synthesis. These inhibitors do not stop the synthesis of the proteins characteristic of mitochondrial ribosomes, which must therefore be made in the cytoplasm and then migrate to the mitochondria either before or, less likely, after they have associated with mitochondrial ribosome RNA. This immediately raises the question of whether the genes for mitochondrial ribosome protein are in the nucleus or in the mitochondrial DNA. Estimates of the total amount of genetic information in mitochondrial DNA make it extremely unlikely, if not impossible, that mitochondrial DNA specifies the proteins of mitochondrial ribosomes. There just is not enough DNA to code the necessary information. The RNA component of mitochondrial ribosomes, on the other hand, is apparently specified by the mitochondrial DNA.

Küntzel has also recently characterized the subunits of the two classes of ribosome (*J. Mol. Biol.*, **40**, 315; 1969). The intact cytoplasmic and mitochondrial ribosomes sediment at 77*S* and 73*S* respectively. The large subunits can also be differentiated in the ultracentrifuge, where they sediment at 60*S* and 50*S* respectively. The small subunits, however, have the same sedimentation coefficient, 37*S*. But this is a coincidence; it does not, of course, mean that the two small subunits are structurally or functionally identical. In fact, as Küntzel has shown, they contain quite different proteins and, as Reflein *et al.* reported in 1967, their RNA differs in sequence and base composition.

Two new species of RNA, apparently associated with mitochondria, have been detected in HeLa cells by Vesco and Penman (*Proc. US Nat. Acad. Sci.*, **62**, 218; 1969). The RNAs sediment at about 21*S* and 12*S* and have several unusual characteristics. They are unmethylated, have a slow rate of turnover and their synthesis is at least five times more resistant to ultraviolet light than the synthesis of nuclear RNA. The function of 21*S* and 12*S* RNA, which are not found associated with polysomes and therefore probably are not involved in protein synthesis, remains obscure.

The list of mitochondrial enzymes which are specified by nuclear rather than mitochondrial DNA also continues to grow. Longo and Scandalios (*Proc. US Nat. Acad. Sci.*, **62**, 104; 1969) have recently reported that genes for malic dehydrogenase isozymes of mitochondria of maize are inherited in a classical Mendelian fashion, which must mean that they are in the nucleus, not the cytoplasm.

ENZYME BINDING

Perturbed Coenzymes

from our Enzymology Correspondent

THERE are already several ways of following the binding of redox coenzymes to their dehydrogenase partners. Two approaches have been especially fruitful: analysis of the effects of enzyme binding on coenzyme fluorescence and absorption, and analysis of the effects of coenzyme binding on enzyme optical rotatory dispersion spectra. A new technique has recently come to the fore. Charles Blomquist, of the Los Alamos Scientific Laboratory, has followed the binding of liver alcohol dehydrogenase to NADH by observing the effects of enzyme binding on the D_2O perturbation of NAD fluorescence (*J. Biol. Chem.*, **244**, 1605; 1969).

Dr Blomquist records that the fluorescence emission spectrum of NADH is boosted by D_2O. The fluorescence increase turns out to be a linear function of D_2O concentration, and in 75 per cent D_2O it is 20 per cent. To eliminate the possibility that the fluorescence enhancement reflects a D_2O-induced shift in the distribution of NADH into its open and folded conformations, Dr Blomquist looked for solvent effects in phosphodiesterase-hydrolysed NADH and *N*-methyldihydronicotinamide. Both compounds showed the same fluorescence enhancement in D_2O as intact NADH.

The effect is thus presumably a matter of energy transactions within the coenzyme's nicotinamide chromophore. One possibility is that D_2O may be exerting a kinetic isotope effect on proton transfer reactions in nicotinamide's excited state. The nicotinamide amino group cannot be very deeply involved in this, however, because an analogue of NADH which has a methyl group standing in for the ring amino displays the same D_2O effects as NADH.

When liver alcohol dehydrogenase is added to the coenzyme, the fluorescence enhancement subsides. If isobutyramide, which forms a very tight ternary complex with NADH and the enzyme, is added as well, the fluorescence enhancement is totally extinguished. This change must reflect a direct interaction between the reduced nicotinamide moiety of the coenzyme and some portion of the enzyme molecule.

That there is such an interaction is of course no news; it has been known for more than a decade as a result of kinetic and spectral studies. But nonetheless the D_2O perturbation method holds fair promise for the future. Dr Blomquist's experiments did not indicate whether binary and ternary complexes differ in their solvent sensitivity. It would be interesting to find this out, and it would be interesting to learn the underlying mechanism of this perturbation of NADH fluorescence.

CELLULAR ORGANELLES

Activity inside Cells

from a Correspondent

AT a symposium on the biogenesis of mitochondria and plastids held by the British Society for Cell Biology on March 27, Professor D. J. L. Luck (Rockefeller University) emphasized the autonomy of mitochondria in *Neurospora crassa* by showing that mitochondrial DNA extracted from a mutant strain of cells containing defective mitochondria can induce defective mitochondria in wild type cells which originally contained healthy mitochondria. He also presented evidence that mitochondrial DNA contains the information necessary to synthesize both ribosomal RNA and ribosomal protein. Mutations resulting in defective mitochondria produce defective mitochondrial ribosomes. Drs A. Jurand and G. G. Selmon (University of Edinburgh), discussing the replication of different organelles during the fission cycle of *Paramecium aurelia*, described how each organelle seems to have a different mode of formation. They suggested that new mitochondria are formed by vesicles budding off from the outer mitochondrial membrane.

Dr T. S. Work (National Institute for Medical Research, London) showed that in experiments using isolated mammalian mitochondria he was able to synthesize only a single species of protein which was identified as an acidic glycoprotein associated with the structural elements of the inner mitochondrial mem-