THE ACTIVITY OF SLUGS

I. THE INDUCTION OF ACTIVITY BY CHANGING TEMPERATURES

BY BARBARA H. DAINTON*

Zoological Laboratory, University of Cambridge

(Received 30 March 1953)

Slug activity is associated with conditions of high humidity, and this association has, not unnaturally, led to the hitherto untested assumption that it was in some way induced by the necessarily damp conditions. The experimental examination of this phenomenon, described in this paper, has shown that certain changes in temperature, and not conditions of high humidity, induce activity. Such temperature changes occur in the situations inhabited by slugs on just those occasions when slug activity is observed in the field.

It seems desirable to define what is implied by the term 'activity'. The natural historian, when describing an animal as nocturnally active, means that it can be found at night moving about, presumably in search of food and mate, whilst during the day it is found at rest. Where this behaviour has been investigated in the laboratory (Szymanski, 1920; Gunn, 1940; Bentley, Gunn & Ewer, 1941), activity has usually been taken to mean locomotor activity because it is possible to obtain a measure of this, and it necessarily accompanies the acquisition of food and mate. In the case of slugs, where food and mate may be present in the environment during the day as well as during the night, feeding and mating occur normally only at night and follow the onset of nocturnal locomotor activity. Consequently the investigations have been directed to the study of this aspect of activity, which is regarded as the usual precursor of other activities. Unless otherwise stated, activity means locomotor activity. This usage of the term has the additional advantage of making the work directly comparable with that of other authors on different animals (Szymanski, 1920; Gunn, 1937, 1940; Bentley et al. 1941).

Most of the work has been done on the grey field slug *Agriolimax reticulatus* (incorrectly named *A. agrestis* by some authors), which is a pest of root crops, cereals and garden vegetables in this country. This species was chosen because it was conveniently abundant. Other species have been used occasionally, in which case this is stated in the text.

ATMOSPHERIC HUMIDITY

The activity of slugs was recorded after introduction into chambers maintained at different relative humidities. The method was similar to that used by Gunn (1937) for his investigation of the effect of atmospheric humidity on activity in the woodlouse.

* Present address: 16, North Parade, Leeds, 6.
The humidity chambers, which were in all essentials like those described by Gunn & Kennedy (1936), consisted of round glass dishes fitted with airtight lids. Inside, a false floor was fixed above a solution of sulphuric acid known to be in equilibrium with a certain atmospheric humidity (Wilson, 1921). Cellophane, stretched on an embroidery hoop, was substituted for the original perforated zinc floor because the slugs avoided moving over the zinc. A rim of perforated zinc, fitting inside the hoop, prevented the slugs escaping over the edge. The chamber was left at the temperature of the experiment for at least 2 hr. to ensure that equilibrium between atmosphere and solution had been reached. This was twice the time found by Gunn to be necessary in a similar apparatus. The animals were introduced through a small hole in the lid which was covered when not in use by a greased glass slide.

Slugs were introduced singly into the apparatus, partly because it was felt that, in view of their wet bodies, they would, if introduced in any numbers, materially raise the humidity in the chamber, and partly because it was decided to measure the activity by the extent of the mucous trail left by the slugs instead of by direct observation. This made it possible to perform the experiments in the dark and thus eliminate any possible complication due to illumination. The animal, which was introduced through the hole in the lid, was removed after 30 min., and the trail rendered visible by immersion of the cellophane false floor in a 1% aqueous solution of Grühler's methylene blue for 5-10 min. After washing in water, the sheets of cellophane were dried and the trails examined. Introduction into the chamber is liable to induce a short period of activity due to mechanical irritation

* This method was suggested by Dr D. L. Gunn.
The activity of slugs

which was unavoidable. Since the likelihood of such stimulation was the same at all humidities, it could not have obscured any effect which atmospheric humidity might have had on activity.

Table 1. Types of slug trails made during 30 min. at different relative humidities

<table>
<thead>
<tr>
<th>Group</th>
<th>Temp. (°C)</th>
<th>R.H. (%)</th>
<th>No. of trails of type</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>A</td>
</tr>
<tr>
<td>I</td>
<td>15.5</td>
<td>40</td>
<td>13</td>
</tr>
<tr>
<td></td>
<td></td>
<td>100</td>
<td>2</td>
</tr>
<tr>
<td>II</td>
<td>17.5</td>
<td>40</td>
<td>12</td>
</tr>
<tr>
<td></td>
<td></td>
<td>100</td>
<td>11</td>
</tr>
<tr>
<td>III</td>
<td>24</td>
<td>40</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td></td>
<td>100</td>
<td>13</td>
</tr>
</tbody>
</table>

130 specimens were introduced individually into chambers, half of which were at 40% and half at 100% relative humidity. After 30 min. activity had ceased, which alone showed that if the humidity was affecting the activity, the effect was extremely short-lived and quite unlike the phases of activity observed in the field. After staining, it was found possible to classify the mucous trails into three types termed A, B and C. Type A showed considerable activity, type B activity over a small area and involving much turning, and type C showed little or no activity. Tracings of typical trails are shown in Fig. 1. The number of trails of each type obtained at each humidity was counted. The results are summarized in Table 1. Three groups of experiments were performed in three rooms maintained at different approximately constant temperatures. Examination of the table shows that there was no correlation between type of trail and humidity at 17.5 or 24°C, but at 15.5°C the activity was greater at the low humidity. A similar result can be obtained by measuring the slug trails with a map measurer and treating the distance travelled in the different groups statistically, when a significant increase in activity is only found in the dry air at 15.5°C. This increased activity following introduction into dry air is probably due to the fall in temperature resulting from loss of water by evaporation. Later experiments described in this paper show that a fall in temperature below about 21°C stimulates activity. The actual fall necessary for stimulation varies with different individuals, which may account for the fact that the difference is not more striking, and also for the fact that no such effect was observed in group 2 at 17.5°C. (Group 3 was at 24°C, and, therefore, no such effect should have been observed.) In any case the temperature control and measurement in these experiments was hardly accurate enough, in view of later results, for deductions of this kind to be made. No further investigation was undertaken because it was apparent that this slight response to low atmospheric humidity bears no relation to activity in the field which occurs when the atmosphere is damp. It was clear from the above experiments that high atmospheric humidity does not stimulate activity. A similar result was obtained using *Arion subfuscus* instead of *Agriolimax reticulatus.*
TEMPERATURE

Slug activity is observed in the field at night and following showers or mist by day. These are both occasions, though not necessarily the only occasions, on which the temperature of the earth's surface is falling, which suggested that activity might be related to a low or falling temperature. Since slug activity occurs at all seasons (except in frosty conditions), during which the range of temperature fluctuations varies considerably, it seemed unlikely that slug activity was related to any particular temperature; but it was considered desirable, nevertheless, to start by measuring the activity of slugs at different constant temperatures because such information would clearly facilitate the interpretation of the results obtained at fluctuating temperatures.

Crozier & Pilz (1924) showed that the speed of locomotion in *Agriolimax campestris* varied directly with the temperature. This effect must be clearly distinguished from the responses described below in which inactive animals are stimulated, and the amount rather than the speed of activity is measured. In all experiments, the effect described by Crozier & Pilz was confirmed for *A. reticulatus*, the animals moving faster at high than at low temperatures.

The behaviour of the slugs was studied under conditions of constant or fluctuating temperature in an apparatus which ensured damp conditions and a constant light intensity of 40 foot-candles. Experiments described in a later paper showed that the light was not affecting the results provided 1 hr. was allowed for light adaptation after introduction into the apparatus. The use of light enabled the activity to be recorded by observation. This meant that a far greater number of animals could be employed than was possible if the activity of individual animals was recorded in the dark, using the slime-trail technique described above, or some form of mechanical recording used in a few later experiments described below.

The slugs were placed in Petri dishes which were immersed in a water-bath fitted with heating and cooling systems and, when required, with a thermostat which maintained the temperature constant to within ±1° C. When fluctuating temperatures were in use a record was kept by making 5 min. readings from a thermometer immersed in the bath.

Three slugs were placed in each Petri dish (it was found that they did not interfere with one another), the larger side of which was lined with filter paper. This was done by placing a sheet of filter paper of slightly greater diameter than the Petri dish, over the lid which was placed upside down on a table. The slugs were placed on the filter paper and the bottom of the dish pressed into position. The Petri dishes were secured upside down to a wire frame by rubber bands, and the frame fixed 6–8 cm. below the surface of the water. The water was stirred by a stream of air bubbles which was turned off for each observation, so that the animals were readily visible through the water. The filter papers became damp because their edges protruded from the Petri dishes, whilst the dishes retained their air because their two halves were firmly held together by rubber bands. The air inside the dishes was thus conveniently maintained at 100% relative humidity. The fact that
The activity of slugs

The animals were in good condition after periods in the apparatus twice as long as the duration of any experiment was taken as adequate evidence that limitation of air supply was not affecting the results.

At each observation the animals were found to be in one of three conditions: either actually moving, resting with one or more tentacles extended, or resting with tentacles retracted. The three conditions were named 'active', 'extended' and 'inactive' respectively, and the following criteria adopted for their definition. In an 'active' animal the tip of the tail could be observed to be moving; in an extended animal one or both of the dorsal pair of tentacles were visible, the animal either holding its head still or waving it from side to side; whilst in an 'inactive' animal the tentacles were withdrawn. At each observation the number of animals in each condition was recorded, thus giving an estimate of their activity. The number of animals either active or extended, and the number of active animals, were expressed as a percentage of the total numbers. In experiments involving fluctuating temperatures each reading was plotted in the form of a histogram so as to represent the condition of the animals during 5 min., the reading being taken in the middle of this period. It was found that an 'extended' animal was nearly always either about to move or settling down after a period of locomotion. Animals in such a condition would not produce either a slime trail or a mechanical record, and thus only the record of 'active' animals is strictly comparable with results obtained by the other methods of recording activity mentioned above. In the majority of experiments an apparatus holding ten dishes, and therefore thirty slugs, was used in which it took about 30 sec. to note the condition of the animals. In some experiments at constant temperature a larger apparatus holding seventy-two animals was used and the observations then took over a minute. The smaller apparatus was found to be more satisfactory for fluctuating temperatures because it was easier to control the temperature of the smaller volume of water, and because, when heating or cooling was in progress, it was clearly desirable to turn off the stream of bubbles used for stirring for as short a time as possible when observations were made.

After introduction into the apparatus the animals, even if previously light adapted, showed an initial burst of activity due to mechanical irritation. This was increased if the temperature change resulting from introduction was such as to stimulate activity. No observations were included in the results until the activity had fallen to a constant value. One hour was found to be sufficient for this in all except a very few experiments in which a further half-hour was allowed. In the majority of experiments readings were made at 5 min. intervals, though in some of the longer experiments using constant temperatures they were made at 15 or 60 min. intervals. The duration of the experiments varied from 30 min. to 4½ hr. The average reading at constant temperatures was found to be unaffected by the duration of the experiment, and in later experiments, including those involving fluctuating temperatures, 5 min. readings were made for periods of 1–3 hr.

Preliminary experiments indicated that the activity of the animals was similar at all constant temperatures, though, when they did move, they did so much more quickly at higher temperatures. The results involving changing temperature were
much more striking. Activity rose sharply immediately the temperature began to fall below about 21°C, and a similar increase occurred when it was rising above this value. The increase subsided as soon as the temperature was maintained constant at any value. Temperature changes in the reverse direction had no such immediately obvious effect; as at constant temperature the activity was low when the temperature was rising below 21°C or falling above this value.

**CONSTANT TEMPERATURE**

Temperatures were employed varying between 10 and 30°C. Owing to experimental difficulties, records were not made at lower temperatures, and in view of the results obtained between 10 and 30°C, it seemed unnecessary to make further attempts to obtain satisfactorily constant lower temperatures.

Table 2. *The activity of Agriolimax reticulatus at different constant temperatures*

<table>
<thead>
<tr>
<th>Temp. (°C)</th>
<th>% active animals</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
</tr>
<tr>
<td>10.5</td>
<td>1.5</td>
</tr>
<tr>
<td>15.3</td>
<td>2</td>
</tr>
<tr>
<td>20</td>
<td>1</td>
</tr>
<tr>
<td>20</td>
<td>2</td>
</tr>
<tr>
<td>24</td>
<td>2</td>
</tr>
<tr>
<td>24</td>
<td>2</td>
</tr>
<tr>
<td>30</td>
<td>1</td>
</tr>
</tbody>
</table>

A series of typical experiments are summarized in Table 2, which gives the average percentage of 'active' animals recorded at different temperatures, together with the variation observed in each instance. There is very little activity at all temperatures between 10 and 30°C and at 20°C, where the activity of three batches of animals was recorded, the range in activity is comparable with that observed in the whole series. It may thus be concluded that high activity is not associated with any particular temperature between 10 and 30°C. For reasons already stated, no extensive experiments were made below 10°C, but preliminary observations indicated that this conclusion also holds for temperatures between 4 and 10°C, and therefore throughout the whole viable range.

**THE STIMULATION OF ACTIVITY BY FALLING TEMPERATURE**

Since it was found impossible to repeat exactly the temperature variation obtained in any one experiment, it was necessary to plot the results of each experiment separately, and the histograms recorded here may be regarded as those made from typical experiments. In a few instances, particularly at very low temperatures, only one experiment was performed. This result can, however, be taken as conclusive, since it involved thirty animals, and in other experiments significant differences were never observed between different batches of animals.
The activity of slugs

Below about 21°C, slugs respond to falling temperature by a very marked increase in activity. The response is shown, without exception, by every animal, and to surprisingly low rates of fall. Fig. 2 illustrates a series of six experiments each involving thirty animals. The rate of fall varied from 0.1 to 19°C per hour, and in each case it was accompanied by a marked rise in activity. It should be emphasized that in each experiment all thirty animals responded, but since they did so at different time intervals, and since, once established, slug activity is intermittent, 100% activity was never recorded. At all rates of fall, the increased activity, once initiated, was maintained as long as the temperature was falling. If the rate of fall was slow, this activity fell off immediately the temperature was maintained constant. If the rate was fast the response was maintained for some time after the temperature became constant. The fall need not be sustained to induce activity. In one experiment an increase in activity was observed following a fall of 0.3°C in 5 min. If the fall in temperature was sustained all animals, without exception, responded.

Examination of records kept at, or near, the earth’s surface (see The Observatories Year Book, 1939; Keen & Russell, 1921; Conway, 1936*) show that, in the slug's

* Dr V. M. Conway kindly supplied me with her original complete records.
habitat at all seasons, the temperature falls below 21°C only in the late afternoon, evening and early night, or following showers earlier in the day; in fact, on just those occasions when slug activity is observed in the field. The only qualification it is necessary to make is that the temperature begins to fall earlier in the afternoon than the slug activity appears. Examination of the detailed records of diurnal temperature variation among and beneath vegetation made by Conway (1936), reveal the existence of marked temperature gradients within the vegetation when the temperature first begins to fall. A study of the behaviour of slugs in such a gradient, which is described later in this paper, shows that, once active they would move up the gradient, thus counteracting the fall, and immediately coming to rest again. Only when, as shown by Conway's records, the whole of the vegetation becomes uniformly cool does slug activity of any duration appear. The time at which this occurs in Conway's records is in the evening and agrees remarkably well with the time of the appearance of slug activity in the field.

Fig. 3 illustrates the type of field observation which can be made in warm weather, in which marked activity was noted following a shower of rain. There is no such delay in the appearance of activity here, because the cool rain lowers the temperature of all the vegetation rapidly.

In the experiments described above the temperature never fell below 10°C. Lower temperatures do, of course, frequently occur in the slug's habitat, and exploratory experiments indicated that falling temperatures over this lower range also stimulated activity. Experimental work was not easy at these low temperatures, but a few experiments, each involving thirty animals, were made. Similar results...
The activity of slugs

to those described above were obtained. The burst of activity which followed the fall in temperature took longer to make its appearance than at higher temperatures. Observation of the animals suggested that this was, at any rate in large measure, due to the extreme slowness of the animals at these low temperatures. It took noticeably longer for the tentacles to be extended, and when movement did occur it was much slower.

![Graph of temperature variation and activity](image)

Fig. 4. The effect of falling temperature above 21°C on activity.
Rates of fall: in A = 4.2°C per hour; in B = 12.8°C per hour.

Above 21°C, fall in temperature has no such effect (Fig. 4), and may even reduce activity below the level observed at constant temperature.

THE EFFECT OF RISING TEMPERATURE ON ACTIVITY

Above about 21°C, rising temperature is accompanied by increased activity which falls off immediately the temperature is maintained constant as shown in Fig. 5. The response is not nearly so sensitive as that to falling temperature below 21°C, and no response was obtained to a rise of 1.7°C per hour. Sudden increases in temperature over this range do occasionally occur in the slug’s habitat. A stone or log beneath which slugs have come to rest at dawn may be suddenly exposed to the midday sun, causing the temperature to rise sharply, and in hot weather to reach proportions well above those which are lethal to slugs (30–32°C; see Carrick, 1942). However, before such temperatures are reached, the slugs respond to the rise by becoming active, and once active they move, as described later in this paper, down the temperature gradient and thus towards some cooler, less exposed resting place.

Below 21°C, rising temperature does not stimulate activity. On the contrary, under such conditions activity is extremely low and in some experiments was
practically nil (see Fig. 6). Such a rise in temperature occurs in the slug's habitat after dawn and throughout the early part of the day, a time when slug activity is never observed in the field except in showery weather, when the rain cooled in the upper atmosphere produces a temporary fall which stimulates a short period of activity.
THE ACTIVITY OF SLUGS

THE EFFECT OF CONSTANT TEMPERATURE ON THE DIURNAL RHYTHM OF ACTIVITY AND REST

If slug activity is controlled solely by the temperature changes in the environment, the diurnal rhythm of activity and rest should disappear when the animals are kept at constant temperature. In order to obtain continuous records over several days, a mechanical form of recording the activity of individual slugs was employed. Szymanski (1920) coined the word 'aktograph' for a number of pieces of apparatus which he designed for this purpose, and one of these was adapted by Gunn & Kennedy (1936) and later by Gunn (1940). A similar, but modified aktograph was constructed and placed in a room maintained at 18° C. Records of the activity of individual slugs were kept over periods of several days.

The aktograph was essentially similar to that described by Gunn (1940). The box itself was, however, only half the size of the one in Gunn’s aktograph, being 10 cm. long, 5 cm. wide and 3 cm. deep. The reduction in size was made because the slugs were not only smaller, but also much slower in motion than the cockroaches used in Gunn’s experiments. It was possible for the slug to move transversely without tipping the box, but this was never observed to happen because slugs do not move in straight lines. Moreover, since the aim of the experiment was to determine when locomotory activity occurred, and not the distance traversed, the possibility that a small portion of the movement was unrecorded could be ignored. The box was made of tin and lined with paraffin wax to prevent rusting. The box in Gunn’s aktograph was fitted with a false floor beneath which humidity-controlling solutions were disposed. No false floor was fitted in these experiments, but the box was lined with a double layer of thick wet filter paper which provided a damp surface for locomotion, as well as ensuring maximum humidity in the box when it was closed with a glass lid, which slid into position beneath two flanges. The lid was also lined with a double layer of wet filter paper cut slightly wider than the lid so that it projected at one side. This exposed filter paper was damped when necessary (usually daily) with water kept at the same temperature, and dropped on to it from a pipette. This could be done during a recording and caused only a very slight movement of the lever which is recognizable in the recording and can, therefore, be eliminated from the result. In this way, the slug inside the box was kept damp without upsetting the record. The base of the writing lever was made of tin and was soldered to one end of the box and an adjustable counterpoise to the other end. A long flexible celluloid lever was attached to the tin base, and this was sharpened to a point which recorded on a smoked drum. This type of lever was adopted after experiments with many other types, because it was flexible horizontally and could thus be placed firmly on the smoked drum, without greatly increasing the resistance between it and the paper, and thus reducing the sensitivity of the recording. Moreover, it was not flexible vertically and therefore gave a true record of the vertical movement of the box.

Freshly collected animals were introduced into the box one at a time, and their activity recorded under known temperature conditions for periods up to a week in
duration. All records were made in continuous darkness, the glass lid of the aktograph being covered with black paper. When a constant temperature was required the aktograph was placed in a constant-temperature room at 18°C; when a diurnally varying temperature was required it was placed in an unheated room, or beneath a shelter on the roof of the laboratory or on the grass outside. A thermograph was placed adjacent to the aktograph so that the temperature changes throughout the experiment were recorded.

Under conditions of diurnally varying temperature, the animals showed a period of activity which commenced soon after the temperature began to fall below 21°C, and terminated soon after it began to rise. When maintained at constant temperature a diurnal rhythm of activity and rest persisted for several days, but the activity was dispersed over a longer period each day until, after 4 or 5 days at constant temperature, it was more or less evenly distributed throughout the 24 hr. A record of this type is shown in Fig. 7.

If, after such a period at constant temperature, the slug is reintroduced to a diurnally varying temperature, the diurnal rhythm of activity and rest is immediately re-established, the activity occurring when the temperature is falling below 21°C. Fig. 8 shows a record made in this way.

These results are very similar to those obtained by Gunn (1940) in similar experiments on the light-controlled diurnal rhythm of activity and rest in the cockroach. In Pithus, Bentley et al. (1941) found a similar diurnal rhythm but controlled in this case by light and temperature.

The persistence of the rhythm in an imperfect form at constant temperature is of interest, although such conditions never occur in the slug’s habitat. It is necessary to postulate some form of internal stimulation in order to explain it. This internal factor may play some part in the appearance of regular nocturnal activity, but it cannot be maintained without regular diurnal temperature changes. Moreover, the rhythm gradually deteriorates and even shows some falling off during the first 24 hr. It seems, therefore, that the active phase is initiated in nature by the falling
The activity of slugs

177
temperature and terminated when the temperature begins to rise, as shown to occur experimentally. The persistence of an imperfect rhythm at constant temperature is largely of interest in view of any light it may throw on the nervous mechanisms involved.

![Graph showing activity of slugs](image)

**Fig. 8.** Recording of the activity of one slug reintroduced into a diurnally varying temperature after 5 days at constant temperature. The thermograph recording the temperature is placed beside the apparatus, and is not near the slug, and this may account for the fact that activity does not appear to start immediately the temperature begins to fall. The cessation of activity while the temperature is still falling is probably due to loss of water in mucus production. When this was regained the temperature was constant and activity recommenced as soon as it began to fall again, and ceased once it began to rise.

**TEMPERATURE PREFERENCE**

It has been shown that temperature change is all important in determining activity in slugs. The slug's habitat is, moreover, situated near the earth's surface, a region of maximum temperature change and therefore of marked and changing temperature gradients. Indications of the extent of this gradient and its variation at different times of the day and at different seasons can be seen by examining the records kept on open ground at Rothamsted (Keen & Russell, 1921) and beneath vegetation at Wicken (Conway, 1936). The position in which slugs come to rest in this gradient and their behaviour towards it when active, will clearly affect the temperature changes to which they will become subjected. An investigation was, therefore, made into the behaviour of slugs in a linear temperature gradient.

The apparatus used to obtain a suitable temperature gradient between 0 and 30°C is shown in Fig. 9. A copper trough (t) was made from a single sheet of copper just under 1 mm. thickness and 58 cm. long. The trough was roofed in copper for 5 cm. at each end, and each end of the roofed portion was closed with copper sheeting. This left a trough, between the two covered ends, measuring $48 \times 2.5 \times 2.5$ cm. The sides and floor of this trough, and of the enclosed 5 cm. at each end, were one continuous sheet of copper, thus ensuring efficient conduction. The edges were
bent over to form a flat flange on which a glass lid (gl.) rested. Flexible metal tubing was wound seven times round each end, and was in contact with the copper. Hot water was circulated through one end and cold water through the other. In some experiments ice was packed round the cold end. The trough was placed in a wooden box packed with cotton-wool and was covered with cotton-wool during the experiments. It was illuminated by a lamp, fitted with a water filter to absorb heat rays. The resulting temperature gradient in the trough varied slightly from experiment to experiment, depending on the exact temperature of the water used, but it usually covered the range 1–30° C, although in some it did not go below 5° C. The gradient was determined by seven thermometers which passed through holes in the glass lid and which were placed so that their bulbs were just in contact with the surface over which the slugs were moving. The holes for the thermometers were 7.5 cm. apart, the end ones being 1.5 cm. from the ends of the trough. The trough was lined with graph paper on which the scales were made readily visible with Indian ink. In order to avoid the defect in such an apparatus pointed out by Gunn (1934) by which a gradient exists between the bottom and top of the apparatus as well as along it, the animals were kept within 1 cm. of the bottom by enclosing them in a ‘sausage skin’ of cellophane kept distended by flattened wire loops and a strip of cardboard. They were thus largely restricted in movement to a horizontal plane. The cellophane was wet to allow the animals a damp surface for locomotion.

Twenty slugs were distributed evenly along the length of the cellophane trap and the whole dropped into the copper trough. The lid was placed on the trough and the thermometers passing through it adjusted so that they were in contact with the cellophane. No difference in temperature was detected between the top and bottom of the cellophane trap. The slugs could, and did, move freely up and down the gradient, but they could not escape from the trap and so could not move towards the lid of the trough. After 1 hr. the thermometers were read, the cotton-wool and lid removed, and the position of the head of each animal on the scale lining the trough was noted. The temperature gradient was plotted for each experiment, and from this was read the temperature at the position on the scale at which each animal was situated.

Fig. 9. Apparatus used to obtain a temperature gradient between 0 and 30° C.

- t. copper trough; gl. glass lid; ct. cellophane tube.
The activity of slugs

Two hundred animals were used in ten experiments, five of which were performed by day and five by night. In the daytime experiments activity was stimulated by introduction, but at the end of the experiment practically all the animals were at rest. In the night experiments they were sometimes still moving. Moreover, in the night experiments it was very hard to get an even distribution of the animals along the gradient at the start of the experiment because of the high initial activity.

The results are summarized in Fig. 10, in which the number of animals recorded at each temperature is plotted in the form of a histogram. A is plotted from the results obtained by day when the animals were in their inactive phase, and B from those obtained at night when the animals were active. In C the two sets of results are plotted together. Both by day and night the animals aggregate about a preferred temperature of 17–18° C. The temperature appears to be slightly higher by day. There is an additional small aggregation below 8° C. The reason for this was ascertained in some further experiments in which the animals were observed through
the glass roof after introduction into the apparatus. Animals introduced at very low
temperatures were seen to start moving up the temperature gradient only to be
 overtaken by cold torpor.

These results involving a preferred temperature resemble those obtained by other
authors on different animals, and in particular those obtained by Gunn & Walshe
(1942) on *Ptinus tectus*, where there is also a tendency to an additional minor
aggregation at very low temperatures due to cold torpor.

The movement of slugs in a temperature gradient towards a preferred tempera-
ture of 17-18° C. has a number of important results which affect their behaviour
in the field.

In summer the temperature begins to fall below 21° C. some hours before
activity is observed in the field. Examination of the records kept by Keen & Russell
(1921) and Conway (1936) show that in summer, as would be expected, very con-
siderable temperature gradients develop during the day near the earth's surface.
Should an animal be subjected to a fall in temperature which stimulates activity,
the resulting movement up the temperature gradient towards 17-18° C. (and even
in very warm weather the records referred to above show that the surface of the
earth where the slugs are resting does not rise above this value) would result in the
slugs being subjected to a rise in temperature which, it has been shown, immediately
terminates the activity. Only in the evening, when the temperature of the vegetation
falls to the same value as the surface of the earth (see Conway, 1936) and there are,
for a short period, no such temperature gradients, will the activity really start and
the animals move out from their resting places. They are then found moving about
on the surface of the vegetation. Before dawn the temperature of this vegetation
may, on a clear night, fall below that of the surface of the earth beneath, in which
case activity will be terminated by the movement of the animals towards the warmer
earth, but since the temperature of the earth's surface is also falling the termination
will only be temporary, and the activity will only finally cease at dawn when the
temperature of earth and vegetation both begin to rise. At this moment the earth's
surface is warmer than the vegetation and therefore the slugs come to rest near the
surface, which is where they can be found during the day.

The behaviour of slugs in a temperature gradient is also very important to their
survival in cold weather. Carrick (1942) has shown that, while slugs can survive
exposure to one or two degrees of frost, oviposition and normal development do not
occur below 5° C. In winter when the soil is warmer than the surface debris the
slugs will penetrate cracks in the soil moving towards their preferred temperature,
and thus avoid heavy frosts. In summer the gradient is in the opposite direction
and the slugs will be led to the surface vegetation and its food supply.

The effect of vegetation will also produce horizontal gradients between open
ground and that with a plant cover. In very hot and very cold weather the slugs
will frequent the less exposed ground beneath vegetation by moving towards their
preferred temperature.
Howes & Wells (1934b) noted weight fluctuations due to variations in water content in individual slugs (*Arion ater* and *Limax flavus*), kept under certain laboratory conditions, which are similar to those found in snails (Howes & Wells, 1934a). The fluctuations occurred over periods of several days, and, in snails, low weight was correlated with an inactive condition (either withdrawn or aestivating), but no such correlation was made in slugs. Wells (1944) concludes, as a result of further experiments on snails, that high water content is not the immediate cause of activity which is due rather to some form of sensory stimulation, the water content simply determining whether response to such stimulation is possible. The present author has reached a similar conclusion with regard to activity in slugs. Animals of high water content show no more evidence of spontaneous activity than animals of low water content.

Slugs may lose water either by evaporation or by mucus production. Experiments were performed to determine the magnitude of the loss in each case and its effect on activity following stimulation.

The rate of loss by evaporation from the slug’s body into an atmosphere of 45% relative humidity was found to vary between 3 and 5% of the original weight of the animal per hour, depending on the size of the slug (and therefore on the surface area/volume ratio). It was similar in all species if size was taken into consideration. The species observed included *Arion ater*, *A. subfuscus*, *Limax maximus*, *Milax sowerbyi*, and *Agriolimax reticulatus*. The rates were measured in a wind tunnel (Ramsay, 1935) in moving air (347 m. per hour). Similar values were obtained in still air provided the vessel in which the slugs were dehydrated was not too small.

Experiments were performed to find whether such loss of water by evaporation resulted in reduced activity following further stimulation. Activity was induced by placing the slugs on their backs which resulted in a righting reaction and locomotion. A loss by evaporation of 4% of the original weight of the animal had no effect on the amount of activity following this stimulation, but a loss of 17% had a very marked effect. Loss of water by evaporation to this extent must rarely happen in nature, for slug activity occurs under damp conditions, and it would take between 3 and 5 hr. to produce such a loss by exposure to air of 45% relative humidity, and considerably longer in damper air. Moreover, as is shown later, slugs can absorb water from damp surfaces over which they may move, and thus further reduce the rate of loss.

Slugs produce abundant mucus during locomotion. Slug mucus contains 98% water by weight. Kunkel (1916) concluded, from a study of mucus produced by slugs in varying stages of dehydration, that the glands produce a secretion which swells rapidly on contact with water, and that in normal animals of high water content the bulk of the swelling is done by absorption of water at the expense of the animal. Mucus production, therefore, involves considerable loss of water by the animal. The present author found that specimens of *A. reticulatus* lost about
17% of their original weight through mucus production during 40 min. locomotion in a saturated atmosphere, and thus over damp surfaces.

Experiments were performed to find whether this loss of mucus during locomotion results in reduced activity in response to further stimulation. The same form of mechanical stimulation was used as in the experiments involving loss of water by evaporation. After a loss in weight of 17% due to mucus production, the slugs (A. reticulatus) were left resting for 20 min. and then stimulated again. This time activity was considerably reduced and the loss in weight during 40 min. was only 6% (of the original weight). A further repeat resulted in a loss of about 2%, and the activity was practically nil in spite of strong stimulation. The intervening periods of rest were included to allow the animals time for sensory adaptation or to recover from muscular fatigue. They were kept in a dry atmosphere during this time in order to prevent absorption of water from damp surfaces. It was known from the earlier experiments that the water thus lost by evaporation would be of a very small order compared with that lost in mucus production. Similar reductions in activity were obtained for Limax maximus and Arion ater following mucus production caused by stroking the animal with the handle of a scalpel. The fact that a similar reduction in activity was produced by an equivalent loss of water through evaporation suggested that the reduced activity following mucus production was due to loss of water and not loss of mucoprotein. During later investigations loss of mucus above a certain value, or of an equivalent amount of water by evaporation, was found to reduce activity in response to forms of stimulation other than the mechanical one used in these experiments.

Experiments were also performed to determine whether water loss could be made good by adsorption of water from damp surfaces, and whether, with increased water content, the ability to become active in response to suitable stimulation was regained. In all the three species investigated (see above), the animals survived a loss of 50% of their original weight and recovered their full activity, and most of their weight after some hours on damp filter paper or soil. A little weight was often lost permanently through the production of faeces.

Slugs were never observed to drink water and, though some must regularly be taken in with their food, it was clear that the bulk of this water absorption must occur through the body surface. The following experiment showed that this was possible. A specimen of Limax maximus weighing 9.1 g. was stimulated to produce mucus by stroking with the handle of a scalpel until it weighed only 6.6 g., after which it was in a very inactive condition. It was then suspended in a beaker of water by a thread passing through its sole, in such a position that the mouth, pneumatophore and anus were all above water-level. This meant that any gain in weight which resulted was due to absorption of water through the surface of the skin. The slug was weighed at intervals by suspending it from one arm of a balance, and it was found that practically the whole weight was regained within 2 hr. The animal was then in a very active condition. When a similarly treated animal was kept on damp soil it showed a similar recovery.

It is clear from the above experiments that water loss by evaporation must only
The activity of slugs

very rarely reach proportions great enough to limit activity, whereas water loss by mucus production must frequently reach such proportions. Such water loss can, given time, be made good by absorption of water through the skin quite apart from any the animal may gain with its food. As previously mentioned, in experiments involving a number of slugs 100% activity was never recorded, even though all animals responded to the stimulus applied by becoming active; it was noted that, during a phase of activity the slug is not continuously active but pauses frequently, coming to rest and withdrawing its tentacles. It seems possible that such period of rest may occur when the water content has fallen due to mucus production, and activity does not recommence, even in the presence of the original stimulus, until the water content has risen again. The lengths of the periods of activity which occur in response to continuous stimulation will, of course, depend on the rates of mucus production and water absorption, and these will be influenced by the nature and dampness of the surface over which the slug is moving. As would be expected, the period of activity is reduced when the animal is moving over a dry surface. It should be emphasized that high water content does not, in itself, result in activity, but only enables the animal to respond to some other stimulus. This result is in agreement with the conclusion reached by Wells in the case of the snail *Helix pomatia* (Wells, 1944).

**DISCUSSION**

Slugs are normally active at night, but daytime activity can sometimes be observed in damp or showery weather. Since atmospheric humidity rises sharply at nightfall, and since daytime activity occurs only under damp conditions, it seemed at first sight reasonable to suppose that the activity of slugs was a response to high humidity. Experiments proved that no such response existed, and this result was confirmed by field observations which showed that while daytime activity occurs only in damp weather, it does not occur in all damp weather. Moreover, since slugs are found at rest in damp situations beneath vegetation and stones, it is hardly possible that they could be aware of humidity changes occurring in the air above, and it is also difficult to envisage, on such a wet skinned animal, any form of hygroreceptor, which could register anything but saturation. Slugs are, however, very sensitive to certain changes in temperature, and, since they live near the surface of the earth, they are subject regularly to a marked variation in this respect. They respond readily to a fall in temperature below 21°C. by becoming active, and such a fall is experienced in the evening and following showers and mist by day, in fact on just those occasions when slug activity occurs in the field. Rising temperature below 21°C. induces inactivity, and such conditions occur during the day when the slugs are found at rest beneath stones and vegetation. Thus the regular diurnal rhythm of activity and rest follows the diurnal rhythm of temperature change. Minor fluctuations may be superimposed on this diurnal temperature rhythm by precipitation of rain cooled in the upper atmosphere, and, as this results in a short fall in temperature below 21°C., the slugs respond by a short burst of activity which is terminated when the temperature begins to rise again. On continuously rainy days, when the tempera-
ture rises steadily throughout the day, no such activity occurs, though clearly
conditions with respect to dampness are ideal for such activity.

Activity is also stimulated by a rise in temperature above 21° C. This response is
not nearly so sensitive as that to falling temperature below 21° C., in which case a
response was noted to a fall of 0.1° C. per hour. It does, however, enable the
animals to escape a rapidly rising temperature which might be caused by the
midday sun, and which might soon reach lethal proportions. Once active the
animal will move down the temperature gradient towards some cooler resting
place, and the resulting fall in temperature will soon bring it to rest, because
falling temperature above 21° C., like rising temperature below 21° C., induces
inactivity.

In a temperature gradient active slugs aggregate about a preferred temperature
of 17–18° C. This behaviour has been shown to account for the exact time at which
prolonged slug activity appears in the field. In the early afternoon when the
temperature of the earth’s surface first begins to fall no slug activity is apparent
because, at this time, very considerable temperature gradients exist between the
vegetation and the earth’s surface, and any movement up such a gradient would
immediately counterbalance the fall in temperature which had induced the activity,
and bring the slug to rest. Only in the evening when the temperature of the vegeta-
tion falls to the same value as the surface of the earth, will prolonged activity begin.
Behaviour in such a gradient also accounts for the fact that, once stimulated to
activity by a rise in temperature above 21° C., the slugs move away from the region
of high temperature. It also accounts, at any rate in large measure, for the fact that
when slug activity subsides, the slugs come to rest beneath surface debris or
vegetation and not in the open, and thereby exposed to possible desiccation or to
bird predators. This response may, as described in a later paper, be augmented by
a response to air movements.

Conditions of constant temperature are not experienced in the field except in
heavy frost when slug activity does not occur, the slugs having retreated to warmer
crevices in the soil. When maintained in the laboratory under conditions of con-
stant temperature slugs show a persistent but immediately and steadily deteriorating
diurnal rhythm of activity and rest. The persistence of such a rhythm is similar to
that found by Gunn (1940) in the cockroach, and Bentley et al. (1941) in Ptinus
tactus. When the activity of large numbers of slugs was measured at different constant
temperatures it was found to be low in value and similar at all temperatures. The
fact that slug activity is not associated with any particular temperature agrees with
the field observation that it is observed at all seasons. The low level of activity
found in the laboratory at constant temperature may be due to the small fluctuations
necessarily inherent in any ‘constant’ temperature maintained by a thermostat.
As already pointed out, slugs are extremely sensitive to certain types of temperature
change. Or, it may be due to the initial spreading of the period of activity in the
inherent rhythm described above. Such conditions are not experienced in the field
where the diurnal rhythm of activity and rest is maintained by the diurnal rhythm
of rising and falling temperature.
The activity of slugs

Humidity, while not responsible for the appearance of activity, may influence the amount of such activity once it occurs by determining the water content of the body. Animals of high water content are not spontaneously more active than animals of low water content, but they respond more readily to stimuli, such as mechanical irritation or certain temperature changes, which induce activity. This result is in agreement with a similar conclusion reached by Wells (1944) for the snail. Moreover, the water lost in mucus production during locomotion may temporarily reduce the water content of the body to such a level that activity ceases even in the presence of a suitable stimulus. In damp surroundings such loss is made good by absorption through the skin, and if the stimulus is still present when this reaches the necessary proportions, activity is resumed. Such an explanation may account for the observed intermittent nature of slug activity in damp conditions and under constant stimulation, both in field and laboratory.

Difficulties arise when an attempt is made to describe the temperature responses of slugs using the terminology evolved by Fraenkel & Gunn. As already stated, Crozier & Pilz (1923-4) showed that the speed of locomotion in Agriolimax campestris varies directly with the temperature, and this effect has been confirmed by the author for other species. Using Fraenkel & Gunn's terminology (see Fraenkel & Gunn, 1940) this is clearly a case of orthokinesis in response to temperature. If we consider the other form of orthokinesis defined by these authors, namely, the occurrence rather than the speed of locomotion, then it has been shown that there is no orthokinesis, for the activity of slugs as measured by the percentage of animals found active at any one instant is the same at all temperatures.

It has, however, been shown that rising temperature above 21° C. stimulates activity. This can also be described as an increase in activity to high and low temperatures which shows very quick adaptation. Thus it could be said that high and low temperatures result in an orthokinesis which shows almost immediate adaptation, provided the temperature is reached by the correct rise or fall, and provided this orthokinesis is distinguished from that described by Crozier & Pilz. The terminology is, however, by this time, becoming so cumbrous that it is well nigh useless. It is possible to simplify its use by making a certain change of temperature; instead of temperature, the stimulus: and thus stating that rising temperature above 21° C. or falling temperature below this value stimulates an orthokinesis. This is possibly the most satisfactory solution if this terminology must be used, but it does not distinguish this orthokinesis from that involving speed of locomotion, nor does it take any account of the effect of temperature changes in the reverse direction. In fact, an attempt to apply this terminology to these responses is certainly not satisfactory, and it is a great deal simpler and clearer to describe the responses in simple language as follows. Activity at all constant temperatures is similar and low. Rising temperature above and falling temperature below 21° C. stimulate activity. Temperature changes in the reverse direction have a depressing effect on activity and may even reduce it below the low value observed at constant temperature.
1. The effect of moisture and temperature on the activity of slugs has been investigated in the laboratory and in the field.

2. Atmospheric moisture has no direct effect on activity, though it may limit the duration of activity by influencing the water content of the body. Animals of high water content are not inherently active but they respond more readily to other stimuli than animals of low water content.

3. Between 4 and 20° C. activity is induced by falling temperatures and suppressed by rising temperatures. Temperature changes as slight as 0.1° C. per hour are perceived.

4. The daily rhythm of activity and rest thus follows the normal diurnal rhythm of falling temperature by night and rising temperature by day, except when daytime mists and showers superimpose a minor fluctuation and result in daytime activity. No activity is observed on continuously wet days which are normally without such fluctuations.

5. Between 20 and 30° C. activity is induced by rising temperatures and suppressed by falling temperatures. The ecological significance of this is discussed.

6. When maintained at constant temperature slugs show a persistent but immediately and steadily deteriorating diurnal rhythm of activity and rest which could not of itself account for the regular appearance of nocturnal activity in the field.

7. In a temperature gradient slugs aggregate about a preferred temperature. The ecological significance of this is discussed.

8. The difficulty of describing these reactions in the terminology put forward by Fraenkel & Gunn (1940) is considered.

I am indebted to Dr C. F. A. Pantin, F.R.S., for his constant advice and encouragement and to Prof. J. Gray, F.R.S., for affording experimental facilities. I am also grateful to Newnham College, Cambridge, and the Department of Scientific and Industrial Research for financial aid.

REFERENCES


The activity of slugs


