sumably determined by interactions with side chains in the protein. In one unrelated dehydrogenase, different perturbations were observed, and Lee *et al.* suggest that the cofactor may be differently bound.

To come by definitive structures for nucleotide cofactors of lactate dehydrogenase, Rossmann and his colleagues (Chandrasekhar *et al.*, *J. mol. Biol.*, **76**, 503 ; 1973) have applied the constraints on dihedral angles that have been deduced from surveys of nucleotide and polynucleotide X ray structures. By this means the NAD structure in the ternary complex with pyruvate has been extracted and compared with bound NAD^+ in the binary complex and with adenosine and its mono- and diphosphates. The precise geometry of the phosphate groups is evidently critical in determining the binding, and the induced conformational change in the protein, involving the movement of a loop of the polypeptide chain across the substrate cavity, can evidently be touched off by a change in phosphate ionization, for crystals of the apoenzyme disintegrate if the nucleotides are allowed to enter at a *p*H below the phosphate *p*K. Because adenosine mono- and diphosphates, when they bind at the cofactor site, elicit the same conformational switch as does NADH or NAD^+, a direct comparison of their structure *in situ* is possible. The nucleotide structures are of the standard form, with the anti-configuration about the glycosidic bond, and a C_3-*endo* pucker in the sugar, but small differences of detail are apparent between the different ligand and their environment, in the protein. The second phosphate of ADP is known not to contribute appreciably to the interaction energy, which fits with the new observation of two alternative, partly occupied, locations for this group. Shifts of up to 2 Å or so between phosphate groups in the different ligands can be seen.

Cognoscenti of dehydrogenases will also wish to give close attention to the accompanying paper by Adams, Liljas and Rossmann (*ibid.*, 519), who have compared the lactate dehydrogenase structure in crystals grown in sulphate and in citrate solutions. Two specific anion binding sites are present, one of them in the active centre cleft, and the interactions between the ionized and protonated carboxyl groups of the citrate ion with its surroundings in the protein have been defined. The second site is at a subunit interface, separated from the active centre by two arginine side chains, projecting from either side of an α helix. Adams *et al.* tentatively link this secondary anion binding site with a second substrate binding site, and with a stabilizing effect on the tetramer. Since citrate is known to inhibit the breakdown of the protein to dimers, the site identifies the presumptive interfaces for this dissociation. The stable dimer unit is also thereby defined, and is structually analogous to the dimeric malate dehydrogenase molecule (Rossmann *et al.*, *ibid.*, 533).

FISH BIOLOGY

Well Equipped Iniomi

from our Marine Vertebrate Correspondent

FISHES of the deep-sea genus *Benthalbella* are poorly known in general even though they are found in all tropical and warm temperate oceans. Their biology has been little studied beyond general observations concerning their bathymetric range ; even their systematic relationships are not clear.

A study on *Benthalbella infans* by Merrett, Badcock and Herring (*J. Zool., Lond.*, **170**, 1 ; 1973) goes a long way towards rectifying the deficiencies in knowledge and clarifies an outstanding taxonomic problem. *Benthalbella infans* was described in 1911 by Zugmayer from specimens collected by the Prince of Monaco's yacht Princess Alice ; this original specimen was small as its name suggests, and Zugmayer realized it was a juvenile. Relatively few specimens have been reported since, but in 1955 a second species, *B. dubius*, was described from a larger specimen and at the time it was suggested that this might be the adult of *B. infans*. Merrett and his colleagues now report on ninety-seven specimens of *Benthalbella* caught by RRS Discovery over recent years. These new specimens nicely span the length range between Zugmayer's post-larval *infans* and the adult specimen of *dubius* ; they show that the latter is indeed the adult *B. infans*.

The Discovery series of specimens provides much basic information on the developmental stages of *Benthalbella*, of which the changes in the eye are of great interest. The eyes are essentially tubular, except that the early post-larvae have lateral eyes which attain the adult condition first by a dorsal movement of the lens in respect of the eye cup, and as the eye grows a rotation, so that it is dorsally directed. This leaves the eyes as tubular organs forever looking upwards.

Also during development the body posture changes considerably. Young fish are hump-backed in the region of the forebody, but adults are straight-bodied or are even hollow-backed so that the top of the head is at a higher level than the midpoint of the back. Furthermore, the anterior vertebrae are unossified so that the head can be presumed to be unusually mobile in a vertical plane. Merrett *et al.* associate these structures and modifications with the deduced feeding behaviour of *Benthalbella*. Tubular, upwardly-directed eyes are presumed to be associated with detecting downcoming light from lumi-

Sex Hormone Binding Proteins in Tissues

TARGET tissues for the sex hormones contain specific proteins which by interacting with the hormone are responsible for its uptake by the tissue and which are also involved in the early actions of the hormone. Binding proteins exist both in the nucleus and in the cytoplasm. In the case of the uterus the nuclear-binding protein has a sedimentation coefficient on ultracentrifugation of 4.5S, a molecular weight of about 60,000 and is relatively unstable, particularly in the absence of oestradiol. The cytoplasmic-binding protein is much larger (molecular weight of about 240,000, sedimentation coefficient 8.6S). The cytoplasmic protein can be broken down by exposure to buffers of high ionic strength to a smaller protein which in turn can be converted into the nuclear-binding protein by a factor present in the cytoplasm. Purification of the binding proteins has been difficult not only because of their low concentration in tissues but in the case of the nuclear-binding protein because of its instability.

Only poor yields and small degrees of purification have been achieved by conventional procedures or by attempts to purify the binding proteins chromatographically using columns containing oestrogens or oestrogen derivatives. Recent developments in affinity chromatography have been more successful. In *Nature New Biology* next Wednesday (July 11) Sica *et al.* describe the purification of the oestradiol-binding protein from calf uteri by affinity chromatography using an adsorbent oestradiol-17-hemisuccinate coupled by the carbodiimide procedure to an agarose derivative. In the purification of material present in such low concentration special care must be taken to remove non-covalently-bound oestradiol from the solid phase before chromatography and the methods must be sufficiently sensitive to detect the elution of the material.

The oestrogen-binding protein eluted from the adsorbent had similar physical characteristics to that of the nuclear receptor. The factors involved in the selective absorption and elution of the receptor proteins are also discussed in some detail. Although in these particular experiments the degree of purification achieved was not large, the yield obtained in most experiments was satisfactory. The authors suggest that by modification of the adsorbent more than a 10,000-fold purification of the receptor protein can be obtained but would still be only 20–40% the purity.