1 Holbrook, J. J., Liljas, A., Steindel, S. J., and Rossmann, M.G., *The Enzymes*, 11 (edit. by Boyer, P. D.), 191–268 (Academic, New York, 1976).
2 Everse, J., and Kaplan, N. O., *Adv. Enzymol.*, 37, 61–133 (1973).
3 Taylor, S. S., and Oxley, S. S., *Fedn Proc.*, 34, 2335 (1975).
4 Suzuki, K., and Taniguchi, Y., *Soc. exp. Biol. Symp.*, 26, 103–124 (1972).
5 Dawson, T. G., and Hulbert, A. J., *Am. J. Physiol.*, 218, 1233–1238 (1970).
6 Smyth, D. M., *Comp. Biochem. Physiol.*, 45A, 705–715 (1973).
7 Schmidt-Nielsen, K., Dawson, T. G., and Crawford, E. C. Jr, *J. cell. comp. Physiol.*, 67, 63–72 (1966).
8 Bartholomew, G. A., and Tucker, V. A., *Physiol. Zool.*, 37, 341–345 (1964).
9 Baldwin, J., Storey, K. B., and Hochachka, P. W., *Comp. Biochem. Physiol.*, 52F, 19–23 (1975).
10 O'Carra, P., and Barry, S., *FEBS Lett.*, 21, 281–285 (1972).
11 Mustafa, T., Moon, T. W., and Hochachka, P. W., *Am. Zool.*, 11, 451–466 (1971).
12 Hochachka, P. W., and Somero, G. N., *Strategies of Biochemical Adaptation*, 233 (Saunders, Philadelphia, 1973).
13 Atkinson, D. E., *Br. biochem. Soc. Symp.* (in the press).
14 Subramanian, S., Stickel, D. C., and Fisher, H. F., *J. biol. Chem.*, 250, 5885–5889 (1975).
15 Low, P. S., and Somero, G. N., *Comp. Biochem. Physiol.*, 49F, 307–312 (1974).
16 Borgmann, U., Laidler, K. J., and Moon, T. W., *Can. J. Biochem.*, 53, 1196–1206 (1976).
17 Lumry, R., and Rajender, S., *Biopolymers*, 9, 1125–1227 (1970).
18 Cohen, S. G., Vaidy, V. M., and Schultz, R. M., *Proc. natn. Acad. Sci. U.S.A.*, 66, 249–256 (1970).
19 Hochachka, P. W., *Comp. Biochem. Physiol.*, 52B, 25–31 (1975).
20 Low, P. S., and Somero, G. N., *Proc. natn. Acad. Sci. U.S.A.*, 72, 3305–3309 (1975).

Possible Gibbs–Donnan mechanism for cation uptake

MANY freshwater animals take up metal ions from solution in the surrounding water, often by active transport against a concentration gradient. The uptake of ions would be helped, and their loss opposed, by any means which increases the electrochemical potential of the ions in question in the external medium. One such means would be for the animal to secrete into the water a cationic substance to which the skin is not permeable; this would set up a Gibbs–Donnan effect (whereby an unequal distribution of diffusible ions is established at equilibrium on the two sides of a membrane if one side contains a non-diffusible ion) on the ions which are to be absorbed. The presence of such a mechanism in nature would be evident from the accumulation in the growth medium of cationic substances whose effect on the uptake of metals could be demonstrated experimentally.

For instance, the pulmonate snail *Biomphalaria glabrata* was shown by Thomas *et al.*[1,2] to secrete a substance which might act this way. The growth of the snails in these experiments was on the whole limited by the amount of calcium which could be absorbed, but the snails' secretion increased their growth rate and lowered the minimum concentration from which net uptake of calcium could be achieved. When the substance was allowed to accumulate in a limited volume of water (< 200 ml) for 3 d, the calcium concentration was reduced by the snails' uptake to $\sim 2.5\times10^{-5}$ M. But when the secretion was prevented from accumulating by continuous replacement of the medium, net uptake was not possible from solutions $\lesssim 2.5\times10^{-4}$ M. Addition of the partially purified secretion of other snails to the continuously replenished medium lowered the limiting calcium concentration from which net uptake was possible.

This model implies that the secretion of *B. glabrata* could increase the electrochemical potential of calcium tenfold. It is possible to calculate the effective concentration of non-permeant cations required to produce this effect, assuming that no other cations are present in significant amount. We can treat the system as though Ca^{2+} were partitioned at equilibrium between two compartments separated by a membrane permeable only to Ca^{2+} and to a common anion such as HCO_3^-. The molar calcium concentration in the compartment from which the non-permeant cation is absent (representing the interior of the snail) would then be $m'_{Ca} = 2.5\times10^{-4}$ and in the compartment where the non-permeant cation is present (representing the exterior medium) the calcium concentration is $m_{Ca}=2.5\times10^{-5}$. If the substance in question has molecular charge z, the molar concentration, m_p, required will be given by the Gibbs–Donnan equation

$$\gamma_\pm{}^3 m_{Ca}(2m_{Ca}+zm_p)^2 = 4\gamma_\pm{}'^3 m'_{Ca}{}^3$$

where $\gamma_\pm$, $\gamma_\pm'$ are the activity coefficients of the calcium salt in the compartments with and without the non-permeant cation, respectively. Substituting the calcium concentrations given above, and assuming the activity coefficients equal, we obtain the value $zm_p = 1.6\times10^{-3}$, which seems reasonable.

The Gibbs–Donnan effect will operate on all permeant cations, to an extent depending on their relative concentrations and their valence. (There is more effect on multivalent than on univalent ions.) The presence of an excessive concentration of another permeant ion such as Na^+ would reduce the enhancement of the electrochemical potential of Ca^{2+}. Other things being equal, the uptake of permeant cations would be favoured in approximate proportion to their external concentrations. Other non-permeant ions would enhance or reduce the Gibbs–Donnan effect depending on their concentrations inside and outside the animal and on their net molecular charges.

Apart from the identification of cationic macromolecular substances in the secretions of freshwater animals, this model could be confirmed by reproducing the Gibbs–Donnan effect with cationic polymers such as diethylaminoethyl dextran or polylysine. The presence of these in the water at quite modest concentrations ought to assist the uptake of calcium and stimulate the growth of animals as does the substance studied by Thomas.

It is not to be expected that such a mechanism would be useful to all freshwater animals, but only to those living in limited volumes of still water in which the hypothetical substances could accumulate. Because the mechanism would act equally well to retard loss as to promote uptake of ions, it would be of greater value to animals vulnerable to such loss; for example, it would be more likely to occur among gastropods, which are liable to dissolution of the shell, than among crustaceans, whose calcareous structures are protected by an impermeable cuticle.

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1 Thomas, J. D., Benjamin, M., Lough, A., and Aram, R. H., *J. Anim. Ecol.*, 43, 839–860 (1974).
2 Thomas, J. D., Goldsworthy, G. J., and Aram, R. H., *J. Anim. Ecol.*, 44, 1–28 (1975).