

Site of water vapor absorption in the desert cockroach, *Arenivaga investigata*

(threshold relative humidity/hygroscopic secretion/insect mouth parts/surface temperatures)

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ABSTRACT The desert cockroach, *Arenivaga investigata*, can gain weight by absorption of water vapor from unsaturated atmospheres above 82.5% relative humidity. Blocking the anus or the dorsal surface with wax does not prevent water vapor uptake, but interference with movements of the mouthparts or blocking the mouth with wax prevents such uptake. Weight gains are associated with the protrusion from the mouth of two bladder-like extensions of the hypopharynx. During absorption these structures are warmer than the surrounding mouthparts, their surface temperature increasing with relative humidity. This suggests that the surfaces of the bladder-like structures function at least as sites for condensation of water vapor, but the precise location of its transfer into the hemolymph has not yet been identified.

Certain arthropods are able to absorb water from unsaturated air and thus increase the water content of their bodies. This uptake of water vapor can best be demonstrated if the animal has first been water-depleted in a very dry atmosphere and then returned to more humid, yet still unsaturated, air.

Studies of this water vapor absorption have in recent years (1-5) shown that localized sites are involved in the process, rather than the entire external surface as was previously postulated (6-9). Mouthparts have been implicated in ticks (1), while insects such as *Tenebrio* larvae (2-4) and *Thermobia* (4, 5) use mechanisms involving the rectum.

Arenivaga investigata, the desert cockroach, was shown by Edney (10) to be capable of water vapor absorption in humidities above 82.5% relative humidity (R.H.). This limit appears to be temperature-independent. The present work deals with the site of absorption in *A. investigata* and presents preliminary evidence for the involvement of a nonrectal location, the hypopharynx, for absorption in insects.

METHODS

Cockroaches were collected in the deserts near Riverside, CA, and shipped by air to Toronto. They were maintained either in containers of sand or in 15-ml vials. Ground "Purina Lab Chow" was used for food, and the animals' water needs were supplied by exposing them to near-saturated humidity conditions for 3-4 days/week. No free water was given to the animals. Nymphs weighing 100-500 mg or adult females weighing 400-700 mg were used in the experiments. prior to each experiment, animals were desiccated for 2-3 days over silica gel, which reduced their body weight by 10-20% from the initial value.

Various surfaces of the animal could be sealed off from the atmosphere by application of a melted beeswax/resin mixture at 60° (11). Continuous weight recordings were made with a Cahn RH or Mettler ME22 electronic microbalance. Humidity and temperature control in the animal chamber of the Cahn balance has been described (12). Surface temperatures were

Abbreviation: R. H., relative humidity.

Table 1. Weight changes at 25° and 96% R.H. in animals with part of body surface covered with beeswax/resin mixture

Surface waxed	Rate of weight gain (% initial body weight/day)*	SE	n
Control, no wax applied	3.59	0.51	25
Anus	0.87	0.13	18
Dorsal	1.72	0.48	7
Mouth	-1.24	0.15	16
Mouth, wax removed	0.44	0.32	11

* A minimum of three daily weight measurements were recorded for each animal.

determined as potential differences between a thermocouple junction placed on the surface and another junction in the air above it. The thermocouples were constructed from 40 gauge copper and constantan wires fused to form a bead 0.2-0.3 mm in diameter. Potential differences were measured on a Keithley 155 microvoltmeter.

RESULTS AND DISCUSSION

Blocking the anus or covering the dorsal surface with the beeswax/resin mixture did not prevent water absorption but resulted in decreased absorption rates relative to control animals (Table 1). Continuous recordings of weight changes in control animals showed periods of weight gains interspersed with short intervals during which small weight losses occurred. In animals with the anus blocked, uptake rates during periods of continuous weight gain were unaffected, but this treatment increased the frequency or duration of the interruptions of uptake, apparently without affecting the absorption mechanism itself. Similarly, the humidity at which water uptake was balanced by evaporative losses was unchanged by anal blocking. For seven animals the mean threshold relative humidity at 25° was $82.5 \pm 2.96\%$ (SE) prior to blocking and $82.6 \pm 1.24\%$ (SE) after blocking; the difference is not significant. These results also indicate that blocking the anus does not prevent functioning of the absorption mechanism.

In addition, studies of the weight changes of newly voided fecal pellets in absorbing animals precluded the rectum as the site of absorption. Weight traces of recently fed animals using a balance pan that prevented the loss of fecal pellets changed immediately from gaining weight to losing weight upon defecation. The rate of loss decreased progressively, returning to predefecation uptake rates within 4-6 hr. This loss of weight suggests that the fecal pellets were equilibrated to a rectal humidity that was much higher than the humidity from which the animals were absorbing. If the rectum were involved in water absorption, it would probably also produce fecal pellets

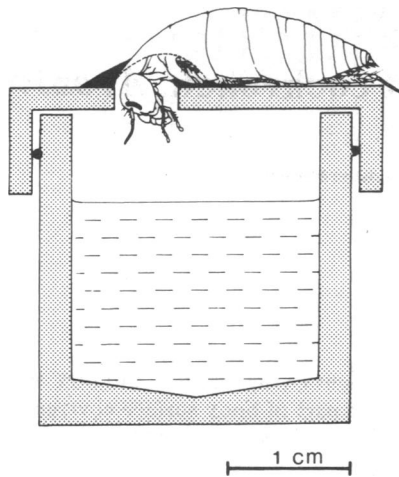


FIG. 1. Apparatus for determining exchanges by localized parts of the body in differing relative humidities. The lid fits around a Neoprene O-ring (black circles) lying in a groove of the lower section of the chamber. The animal is attached with wax (black shading) around a hole in the lid.

that would themselves take up water from the ambient air. Data in Table 1 further show that blocking the mouth with wax resulted in weight losses. However, the blockage did not have to be complete; prevention of movements of the labium or labrum with wax or tying a strand of surgical thread across the mouth

Table 2. Rates of weight gain with the thorax and abdomen in a different humidity from the head

% R. H.		Rate of weight gain (% initial body weight/day)	SE	n*
Around head	Around thorax and abdomen			
96	64	1.14	0.23	26(8)
64	96	-1.04	0.14	24(12)

* Total number of daily measurements with number of animals in parentheses.

produced similar inhibition of absorption. If the wax was carefully removed, the animal's ability to absorb water vapor was partially restored. Uptake rates were more fully restored in animals with the thread removed.

It has been suggested that covering parts of the animal with wax could interfere with an animal's sensory input and behavior in such a way as to indirectly inhibit the absorption process (13). Evidence to the contrary was provided by techniques in which localized regions of the animal's body were reversibly exposed to conditions favorable to uptake. The head of the animal was exposed to a humidity different from that of the thorax and abdomen by forming a wax collar around the back of the neck and attaching the collar with more beeswax to the lid of a stainless steel chamber (Fig. 1). With the lid on the chamber, the head of the animal was exposed to a humidity controlled

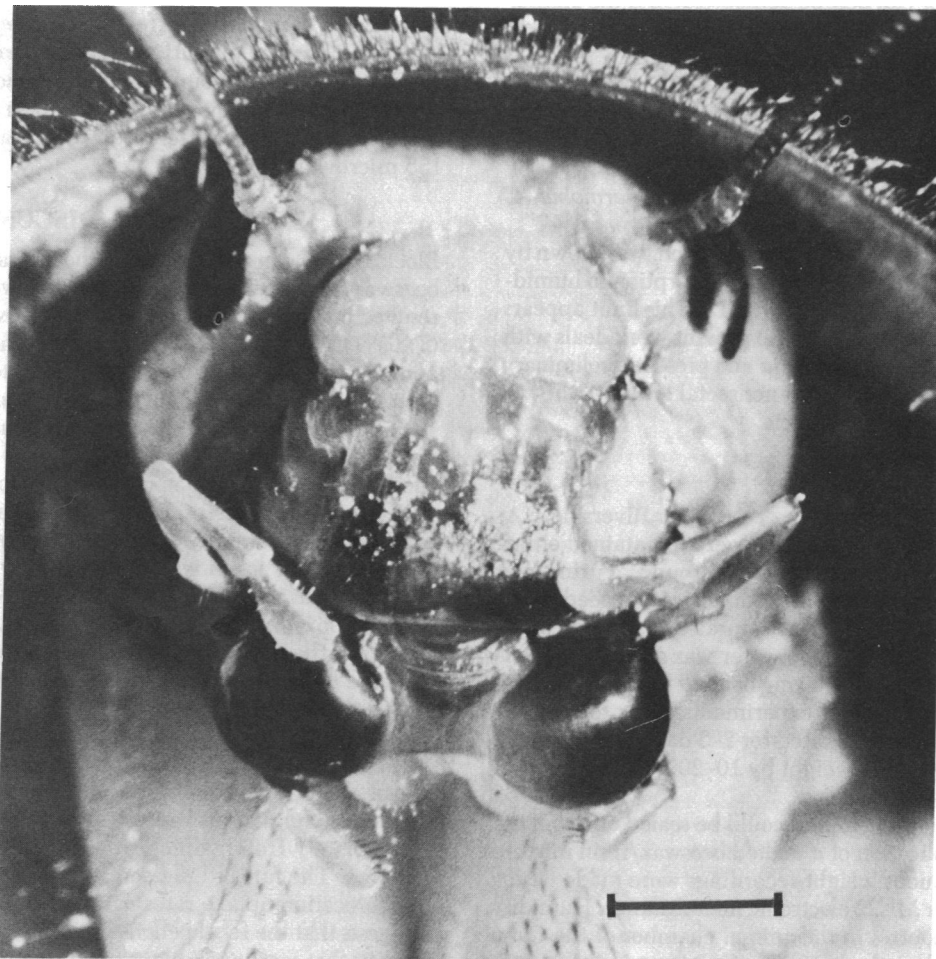


FIG. 2. Photograph of the head of a desert cockroach. The bladders are the two dark spherical structures protruded from the mouth (center). Scale = 1 mm.

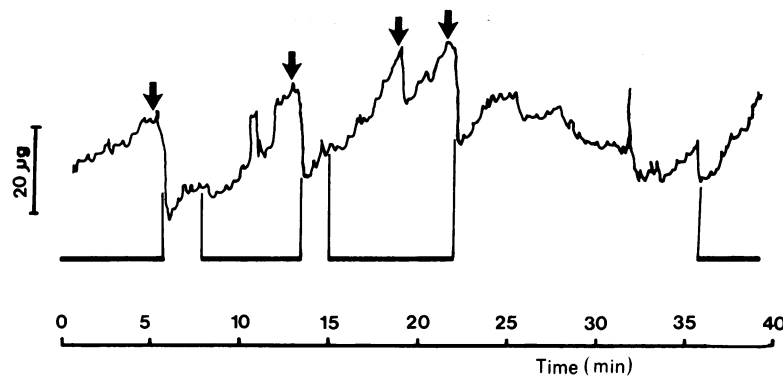


FIG. 3. Continuous recording of weight changes of a desert cockroach exposed to various atmospheric humidities. Bladders protruded from the mouth during the periods marked by thickened lines beneath the weight trace. Relative humidity was lowered several percent from 95% R.H. for 10–15 sec at the lines marked by the arrows.

by one saturated salt solution, while the thorax and abdomen were outside the chamber and could be subjected to another humidity. The lid and attached animal were then removed and weighed to the nearest milligram at 24-hr intervals. Table 2 shows that net absorption of water vapor occurred only when the head was exposed to high humidity, even though the much larger surface area of the thorax and abdomen was in low humidity. In the reverse situation, the head in low humidity and thorax and abdomen in high humidity, there was a net loss of weight. In further experiments with the thorax and abdomen in 96% R.H. at 25°, 1 out of 15, 3 out of 21, and 5 out of 7 animals gained weight with the head exposed to 71.5%, 75.5%, and 80% R.H., respectively. The threshold of the absorption mechanism is therefore about 7% R.H. lower than the whole animal's threshold of 82.5% R.H. The fact that the proportion of animals showing net weight gains is humidity-dependent suggests that there is variability either in the physiological mechanism for absorption or in the animal's response to humidities favorable to uptake. Unfortunately, similar variability in passive losses in subthreshold humidities, presumably of respiratory origin, have compounded the difficulty in establishing whether the absorption mechanism has a fixed physiological threshold of the type described by Beament (14).

Animals in high relative humidity observed at half-hour intervals over 12 hr showed a significant correlation between gain in weight and the proportion of time during which two bladder-like structures (Fig. 2) were everted from the mouth. These structures consist of lateral outcroppings of the hypopharynx and are inflated by hemolymph pressure, presumably increased by contraction of the thoracic and abdominal musculature. Individual animals could be suspended from the beam of the electronic balance by a loop of wire waxed onto the dorsal surface. This allowed observation of the head and mouthparts of the animal while weight changes and humidity were recorded. A typical weight trace is shown in Fig. 3. Weight gains occurred only when the bladders were protruded from the mouth. Other traces showed that the gains occurred as long as the bladders remained protruded, indicating a continuous rather than an oscillatory nature of the absorption process. If the humidity was sharply reduced for 10–15 sec, a loss of weight occurred (10–20 µg), and the animal often withdrew the bladders. Gains ceased while the bladders were unexposed. The animal salivated copiously and groomed the bladders with its maxillae and mandibles prior to protruding them; small losses or no change in weight occurred for about a minute after this, followed by a constant rate of uptake. This behavior suggests that the salivary fluid used to groom the bladders is not hygroscopic.

A total of 53 measurements of the surface temperature of bladders for seven animals indicated that they were significantly warmer (0.13–0.21°) than the atmosphere and 0.08–0.16° warmer than other parts of the body (labrum, labium, foreleg femur, or abdomen). The temperature at any one point increased significantly with relative humidity, suggesting that the primary source of heat was the condensation of atmospheric water vapor. The shiny wet appearance of the bladder surfaces during these measurements indicated the presence of a fluid layer. If one of the bladder surfaces became dry, its temperature was not significantly different from that of other parts of the body. These observations suggest that condensation occurs onto the layer of fluid and not onto the dry bladder surface itself. Scanning electron microscopic studies show the surface of the bladders to be provided with a covering of densely packed hairs, 0.5–0.8 µm in diameter and 80–100 µm long (15). These apparently serve to hold a layer of fluid onto the bladder surface but may also be involved in maintaining its flow.

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