

group (Herner *et al.*, *ibid.*, 1335) also report the opposite action of sparsomycin, which strongly stabilizes the initiation complex (compare Černá *et al.*, *ibid.*, 27). Herner *et al.* suggest that the translocase is inhibited, so that the peptidyl-*t*RNA is unable to leave its first site.

The steroidal antibiotics, such as fusidic acid, which has been the subject of recent work by Pestka, have also been examined by Tanaka *et al.* (*J. Biochem., Tokyo*, **65**, 459; 1969), who obtained concordant results. These compounds again inhibit translocation, and have now been shown to do so by interacting with the relevant transfer factor (G): the loss of GTPase activity has been demonstrated.

MEMBRANES

Around the Algal Chloroplast

It is evidence of a healthy development in biochemistry when a single investigation can embrace topics so traditionally distinct as membrane structure, porphyrin metabolism, bioenergetics and protein synthesis. Biochemists are not commonly masters of many disciplines and too many specialists dare not, or even feel they may not, trespass beyond the confines of a single molecular process, thereby divorcing it unnaturally from the rest of the subject. So it is refreshing to see an increasing number of papers such as that by J. K. Hooper, P. Siekevitz and G. E. Palade (*J. Biol. Chem.*, **244**, 2621; 1969) which, although basically concerned with protein synthesis in algae, necessarily involves an understanding of chlorophyll biosynthesis, photosynthetic electron transfer, and the structure of chloroplast membranes and grana.

This group, from the Rockefeller University, wished to determine the role of the two types of ribosome known to exist in algae—the 68S particles in the chloroplast and the 80S units in the cytoplasm. Attention was concentrated on the proteins of the chloroplast membranes which go to make up the grana, and on the enzymes concerned with chlorophyll synthesis. Rather than use normal cells in which many sorts of protein are being synthesized simultaneously, Hooper *et al.* studied a mutant alga (*Chlamydomonas reinhardtii* γ -1) which does not produce chlorophyll—and, as a consequence, chlorophyll-containing membranes—in the dark. By growing the organism in the dark and then following its restoration to normal growth in the light (“greening”), the synthesis of membrane proteins could be followed and distinguished from the biosynthesis of most other proteins. Moreover, by studying the effects of two antibiotics on the greening it was possible to differentiate these processes still further and to come to a conclusion regarding the roles of the two ribosomal systems.

The studies were, however, not restricted to observations of amino-acid uptake: the production of chlorophyll and the parallel rise in electron transport activity were followed, and the formation of chloroplast disks and grana was observed by electron microscopy.

The combined results point to the involvement of two protein synthetic systems in the elaboration of chloroplast membranes, and indicate that the formation of disk components and their fusion into grana is a multi-step process. These findings may be correlated with the two or more types of DNA known in this alga, and with the two ribosomal populations. This is of

particular significance, for it demands that part of the chloroplast proteins originates in the cytoplasm—a situation already known in connexion with mitochondria—and requires that large macromolecules cross a double membrane barrier—a phenomenon that invites further analysis.

NUCLEIC ACIDS

Dissecting Transfer RNA

from our Cell Biology Correspondent

THE transfer RNA molecule functions in protein synthesis by picking up an amino-acid molecule and transferring it to the ribosome. Inside the ribosome, part of the *t*RNA molecule, the three nucleotide bases (forming the anticodon) pair with three complementary bases (a codon) in the messenger RNA molecule and thus ensure that only the amino-acid specified by that particular codon is added to the growing polypeptide chain. There is, therefore, one species of *t*RNA for every codon of the genetic code, and because the code is degenerate, using several codons to specify the same amino-acid, there is more than one *t*RNA species for each amino-acid.

The amino-acids do not, of course, attach themselves to a *t*RNA molecule but are added by enzymes specific for each kind of *t*RNA and amino-acid. Each of these amino-acylating enzymes presumably recognizes some unique structural feature of its corresponding *t*RNA, but what structure? On page 1147 of this issue of *Nature*, Imura, Weiss and Chambers report an ingenious experiment which involves dissecting a species of *t*RNA into “quarter” pieces with ribonucleases and then testing the fragments for amino-acid acceptor activity. In other words, they test to see which fragments or combinations of fragments can be recognized by the corresponding amino-acylating enzyme and charged with the amino-acid.

With yeast alanine *t*RNA, it turns out that none of the three fragments into which the molecule is split has acceptor activity, but, if the fragments which include the two ends of the molecule are mixed together, the resulting complex does have acceptor activity. In the intact molecule these two end sequences of the *t*RNA chain are held together by base pairing and the molecule has a cloverleaf structure with a stem made of the two ends (see page 1147). Apparently the amino-acylating enzyme can recognize with great precision some structure, or sequence of bases, in the stem of the molecule. In fact, Imura *et al.* suggest that the crucial bases are the fifth, sixth and seventh counting from the 3' end of the molecule to which the amino-acid is attached. These bases would have to carry the necessary chemical information for specific recognition by the corresponding amino-acyl synthetase enzyme and would have to be unique for each *t*RNA. Obviously the fact that all seven species of yeast *t*RNA of which the nucleotide sequence has been determined have unique sequences at these three positions supports the idea. But the suggestion that the stem sequence and structure determine amino-acid acceptor activity does not necessarily demand that in all *t*RNAs of all organisms it is bases 5, 6 and 7 that are crucial or, for that matter, the crucial bases need to be in linear sequence. All it demands is that some part of the stem is unique to each *t*RNA species.