Rainer Rosengren Foraging strategy of wood ants (Formica rufa group).
II. Nocturnal orientation and diel periodicity

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FORAGING STRATEGY OF WOOD ANTS
(FORMICA RUFA GROUP)

II. NOCTURNAL ORIENTATION AND DIEL PERIODICITY

Rainer Rosengren
Abstract


In colonies of the wood ant Formica polyctena foragers leave the mound during warm nights in late summer. Nocturnal foraging does not require the presence of the moon and also takes place when the sky is completely overcast. The fidelity of forager groups to their own specific routes (Ortstreue) is as high in the night as in the light period. On the other hand, observations of colour-marked foragers surviving the hibernation period indicate that spatial fidelity is based on a persistent memory of visual landmarks. These results can be explained by assuming that wood ants use different orienting mechanisms in the day and the night. The nocturnal forager population is largely the same as the diurnal one, which means that the same individuals must possess orienting mechanisms for both nocturnal and diurnal foraging. The results of marking foragers in the field with different colours for different parts of the day disproved the existence of forager groups separated by differences in circadian periodicity. These results were confirmed by machine counts of foragers departing from an artificial nest. Attempts to condition foragers to a certain feeding time failed completely. There was, however, clear evidence that a light/dark-cycle (LD) or a temperature cycle induces a general activity rhythm in the ant population. Peak activity occurs at the end of the scotophase and/or at the beginning of the photophase. Both the occurrence of nocturnal foraging and the lack of time conditioning may be seen as adaptations to the omnivorous foraging strategy of the species.

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I. INTRODUCTION

A. Nocturnal orientation

It is well known that the orienting aids used by wood ants include both visual cues from the sky (sun and polarization pattern) and terrestrial landmarks (Jander 1957). Wood ant foraging has, however, also been observed in the night (Bruns 1954, Horstmann 1970, de Bruyn & Kruk-de Bruin 1972, Atanassov 1974). Snelli (1964) found that the traffic of F. lugubris ants leaving the mound during warm nights in the middle of August was about 30% of the daytime traffic. On those occasions the light intensity within the forest was too low to be measured with an AEG light-recorder. Jander (1957) was able to show that wood ants can use the moon as an orienting cue (see also Duelli 1972 for ants of the genus Cataglyphis), but moon orientation cannot explain my present findings. Experienced wood ant foragers show a strong and very persistent fidelity to route and place (site tenacity or Orts­treue), i.e. each individual returns to the same specific foraging ground by the same route (e.g. Ökland 1931, Otto 1955b, Dobrzanska (1958). This feature can be used to study the accuracy of direction-finding under different natural or semi-natural conditions. Previous experiments have shown (Rosengren 1971:55—57) that during the light period of the day fidelity to feeding places is mainly due to a memory of the position of trees and other large visual landmarks (see also Hölldobler 1974 for ants of the genus Pogonomyrmex). But these experiments, carried out in a very large open-air arena with movable "trees" of natural size (the ants, entering the arena through a central opening, were marked with different colours for different feeding directions), showed very clearly that the foragers did not react to a shift in the position of trees during the night (Rosengren 1971: 56, fig. 26, histograms 10 and 22). On the other hand, observations of marked forager groups in natural colonies indicated that the fidelity of foragers is retained at night in August and September (Rosengren 1971:15 and 33). It thus seems that fidelity at night may be due to other cues than fidelity in the light period. Olfactory cues immediately suggest themselves as an explanation, because there are indications that wood ants, like other Formica species, may use olfactory trails (Hangartner 1967, Horstmann 1975b, 1976, Elgert & Rosengren 1977; see also Möglich and Hölldobler 1975 for F. fusca ants). However, this would require the additional assumption that olfactory trails smell differently on different routes, because each ant must be able to identify its own specific route among a number of alternatives. The difficulties involved in explaining nocturnal orientation in wood ants according to known mechanisms motivate a closer investigation of the phenomenon. One question which arises is whether the same individuals possess orienting mechanisms for both nocturnal and diurnal foraging.

B. Activity rhythms

The question of nocturnal foraging is related to the question of activity rhythms and the influence of light on activity in wood ants. Is the nocturnal population of foragers different from the diurnal population, e.g. owing to differences in the basic reaction to light, or owing to differently "set" circadian clocks? Honey bees show both a light/dark-induced circadian rhythm and a time memory synchronized with the diel rhythm of flowers or with
an artificial feeding time (see Renner 1958, von Frisch 1967 and Koltermann 1973 for review). Some foraging objects utilized by wood ants show a diel periodicity (e.g. an aphid species reported by Horstmann (1970) to have a maximal production of honeydew in the night). Wood ant foragers gradually learn to compensate for the apparent movement of the sun and the polarization pattern, which means that they possess an adjustable "inner clock" (Jander 1957). A memory of the feeding time has been suggested in wood ants, but later experiments with time conditioning have yielded no evidence of its existence (see Renner 1958 and Wilson 1971:215 for review). A particularly surprising result is the failure to demonstrate a light/dark induced activity rhythm in wood ants (De Bruyn & Kruk-de Bruin 1972), because, unlike time conditioning, this feature is very common. Camponotus has a marked light/dark-induced circadian periodicity in both the male and worker caste (Mccluskey 1965) and a light/dark cycle at constant temperature also leads to a marked periodicity in foraging activity in some Canadian Formica species (Finnegan 1973). I have therefore found it necessary to reinvestigate this question in wood ants of the Formica rufa group.

Acknowledgements. I wish to thank Miss Christina Elmgren for examining samples of ants and Mrs. Anna Damström for revising the English. Mr Veikko Matila constructed the photo-electric actograph. This work was supported by grants from the Central Board of Finnish Research Councils and the Foundation for Research on Natural Resources in Finland.

II. MATERIAL, METHODS AND SOURCES OF ERROR

A. Field work

1. Study colony

A large colony of Formica polyctena (mound B, Sunnanvik, Sjundåa rural district about 40 km west of Helsinki) was used for the present experiment (see Rosengren 1977 for taxonomic problems). The mound (94 cm high and 750 cm in circumference) was situated in coniferous forest with climax type vegetation (Vaccinium and Calluna, a continuous moss layer). Two of the routes (7 and 8 fig. 1) led to vegetation of a more luxuriant type (some deciduous trees and herbs and grasses). These two routes (for the concept of route, see Rosengren 1971:7) were difficult to follow, owing to the type of vegetation, and were less well defined than the other routes of the system, especially in spring. Route 6 ran towards a dry, rocky habitat with patches of Cladonia lichens. The situation in a transition area between different types of vegetation seems to be fairly typical of wood ant mounds in southern Finland (Oinonen 1956). The very simplified picture of the route system presented in fig. 1 does not in-

clude small, often temporary, branch paths running to numerous "aphid trees" (generally not visited before mid May).

Some of the routes could be followed for more than 100 m through the mossy terrain. Near the mound several of the roads formed about 30 cm broad, completely naked furrows through the rough moss cover (mainly Pleurozium schreberi and Hylocomium splendens). None of the trunk routes or the larger branch routes terminated in a particular "aphid tree", as has been reported to be the rule on the continent. The forager population of the colony was so large that traffic counts could be made on the routes only in early spring and autumn. The number of foragers probably exceeded 1 million during the peak of the active season (the estimate is based on comparison with a small mound in the vicinity with a forager population of known size, see Rosengren 1977).

The large size of the colony was an obvious advantage in the present case because the removal of large samples of foragers from the routes of smaller colonies may seriously deplete the population on the sampling routes and thus affect the results.
2. Colour markings with aerosol paints

Spray paints provide an excellent means of marking a large number of foragers and are essential if the aim is to carry out extensive marking during a limited period of the day. Five leather paints (the "Magi Dye" series) and two paints for metals ("Sparvar") were used in the present case. The best way to apply the paints is to press the button so lightly that separate small droplets are formed. It is important to use paints with a large amount of light pigments, avoid "gold colours" which oxidize in time, and avoid sprays that are too thin to cover an object if applied in a single layer. None of the workers in a laboratory colony marked with "Sparvar" chrome-yellow, and white and orchid from the "Magi Dye" series lost their markings completely during a month. This (and my field observations) indicates that the aerosol paints used here are several times more durable than the manually applied paints used by some other ant students (cf. Horstmann 1973). The variation in the intensity of spray markings may be a source of error in the field (e.g. very lightly marked individuals are not observed at twilight). Another difficulty is the presence of individuals with double markings. Those errors were of less importance in the present case, because the ants were all examined under a binocular microscope. Two of the colours (pink and beige of the Magi Dyes) were nevertheless rather difficult to differentiate in lightly marked individuals.

3. Marking procedure

The experiment was carried out in mid August 1972. Routes 3a, 3b and 6 (see fig. 1) were used for marking. Route 3a had by far the largest volume of traffic, route 6 had a medium volume and branch route 3b was clearly the least frequented. The ants were marked on 9.VIII and 10.VIII at 01.00—02.00 and 10.00—11.00 on all the three routes (partly overcast sky, about 14—16°C during night marking). Route 6 was also marked at 17.00—18.00 on the same days.

The routes were spray-marked on a given stretch (about 10 m for the day groups and 20 m for the night groups) whose proximal limit 10 m from the mound on route 6 and about 2 m beyond the forking point (fig. 1) on routes 3a and 3b. Marking was started from the distal limit of the
marking stretch and progressed at about 5 minute intervals towards the mound. This procedure makes use of the fact that foragers leaving the mound hesitate and aggregate before the marking front and may thus be marked in large numbers. At the same time the stream of foragers returning to the mound is delayed by a growing stretch of marked route surface. This inhibition of the return stream is rather shortlasting but allows a certain selectivity of marking with respect to departing and returning streams. Most ants marked in the present case were thus leaving the mound.

It was considered advisable to wait about two days after marking to allow blinded and confused ants to be eliminated from the system (through loss in the terrain and active cleaning of paint from eyes and antennae). However, this was not done in the comparison described on page 15 (tables 3 and 4), because a difference in periodicity between different forager groups may be shortlasting.

4. Sampling

Ants found on the previously spray-coloured stretches of the routes were placed rapidly in jugs of water. It was unfortunately not possible to keep the departing and returning ants separate. Practically all the ants found on the marking stretches of routes 6 and 3b were removed, but only spot samples could be obtained from route 3a, owing to the large volume of traffic on that route. Samples were taken at 10.00-11.00, 17.00-18.00 and 24.00-02.00, between 1972-08-11 and 1972-08-20 and in late April 1973. The spring samples had to be taken closer to the mound owing to disturbance of the traffic on branch routes 3a and 3b by timber felling in the previous winter (cf. page 12).

The moon was absent during the night sampling (1972-08-15, 1972-08-19, 1972-08-20) and in addition one of the nights was completely overcast (1972-08-19). It was necessary to use a flashlight during night sampling. However, new ants from the mound would not be able to reach the distant sampling places during the rapid sampling procedure (the ants were of course not collected one by one, but raked together with leaves, twigs, needles and debris from the sampling stretch). The ants were killed and stored for some time in a dried condition (no bleaching of the colour was observed).

B. Laboratory work

1. System a

A colony of *F. polyctena* (several thousand workers, including a large number of so-called "Speichertiere" storing fat in the gaster, and 6 queens) was kept in a laboratory nest from the beginning of February onward. Moulded around the basal part of the nest (a hollow cylinder of spruce wood filled with natural nest material) was a large block of plaster of Paris, which was kept moist. The nest was situated in a circular drum of white cardboard covered on the inside with an oil-smearcd sheet of acrylic plastic. The 70-cm-high drum was lighted centrally with a 100 watts incandescent reflector bulb hanging 87 cm above the 3-mm-thick milky acrylic plate that covered the drum. The light could be switched on and off at regular intervals. The light intensity on the floor of the drum was normally about 700 Lux. This intensity could be decreased with the aid of large gray filters of known optical density placed in a frame between the bulb and the milky roof of the drum. A 10-cm-high acrylic partition smeared with paraffin oil divided the plaster floor of the drum (100 cm in diameter) into a nest field and a foraging field (fig. 2 system a). To reach the foraging field, the ants had to cross a glass bridge (5 mm broad), permitting traffic in only one direction (see fig. 2 system a). Ants returning from the foraging field had to cross another identically constructed one-way bridge. The ants entered the glass bridges by climbing up black wooden cylinders, which also functioned as visual beacons. Remarkably few ants tried to enter the bridges from the "wrong" end, but some hesitated at the pointed glass rod terminating the bridge and returned to the nest. This source of error was especially disturbing during low traffic intensity, but was practically absent during strong traffic, e.g. due to recruitment to food. It was possible to minimize this error by carefully regulating the gap between the point of the rod and the floor of the arena (the gap should not exceed about 3 mm, but on the other hand it should not be so narrow that the ants are tempted to enter the bridge from the wrong direction). It seemed as if training may have had some influence, because the traffic over the bridges progressed with less hesitation when the ants had been using them for some time. The traffic on the bridge leading to the foraging field was automatically recorded with an infrared-
sensitive phototransistor coupled to a printing counter (Sodecoprint). The transistor above the bridge was combined with a 900 nm infrared emitter situated underneath the bridge (see fig. 2). The printer added impulses separately for each 5 minute period (setting to zero with a synchronous motor). When properly adjusted this device counted 80 % or more of the ants crossing the bridge on their way to the foraging field.

This recording device could count ants crossing the bridge in complete darkness. Counting in darkness was avoided, however, during my initial experiments with system a, because the nest population, mainly consisting of young so-called "Innendiensttiere", was overcrowded and the ants were showing a tendency to aggregate and even deposit brood outside the nest during the dark period of the light-dark cycle. This activity made it impossible to obtain a reliable measure of foraging activity during the dark period. "Nest expansion" disappeared later and the activity along the bridge could then be measured in complete darkness as well (fig. 5 C).

The humidity of the light-proof room was kept at about 60 % RH with the aid of a continuously working humidifier (Siemens) based on the evaporation principle. The temperature within the drum rose to a maximum of about 26°C during the light period and sank to a minimum of about 23°C when the light had been switched off (temperature and humidity controlled with a Lamprecht thermohygrometer). During constant light the temperature within the drum stayed at about 25°C±1°C. A pneumatic feeding vessel (see von Frisch 1967:18) filled with a solution of syrup gave a constant supply of sugar within the foraging field (fig. 2), but only some of the ants entering the bridge visited the vessel. Others carried dead nestmates or pieces of pupal husks to certain sites within the foraging field. Most of the "foragers", however, just seemed to "explore" the arena. The ants were fed with Tenebrio larvae or fresh beef at longer or shorter intervals. For further details of the experimental procedure see pp. 17—19.

2. System b

An improved version of the equipment just described was used for my later recordings (fig. 6 and 7). The main differences were (fig. 2 system b):

Fig. 2. System a. I. Arena with nest A and a pneumatic feeding vessel B. 1: one-way bridge leading from the nest field to the foraging field. 2: one-way bridge leading from the foraging field to the nest field. 3: partition between the nest field and the foraging field.

II. 1: black cylinder. 2: glass bridge leading from the nest field to the foraging field. 3: partition between the nest field and the foraging field. 4: pointed glass rod terminating the bridge. 5: cylinder holding the infrared emitter. 6: phototransistor coupled to a printing counter.

System b. I. Arena seen from above with the nest field (A) and the circular foraging field (B). Entrance to the nest in the centre of the nest field. One-way bridges between nest field and foraging field indicated by arrows.

II. Cross-section of arena within circular wall. A: nest field. B: foraging field. C: nest. D: rubber tube connecting nest with centre of nest field. E: Hg mixed light lamp within ball of opal glass (this lamp was replaced by a fluorescent tube during some experiments). 1: recording bridge leading from A to B. 2: infrared emitter and phototransistor. 3: printing counter (Sodecoprint).

1. The nest, made of partly transparent acrylic plastic filled with natural nest material, had a
much larger volume (41 cm in height, 55 cm in diameter) and was situated under the arena. The ants reached the circular nest field of the arena through a 0.5 m-long rubber tube. No tendency to deposit nest material or brood in the nest field during the dark period was observed in this case.

2. The arena was larger and was divided by a circular acrylic partition into a round "nest field" and a circular "foraging field" (fig. 2b). The ants used the circular foraging field as a kind of "running track". This "release of locomotor behaviour" in the foraging field combined with the long way from the nest to the recording bridge seemed to stabilize the traffic over the bridge, so that irregular activity peaks were less common.

3. The arena was surrounded by a circular wall made of white cardboard but was not situated in a closed drum as in system a, so that the heat from the lamp was not "trapped" within the drum. This and the use of a Hg mixed-light bulb (160 watt), which produced less heat than the incandescent lamp previously used, gave a lower temperature within the arena (22 ± 1°C), and made the temperature difference between the light and dark period smaller (p. 8). During part of the recordings, the Hg mixed-light bulb (giving 175 Lux in the arena) was replaced with fluorescent tubes (giving 350 Lux in the arena).

4. Both the Hg lamp (in a ball of opal glass) and the fluorescent tube contrasted sharply with the black-painted ceiling of the laboratory room. The electronic recording device and the bridges were, with small modifications, the same as in system a. Relative humidity was kept at about 60% with a continuously running evaporator.

C. Sources of error

1. In the field

The long mean journey time (see Holt 1955, Horstmann 1974) and variations in the journey time of each forager may be expected to blur original timing differences between groups (cf. page 17). This makes it difficult to draw any conclusions from negative results.

A criticism that can be made of my results showing retention of fidelity during nocturnal foraging (pp. 10-41) is that I cannot prove that all the ants captured during the night had departed from the mound during that period. It can be argued that some of them left the mound in the evening when the light conditions still allowed visual orientation and then returned in the dark period along scented trails. I confirmed, however, during each night sampling (24:00—02:00) that streams of foragers were moving away from the mound along the sampling stretches. Thus if visual orientation were a deciding factor, fidelity could be expected to decrease.

2. In the laboratory

Variations in temperature

It was not possible to control the temperature of the laboratory exactly but it was checked continuously with thermographs. In the closed cylinder of system a, the temperature differed too greatly between the light and the dark period to permit firm conclusions concerning the specific effect of light. The Hg lamp of the open b system gave a maximum difference of about 0.5°C—0.6°C between the light and the dark period, and this was originally considered "safe", because the switching on and off of an electric heater (12:12 cycle) giving about the same cyclic difference in temperature during constant light (LL) induced no clear activity rhythm in the population (in this pilot study, the colony had not been subjected to any regular light cycle before the experiment). Later studies showed, however, that a maximum difference of only about 1°C can maintain a very clear activity rhythm in complete darkness (DD), at least if the population has previously been subjected to a LD rhythm (fig. 6). The fluorescent tube used in system b gave no measurable difference in temperature between the light and dark periods, but nevertheless induced a clear activity rhythm in the population. But even in the latter case, a thermal effect cannot be completely ruled out, because the bodies of the ants may absorb some thermal radiation from the light source.

Variations in humidity

It is obvious that variations in the relative humidity of the air may be a serious source of error when foraging activity is studied as a response to other factors (see p. 25), and the relative humidity of the laboratory was checked with a hygrograph. A hygrostat was used initially to obtain a constant RH level, but it had to be removed, be-
cause it caused a regular pattern of small oscillations in RH. The continuously running evaporator was found to give a smoother humidity curve in the closed room. The RH level did indeed change gradually over several hours or days, but these changes did not show any diel pattern.

Variations in sounds and vibrations caused by human activity

The laboratory and the nest was not shielded from vibrations, e.g. those due to car traffic on surrounding streets. It is easy to demonstrate that wood ants react to strong vibrational stimuli with increasing locomotor activity (unrest), and the vibrations from human activities followed approximately the same diel "rhythm" as the light-dark cycle used through some of the experiments. This source of error was nevertheless less serious than may appear at first glance. This is because my recording device did not measure general locomotor activity but very specific, probably learned, behaviour (climbing to the top of a cylinder and thereafter crossing the bridge to the foraging field).

The difference between random unrest in the population and goal-directed traffic to the foraging field was especially evident in experiments where the RH level was raised. Low atmospheric humidity increases random-type locomotor "restlessness" of wood ants, while increased humidity has a "calming" effect (my unpublished observations). In spite of this calming effect of high humidity, the traffic along the bridge regularly increased when the humidity level of the air was raised at the temperature used during the experiments (see p. 23).

Moreover, after a prolonged period in continuous light or continuous darkness, the ants' periodicity faded out although, if the vibratory stimuli served as a "Zeitgeber", the periodicity might have been expected to continue. Experiments with displaced LD cycles (fig. 7) also indicated that vibrational stimuli hardly can have been a serious source of error.

Variations in locomotor speed

My recording device measured the number of ants crossing the bridge but not the running speed. Speed is mainly dependent on temperature (WELLENSTEIN 1954) but may also be influenced by other factors (ROSENGREN 1971:45). The speed of wood ant foragers has been reported to show a weak diel periodicity (DE BRUIJN & KRUK-DE BRUIJN 1972) and a description has recently been given of a "TV-scanning line comparator", which records both number and speed of goal-directed insects (KRUK-DE BRUIJN & TISSING 1975).

III. RESULTS

A. Orientation and fidelity to routes

1. Trunk routes

Like _F. rufa_ L. and _F. lugubris_, _F. polyctena_, shows a route fidelity between about 85% and 95% in foragers marked late in the summer (ROSENGREN 1971:15, 33 and 46) and observed through the rest of the active season. This means that a maximum of about 15% of the foragers mass-marked with a different spray colour for each route of a system can be observed to change their routes during an observation period of two months. The route fidelity of foragers showed only a very small, probably insignificant, decrease as a function of time (ROSENGREN 1971:33). If the ants are marked on foraging grounds outside the routes, however, a decrease in _Ortstreue_ with time can be observed in a given batch of marked hunters (HORSTMANN 1973). My previous estimates of the pooled fidelity of route systems were based on examinations of _all_ the trunk routes of my study colonies (for methods see ROSENGREN 1971:5–7). The screening was less effective in the present case because only the routes where marking had been carried out were
Table 1. The pooled results of samplings in August 1972 and April 1973 of ants marked on 1972-08-09-10 on two trunk routes. Column "marked C-ants": ants showing fidelity to the marking route. Column "marked D-ants": ants found on the "wrong" route. The values for the cold night of 15.VIII (11°C) have been excluded from the two last columns: "range of temp." and "range of %".

<table>
<thead>
<tr>
<th>trunk route</th>
<th>time of sampling</th>
<th>no. of samples</th>
<th>marked C-ants</th>
<th>marked D-ants</th>
<th>total no. of ants</th>
<th>range of temp.</th>
<th>range of % marked per sample ± 1.96 ( \delta_p )</th>
</tr>
</thead>
<tbody>
<tr>
<td>6</td>
<td>1972-08-13 - - 20</td>
<td>6</td>
<td>1446</td>
<td>1</td>
<td>3879</td>
<td>14°C to 20°C</td>
<td>34.2 ± 3.6 to 37.3 ± 5.3</td>
</tr>
<tr>
<td>3</td>
<td></td>
<td>8</td>
<td>1128</td>
<td>0</td>
<td>3235</td>
<td>-</td>
<td>34.0 ± 4.0 to 38.0 ± 3.9</td>
</tr>
<tr>
<td>( \Sigma )</td>
<td></td>
<td></td>
<td>2574</td>
<td>1</td>
<td>7114</td>
<td>-</td>
<td>24.6 ± 6.8 and 34.6 ± 3.5</td>
</tr>
<tr>
<td>6</td>
<td>1973-04-20 - - 22</td>
<td>5</td>
<td>78</td>
<td>0</td>
<td>1928</td>
<td>6.5°C to 10.5°C</td>
<td>2.1 ± 1.5 to 6.3 ± 2.0</td>
</tr>
<tr>
<td>3</td>
<td></td>
<td>4</td>
<td>56</td>
<td>1</td>
<td>1767</td>
<td>-</td>
<td>2.5 ± 1.7 to 3.4 ± 2.1</td>
</tr>
<tr>
<td>( \Sigma )</td>
<td></td>
<td></td>
<td>134</td>
<td>1</td>
<td>3695</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

included; five routes of the system were left unexamined.

On August 9 and 10, the ants were marked on three routes of the system (fig. 1), a different marking colour being used for each route and time group (see page 5). The percentage distribution of marked groups in each separate sample is seen in fig. 4. The pooled values given in table 1 (rows for sampling in 1972) have been used for the present estimate. The table is based on the same samples as fig. 4, but the values from branch routes 3a and 3b have been pooled and 46 marked ants changing their direction at the fork of 3a and 3b have been included. Of a total of 2575 marked ants sampled in 1972 (table 1) only one ant (captured in the light period) was found on a "wrong" route, i.e. it did not show fidelity. Of those marked ants, 1472 had been sampled during the light period and 1103 in the night (24.00—02.00, no moon present). It can be concluded that both the night groups and the day groups showed a fidelity of nearly 100% with respect to the examined routes. The exceptionally high fidelity found in the present case was probably due both to the large angular distance between the examined trunk routes (about 70 degrees) and the fact that the ants had been marked far away from the mound (see page 5). In addition, as in most of my study colonies, the naked furrows of the trunk roads were isolated from each other by a dense and almost impassable moss cover (mainly Pleurozium schreberi). All this means that the trunk system examined may have been too crude an instrument to reveal small differences in direction-finding capacity between day foragers and night foragers.

2. The fork system

There was, however, a more sensitive system, namely the fork of route 3. A small number of foragers which had been marked on route 3a regularly appeared in the samples from branch route 3b. The pooled day samples from branch 3b (1972-08-13-20) contained 1532 ants in all (marked + unmarked), 19 (1.24 %) of which had been marked on the other branch. The corresponding value for the night samples from this route (941 ants in all) was 14 (1.49 %). The difference is not significant \( (x^2 = 0.10 \quad 1 \text{ d.f. } P = 0.75) \). The fork system was used for a more rigorous test, in which samples taken during the heavily overcast night of 19.XIII (about 14°C, no moon, sampling time: 24.30—01.15) were compared with those taken on the partially overcast day of 1972-08-20 at about 11.00.

The results are presented in table 2. If we divide the material into two categories only (ants faithful to the marking route versus ants unfaithful) and pool the numbers for both the branches, we obtain the
### Table 2.

<table>
<thead>
<tr>
<th>Marking period and route</th>
<th>Night sample</th>
<th>Day sample</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1972-08-19</td>
<td>1972-08-19</td>
</tr>
<tr>
<td>Branch 3a</td>
<td>33</td>
<td>2</td>
</tr>
<tr>
<td>Branch 3b</td>
<td>21</td>
<td>40</td>
</tr>
<tr>
<td>Day-marked 3a</td>
<td>86</td>
<td>22</td>
</tr>
<tr>
<td>Night-marked 3a</td>
<td>13</td>
<td>2</td>
</tr>
<tr>
<td>Double-marked 3a</td>
<td>108</td>
<td>28</td>
</tr>
<tr>
<td>Day-marked 3b</td>
<td>37</td>
<td>22</td>
</tr>
<tr>
<td>Night-marked 3b</td>
<td>14</td>
<td>10</td>
</tr>
<tr>
<td>Double-marked 3b</td>
<td>14</td>
<td>10</td>
</tr>
<tr>
<td>Total number of ants examined (marked + unmarked)</td>
<td>188</td>
<td>118</td>
</tr>
</tbody>
</table>

following result (95% confidence interval $p \pm 1.96 \delta p$):

1. Night group (1972-08-19): 94.3% ± 4.4% of 105 marked ants faithful to the marking route.
2. Day group (1972-08-20): 94.1% ± 2.6% of 324 marked ants faithful to the marking route.

It can be concluded that the fidelity of the foragers to the examined branch routes remained unaltered during the night, although the sky was completely overcast and the moon was absent during the time of sampling. It is possible that the clouds may have reflected some light from human settlements some kilometres away, but the light conditions within the spruce-dominated forest were too poor for the visual orientation of a human being.

3. The situation after the hibernation period

The following results confirm my previous conclusions about retention of fidelity through the winter (ROSENGREN 1971), but also show that visual changes due to cutting of trees during winter may lead to a complete breakdown of the previous foraging tradition at the points exposed to the disturbance. At least part of the memory surviving the hibernation period and normally leading to a restoration of the previous route system (even before the activity of aphids, see ROSENGREN 1971 and 1977) is thus very probably attributable to the visually stable environment provided by a coniferous forest. It seems likely that the deciduous habitats (oak-hornbeam forests) studied by HORSERMANN (1975a) did not admit visual recognition of previous feeding places in the spring because the trees had lost their leaves during the winter. These questions have been analysed in greater detail in a more systematic investigation of the effect of thinning and clear cutting in forests on wood and foraging (ROSENGREN & PAMILO 1977). The most important point in the present context is the implication that nonvisual clues used during nocturnal foraging either are not preserved through the hibernation period or are insufficient for the restoration of the previous foraging tradition in early spring.

### a. Mortality during hibernation

The hibernation period during winter 1972—1973 kept the ants isolated within the nest for about 5 months. A crude estimate based on table 1 indicates that 10% of the forager population marked on 1972-08-10 survived to late April 1973 (This is clearly an underestimate because marking losses are not taken into consideration). This means that part of the forager population of late summer survived for at least a further 8 months. Foragers marked at the beginning of the season (May) may survive up to 5 months (my unpublished observations). The present marking gives no estimate of the real mortality of foragers during hibernation, especially because the ants had been marked about 2.5 months before the end of the active season. Markings carried out in another colony of *F. polyctena* in late September indicated that actual losses during the hibernation may be much smaller (29.4% marked foragers, $n = 300$ in late October immediately before the end of the active season compared with 8.6%, $n = 995$ and 21.1%, $n = 290$ during different samplings in April the following year). This indicates that at least a considerable part of the forager
population still active in late autumn may survive the hibernation period in southern Finland.

b. Variations in the proportion of marked ants in samples from early spring

In samples taken in late August 1972, the percentage of marked ants was rather uniform, if the values for the cold night of 1972-08-15 are excluded (table 1 and fig. 4). The spring samples were caught during a more restricted period (1973-04-20 - 22 between 09.50 and 15.00) and under more uniform conditions (clear weather, 6.5° C to 10.5° C).

Nevertheless the five samples caught on route 6 showed statistically significant differences in their proportions of marked ants ($\chi^2 = 15.7$, 4 d.f. $0.005 > P > 0.001$). The reason for these variations may be a difference in traffic behaviour in early spring between foragers of different age (see Rosengren 1977). The overlap of the extreme values in the samples from route 3 (table 1) may be due to the fact that the blocking of this route (see page 13) caused a more thorough mixing of different age fractions (the traffic stream on route 6 extended about 60 m from the mound along the route).

c. Retention of fidelity through the winter

The pooled spring samples from route 6 included 78 marked ants, all of which had been marked on that route (table 1). The corresponding value for trunk route 3 was 57 ants; all these had been marked on either of the branches of trunk route 3, except one ant originally marked on route 6. The ants of the examined trunk route system had thus retained most of their fidelity through the winter. My previous results indicate a small but statistically significant decrease in fidelity during the hibernation period (Rosengren 1971:70), but it was not possible to demonstrate this effect in the present small material.

d. Effect of visual disturbance

Previous results indicate that fidelity is also retained through the winter with respect to the branches of a fork situated several metres from the mound (Rosengren 1971:73).

Part of the forest on the right side of route 3a as one progressed from the forking point (see fig. 1) had been cut during winter 1973. The timber felling produced deep gaps in the forest, which greatly altered the light conditions and the forest contour as seen from the forking point. However, the change did not involve the visual prospect from the mound or the forest contour on either side of trunk route 3 along an 11-m stretch between the mound and the fork. Nor did the clearing involve the swampy part of the forest in which route 3a terminated. The foraging territories of the study routes thus remained practically untouched. All slash (branches and twigs) from the timber felling had been carefully removed from the area during the winter.

The foragers of this colony clearly followed the routes right from the beginning of activity in early spring (see page 5), and the traffic along the trunk routes of the system appeared quite normal (distinct traffic streams were found on routes 3, 4, 5 and 6 between 1973-04-20 and 1973-04-22). The ants travelled along trunk route 3 in especially large numbers (this trunk and branch 3a had the largest traffic volume in late summer 1972). About 1—2 m before the forking point (3a, 3b) the stream along trunk 3 dissolved, however, and the ants spread and clustered in the terrain surrounding the fork (at the same time, the traffic streams on the undisturbed routes of the system advanced about 50—60 m from the mound). A traffic count (1973-04-21 at 10.30 to 11.30, 10° C to 11° C) on trunk route 3 between the mound and the fork showed that ants were proceeding from the mound along this trunk at the rate of 680 per 5 min (the value for route 6 was 138 ants/5 min). Counts on the other side of the fork gave 11 ants/5 min for branch 3a and 6 ants/5 min for branch 3b. Furth-
er observations during this period (1973-04-20 - 22) revealed that there was very weak traffic along much of the previous length of route 3b, but that the traffic on route 3a was only apparent; the ants counted when they crossed a given line on branch 3a did not actually follow the route but reached the counting-line more or less "at random" from the surrounding terrain. The failure to follow route 3a was especially remarkable because the route formed a bare 10—15-cm-broad furrow through the moss all the way from the forking point to the intact part of the forest (not seen in fig. 1). It seemed as if the foragers "swarming" or clustering in the moss from the forking point onward had a preference for the sunny gaps created by the cutting, and far more foragers were found on the right side of route 3a than on the left side. It also seemed as if the ants clustering in this area had difficulty in returning to the mound. Some of the clusters were still present in the terrain in the evening, when ants of the other routes had already regained the mound. Further evidence of disorientation was the unusually intense carrying of nestmates from the disturbed area (cf. Rosengren 1977). In several wood ants species, ants may pass the winter in hibernation chambers situated in hillocks outside the mound (Kn"etzt 1964, Rosengren 1969, Rosengren 1971: 9) and are then carried back to the mound by nestmates in early spring (e.g. Rosengren 1969). However, the present carrying had nothing to do with hibernation outside the mound; no carrying from the mound was observed the previous autumn and no hibernation chambers were found when the earth was dug under various small elevations observed around the forking point. It should be pointed out that my observations about the disturbing effect of timber felling were made before the active period of aphids (no traffic to aphids was observed anywhere in the colony during this time).

The "swarming" within the fork area continued for some days but disappeared at the beginning of May, with an obvious decrease in the initially very high traffic intensity on trunk route 3. The traffic increased on branch 3b during the spring, but 3a remained abandoned until summer, when very weak but regular traffic was observed on this route. A wholly new route (broken line in fig. 1) developed during the same spring toward the rich foraging area of route 3a.

During 1975 this was the most frequented route in the colony, while route 3a remained the least frequented of the previous main routes.

On 1973-04-20 I collected all the marked ants found on trunk route 3 between the mound and the forking point, and the marked ants found within two rectangular areas of the terrain roughly corresponding to 3a and 3b (fig. 3). The results show that the ants captured within the 3b area had retained a statistically significant preference for this left-hand direction in spite of their failure to locate the exact position of the route ($\chi^2 = 6.41$ d.f. 0.025 > P > 0.01).

The sample from the area corresponding to route 3a is more difficult to interpret.

![Diagram](image-url)
The proportion of ants marked the previous summer in this branch appeared to be lower (61%) than on the trunk route (71%), but the difference was not statistically significant ($\chi^2 = 0.8$, 1 d.f., 0.50 > $P > 0.30$). It may be that all the marked groups deviated to the right, attracted by the open gaps in the forest (as was actually observed). This tendency may have been stronger for ants marked on the more disturbed route 3a. It is known that wood ants may "regress" from a learned menotaxis to a basic phototaxis if the situation becomes too confusing (Jander 1957). This explanation is offered very tentatively in the present case, but two conclusions seem quite clear:

1. Tactile or olfactory cues from the cleared road surface do not make an important contribution to the spatial memory or fidelity preserved through the winter (not a single marked ant captured within the sampling areas seen in fig. 3 was found on either of the branches).

2. Timber felling may lead to an almost complete breakdown of the previous foraging tradition before aphids can be utilized (the foraging grounds in which the routes terminated had not been affected at all by the clearing), and the most likely explanation of this effect is a lack of familiar visual cues. For further evidence on this point, see Rosengren (1977) and Rosengren & Pamilyo (1977).

4. Concluding remarks

Nocturnal foraging in wood ants does not require the presence of the moon and also takes place under a completely overcast sky (midnight in late August within a coniferous forest). Under these conditions, the individual ant is still able to locate its "own" specific route among a number of alternative similarly trafficked routes. Measured by route fidelity, the spatial discrimination of the ants is not lower during the night than during the light period. This was also the case in tests with a fork system situated about 14 m from the mound. Yet previous results have shown that under the light conditions prevailing during late summer nights, wood ants do not react to visual landmarks.

Marked foragers surviving an uninterrupted hibernation period of 5 months are able to recognize their previous foraging directions after winter. Observations of the disturbing effect of timber felling make it seem likely that the ants remember a configuration of visual landmarks through the hibernation period. If the visual environment has been altered, the ants are not able to follow a cleared road furrow in early spring, or not, at least, for any great distance from the mound. This accords with previous findings that the site tenacity shown by wood ant foragers in the light period is mainly due to a memory of visual landmarks.

These results can be explained either by assuming that fidelity to route and site is due to different orienting mechanisms in the same individual forager, one mechanism operating in light and the other in darkness, or by assuming that there are two populations of foragers, one active in the day and the other in the night.

B. Activity rhythms

1. Are night foragers and day foragers differently "phased" populations?

The results summarized in fig. 4 indicate that the differences between night samples and day samples are so small that they may lack ecological importance. On the whole, night foragers and day foragers do not constitute clearly separate populations. Some differences were, however, found. In the samples taken from both routes on the cold night of 1972-08-15, the proportion of unmarked ants was lower than in the preceding samples, the difference being statistically significant, and the proportion of night-marked ants was higher. That this effect may have been due to low temperature rather than to poor light conditions is evident from the fact that the samples taken on the warmer nights (1972-08-19 and 1972-08-
Fig. 4. Samples from route 6 (upper row) and route 3 (lower row). Ordinate: percentage of marked and unmarked ants within a sample. Abscissa: time of the day, temperature and cloudiness (O = clear weather, • = completely overcast sky). Black columns: ants marked in the night (01.00—02.00). Small white columns: ants marked in the day (upper row at 10.00—11.00 and 17.00—18.00; lower row at 10.00—11.00). Tall white columns with lines showing 95 % confidence interval: unmarked ants. Hatched columns: double marked ants (included only for samples from route 3). In the lower row, both routes 3a and 3b are represented in the histograms for 19.VIII and 20.VIII, but only route 3b in the others.

20) from routes 6 and 3b did not show such differences. The night sample from route 3a (1972-08-19) showed a significantly lower percentage of unmarked ants than the corresponding day sample (1972-08-20) ($\chi^2 = 6.7$ 1 d.f. $0.01 > P > 0.005$) and a specific increase of ants marked in the night (fig. 4).

Tables 3 and 4 compare pooled groups of ants marked at different times (in addition to the material in fig. 4, samples taken on 1972-08-11 and 1972-08-12 have been included in this comparison, see page 6). It can be concluded that the percentage of night-marked ants was larger in night samples than in day samples (differences statistically significant in tables 3a, 3b and 3d, but not 3c). Route 3b (table 3c) may have had a more uniform forager population owing to its low traffic frequencies (see page 5). This is supported by my current finding that ants of the young recruit group are more inclined to join routes with a large traffic volume than routes with a small traffic volume (Rosengren 1977). This means
Table 3.

<table>
<thead>
<tr>
<th></th>
<th>ants marked at 10.00—11.00 (route 6)</th>
<th>ants marked at 01.00—02.00 (route 6)</th>
<th>Sum</th>
</tr>
</thead>
<tbody>
<tr>
<td>a. samples from 10.00—11.00</td>
<td>210 (51.0 %)</td>
<td>202 (49.0 %)</td>
<td>412</td>
</tr>
<tr>
<td></td>
<td>162 (36.7 %)</td>
<td>279 (63.3 %)</td>
<td>441</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>b. samples from 17.00—18.00</td>
<td>124 (44.9 %)</td>
<td>152 (55.1 %)</td>
<td>276</td>
</tr>
<tr>
<td></td>
<td>149 (34.8 %)</td>
<td>279 (65.2 %)</td>
<td>428</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>c. samples from 11.00—17.00</td>
<td>137 (30.5 %)</td>
<td>312 (69.5 %)</td>
<td>449</td>
</tr>
<tr>
<td></td>
<td>91 (29.1 %)</td>
<td>222 (70.9 %)</td>
<td>313</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>d. samples from 10.00—11.00</td>
<td>92 (71.9 %)</td>
<td>36 (28.1 %)</td>
<td>128</td>
</tr>
<tr>
<td></td>
<td>33 (56.0 %)</td>
<td>25 (43.1 %)</td>
<td>58</td>
</tr>
</tbody>
</table>

\( \chi^2 = 16.9 \) 1 d.f.  \( P < 0.0005 \)

\( \chi^2 = 6.8 \) 1 d.f.  \( 0.01 > P > 0.005 \)

\( \chi^2 = 0.13 \) 1 d.f.  \( 0.80 > P > 0.70 \)

\( \chi^2 = 4.09 \) 1 d.f.  \( 0.05 > P > 0.025 \)

that intensely frequented routes (such as route 6 and especially route 3a in the present case) probably have a more variable age composition than routes with weak or declining traffic.

This interpretation is supported by the following calculation. The ants on route 6 had been marked with three different spray colours and we would therefore expect a larger number of double or triple marked ants on this route (these groups were not included in fig. 4 upper row but can be calculated from the figure as 100 — sum of included columns). This was not, however, the case. Route 3b had a larger percentage of double marked ants than any of the other routes (route 3b: 14.3 % double marked, all marked ants = 886, route 6: 11.1 % double or triple marked, all marked ants = 1446, route 3a: 3.6 % double marked, all marked ants = 193). These differences are statistically significant: route 3b vs. route 6 (\( \chi^2 = 5.5 \) 1 d.f.  \( 0.025 > P > 0.01 \)) route 3b vs. 3a (\( \chi^2 = 15.7 \) 1 d.f.  \( P < 0.0005 \)). The probability that day-marked foragers will also be marked in the night is very probably higher for regular foragers (with a stronger urge to forage) than for groups which leave the mound only during optimal conditions and/or during mass recruitment.

Old, "experienced" foragers seem to have a higher motivation for foraging
than younger categories. This is seen from the fact that old foragers (e.g. foragers marked in the autumn and observed in spring the following year) start the traffic from the mound as "pioneers" when the conditions are still suboptimal (ROSENGREN 1977). Ants marked in the night (especially at low temperatures) probably belong to the same category. The old forager category is also probably the most "experienced" and thus may have the greatest ability to master subtle cues during nocturnal orientation.

The fact that some of the routes showed a clear difference in the composition of the forager populations during the day and the night could also be explained by assuming a difference in periodicity, due to an individual sense of time, between different forager groups. It is therefore necessary to analyse in some detail the question of time memory and circadian rhythm in wood ants.

The ant leaving the mound along route 6 had been marked with two different colours at two different times of the light period. The results for all the samples from 11.VIII onwards have been summarized in table 4. It can be seen that the proportions of marked groups visiting the foraging field at different times were almost equal. ($\chi^2 = 0.0123$, 1 d.f. 0.95 > $P$ > 0.90).

It can be seen from the table that the proportions of marked groups visiting the foraging field at different times were almost equal. ($\chi^2 = 0.029$, 1 d.f. 0.90 > $P$ > 0.80). Thus no evidence was obtained of an individual memory of the feeding time.

The total traffic on the one-way bridge leading to the foraging field was measured automatically by the photoelectric counting device (page 7) through out the

<table>
<thead>
<tr>
<th>Table 4.</th>
<th>ants marked at 10.00—11.00 (route 6)</th>
<th>ants marked at 17.00—18.00 (route 6)</th>
<th>Sum</th>
</tr>
</thead>
<tbody>
<tr>
<td>samples from 10.00—12.00</td>
<td>210 (51.7%)</td>
<td>196 (48.3%)</td>
<td>406</td>
</tr>
<tr>
<td>samples from 17.00—18.00</td>
<td>137 (52.5%)</td>
<td>124 (47.5%)</td>
<td>261</td>
</tr>
</tbody>
</table>

2. Conditioning to a feeding time?
My laboratory colony (p. 6, system a) was kept without insect food from the beginning of March (12:12 hour LD cycle, light period: 09.00 to 21.00). For a week from 11.III onwards, the ants were offered pieces of Tenebrio larvae in the foraging field (fig. 2) each day between 17.00 and 18.00. During the last meal, 200 ants visiting the foraging field were marked with green colour patches. Another group of 200 ants visiting the foraging field in the morning between 11.00 and 12.00 was marked with white patches. During 3 days the numbers of differently marked ants visiting the foraging field were counted through a window in the wall of the drum at 11.00—12.00 and 17.00—18.00. Table 5 gives the pooled results for the 3 days.

<table>
<thead>
<tr>
<th>Table 5.</th>
<th>ants marked at 11.00—17.00</th>
<th>ants marked at 17.00—18.00</th>
<th>Sum</th>
</tr>
</thead>
<tbody>
<tr>
<td>(white)</td>
<td>(green)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>visits to foraging field</td>
<td>visits to foraging field</td>
<td></td>
<td></td>
</tr>
<tr>
<td>at 11.00—12.00</td>
<td>77 (47%)</td>
<td>87 (53%)</td>
<td>164</td>
</tr>
<tr>
<td>at 17.00—18.00</td>
<td>90 (48%)</td>
<td>98 (52%)</td>
<td>188</td>
</tr>
</tbody>
</table>
FIG. 5. Ordinate: number of ants crossing the bridge per hour (1 = 100 ants). Right ordinate in C: relative log intensities of light = height of areas within dashed line. Abscissas: the time of the day in hours (complete darkness between 21.00 and 9.00 on days 1–3 in A, but continuous light during days 4–10 in B). Arrows in A indicate the feeding time on the days before the recording. Small arrows at 17.45 in fig. D (days 2 and 3) indicate feeding with Tenebrio larvae during the recording. For further explanation, see the text.
light period during the 3 days that the marked ants were watched. The records (fig. 5 A days 1—3) show no clear activity peak at the previous feeding time (17.00—18.00).

The LD cycle was then replaced by continuous, diffuse light (fig. 5 B days 4—10). No clear activity peaks were observed at the previous feeding time (arrows pointing downwards). Some indications of activity rhythms probably related to the previous LD cycle were found during the first 3 days, but this periodicity seemed to fade out rapidly. It is difficult to draw any definite conclusions, because the total level of activity was greatly reduced.

Oscillations in the activity level initiated by the sudden switching on or off of the light may have masked weak activity peaks due to a memory of the feeding time (fig. 5 A and B). I studied this point in a population (system a) kept for about two weeks in constant light (after the same LD cycle as that mentioned previously). The starved colony was fed with pieces of Tenebrio larvae at 17.45 during two successive days. Strong recruitment peaks (for a closer analysis of similar recruitment peaks see fig. 8 B) were observed during feeding and carrying of the insect pieces to the nest (fig. 5 D days 2 and 3 at 18.00—19.00), but no activity peaks attributable to an induced feeding rhythm were observed at the previous feeding time during the following days (fig. 5 D: days 4 and 5 after feeding compared with days 1 and 2 before feeding). The activity curve both before and after feeding showed similar, very slight, indications of an afternoon peak of unknown origin.

3. The effect of light and temperature cycles on the diel activity pattern

The recordings reported so far were restricted to the light period of the LD cycle and thus gave a very incomplete picture. We may ask whether the marked morning peaks seen in fig. 5 (days 1—4) represent an exogenous or "startle" reaction associated with the sudden onset of light (see Saunders 1976:12). The sudden removal of some of the optical density filters placed between the bulb and the milky roof of the arena (system a, p. 6) caused clear activity peaks (fig. 5 C), which supports the interpretation that the reaction was exogenous. This cannot, however, be the whole explanation, because during a continuous LD cycle the activity begins to rise well before the actual onset of light (fig. 6 and 7 based on recordings in system b, p. 7). The activity builds up some hours before the onset of light and may actually reach its maximum within the scotophase. During the summer, the lowest level of activity is found during the first hours of the scotophase (fig. 7). Recordings from late autumn suggests that during this season the ants are generally less active in the light period than in the dark period (the time of peak activity regularly occurs well before the onset of light). This results seems to be in accordance with my observation that in natural colonies nocturnal foraging is more common toward the end of the active season (p. 27) but the mechanisms probably involved in this case (see later) appear to be so complicated that conclusions on this point seem premature.

Light or temperature?

However, the recordings presented in figs. 5 and 7 do not prove that the activity rhythm observed in the population was induced by the light cycle per se, because the light sources used during those experiments also had a heating effect (see also p. 8). In the experiments with the mixed Hg bulb (fig. 7), the maximum temperature difference between the light and the dark periods was about 0.6°C. The diel temperature curves caused by the bulb (fig. 7) and by an electric heater (fig. 6) had the same general shape, but the amplitude was larger with the heater (maximum about 1.5°C). The fluorescent tube used with system b had no measurable effect on the ambient temperature of the arena and it was thus possible to compare activity in a relatively "pure"
LD cycle with activity in a temperature cycle (fig. 6).

An LD 12:12 cycle (fluorescent tube) resulted in clear diel periodicity with maximum activity at, or slightly before, light on (fig. 6). When the light cycle was replaced with a temperature cycle of the same period (12:12) in continuous darkness (DD), the activity of the population retained a marked periodicity, but the pattern was different, because the peak of activity shifted from early "morning" to the middle of the "day". In continuous light (LL), the rhythm induced by the temperature cycle was much weaker and the periodicity seemed gradually to fade out. Replacement of the temperature cycle (LL) with the original LD 12:12 cycle restored the original pattern with a peak in early "morning" (fig. 6).

Thus, contrary to previous claims (DE BRUYN & KRUUK-DE BRUIN 1972), it seems that light alone can induce an activity rhythm in the population, although the evidence for this is perhaps not entirely conclusive (absorption of thermal radiation by bodies of ants, see p. 8). A strong activity rhythm can also be maintained in the population by a temperature cycle, especially in continuous darkness. The smallness of the temperature differences needed for this effect suggests that temperature is here functioning as a "Zeit-
FIG. 7. Recordings of the diel activity in system b (p. 00). Ordinate: no. of ants crossing the recording bridge per hour (1 = 100 ants). Abscissa: time of day in hours. A 12:12 LD period was used in all cases, but the onset of the dark period (black stretches) was quite different during different recording periods (months). The ant population was the same in all recordings. The population was kept in LL for at least one week before a change in the LD cycle. A Hg mixed-light bulb was used in all the experiments (maximum temperature difference between light and dark period: 0.5—0.6°C). Arrows: ants fed with proteins.

geber" rather than influencing activity directly. This interpretation is supported by the fact that activity begins to increase well before the switching on of the electric heater (fig. 6). When the experiment illustrated in fig. 6 (replacement of LD
cycles with temperature cycles) was repeated, the results confirmed the above conclusions.

Wood ants are considered to be typical diurnal animals. The present observations indicate that this cannot be explained by the "stimulating" effect of light on locomotor behaviour. Darkness does not generally have a suppressing effect on activity (figs. 5C, 6 and 7). The ants visited sugar and meat and carried dead nestmates and pupal husks to the foraging field during the dark phase as well. In nature the LD cycle is generally coupled to a temperature cycle. The function of entrainment by the light cycle is probably to synchronize the activity of the foraging population more exactly than is possible by temperature alone (see p. 27 for discussion).

Some characteristics of the diel rhythm

Fig. 7 shows that it is possible to entrain the same population to new 12:12 cycles not corresponding to the natural cycle (before entrainment to a new cycle the population was kept in constant light and temperature for at least 1 week).

A characteristic feature of some of my recordings is that the amplitude of the rhythm increases with the degree of starvation of the colony (the ants obtained no protein during days 1—9 fig. 7). Another common feature is the "depressing" effect of continuous light (LL). Not only was there a more or less rapid decrease of former activity peaks in LL (figs. 5 and 6), but the mean activity reached a very low level (see also fig. 7).

Continuous darkness (DD) could not be observed to have a depressing effect, and activity in DD actually appeared to be greater than the total activity recorded through a period of LD cycles (fig. 6 and 7), at least for the ant populations studied in late autumn and winter. This curious result becomes understandable if we assume that negatively phototactic ants of the intranidal (nurse) group, which are normally prevented by light from leaving the nest (see Kneitz 1964), may visit the running field during the dark period. This effect could not be observed during my field experiments and may thus be a "laboratory artefact" (small dimensions of the laboratory arena compared with the foraging range in natural colonies, high temperature through the dark period). In nature, the dark period is also a period of low or decreasing temperature and temperature may be even more crucial than light for ants of the intranidal group. The dual nature of the population with respect to light reactions makes it especially difficult to draw general conclusions from this type of laboratory experiments. Light may stimulate the activity of part of the population (which may explain the recordings of fig. 5C) while inhibiting the activity of another part. The final result (net increase or decrease in activity) will be determined by the composition of the population (probably mainly its age distribution).

The depressing effect of continuous light was evident with both system a (incandescent light penetrating the milky roof of the drum) and system b (point source Hg lamp or fluorescent tube contrasting sharply with the black ceiling). However, a difference was apparent between the systems; with system a (fig. 5) there was some, though rather slight, evidence of the continuation of the activity rhythm for two or three days at constant light (LL) and temperature, whereas this effect could not be detected at all with system b (Hg lamp, fig. 7). Recordings at constant temperature in continuous darkness (DD) showed fairly large fluctuations in activity, but the peaks were too irregular to permit any conclusions. Definite proof of the true circadian or oscillatory nature of the rhythm (cf. the alternative hour glass principle in Saunders 1976) is thus still lacking. Some of the recordings showed slight evidence of an activity peak at light off (fig. 6 and 7).

4. The relation between activity and relative humidity

A sudden rise in the relative humidity
of the air at 25°C to 26°C, caused by switching on a rapid Defensor air humidifier, very clearly increased the number of ants leaving the nest for foraging. This effect cannot have been due to the vibration of the Defensor, since it was not observed with an empty but vibrating humidifier. The example given in fig. 8A is an extreme case but the same trend could be seen in several repetitions of the experiment, especially with system a. It seems likely that the high temperature within the drum (system a) had an inhibitory effect on the traffic activity; HORSTMANN 1970 and DE BRUYN & KRUKDE BRUIN 1972 consider that the optimal temperature for foraging traffic in F. polyctena is about 22.5°C under field conditions. It appears that the rise in humidity removed the inhibitory effect of high temperature, which suggests that the temperature optimum for foraging is not fixed but may be a function of the relative humidity. An increase in the upper limit of temperature tolerance with increasing relative humidity would be
advantageous, because the temperature optimum for catching insect booty (Buttner 1974) and for running (Wellenstein 1954) is higher than the temperatures reported to prevail when maximum numbers of workers were present on the track. Temperatures considerably above +23°C can hardly be injurious per se, because the "target" temperature within the mound at the beginning of the summer is +27°C to +29°C (Kneitz 1970 and my unpublished measurements in southern Finland). However, outside the nest, high air temperatures are often associated with very low levels of relative humidity (which are clearly deleterious to wood ant foragers, see Kirchner 1964). If the temperature optimum rises with increasing humidity, the maximum benefit can be obtained from hunting at the minimum cost. However, it seems as if the behavioural humidity effect may be a response to a rapidly rising RH level and may decline as soon as the level becomes fixed, having the character of a "startle" reaction. We must, therefore, beware of drawing too far-reaching ecological conclusions. The humidity response could be demonstrated in system b as well (23°C), but the effect was much weaker.

5. Concluding remarks

One conclusion to be drawn from my field experiments is that the nocturnal forager population is largely the same as the diurnal one. Some statistically significant differences suggest that the nocturnal population (especially during cold nights) may represent a selected part of the diurnal population. This may be due to differences in foraging motivation, which is probably correlated with the age of the foragers (cf. Rosengren 1977). No evidence was found in the field of forager groups separated by differences in diel periodicity. The results of my laboratory experiments agreed with this observation: the ants of a protein starved colony reacted to food with clear activity peaks (suggesting recruitment) but showed no indications of a memory of previous feeding times.

Clear indications were found of general diel periodicity of the population with an activity maximum prior to the onset of light (12:12 LD cycle). The periodicity could not be explained as a "startle" reaction (due to the sudden change from light to dark and vice versa) but the influence of the heating effect of the light source was difficult to exclude completely. It was repeatedly observed that continuous light has a "depressive" effect on activity.

The observation that the activity level may be higher in the dark period than in light is probably an "artefact"; under laboratory conditions, young intranidal workers, which are prevented from leaving the nest by the light, may leave it in the dark period. No evidence for this effect was found in the field.

The experiments gave no clear-cut evidence of a rhythm persisting through LL or DD and it is still an open question whether it represents a true circadian (oscillatory) phenomenon or can be attributed to other mechanisms (see Saunders 1976). The number of ants leaving the nest for foraging at a high temperature increased with a rise in the relative humidity of the air.

IV. GENERAL DISCUSSION

A. Nocturnal Orientation

The wood ant has an apposition eye rather similar to that of the bee, with fused and relatively narrow rhabdoms apparently containing small amounts of visual pigments. Thus its eye differs anatomically from that of typical night-
orienting insects. My observation that fidelity to routes does not decrease at all in the night strongly suggests that orientation in the dark period is not visual, because we would expect that diminished visual discrimination would cause at least some decrease in fidelity. Clear evidence that nocturnal orientation is not visual was provided by my previous experiments (ROSENGREN 1971), in which the ants in an open-air arena did not react to a shift in the position of landmarks during the dark period, although the arena was located on top of an open hill, where the light conditions were better than in the present case. Another result of my previous arena experiment also deserves attention in the present context. When an artificial "forest" was shifted by 180°, ants marked with different colours for different feeding directions showed a high degree of "reversed" fidelity during the light period (ROSENGREN 1971:56-57). But, directly after the shift, the distribution of marked ants visiting the feeding vessels in the dark period was random. Nocturnal sampling was repeated 1.5 months later (overcast sky in the middle of October at midnight). Two days earlier the "forest" had been turned by 180°. The foragers by then showed indications of site tenacity, which did not, however, correspond to the shifted position of the landmarks but to the normal (non-reversed) fidelity pattern existing before the shift (ROSENGREN 1971: fig. 26, compare histograms 10 and 22). The results thus suggest that the ants gradually "learned" to use some type of non-visual directing cues during the dark period.

It might be argued that foragers belonging to different routes may occupy different sites within the mound and thus only have to join the scent trail nearest to this site to find their specific route in the night or after the hibernation period. This explanation is disproved by the following observations:

1. Foragers show fidelity to branch routes situated far away from the mound (page 17).

2. The mound and the earthnest under the mound can be destroyed completely by digging up the nest and mixing the nest material. This process usually leads to migration from the old site to one or several new nest sites, in which cases the fidelity of previously marked foragers seems to decrease considerably (ROSENGREN 1971:44). I have, however, observations from a case where complete destruction of the mound and earthnest led to only short migratory unrest (carrying of workers from the destroyed mound and back again) followed by rebuilding of the old mound. The forager population, which had been extensively marked with different spray colours on four routes of the system, showed practically no decrease in fidelity to the routes (my unpublished material).

3. Marking experiments with a seminatural, revolvable "route system" revealed a clear fidelity, although the ants reached the "routes" from the top of the mound (ROSENGREN 1971:49—51).

4. Foragers hibernate in clusters underneath the mound and thus do not stay in the same parts of the nest as during summer.

Possible mechanisms of nocturnal orientation:

1. The possibility that the different wood ant routes in a system may smell differently deserves serious consideration. Wood ants and related species use faecal deposits when laying a scent trail (MOGLICH & HÖLLOBLER 1975, HORSTMANN 1976), and it is thus theoretically possible that forager groups visiting different kinds of food may give distinct scents to different routes, which allow a wood ant forager to recognize its "own" specific route in the dark period as well. One of my previous experiments seemed to exclude scent trails as a cause of fidelity (ROSENGREN 1971:49—51), but in it the ants of a seminatural, revolvable "route system" were fed with the same type of food on all the routes. However, the hypothesis that trails are marked with differently scented faecal matter can hardly explain the stable character of site tena-
city in wood ant foragers. The kind of food found along different routes is likely to change through the season, while the fidelity of foragers to their routes remains practically unaltered during at least two months of the active season (RosENGREN 1971:15 and 33). Current experiments with tracer-marked sugar indicate that forager groups caught on the different routes of a colony exchange food freely between each other when brought together in a laboratory nest (GYLLENBERG, LUNDQVIST & ROSENGREN in preparation). Thus no clear feeding barriers can exist within the nest, although COURTOIS & LECOMTE (1962) found that tracer-marked sugar fed to the ants on a natural route did not spread to the same extent to foragers on other routes. Possible differences between routes in the odour of faecal deposits (e.g. due to differences in the honeydew of different aphid species) are likely to be blurred by the exchange of food.

JAISSON (1975) showed that the characteristics of the pupal cocoons of the colony or species are learnt during a sensitive period in the early life of wood ants, the process resembling imprinting. He was able to demonstrate that this memory lasted for several months in ants isolated from the pupae, and it seems likely that it is at least partially olfactory. The failure of the foragers at the visually disturbed fork area (page 12) to locate or follow the previous roads after the winter indicates that the memory persisting through the hibernation period was neither olfactory nor tactile (stimuli from the cleared road furrow), which accords with my previous observations (ROSEN GREN 1971:79). There is, however, some evidence that auxiliary cues from the road surface may be used in the summer season if visual cues are disturbed (ROSENGREN 1971:43).

2. The desert ant Cataglyphis bicolor is able to use the direction of the wind as an orienting cue during moonless nights (DUELLI 1972), but such a mechanism could not account for the stable fidelity pattern observed in the present case (moreover, the wind can hardly be a reliable cue in a forest).

3. Kinaesthetic or idiothetic cues originating from inner states (e.g. proprioceptive inputs) participate in a variety of orientation mechanisms (for references, see SCHÖNE 1975). The role of kinaesthetic cues in direction finding and homing over longer distances is, however, the subject of some controversy. Kinaesthetic homing has been described in spiders (e.g. GOERNER 1973) and in some vertebrates (DUELLI 1975). JANDER (1957) found no indication of kinaesthetic orientation in the wood ant, but we cannot dismiss the possibility that ants reaching the forking point at trunk route 3 were able to discriminate between the left and the right branch of the fork with the aid of kinaesthetic cues.

4. Geomenotaxis based on gravitational sense organs in the neck has been demonstrated in the wood ant (MARKL 1964). The use of the slope of the terrain as a means of orientation to a gravitational vector would be a possible way of finding the direction in the night. There was indeed a clear difference in slope between trunk 6 and trunk 3, but the terrain at the fork of branches 3a and 3b was practically level.

5. The evidence that nocturnal foraging may require a long "learning" or setting period (see also SNELI 1964) is compatible with the possibility that geomagnetic cues may be used. Being a very weak stimulus, geomagnetism seems to require a period of adaptation (e.g. LINDAUER & MARTIN 1968). There is, however, no real evidence for geomagnetic orientation in the present case.

6. As the ground temperature certainly differs in different parts of the terrain and on different routes, especially at the beginning of the night, the possibility also exists that the ants find their specific routes and feeding grounds with the aid of a "thermic pattern".

It is not yet possible, however, to assess the contribution (if any) of the above mechanisms in the nocturnal orientation
of wood ants, and we may conclude that this problem still remains to be solved.

B. Nocturnal foraging and activity rhythms

As Sudd (1967:77) has pointed out, all the items found in the diets of the more specialized ant feeders are eaten by wood ants, and the development of mechanisms for nocturnal orientation may be seen as a consequence of this omnivorousness. Many prey insects, like the aphid species already mentioned, have their active period in the night, but the hunting of living prey may be seriously limited by poor light conditions. It is known that wood ant foragers in the vicinity of a moving insect detect and locate the prey by visual "kinopsis" (Stäger 1931, Ayre 1963), and the ability of wood ants to locate and capture living prey is clearly reduced if the light intensity drops below 0.1 Lux (Buttner 1974). It should, however, be observed that wood ants may find their prey insects by chance. In my study colonies, most of the booty carried by ants to the mound in late autumn consisted of small frost-killed or cold-immobilized insects, mainly midges, and midges also made up the greater part of the booty carried to the mound during the nights of late August. Moreover in good light conditions there is the risk that some prey insects may react by visual "kinopsis" to the moving predator, and fly away.

It would thus be premature to conclude that insect booty is without importance during the dark period (the main limiting factor is probably temperature rather than light, see Wellenstein 1954). It is quite evident that wood ants continue to visit aphids throughout warm nights.

Thus, it is not very surprising if a non-specialized feeder of this kind develops alternative means for direction finding and homing.

The need to prey on a large number of different insect and other invertebrate species may also explain why foraging rhythms of an endogenous nature seem to be rather weak in wood ants. As pointed out by Reichle (1943) since the different prey insects have different activity periods, strong periodicity in the predator would seriously impair the efficiency of foraging. This disadvantage could be overcome by developing forager groups with differently adjusted circadian clocks, synchronized to the circadian periods of different food items, and such a strategy, based on a circadian memory, has obviously been utilized by the honeybee (see Renner 1958, von Frisch 1967 and Koltermann 1973 for review), but in the present investigation wood ant foragers did not show any circadian memory based on time conditioning.

I have made some preliminary field observations which suggest that wood ant activity is more restricted to the light period at the beginning than at the end of the season. Although nocturnal foraging has been observed at the end of May in wood ant colonies north of the Arctic Circle (Snelli 1964) at this time of the year, the ants of my study colonies regularly return to the mound at sunset. This is partly, but not entirely, an effect of temperature. According to my notes, the sudden return of almost the whole forager population to the mound at sunset occurs at a higher ground temperature than that at which traffic starts in the morning. According to Snelli (1964:41) the very intense activity peak seen in natural wood ant colonies in the morning cannot be correlated with temperature and may thus have an endogenous component. According to my notes, this morning peak is also more sharply defined at the beginning of the active season (spring and early summer) than later. A stronger activity rhythm at the beginning of the season would be adaptive, because in Finland frost is common during the night in spring and early summer. Some conflict may thus exist between the omnivorous foraging strategy (which may favour nocturnal foraging and relaxation of a light-coupled activity rhythm) and the need to avoid being caught by low temperature while outside the mound. It is however very difficult to correlate these field observations with my present laboratory
experiments. The natural activity rhythms probably result from a complicated interaction between light and temperature, acting as both endogenous timegivers and as exogenous (stimulating or inhibiting) factors.

**Summary**

1. In the wood ant *F. polyctena*, nocturnal foraging has been observed under the extremely poor light conditions prevailing in coniferous forests in late summer (no moon and overcast sky).

2. The ability of foragers to locate their own routes was examined by spray marking foragers with different colours for different routes. Spatial discrimination, as evidenced by fidelity to routes did not decrease in the night, even in the case of a fork system situated in level terrain about 14 m from the mound (the foragers on each branch of the fork were marked with different colours).

3. Previous results (Rosengren 1971) show that during the light part of the day route fidelity is mainly due to the memory of a configuration of visual landmarks. In the night, wood ants did not react to landmarks at all. Consequently, the retention of fidelity in the night cannot be attributed to visual orientation. Alternative mechanisms of nocturnal orientation are discussed, e.g. the possibility that wood ants may mark their routes with differently smelling faecal deposits.

4. Colour-marked foragers surviving an uninterrupted hibernation period of 5 months are able to recognize their previous foraging directions after the winter. Timber felling has been observed to have a disturbing effect, which makes it seem likely that the ants remember a configuration of visual landmarks through the hibernation period. The ants were not able to follow a cleared road furrow in early spring if the visual environment had been altered. This indicates that the memory preserved through the hibernation period is neither olfactory nor tactile.

5. Ants foraging in the dark period were marked with other colours than ants foraging in the light. Samples taken from the routes at different times in the natural light-dark cycle revealed no marked difference in composition between the nocturnal and diurnal forager populations. Thus, the same individuals must possess mechanisms for both nocturnal and diurnal orientation.

6. Small mean differences in composition were, however, found between the day and night populations (probably due to temperature rather than light). The most likely explanation is that the nocturnal forager population represented a selected part of the diurnal population, owing to differences in foraging motivation between foragers of different age (see Rosengren 1977).

7. The alternative possibility that a wood ant society possesses differently timed forager groups (e.g. owing to conditioning to the diel periodicity of different foraging objects) was not supported by the results of laboratory tests in which ants leaving the nest for foraging were counted automatically when crossing a beam of infrared light. It was not possible to condition the ants to a certain feeding time, although the population reacted to food with very clear activity peaks, suggesting recruitment.

8. There were, however, clear indications that a light-dark cycle or a temperature cycle (under constant light conditions) may induce or maintain an activity rhythm in the population as a whole. In a light-dark cycle (12:12 LD), the activity rises at the end of the scotophase and may in many cases reach its maximum prior to the onset of light, while in a thermal 12:12 cycle, activity reaches its maximum in the middle of the warm period. This, together with the observation that continuous light had a depressive effect, indicates that light per se cannot be said to be "stimulating" for the population as a whole. It seems probable that some of my laboratory results can be explained by the difference in the reaction to light existing between different groups of the population (negatively and positively phototactic ants).

9. A sudden rise in the relative humidity of the air at 25°C to 26°C very clearly increased the number of ants leaving the nest for foraging (for discussion see p. 23). This phenomenon, detected here by pure accident, has not been described before.

10. The development of mechanisms for both nocturnal and diurnal foraging, the lack of a clear circadian memory (conditioning to a feeding time) and the rather weak development of general periodicity may be seen as components of the foraging strategy of an omnivorous species (for discussion see pp. 24—28).
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