is probably an indication that these lipids are located in cholesterol-free regions.

Removal of cholesterol leads to an increase in the fraction of lipids undergoing melting, as well as a shift to higher melting temperatures. This observation lends support to the suggestion, by Marsh et al.⁵, of a preferential segregation of cholesterol at these temperatures. Similar effects with cholesterol-depleted membranes were observed for myelin 15 and erythrocyte ghosts 16 that show no melting at all in the physiological range of temperatures, as long as cholesterol is present. Our experiments indicate that Ca²⁺ at physiological concentrations does not induce any further segregation of membrane lipids, in addition to that already present, in cholesterol-depleted membranes. This does not mean that at the lower degrees of segregation of the lipids in native granule membranes Ca⁺⁺ effects are also absent. However, due to the smallness of the melting peaks in the thermograms of native membranes the measurement of any such effects will be difficult.

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- O.H. Viveros, in: Handbook of Physiology, Endocrinology, p.389. Ed. H. Blaschko, G. Sayers and A.D. Smith. Am. Physiol. Soc., Washington DC 1975.
- 3 E. DeRobertis and A. Vaz Ferreira, Exptl Cell. Res. 12, 568 (1957).
- 4 C.L. Bashford, L.H. Johnson, G.K. Radda and G.H. Ritchie, Eur. J. Biochem. 67, 105 (1976).
- 5 D. Marsh, G.K. Radda and G.A. Ritchie, Eur. J. Biochem. 71, 53 (1976).
- 6 M.A. Martonosi, FEBS Lett. 47, 327 (1974).
- 7 D. Bach, I. Bursuker and R. Goldman, Biochim. biophys. Acta 469, 121 (1977).
- 8 D. Bach, I. Bursuker and I.R. Miller, Experientia 34, 717 (1978).
- 9 J. Hochman and R.L. Perlman, Biochim. biophys. Acta 421, 168 (1976).
- A.S. Schneider, R. Herz and K. Rosenheck, Proc. natl Acad. Sci. USA 74, 5036 (1977).
- 11 J.M. Trifaro and J. Dworkind, Analyt. Biochem. 34, 403 (1970).
- B. Roelofsen and L. L. M. van Deenen, Eur. J. Biochem. 40, 245 (1973).
- 13 Y. Folch, M. Lees and G.H. Sloane-Stanly, J. biol. Chem. 226, 497 (1957).
- 14 H. Blaschko, H. Firemark, A.D. Smith and H. Winkler, Biochem. J. 104, 545 (1967).
- 15 B.D. Ladbrooke and D. Chapman, Chem. Phys. Lipids 3, 304 (1969).
- 16 W.M. Jackson, J. Kostyla, J.H. Nozdin and J.F. Brandts, Biochemistry 12, 3662 (1973).

Desiccation in the black dragon, Hagenius brevistylus Selys1

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Summary. Hagenius brevistylus lost mass by evaporation in a moderately desiccating environment at the rate of 20.4 mg h^{-1} , and died of desiccation in less than 1 day at a body mass of 79.8% of their normally hydrated mass. It was estimated that Hagenius minimally would have to consume the equivalent of 60% of its body mass each day to meet its daily water requirements. This amount of food is equivalent to that necessary to power flight of a dragonfly for 4.6 h.

During a study of the role of posturing in the thermoregulatory repertoire of the black dragon³ (Hagenius brevistylus) Selys (Odonata), we noticed that this large dragonfly, held without water in a large flight cage, would position itself in the environment so that its body temperature would remain low. This seemed to differ from the behavior of freeranging dragonflies which periodically basked in the sun, presumably to raise their body temperatures. We hypothesized that the 'unusual' behavior exhibited by black dragons deprived of water in the flight cage, was due to behavioral hydroregulation to reduce evaporative water loss.

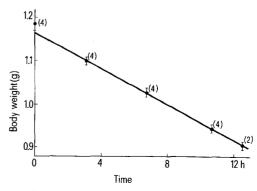
To see if evaporative loss rates in *Hagenius* could be great enough to become 'stressful', we captured 4 dragonflies, weighed each to the nearest tenth mg and hung them up to desiccate under laboratory conditions in bags made of a single thickness of cheesecloth. The laboratory air temperature was 25 °C, relative humidity was 65%, and wind speed was 0-0.5 mph.

Periodic weighings during desiccation (figure) showed that the dragonflies lost mass at a rate of 20.4 mg h⁻¹. This rate led the 4 dragonflies to die in about 12 h at a mean mass of 79.8% (\pm 3.0) of their initial mass.

The conditions under which this experiment was run can be regarded as minimally stressful relative to natural conditions for black dragons which are vigorous fliers and bask often in the sun. However, even under laboratory conditions, these dragonflies lost water at rates equivalent to 48%

of their body mass per day. Indeed, the dragonflies lost water at rates faster than reported for larger insects even during flight⁴. Thus, if these insects receive all of their water from food, they would have to consume the equivalent of 60% of their body mass per day (assuming 80% water in food) minimally.

It should be made perfectly clear that these dragonflies could conceivably consume the equivalent of 60% of their body mass in food each day. Tucker⁵ reports the minimum cost of locomotion for flying animals such that the power



Body mass of Hagenius during desiccation as a function of time.

(or food) required for flight by dragonflies can be calculated from knowledge of the mass of the animal and the speed of flight. If we assume that *Hagenius* (mass = 1.2 g) flies up to 9 m/s (20 mph), the cost of flight would be about 0.179 W. This translates to approximately 0.155 g of food (this assumes that there are 20.92 J/(g dry mass) in the food and that the food contains 80% water) necessary to fuel 1 h of flight, and this rate of intake would net the dragonfly about 0.124 g of water per h. Thus, *Hagenius* could conceivably consume the equivalent of 60% of its body mass in food in about 4.6 h. However, it is still unknown at what rate water would be lost from dragonflies in flight, and thus, it cannot yet be determined if flying animals can obtain enough water from the food they must eat to account for their needs throughout the day. It can be said with considerable certainty, however, that without access to

food or water, *Hagenius* would continually lose water at a rate that could be stressful and could cause these dragonflies to exhibit hydroregulatory behavior directed at retarding desiccation.

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- 3 C.R. Tracy, B.J. Tracy and D.S. Dobkin, Physiol. Zool., in press (1979).
- 4 N.S. Church, J. exp. Biol. 37, 171 (1960).
- 5 V.A. Tucker, Am. Scient. 63, 413 (1975).

The invasion and growth of Babesia bovis in tick tissue culture

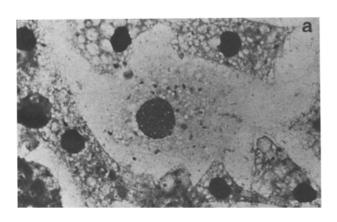
U.K.M. Bhat, D.F. Mahoney and I.G. Wright¹

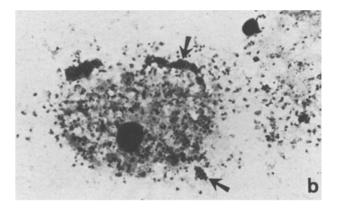
CSIRO, Division of Animal Health, Long Pocket Laboratories, Private Bag No. 3, P.O., Indofoopilly (Queensland, Australia 4068), 26 September 1978

Summary. Erythrocytic forms of Babesia bovis inoculated into cell cultures of the tick Boophilus microplus invaded the tick cells and showed multiplication for up to 48 h after inoculation.

Babesia bovis is a tickborne intra-erythrocytic parasite of cattle that causes economic loss in many countries of the world. A common vector is the one-host tick, Boophilus microplus. Although an attenuated vaccine prepared from the blood of infected cattle is available for the control of the disease², there are risks involved in its use such as the accidental transmission of other diseases and the production of haemolytic anaemia in new born calves³. B. bovis, grown in tissue culture, would be ideal as the basis of an improved vaccine and also as a source of antigens for immunological studies. However, the conditions for the invasion of tissue culture cells by a Babesia and its subsequent multiplication in vitro have not yet been established4. The invasion and multiplication of B. bovis in the cultured cells of the tick Boophilus microplus are reported in this paper and constitute the first successful attempt to grow a member of the genus Babesia in such a system.

Materials and methods. B. bovis was grown in splenectomized calves⁵ and infected blood was drawn using EDTA as anticoagulant. The infected erythrocytes were concentrated by differential lysis in hypotonic saline⁶. Suspensions





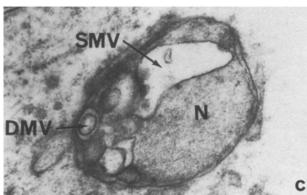


Fig. 1. a The invasion of the tick cells by B. bovis 8 h after inoculation of the culture with infected blood. $\times 730$. b Showing the increase in the number of B. bovis in the cells after 48 h. Arrows indicate the large multinucleate bodies. $\times 730$. c Electron micrograph of B. bovis inside a cultured tick cell, 24 hPI. N: nucleus, DMV: double-membraned vacuole, SMV: single-membraned vacuole. $\times 20,000$.