been speculation, based on spectroscopic evidence, that the structure of the coenzyme itself is altered when it binds: new light on this question also now comes from the X-ray work.

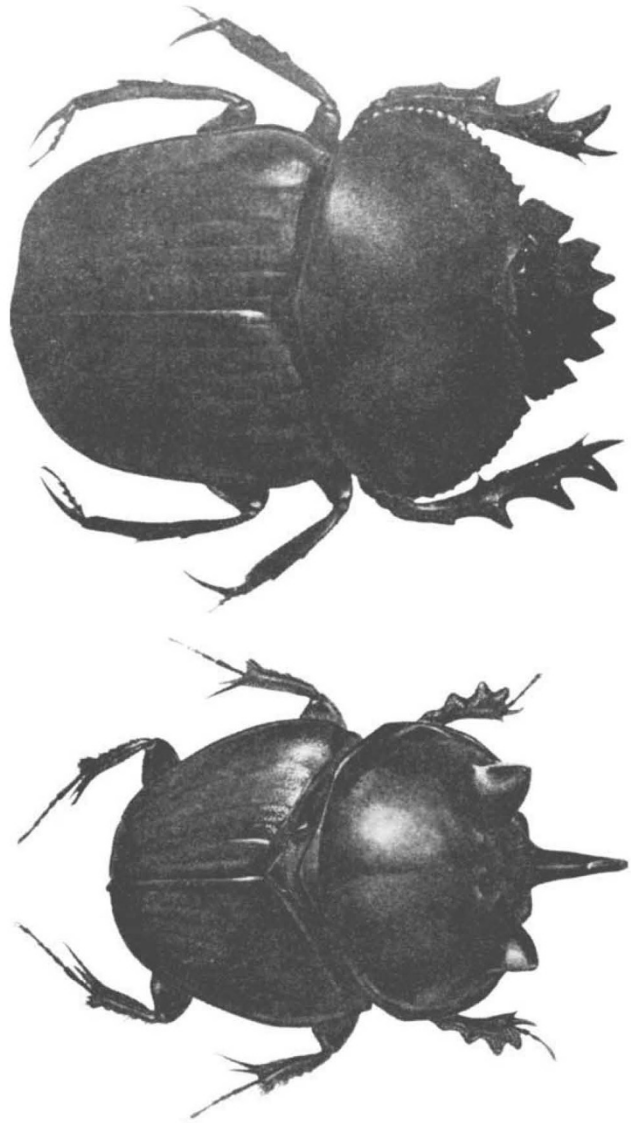
Adams *et al.* (*J. Mol. Biol.*, **51**, 31; 1970) working at a resolution of 5 Å have diffused NAD into crystals of the apoenzyme. A change in crystal symmetry occurs, and two prominent areas of electron density appear in the difference map. One is positive, and corresponds to the co-factor, and the other is negative, arising from a disturbance in the conformation at the junction of the subunits. The NAD density has the form of three maxima, one of which coincides with the single peak that is generated when AMP is introduced into the apoenzyme crystal. Although the 5 Å resolution is insufficient to delineate the co-factor structure in detail, it is possible to arrive at a relatively precise conformation by taking into account the restrictions on permitted angles of rotation that have been derived from the mass of existing data on the structures of nucleotides and nucleic acids. The NAD lies in a groove in the protein, the nicotinamide ring projecting inwards. The structure is an extended one, with the adenine and nicotinamide rings essentially at maximum separation. The reactive hydrogen atom on the nicotinamide approaches to 13·5 Å of the enzymatically essential thiol group, but there is no evidence of direct interaction.

The extended conformation means that the coenzyme undergoes a drastic change (an induced fit, so to speak) on binding, for there is convincing physical evidence in favour of a compact structure with stacked aromatic rings for the free molecule in water. This rests on measurements of fluorescence, of optical rotatory dispersion and, most definitively, proton magnetic resonance. A series of painstaking studies by Sarma and Kaplan (*Biochemistry*, **9**, 539, sqq.; 1970) by high-resolution proton magnetic resonance of NAD and a series of analogues leads to the conclusion that the ring can exist in two stacked forms, one left, the other right handed. In aqueous solution both are present in equilibrium proportions. On heating, or after addition of non-aqueous solvents, the stacking interaction is progressively broken down.

The immobilization of AMP in the lactate dehydrogenase crystal is also an interesting result, and has provoked an examination of the binding of various NAD analogues to the apoenzyme in solution (McPherson, *J. Mol. Biol.*, **51**, 39; 1970), in which the results are related to crystallographic manifestations. Binding constants were determined by competition experiments against NAD: competitive inhibition was established not only for adenine nucleotides, but also for adenosine. Here, however, binding was not accompanied by the switch in crystal symmetry; this therefore is evidently triggered by the phosphate group—an effect that it will be interesting to contemplate more closely when higher resolution X-ray data become available. Nicotinamide mononucleotide alone gave no indication of binding to the apoenzyme, but if AMP was present binding supervened, with a free energy almost equal to the difference between the values for NAD and AMP. It can be inferred that association of lactate dehydrogenase with its coenzyme is a two-step process, in which the AMP portion of the molecule first generates a conformational change in the enzyme to a form with a high affinity for the nicotinamide ring. This makes a diverting new variation on the allosteric theme, which should send practitioners flying to their drawing boards.

ENTOMOLOGY

Mozambique Scarabs



Kheper subaeneus (above) and *Onthophagus tersidorsis*, two dung beetles found in Mozambique. From M. C. Ferreira, *Revista de Entomologia de Moçambique*, **10**, 56 and 476; 1967.

CONTRACEPTION

Pills and Thrombosis

A RECENT survey has shown that the use of contraceptive pills before a surgical operation increases by three to four times the risk of thromboembolism after the operation. In practice this is still a very small risk, but it means that it might be wise for women to give up the pill for a month before undergoing an operation when this is possible.

The evidence for the increased risk comes from a retrospective study by a team at the Radcliffe Infirmary in Oxford, whose results are published in the *British Medical Journal* by Vessey, Doll, Fair-