece of copper tubing by placing tape strips across the wings and between the head and thorax (Menzel et al. 1974). The heads were then immobilized by placing a small drop of molten beeswax, which quickly hardened, between the head and the tape strip behind it. After warming to room temperature, the bees were fed a 1.5 M sugar-water solution until satiated, and then left in a moist, dark environment overnight. Feeding minimized differences among bees in motivational state on the following day, thus minimizing effects due to variation in the bees’ motivational states.

**Fig. 1:** Typical spiking pattern obtained by recording EMGs from the M17 muscle during proboscis extension after stimulation with a conditioning odorant (bar above the spike trace corresponds to length of presentation of the odorant). The graph shows the cumulative number of spikes through time. Bursts of spikes corresponding to licks are evident in the trace; licks are represented by bumps in the graph, which indicate temporary acceleration in spike frequency when each lick occurs. Some of the variables defined in Table 1 are indicated in the graph. Average slope over the entire response reflects the average spike firing frequency and hence is defined in Table 1 as the ‘speed’ of responding.
On the following day the bees were first fed a small droplet of sugar-water. To record the EMGs (Rehder 1987), a small hole was made just behind one compound eye in the dorsal cranial wall of the head of each bee, where the insertion point for an M17 muscle is found. The M17 extends from this point to the ligular arm of the proboscis (Snodgrass 1956). A copper-wire electrode was inserted 1–2 mm into the hole; the indifferent electrode was then inserted into the eye that was, for most recordings, on the opposite side from which the recording was made. Any bee from which only a poor recording could be made was not used in the experiments. All tests with these bees were completed within three hours of electrode insertion.

All recordings from each bee were amplified and viewed on an oscilloscope and simultaneously monitored with a window discriminator. The window discriminator recorded a spike each time the electrical potential changed such that it exceeded a set voltage above the baseline. The total spikes during every 20-ms time window over a 20.48-s recording period were stored, and then passed to a microcomputer diskette. The recording from each bee was then analyzed by Pascal program designed to return several parameters reflecting the bee’s response (Table 1, Fig. 1).

Table 1: Definition of the variables derived from each bee’s electromyogram response.
Variables with a superscripted a are in ms

<table>
<thead>
<tr>
<th>Variable</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Spike</td>
<td>summation of spike activity over the entire 20.48-s recording period during the test.</td>
</tr>
<tr>
<td>Duration</td>
<td>duration during the 20.48-s test that the bee responded with proboscis extension; defined by the spike frequency exceeding 20 Hz during a 20-ms time window.</td>
</tr>
<tr>
<td>Speed</td>
<td>slope of a line drawn from the beginning of a response, proboscis extension, to the point where the proboscis was retracted on a curve plotting cumulative number of spikes through time (Fig. 1). Higher spike frequencies yield steeper slopes.</td>
</tr>
<tr>
<td>Interruptions</td>
<td>number of times a bee extended and retracted its proboscis during the 20.48-s test period. A score of 1 indicates a single extension and retraction with no interruptions. Higher scores indicate an increasing tendency for the response to be interrupted.</td>
</tr>
<tr>
<td>Number of licks</td>
<td>the initial extension of the proboscis is followed by rhythmic extension and retraction of the glossa (Rehder 1987). This activity is reflected in rhythmic bursts of spikes in the EMG recording. A procedure in the analysis program recognizes the beginning, inflection point, and end of each burst. A ‘lick’ was defined as a burst longer than 100 ms and shorter than 500 ms in order to avoid erroneously counting the extension and retraction phases.</td>
</tr>
<tr>
<td>Latency</td>
<td>time from the start of a test until the first lick is recognized. Recorded only if a lick was detected.</td>
</tr>
<tr>
<td>Lick length</td>
<td>mean length of all licks detected from an individual bee. In addition, the program returns the within individual coefficient of variation of lick length.</td>
</tr>
<tr>
<td>Lick separation</td>
<td>average time between the inflection points of successive licks, given that two or more licks were detected.</td>
</tr>
</tbody>
</table>

Measuring Conditioned and Unconditioned Responses

2 to 3 h after insertion of the electrodes bees were exposed for three s to an air-stream containing either cirtal or n-pentanal (CS+). The odorant presentation was followed by activation of a feeding machine that moved a small piece of sugar-water impregnated filter paper (the unconditioned stimulus, US) close enough to the bee such that the antennae could touch the paper. The bee then extended its proboscis and was allowed to feed by licking the filter paper for 6.5 s. Each bee received only one learning trial before testing. The single post-training unrewarded (i.e., no sugar-water feeding) test employed one of the following odorants, presented in an identical manner as the initial CS+: pentanal, cirtal, geraniol, or 2-pentanol. Therefore, in some cases bees were retested with the odorant to which they had been conditioned, while in other cases they were presented with a different, ‘novel’ odorant. The odorants were chosen from a larger variety of odorants tested in a previous experiment (Smith & Menzel 1989). Each bee was tested once, and never used again in subsequent tests.

In some cases the bees received no odorant in the air-stream when the sugar-water was presented. In these cases a test was later performed by touching the antennae with the sugar-water
impregnated filter paper and then recording the response when the bee was not allowed to feed. In this way, effects due to feedback from sugar receptors on the bee’s proboscis were minimized.

Thus the FMP released by the conditioning odorant, a different (novel) odorant, and the unconditioned stimulus could be compared statistically.

Statistical Analyses

For analysis of the results from conditioned and unconditioned response data, nonparametric tests (Kruskal-Wallis and Mann-Whitney-Wilcoxon; Sokal & Rohlf 1981) were employed because of varying distributional properties of the different variables. Furthermore, some of the 8 variables (Table 1) appear correlated with one another, and thus contain redundant information. To determine patterns of closely correlated variables, principal-component analysis of the correlation matrix among all the variables was employed (Harris 1975), followed by varimax rotation of the extracted factors. Two independent data sets were separately analyzed, one from bees tested with the odorant with which they had been trained (conditioning odorant), and the other on data from bees tested with an odorant different from the one that they had been conditioned to (novel odorant). Interpretation of significant patterns of correlation based on the factor loadings was verified by inspection of the correlation matrix for significant correlation coefficients.

Only those bees that responded to the test stimulus by extension of the proboscis were included in the statistical analyses. Thus, in the data below, 100% of the bees responded with proboscis extension; no discrimination among groups would be possible without the EMG recordings. Inclusion of nonresponders (i.e., those that did not learn the association after one conditioning trial) might result in differences among treatments attributable to differing proportions of responders and nonresponders, rather than differences attributable to changes in expression of the FMP. Proboscis

Table 2: Factor loadings for the 8 original variables derived from the EMG recordings from bees tested with the conditioning odorant (A, N = 259 bees) and from bees tested with a novel odorant (B, N = 179 bees). a: variables that are strongly correlated with each factor

<table>
<thead>
<tr>
<th></th>
<th>I</th>
<th>II</th>
<th>III</th>
<th>IV</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. Conditioning odorant</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total spike</td>
<td>0.821*</td>
<td>0.484</td>
<td>-0.025</td>
<td>-0.051</td>
</tr>
<tr>
<td>Duration</td>
<td>0.872*</td>
<td>-0.194</td>
<td>0.036</td>
<td>0.237</td>
</tr>
<tr>
<td>No. of licks</td>
<td>0.842*</td>
<td>0.309</td>
<td>-0.020</td>
<td>-0.083</td>
</tr>
<tr>
<td>Speed</td>
<td>0.168</td>
<td>0.861*</td>
<td>-0.115</td>
<td>-0.115</td>
</tr>
<tr>
<td>Interruptions</td>
<td>-0.064</td>
<td>-0.797*</td>
<td>0.005</td>
<td>0.052</td>
</tr>
<tr>
<td>Latency</td>
<td>-0.319</td>
<td>-0.139</td>
<td>0.816*</td>
<td>0.002</td>
</tr>
<tr>
<td>Lick length</td>
<td>0.374</td>
<td>0.025</td>
<td>0.763*</td>
<td>0.134</td>
</tr>
<tr>
<td>Lick separation</td>
<td>0.099</td>
<td>-0.122</td>
<td>0.092</td>
<td>0.977*</td>
</tr>
<tr>
<td>B. Novel odorant</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total spike</td>
<td>0.820*</td>
<td>0.498</td>
<td>-0.023</td>
<td>-0.034</td>
</tr>
<tr>
<td>Duration</td>
<td>0.906*</td>
<td>-0.195</td>
<td>0.051</td>
<td>0.108</td>
</tr>
<tr>
<td>No. of licks</td>
<td>0.867*</td>
<td>0.291</td>
<td>-0.093</td>
<td>0.006</td>
</tr>
<tr>
<td>Speed</td>
<td>0.126</td>
<td>0.883*</td>
<td>-0.092</td>
<td>-0.186</td>
</tr>
<tr>
<td>Interruptions</td>
<td>-0.083</td>
<td>-0.871*</td>
<td>-0.052</td>
<td>0.028</td>
</tr>
<tr>
<td>Latency</td>
<td>-0.202</td>
<td>-0.032</td>
<td>0.879*</td>
<td>-0.135</td>
</tr>
<tr>
<td>Lick length</td>
<td>0.346</td>
<td>0.009</td>
<td>0.615*</td>
<td>0.388</td>
</tr>
<tr>
<td>Lick separation</td>
<td>0.018</td>
<td>-0.141</td>
<td>-0.008</td>
<td>0.946*</td>
</tr>
</tbody>
</table>
extension was defined as follows: total spike ≥ 30, and speed > 0.00. The speed criterion indicated that the spike frequency exceeded 20 Hz at least one time during the test. Together these conditions met the minimum activity necessary to complete the initial extension phase described by REHDER (1987).

Results

Depending on the experimental series and odorant employed, 50—70% of the bees with electrodes implanted in their heads learned the association between odorant and sugar-water after the first trial, as judged by visual confirmation of proboscis extension to odorant presentation alone. This result corresponds to rates of training success after one-trial odorant conditioning in previous work (MENZEL et al. 1974), and indicates that the surgery had little effect on learning performance.

Correlations among the Measured Variables

Under both test regimes (conditioning odorant and novel odorant) the pattern of correlations among the variables remained the same (Table 2), indicating that the results are general and robust. The distributions of the variables listed in Table 1, however, differed between the two data sets. The distributions for tests with conditioned odorants did not deviate significantly from a normal distribution, but the distributions for tests with a novel odorant tended to be skewed (see below). However, because the two data sets yield almost identical results from principal-component analysis, the distribution differences did not appear to affect extraction of principal components.

In both data sets, over 80% of the total variation observed can be attributed to a few variables that are linear combinations of the original 8. The first factor (PC I) indicates a positive correlation between the total spike count, duration of the response, and number of licks. Together these variables reflect the tendency of bees to respond for different lengths of time. Some bees responded for a long time, producing more spikes. Since the response after the extension phase is primarily due to licking, the number of licks also increased, as expected. In contrast, other bees responded for a shorter period of time and produced few licks.

PC II reflects ‘decisiveness’ (see below) of the response: some bees responded continuously whereas others stopped and then re-started one or more times (see Interruption variable in Table 1) during the 20-s test. The bees that responded continuously, with fewer interruptions, had a higher overall speed of response. They responded faster with increasing spike activity per unit of time. This inverse relationship between speed of response and number of interruptions gave rise to a negative correlation between the two variables. The variables loading on PC II are evidently independent of those loading on PC I, although slight positive loadings for some PC I variables (e.g., total spike count) on this factor are apparent.

Latency of the licking response and the mean length of a lick load on factor III, and the mean separation between licks loads singly on factor IV. These
variables loading on PCs III and IV, are not correlated with variables describing the length and decisiveness of the response, which load significantly on PCs I and II. However, the correlation matrix indicated that the correlation between latency and lick length that generated PC III is weak and nonsignificant. Therefore, there is no reason to believe that factors III and IV indicate significant correlations. We conclude that the three variables vary independently of one another.

The interpretation of the following results was based on the factors described above as length of response (PC I), decisiveness of the response (PC II), and variation in the individual glossal movement parameters involved in licking.

Conditioned and Unconditioned Responses

With regard to some of the variables, responses of bees toward a novel odorant after conditioning differ from responses of bees toward the odorant to which they had been conditioned (Table 3). The total number of spikes was significantly greater when the response was to conditioned odorant than to novel odorant; the means also show that the responses of the bees extended well beyond the three-s odorant exposure. The reason for the difference, with respect to the total spike count, is clear upon inspection of the distributions for both experimental situations (Fig. 2). The distribution for total spike count toward the novel odorant is skewed toward the left, with the most responses falling in the category of 30–60 spikes; i.e., a spike count that would correspond with completion of
the extension phase of the response followed by immediate retraction without a further response. The FMP was much shorter toward a novel odorant than toward the conditioned odorant. Although duration of the response and the total number of licks were correlated with spike count, and the differences in the means are in the same direction as with spike count, the means for the former two variables do not significantly differ from one another across the odorant treatment groups (0.10 < p < 0.05).

The unconditioned response was also significantly higher on all PC I parameters than for either of the conditioned responses (Table 3). Thus after one learning trial a punctuate unconditioned stimulus was still a more salient cue for eliciting the FMP than a stimulus that had been associated with it.

Speed and ‘decisiveness’ (PC II) also varied across treatment groups. The speed of response decreased significantly from the response to sugar-water, to the conditioned odorant response, to the novel odorant response. As expected from the pattern of PC II loadings (Table 2), the number of interruptions was significantly lower in the sugar-water response and increased over the two odorant treatments. Once a bee responded to presentation of the conditioned odorant, there was a much higher probability that it would continue responding than when the response was to a novel odorant. The differences in speed among the treatment groups reflected the negative correlation between the two variables loading on PC II.
Fig. 3: Frequency histograms of the coefficient of variation (CV) of lick lengths for each bee that responded with two or more licks. CVs obtained for lick lengths after stimulation with the conditioning odorant are shown in the top figure, and CVs for lick lengths after stimulation with a novel odorant are shown in the bottom figure. CVs were obtained by dividing the standard deviation of the lick length from each individual's response by the mean lick length for that individual, and then multiplying the product by 100. The distributions are centered on a mean of 39 and do not differ across the two treatments \( p > 0.05 \), Kolmogorov-Smirnov test; Sokal & Rohlf 1981.

Of the three independently varying parameters that describe patterns of glossoal movement, only the latency to the first lick (Latency) showed any marked variation across treatments. The latency decreased as the stimulus became more salient, i.e., as the stimulus was changed from tests with a novel odorant, to tests with the conditioned odorant, to the response toward sugar-water. This result corroborates the results of Rehder (1987), who found that the latency from the time of presentation of the CS+ until the beginning of the extension phase decreased over several learning trials.

The length of the average lick varies remarkably little (Table 2); variation across treatments was not significant, and averages approximately 280 ms in all treatment groups. The distribution of the within-individual coefficient of variation for this parameter does not differ for the treatment groups (Fig. 3; \( p > 0.05 \)), and the mean CVs for both groups were well below 100, which indicates a highly stereotyped behavior (Barlow 1977). Therefore, the mean lick length is relatively stereotypic, at least under these stimulus conditions. In both data sets in which odorants were tested there was more of a tendency to stop the licking response at certain points, which were after the first, fourth, ninth, and thirteenth licks (Fig. 4).

However, mean lick length was the average length of several licks that was then itself averaged over many individual bees' responses. Inspection of the length
Fig. 4: Frequency histogram of the total number of licks recorded from each bee before it stopped responding after stimulation with the conditioning odorant (top) or a novel odorant (bottom). Higher columns indicate that more bees stopped responding after the number of licks associated with that column.

of licks throughout the responses of individual bees that showed moderately long responses indicated considerably more variability within bees than between them. Specifically, the length of a lick varies throughout the response; several long lick lengths are typically interspersed with lick lengths shorter than the group mean. Although the occurrence of these long lick lengths many times appeared to be periodic within individuals, no consistent periodicity across-individuals was apparent.

**Discussion**

We have attempted to examine the usefulness of EMG recordings in the description of the honey bee's FMP under different stimulation paradigms including conditioning. We asked, how can we describe the flow of the behavior through time in terms of predictable, stereotypic components versus less predictable, more variable components? The variability in the temporal expression of the honey bee FMP with regard to length and decisiveness is affected by the bee's previous experience and the nature of the stimulus that releases it. However, the definition of stereotypy proved more problematic. Under the stimulus conditions we employed to release the MAP certain parameters were highly stereotypic. But, as we will discuss, this stereotypy may be limited to the stimulus conditions under which we tested MAP release, thus leading us to the conclusion that the stimulus conditions under which stereotypy is observed must be specified in future studies.
In previous studies of differential olfactory conditioning in honey bees, three
to four learning trials were necessary to achieve a separation between the response
toward the unrewarded odorant and the rewarded one (Bitterman et al. 1983). In
these studies, the response was always measured by visual confirmation of
proboscis extension within a short period after presentation of the odorant; e.g.,
the response occurred, or it did not. Measuring the EMG is clearly a much more
sensitive means of quantifying the FMP; differences in the length of expression of
the FMP were evident after a single conditioning trial. However, several other
muscles besides the M17, from which we recorded, are involved in proboscis
movement. Future work with these other muscles should reveal degrees of
freedom of movement not evident in recordings from a single muscle.

The FMP can be said to be more decisive (Dawkins & Dawkins 1974) when
released by the US rather than by either of the CSs, which is how we have
interpreted the variables loading on PC II. Once the bee has begun responding to
the unconditioned stimulus, there is a higher probability it will continue respond-
ing in a predictable way (rhythmic licking) for a given amount of time than when
the response is toward either of the conditioning stimuli. FMP expression after
presentation of a novel odorant is the least decisive. Once a bee has stopped
responding one has less certainty regarding the bees' response during the remain-
der of the test; many times additional extensions and retractions occurred.

The basis for differing degrees of decisiveness (and probably length of
response, PC I) depends on the associative strength between each stimulus and
the unconditioned stimulus (Wagner 1981). Associative strength is a measure
that determines the likelihood the FMP will be released after stimulation and
probably reflects different efficacies of synaptic interaction between sensory
inputs and neural pathways that trigger motor output. The unconditioned
stimulus is traditionally a stimulus that, over evolutionary time, has been reliably
associated with the presence of some valuable resource, such as the input from
sugar receptors on the antennae of a bee. Because of an innately strong associative
strength, the likelihood that the unconditioned stimulus releases a MAP is
relatively higher than the probability that other stimuli do so. We would expect
that additional rewarded trials with the CS would enhance the associative strength
between the CS and US and lead to a more decisive response toward the CS.

That conditioning stimuli do not release the MAP as reliably as the uncondi-
tioned stimulus reflects a less reliable association over evolutionary time. For
example, floral resources and their olfactory stimuli vary temporally and spatially
throughout the bees' range, whereas sugar perception always indicates nectar.
Experience changes the probability that the FMP will be elicited through tempo-
rnal CS-US associations, and may even shape the expression of the FMP with
regard to the nature of the resource. However, animals have biases that allow
certain conditioning stimuli to become associated with a resource much more
quickly than others because of a much more reliable association between them
over time. A gradient of innate associative strengths may exist between several
potential conditioning stimuli and any stimulus that releases a MAP; the strong
innate association between a US and MAP release may be one extreme of this
associative strength gradient (Gould & Marler 1984). Indeed, using a more extended conditioning procedure, Smith & Menzel (1989) have shown that certain conditioned odorants elicit a stronger response than others after an equivalent amount of conditioning, which may indicate that certain odorants are innately more reliable predictors of nectar.

Qualitative differences between odorant stimuli used when conditioned or novel odorants were tested may reflect differences in associative strength of the odorants due to generalization among the odorants (Smith & Menzel 1989). The less decisive release of the MAP by a novel stimulus may reflect perceptual similarity of the odorants involved (Maes 1984). At another level of explanation, not independent of the first, 'indecisiveness' may reflect an evolved response strategy of the bee in that it is uncertain about the generality of the reward presentation. Unrewarded presentations of the novel odorant would be necessary to confirm that it is not associated with the US. Under this view, the bee must first gather information before making a decision not to respond.

Information regarding the more stereotypic expression of the MAP is contained in the lengths of the licks and the time between them. Separation between licks tended to increase in the novel odorant group, but not significantly. There is no basis for believing that the separation is affected by experience; it may therefore be an unmodifiable expression of the neural structure controlling the MAP. Furthermore, separation is uncorrelated with lick length (Table 2), thus indicating an important component of the FMP that must be explained by any hypothesized neural network that drives the FMP.

The expression of lick length during the FMP is more complex. Although there are no differences with respect to the means across treatment groups (Table 3), and the CVs indicate a highly stereotyped behavioral unit (i.e., one lick), the lick length varied throughout the response such that occasional 'long-licks' occurred (e.g., > 300 ms). The latter information indicates that the lick length is not set at ca. 280 ms, but rather can be varied by the bee.

Under the present stimulus conditions, FMP expression without receptor feedback due to actual feeding, the predominating lick length was ca. 280 ms. However, much more variability in the expression of the lick may be expressed by varying other parameters in the testing situation. For example, our studies attempted to standardize the hunger (motivational) state of the bees by prefeeding with a standard amount of sugar-water on the day before testing. More variability in motivational state among test subjects may increase variability in MAP expression among treatment groups. Additionally, we conditioned bees with a single concentration of sugar-water. Under natural conditions, nectars vary in amount and sugar concentration. Somewhat variable lick lengths may indicate that the bees are able to adjust the expression of the FMP in response to different types of nectar presentation under natural conditions; this ability may allow bees to adjust the FMP to nectars of different viscosities (Roubik & Buchmann 1984).

Interestingly, the response to a trained odorant or to a novel one yielded a rhythmic tendency in the stopping points for the FMP (Fig. 4). This rhythmicity raises the question as to the units of behavior in the FMP, which may not be a lick itself. Although we must be cautious until further experiments are performed, at
least under the present stimulus conditions, it appears that the unit is a multi-lick sequence of approximately three to four licks. Such rhythmicity appears to frame a 'decision unit' of the bee, in that the preferred stopping points are just after certain licks. After the second lick the likelihood that a bee will continue to respond is high, and little new information is obtained by the observation of the third lick. However, after the fourth lick in response to a novel odourant the observer gains relatively more information with regard to the tendency of the bee to continue upon further observation of the FMP.

We do not know the extent to which the plasticity (or stereotypy) of the FMP, and hence the decision strategy of the bee, has evolved to meet evolutionary needs versus the extent to which its expression is constrained by phylogenetic history. Honey bees are highly polylectic; they obtain nectar and pollen from a broad array of floral resources whose morphological structures vary considerably (FAEGRI & VAN DER PILJ 1979). The bees, many times, must first learn how to manipulate the flowers to minimize handling time, which often involves intricate manipulations of the long, slender proboscis. For example, after detecting the nectar well, the proboscis must be moved to a point in space indicated by the position of the antennae or tarsi. Then, depending on how the nectar is presented, e.g., as a droplet or distributed on a mat of trichomes, the nectar must be quickly taken up. Plasticity in the expression of the FMP may be an adaptation for manipulating nectar in these different situations. Future work might examine how the honey bee FMP expresses compromises between adaptation and phylogenetic constraint.

Acknowledgements

We would like to thank the following colleagues for their helpful comments on the manuscript and the work in progress: L. BERNAYS, R. CHAPMANN, W. GIEZ, B. MICHELSION, and V. REHIDER. Additionally, U. GREGGERS provided invaluable assistance during the work; the program used to analyze the data was a joint effort of B. H. SMITH, U. GREGGERS, and B. MICHELSION. The work reported here was funded by an NSF-NATO Postdoctoral Fellowship to B. H. SMITH and a research grant from the Deutsche-Forschungs-Gemeinschaft (DFG #Me365/12) to R. MENZEL.

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