WATER BUDGETS OF CAVE CRICKETS, *HADENOECUS SUBTERRANEUS* AND CAMEL CRICKETS, *CEUTHOPHILUS STYGIUS*

Eugene H. Studier, William D. Wares II, Kathleen H. Lavoie and Julie A. M. Linn*

Department of Biology, University of Michigan-Flint, Flint, Michigan 48502-2186, USA

Abstract—1. Studies of cave and camel crickets from cave entrances in Mammoth Cave National Park have produced estimates of total water budgets and component contributions to water balance.

2. Weight specific (mg/g crop-free live weight/hr) total water loss (3.200 vs 2.220) and water gained in food (2.393 vs 1.902) are greater in cave than in camel crickets, respectively.

3. Weight specific evaporative water loss in both humid, still air (2.269 vs 1.325) and dry, moving air (15.28 vs 9.85) is greater in cave than in camel crickets, respectively.

INTRODUCTION

Vandel (1965) stated that the level of humidity is the most important environmental factor in the life of terrestrial cavernicoles and credited Bedel and Simon (1875) with first recognizing this factor. Data supporting this generalization among cavernicolous arthropods derive primarily from behavioral micro-habitat selection studies (Mitchell, 1971; Edwards, 1971; Bull and Mitchell, 1972; and, Lucarelli and Sbordoni, 1977) and studies of dehydration tolerance (Mitchell, 1971; Bull and Mitchell, 1972; Wilson, 1975). These studies suggest that cave insects tend to select microhabitats that are cool with high relative humidity. Physiologic studies of water relations in cavernicolous arthropods have dealt mostly with measures of evaporative water loss (Herreid, 1969; Vannier, 1977). Hadley *et al.* (1981), compared several measures of water and metabolic relationships between an epigean and a cave spider. Water loss rates were shown to correspond to the environmental stresses each species encountered in their normal habitat. The only aspect of water budgets that has been quantified for cave-dwelling crickets is transpiratory water losses in several Australian species (Campbell, 1980).

This study presents data on all aspects of the water budgets of two cavernicolous crickets and continues our studies of the physiological ecology of these insects (Studier *et al.*, 1986a,b).

MATERIALS AND METHODS

Studies on cave crickets, *Hadenoecus subterraneus*, were conducted during the last week of April and first week of May, 1985, in Floyd Collins Crystal Cave and White Cave in Mammoth Cave National Park ((MCNP), Ky. Data on camel crickets, *Ceuthophilus stygius*, were collected primarily during the last week of June, 1985, in Great Onyx Cave, MCNP, Ky. Studies of evaporative water loss (EWL) in dry

air were performed on both species at the caves indicated above in October, 1985. All *H. subterraneus* studied were adults and the *C. stygius* were adults or penultimate instars (T. H. Hubbell, personal communication).

Data on total water loss, water lost in wastes, water gained in food and metabolic water production were derived from the long term weight loss studies previously described (Studier *et al.*, 1986a). Total water loss was estimated by mean long term total body weight loss minus rate of dry waste lost. Water lost in wastes was calculated from rate of dry wastes produced assuming those wastes had the same fractional water content as cricket carcasses. EWL is estimated as total water loss minus water loss in wastes. Water gained in food was determined from rate of wet crop content disappearance adjusted by fractional water level of crop contents. Metabolic water production was converted from estimated metabolic rate assuming aerobic glycolysis.

Rate of EWL in humid air was determined for *H. subterraneus* on 30 May 1985 in Floyd Collins Crystal Cave. Saturated solutions of specific salts (Winston and Bates, 1960) were poured into the bottom part of plastic "pie savers" (8 cm high × 30 cm diameter) lined with a tight-fitting, lipped, shallow (4 cm high) plastic liner. A circle of screen cloth (1/4 in. mesh) rested on the lip of the liner above the solution. This apparatus was placed inside plastic garbage bags which were sealed leaving ample head room above the screening. When checked in the cave setting (Certified Hygrometer and Temperature Indicator, Model HTAB-176, Abbeon Cal, Inc., Santa Barbara, Calif.), these devices reached constant relative humidities within 1 hr. Containers maintained relative humidities of 71, 84.5, 93.9, 98.5, and 100%. Six crickets (three females, three males) placed in cylindrical cages (4 cm diameter × 15 cm length) constructed of plastic screen cloth (1/16 in mesh), and a control (empty) cage were weighed and placed into the containers described above. After a 7-hr mid-day exposure, at a cave temperature of 13.5°C, caged crickets and the control cage were reweighed to the nearest mg (Ohaus Centogram Balance, Union, N.J.). For those crickets which did not defecate while caged, weight loss is an estimate of EWL.

Determination of EWL in dry, moving air for both species in the previously mentioned caves was carried out in October, 1985. Materials were allowed to equilibrate with the cave environment (100% RH, 13.6°C in Crystal Cave, 100% RH, 14.0°C in Great Onyx). Compressed air, dried by flowing through silica gel, was passed at constant flow-rate (150 cc/min) over individual adult crickets of both sexes in

*Present address: Department of Health Behavior/Education, School of Public Health, University of Michigan, Ann Arbor, MI 48109, USA.

a large (35 mm diameter × 190 mm length) test tube. Plastic screen cloth (1/16-in. mesh) lining the interior of the test tube provided secure footing for crickets. Outflow air was directed through a flow-meter (Matheson, Model No. 62 2SV) for 25 min to ensure that any condensed water had evaporated and that chamber air approximated to 0% relative humidity. Outflow air was then redirected through a pre-weighted (Fisher Scientific Electronic Analytical Balance, Model 2100) column of silica gel for 1 hr. Weight gain by the silica gel represents EWL after correction for any weight changes in the control system which was treated exactly as experimental except that no cricket was contained in the chamber.

Crickets from EWL in dry air studies were sacrificed in chloroform vapor and stored individually in test tubes at 4°C for one week. Length and diameter measurements (accuracy to 0.001 in.) were taken with a caliper (Fowler Max-Cal Caliper No. 54-200-000, Fowler, Inc., Newton, MA). Surface area was calculated for each body segment as if they were cylindrical. Length was taken only on the longest axis of the body segment. Diameter was an average of between two and six measurements taken in various planes of each segment. Measurements were taken to include an equal number of large diameter and small diameter areas. Body segments used in surface area determinations were separated from the thorax/abdomen prior to measurement and included antennae, head, large labial and labral palps, all legs, thorax/abdomen, hind cerci and ovipositors. Total surface area was determined as the sum of the parts. Total surface area in in.^{2} was then converted to mm^2.

RESULTS

Component and total water budgets of cave and camel crickets in still, humid air are given in Table 1.

The only gender related differences are that crop-free live weight (CFLW) and total body water (TBW) of camel cricket males are greater than in females ($t = 3.34$; d.f. = 14; $P < 0.01$; and $t = 3.997$; d.f. = 14; $P < 0.01$). Since CFLW and TBW of cave crickets of both sexes (411 ± 17 mg, $N = 13$ and 300.1 ± 11.9, $N = 13$) is significantly less than CFLW and TBW of female camel crickets ($t = 12.08$; d.f. = 18; $P < 0.001$; and $t = 28.76$; d.f. = 18; $P < 0.001$, respectively), crop-empty camel crickets have greater biomass than cave crickets. Regarding weight specific comparisons (mg/g CFLW/hr) of water balance components between species, cave crickets gain water from food more rapidly than camel crickets (2.393 ± 0.138, $N = 12$ vs 1.902 ± 0.187, $N = 16$; respectively, $t = 2.11$; d.f. = 26; $P < 0.05$). Total water loss in *H. subterraneus* (3.200 ± 0.148; $N = 12$) is greater than in *C. stygius* (2.220 ± 0.069, $N = 16$; $t = 5.99$, d.f. = 26; $P < 0.001$); and, EWL of cave crickets (2.269 ± 0.226; $N = 12$) is more rapid than in camel crickets (1.325 ± 0.172; $t = 3.33$, d.f. = 26; $P < 0.01$).

The short term weight loss study in still air of varying water content shows a significant relation between relative humidity deficit (RHD = 100-test relative humidity) and EWL ($F = 3.28$; d.f. = 3 and 19; $P < 0.05$). The least squares regression analysis gives the equation:

$$\text{EWL (mg/hr)} = 0.116\,\text{RHD}(\%) + 1.26.$$

Since these studies were carried out at constant temperature, RHD can be converted to water vapor

Table 1. Water balance components of male (M) and female (F) cave crickets, *Hadenoecus subterraneus*, and camel crickets, *Ceuthophilus stygius*, estimated from long term weight loss in humid, still air. Crop free live weight (CFLW) and total body water (TBW) are in grams. All other values are in mg/g CFLW per hr. Entries include arithmetic mean and standard error of the mean in parentheses

	H. subterraneus		C. stygius	
Parameter	F; n=7	M; n=5	F; n=8	M; n=8
CFLW	0.426	0.390	1.055	1.304
	(0.012)	(0.037)	(0.051)	(0.055)
Total loss	3.381	2.946	2.239	2.201
	(0.135)	(0.282)	(0.076)	(0.121)
Waste loss	0.955	0.898	1.041	0.750
	(0.194)	(0.177)	(0.134)	(0.217)
EWL	2.492	2.074	1.250	1.451
	(0.314)	(0.343)	(0.207)	(0.285)
Food gain	2.528	2.202	1.896	1.753
	(0.136)	(0.268)	(0.199)	(0.316)
Metabolic gain	0.408	0.451	0.419	0.423
	(0.045)	(0.093)	(0.079)	(0.077)
TBW	0.302	0.298	0.752	0.920
	(0.008)	(0.030)	(0.010)	(0.040)

Table 2. Body parameters and evaporative water loss (EWL) in dry, moving air in cave and camel crickets. Cave crickets were tested at 13.6°C at a water vapor pressure deficit of 11.68 mmHg and camel crickets at 14°C with a deficit of 11.99 mmHg. Asterisks indicate level of significance of t-tests with $* = P < 0.05$, $** = P < 0.01$ and $*** = P < 0.001$. Sample size for each sex of each species is 8. HFL = hind femur length; CFLW = crop free live weight

Parameter	H. subterraneus			C. stygius		
	F	M	t	F	M	t
EWL (mg/hr)	13.24	6.46	2.27*	16.29	14.26	NS
	(2.88)	(0.80)		(1.60)	(1.58)	
HFL (mm)	22.2	20.6	6.40***	22.4	21.2	NS
	(0.2)	(0.2)		(0.5)	(0.7)	
Tot Wt(mg)	642.2	625.6	NS	1890.2	1301.9	3.83**
	(38.4)	(49.7)		(118.4)	(97.8)	
CFLW(mg)	491.9	391.3	6.34***	1216.9	1267.0	NS
	(11.9)	(10.4)		(48.3)	(76.5)	
S.A.(mm^2)	1387.	1244.	2.63*	2293.	1881.	3.27**
	(26.)	(47.)		(84.)	(94.)	

pressure deficit or saturation deficit and the intercept of the equation estimates EWL under water saturated conditions found in these biologically active caves.

Reduced data for EWL in dry, moving air are given in Table 2. CFLW is less than total weight in male ($t = 4.61$; $P < 0.001$) and female ($t = 3.74$, $P < 0.01$) cave crickets as well as female ($t = 5.26$; $P < 0.001$) camel crickets. Comparisons between species show camel crickets to have significantly greater EWL than cave crickets (15.28 ± 1.12 vs 9.85 ± 1.69 mg/hr, respectively; $t = 3.43$; d.f. = 30; $P < 0.01$), significantly greater total weights than cave crickets (1596.1 ± 106.2 vs 633.8 ± 30.4 mg, respectively; $t = 8.71$; d.f. = 30; $P < 0.001$); significantly greater CFLW than cave crickets (1242.0 ± 44.0 vs 441.6 ± 15.1 mg, respectively; $t = 17.2$; d.f. = 30; $P < 0.001$); and significantly greater surface areas (2086 ± 81 vs 1316 ± 32 mm^2, respectively; $t = 8.841$; d.f. = 30; $P < 0.001$).

DISCUSSION

Data derived from the long term weight loss experiment represent the most complete estimates of total water budgets and component contribution to water economy of the crickets studied. These studies also have the greatest biological applicability since they were done in caves under natural conditions. With regard to all estimates derived for *H. subterraneus* it is important to note that the percentage of body water did not change throughout the 5-day experimental period and weight loss during that experimental period was linear (Studier *et al*; 1986a). These data indicate that cave crickets maintained overall water balance and that components of water economy were constant throughout the period of observation. For camel crickets, however, the percentage

of body water of female crickets decreased significantly ($t = 6.56$; d.f. = 14; $P < 0.001$) over the period of observation and weight loss is curvilinear throughout that same period (Studier *et al.*, 1986b) which suggest that female *C. stygius* were in negative water balance during the study. Furthermore, since average values were used to calculate total water budgets and component contributions for *C. stygius*, values shown in Table 1 underestimate levels during the first 2 days of observation and overestimate those same parameters during the last 2 days of the study period.

Weight specific rate of total water turnover (= total loss) of *H. subterraneus* was significantly greater than for *C. stygius* (Table 1). These differences relate to the higher surface area to mass ratio of cave crickets compared to camel crickets (Table 2). This greater observed relative water loss in cave crickets was expected in view of the different roosting habits of the two species. Cave crickets roost individually or in loose groups in deeper cave regions where ambient humidity is higher and more constant than those areas at the cave entrance occupied by camel crickets. Camel crickets often roost in tight clusters or wedged individually in tight cracks in the ceiling. Additionally, the cuticle of cave crickets seems to be thinner, more elastic and appears to be a less effective barrier to water flux than that of camel crickets. Camel crickets are very shiny compared to cave crickets, suggesting the presence of more epicuticular lipids. Camel crickets also routinely forage outside caves more frequently than cave crickets (Campbell, 1976; Leja and Poulson, 1984). Our October observation that male *C. stygius* had empty crops while females didn't (i.e. total weight of males was not significantly different than CFLW, Table 2) is interesting in view of the routine nightly exodus of

Table 3. Evaporative water loss in crickets in dry, moving air at 15 C (14 C for *C. stygius* and 13.6 C for *H. subterraneus*). All but *H. subterraneus* and *C. stygius* are Australian crickets with data from Campbell (1980). Primary habitat designated by symbols as follows: * = cave, + = leaf litter, ○ = soil, and # = burrows. Species arranged from lowest to highest weight related water loss rates. See text for further discussion

Species	n	CFLW mg ± SE	Water loss mg/h ± SE	%/h ± SE	%/h·mmHg^{-1}	mg/cm^2·h^{-1}	μg/cm^2·h^{-1}·mmHg^{-1}
Endacusta sp.#,*	17	187.0 ± 28.2	0.4 ± 0.1	0.29 ± 0.06	0.0227	0.146	11.44
Gryllulus sp.+,#	14	297.1 ± 19.6	1.1 ± 0.1	0.40 ± 0.05	0.0313	0.295	23.08
Teleogryllus commodus +,#	11	649.5 ± 63.1	2.9 ± 0.3	0.46 ± 0.07	0.0360	0.459	35.91
Ceuthophilus stygius *,+	16	1242.0 ± 44.0	15.3 ± 1.1	1.22 ± 0.08	0.1018	0.733	61.14
Australotettix carraiensis*	32	353.1 ± 20.2	7.1 ± 0.4	2.02 ± 0.08	0.1579	1.582	123.69
Pallidotettix nullarborensis*	10	319.0 ± 16.5	6.6 ± 0.1	2.08 ± 0.23	0.1626	1.562	122.11
Hadenoecus subterraneus*	16	441.6 ± 15.1	9.9 ± 1.7	2.19 ± 0.34	0.1871	0.752	64.43
Micropathus tasmaniensis*	10	285.3 ± 14.2	6.7 ± 0.3	2.38 ± 0.17	0.1861	1.694	132.45
Cavernotettix sp.*	15	246.0 ± 9.3	6.0 ± 0.2	2.46 ± 0.20	0.1923	1.656	129.48
Gryllotalpa australis○	10	922.2 ± 175.9	32.2 ± 6.2	3.50 ± 0.25	0.2737	4.045	316.30

this species. Empty crops in males indicate lack of feeding exodus for several days (Studier *et al.*, 1986b). Many camel crickets were observed *in copulo* in October and the active breeding period in *C. stygius* appears to be much more time restricted than in *H. subterraneus* due to the more seasonal appearance of adults. Male *C. stygius* behavior of remaining in the cavern entrance rather than emerging to feed may increase the likelihood of successful copulation with females as they return from foraging bouts.

We have been unable to locate any literature values for arthropods for comparison purposes where total water loss includes more than EWL. Sufficient data in a variety of mammals have been accumulated to yield an allometric equation relating total water loss to body mass (Altman and Dittmer, 1968). Estimated total water loss projected for mammals the size of cave and camel crickets would be 209.9 and 243.4 mg/g live weight per hr, respectively. These are two orders of magnitude greater than values found in crickets (Table 1).

In view of the very low metabolic rates of *H. subterraneus* and *C. stygius* in comparison to expected values of insects for their size (Studier *et al.*, 1986a,b), metabolic water production in these trogloxenic insects may prove to be a lesser source of water gain than in epigean species.

There are marked differences in the contributions of water balance components to the total water budgets in the two species. In cave crickets, EWL is 72.1% of total losses while EWL represents only 60.9% of total losses in camel crickets. Water lost in wastes contributes 29.4 and 40.3%, respectively, to total losses. The significantly greater water gain from food by cave crickets can probably be attributed to the fact that full cave crickets contain a greater mass of crop contents than camel crickets (Studier *et al.*, 1986a,b).

Wharton (1985), in his recent review of water balance in insects, discussed weight loss over time experiments including advantages and disadvantages of utilizing a single group of animals weighed successively as opposed to randomly selected animals exposed for each time period of interest. In that review, Wharton also points out that such weight loss experiments are characteristically performed on experimental animals which are standardized in such a manner that they do not eat, excrete, secrete, grow, or reproduce during periods of exposure to test conditions of controlled ambient temperature and (usually 0%) relative humidity. Although not included in Wharton's standardization list, tested insects are also allowed very limited mobility. Such studies provide a large literature allowing comparison of only EWL in many insects and measure an extreme physiological response to dehydrating conditions which have little direct biological applicability to water balance under natural or quasi-natural conditions.

We have determined EWL for both cricket species in the natural, wet, still air environment existing in caves (Table 1) as well as in moving, dry air conditions (Table 2). EWL was also studied in *H. subterraneus* at a range of high, controlled humidities in still air during the short term weight loss study. Results of an ANOVA of weight loss as a function of exposure relative humidity show that a significant inverse relationship exists. Weight changes of caged crickets during this short term (7 hr) study, however, were quite small and highly variable suggesting this protocol to be of severely limited experimental value. Since the 95% confidence interval for the slope of the least squares regression line includes zero, the regression equation given in the results section is quite tenuous and of highly questionable predictive value. It may well be fortuitous that projected EWL in water saturated air (1.26 mg/hr) agrees well with EWL determined from the long term weight loss studies (0.81–1.06 mg/hr; Table 1), and calculated EWL in dry air (12.9 mg/hr) agrees with values found in moving, dry air (6.5–13.2 mg/hr; Table 2).

The only available comparative data on EWL in humid air appears to be that reported by Campbell (1980) for *Australotettix carraiensis*, an Australian cave-dwelling cricket. He reported a value of $0.28 \pm 0.03\%$/hr for those crickets in moving air at 98% relative humidity at 15°C. Values for cave crickets (0.35 and 0.27%/hr for females and males, respectively) and camel crickets (0.18 and 0.21%/hr for females and males) calculated from the long term weight loss experiment (Table 1) agree closely with Campbell's data. Weight specific EWL of camel crickets in humid air is, however, markedly less than in the other crickets.

Although many references on evaporative (transpiratory) water loss in arthropods exposed to dry air are available (Wigglesworth, 1945; Holdgate, 1956; Mead-Briggs, 1956; Edney, 1971; Vanier, 1977; Arlian and Veselica, 1979; and many others), it seems most reasonable to compare values for cave and camel crickets with other related orthopterans as shown in Table 3. With the exception of values based on surface area, EWL of *H. subterraneus* lie in the middle of the range of EWL values for the four strictly cave-dwelling Australian species while EWL of *C. stygius*, as %/hr, are less than *A. carraiensis* ($t = 7.07$, d.f. = 46; $P < 0.001$) but more than *Teleogryllus commodus* ($t = 7.15$, d.f. = 25, $P < 0.001$). The mole cricket (*Gryllotalpa australis*) exhibits remarkably high EWL in dry air and would appear to be more susceptible to dehydration than the strictly cave-dwelling crickets which, in turn, lose transpiratory water more rapidly than leaf-litter or burrow utilizing crickets.

While weight specific EWL of *H. subterraneus* and *C. stygius* fall nicely into expected levels, area specific EWL values are much lower than expected. Considerable discussion of the problems associated with determination of surface area of insects is available (see Wharton, 1985). Using a method different from ours, Campbell (1980) established a relationship between body mass and surface area for two Australian cricket species (*Endacusta* sp., and *Australotettix carraiensis*) which are morphologically similar to *H. subterraneus* and *C. stygius*, respectively. Based on his equations, the surface area of *H. subterraneus* would range from 503 to 516 mm² and *C. stygius* from 926 to 1043 mm². Use of those calculated surface areas would bring surface area specific EWL parameters for *H. subterraneus* and *C. stygius* in close agreement with Campbell's data. Part of the reason that values projected from his equations are much lower than our

measured values is Campbell's lack of inclusion of antennal and palp surface area in total surface area, however, there appears to be extreme variation in estimates of suface area within and between techniques used. Area specific EWL estimates are, therefore, much more variable and less reliable for comparison than weight specific EWL. It should be mentioned, however, that crop-empty body mass should be used since gut contents may weigh as much as the gut-empty body (Wightman, 1981; Studier *et al.*, 1986a).

In summary, rates of water gain and loss in *H. subterraneus* are significantly greater than in *C. stygius* due primarily to a greater rate of water gain in food and loss as EWL, respectively. These observations correlate to differing behavior patterns in roosting and foraging seen in the two species. *H. subterraneus* roosts in the constant water saturated or near-saturated conditions deeper in caves and forages sporadically outside caves only when low saturation deficits exist. *C. stygius* is able to roost in the more variable (in terms of relative humidity and temperature) conditions at cave entrances and can forage outside the cave refuge under a wider range of environmental conditions.

REFERENCES

Altman P. L. and Dittmer D. S. (1968) *Metabolism*. Fedn Am. Soc. exp. Biol. Bethesda, Maryland.

Arlian L. G. and Veselica M. M. (1979) Water balance in insects and mites. *Comp. Biochem. Physiol.* **64A**, 191–200.

Bedel L. and Simon S. (1875) Liste generale des Articules cavernicoles de l'Europe. *Journal de Zoologie* **IV**.

Bull E. and Mitchell R. W. (1972) Temperature and relative humidity responses of two Texas cave-adapted millipeds, *Cambala speobia* (Cambalidae: Cambalidae) and *Speodesmus bicornourus* (Polydesmida: Vanhoeffenüdae). *Int. J. Speleol.* **4**, 365–393.

Campbell G. D. (1976) Activity rhythm in the cave cricket, *Ceuthophilus conicaudus* Hubbell. *Am. Midl. Nat.* **96**, 350–366.

Campbell G. D. (1980) Aspects of the ecology, environmental physiology and behaviour of several Australian cave-dwelling crickets. Ph.D. Dissertation, University of New South Wales.

Edney E. B. (1971) Some aspects of water balance in tenebrionid beetles and a thysanuran from the Namib Desert of southern Africa. *Physiol. Zool.* **44**, 61–76.

Edney E. B. (1977) *Water Balance in Land Arthropods*. Springer-Verlag, Berlin.

Edwards C. (1971) Relative humidity and temperature preference responses of the ricinuleid *Cryptocellus pelaez* (Arachnida). M.S. Thesis, Texas Tech. University.

Hadley N. F., Ahearn G. A. and Howarth F. G. (1981) Water and metabolic relations of cave-adapted and epigean lycosid spiders in Hawaii. *J. Arachnol.* **9**, 215–222.

Herreid C. F. II (1969) Water loss of crabs from different habitats. *Comp. Biochem. Physiol.* **28A**, 829–839.

Holdgate M. W. (1956) Transpiration through the cuticles of some aquatic insects. *J. exp. Biol.* **33**, 107–118.

Leja W. and Poulson T. L. (1984) Nocturnal cave exodus and return: cave vs camel crickets. Cave Research Foundation Annual Report, 1984, pp. 18–20.

Lucarelli M. and Sbordoni V. (1977) Humidity responses and the role of Hamann's organ of cavernicolous Bathysciinae (Coleoptera: Catopidae). *Int. J. Speleol.* **9**, 167–177.

Mead-Briggs A. R. (1956) The effect of temperature upon the permeability to water of arthropod cuticles. *J. exp. Biol.* **33**, 737–749.

Mitchell R. W. (1971) Preference responses and tolerances of the troglobitic carabid beetle *Rhadine subterranea*. *Int. J. Speleol.* **3**, 249–270.

Studier E. H., Lavoie K. H., Wares II W. D. and Linn J. A. M. (1986a) Bioenergetics of the cave cricket, *Hadenoecus subterraneus*. *Comp. Biochem. Physiol.* **84A**, 431–436 .

Studier E. H., Lavoie K. H., Wares II W. D. and Linn J. A. M. (1986b) Bioenergetics of the camel cricket, *Ceuthophilus stygius*. *Comp. Biochem. Physiol.* **86A**, 91–95.

Vandel A. (1965) *Biospeleology: The Biology of Cavernicolous Animals*. International Series of Monographs in Pure and Applied Biology, Vol. 22, Pergamon Press, London.

Vannier G. (1977) Relations hydriques chez deux especes de Tomoceridae (Insectes Collemboles), peuplant des niveaux ecologiques separes. *Bull. Soc. Zool. Fr.* **102**, 63–79.

Wharton G. W. (1985) Water balance of insects. In *Comprehensive Insect Physiology, Biochemistry and Pharmacology*, (Edited by Kerkut G. A. and Gilbert L. I.), Vol. 4, pp. 565–601. Pergamon Press, Oxford.

Wigglesworth V. B. (1945) Transpiration through the cuticle of insects. *J. exp. Biol.* **21**, 97–114.

Wilson J. (1975) The effect of low humidity on the distribution of *Heteromurus nitidus* (Collembola) in Radford Cave, Devon. *Trans. Br. Cave Res. Assoc.* **2**, 123–126.

Winston P. W. and Bates D. H. (1960) Saturated solutions for the control of humidity in biological research. *Ecology* **41**, 232–237.