same for all transfer RNA species. We have packing arrangements for this model that allow us to account in a satisfactory way for the diffraction results from single crystals of transfer RNA that have been published. In addition to being stereochemically attractive the model accords well with current understanding of biological function.

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Biosynthesis and Maturation of Low-Molecular-Weight Ribonucleic Acid Species

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t-RNA.* Since this small RNA species plays an important role not only in protein synthesis and its control but possibly also in the control of RNA synthesis itself (Maaløe & Kjeldgaard, 1966) it is essential to understand the processes involved in its own biosynthesis.

In cultured animal cells it has been demonstrated that a precursor to cytoplasmic *t*-RNA appears in the cytoplasm within a very short time of synthesis (Lal & Burdon, 1967; Burdon, Martin & Lal, 1967; Bernhardt & Darnell, 1969). This precursor, designated pre-*t*-RNA, is deficient in methyl bases, is somewhat longer than mature *t*-RNA (Burdon & Clason, 1969) and also has a more extended conformation (Burdon, 1967). It is postulated that, as part of a non-conservative maturation process giving rise to *t*-RNA, pre-*t*-RNA acts as substrate for the various specific *t*-RNA methylases that are located in the cell cytoplasm.

Removal of the essential amino acid L-methionine from the culture medium retards both the synthesis of pre-*t*-RNA and its maturation to *t*-RNA. It is thus possible that at least some of the pre-*t*-RNA bases require to be methylated for maturation to proceed and that the methyl-dependent step might be a nuclease attack at a specific site.

The low-molecular-weight RNA component of ribosomes (5s RNA). One molecule of this stable species (120 nucleotides long) is found in every larger ribosomal subunit in animal, plant and bacterial cells. In animal cells 5s RNA has also been detected within nucleolar particles contain the large ribosomal precursor RNA species (Warner & Soeiro, 1967). Although it has been shown in bacterial and in animal cells that 5s RNA and ribosomal RNA are synthesized at the same rate, it appears, however, that 5s RNA is not a part of the large 45s precursor of ribosomal RNA. Molecular hybridization has revealed that the nuclear DNA to which 5s RNA hybridizes can be separated from that to which ribosomal RNA hybridizes (Brown &

Dawid, 1968). Also, it has been demonstrated that the 28s RNA in a newly formed mammalian cell ribosome is not accompanied by a new 5s RNA (Knight & Darnell, 1967). Addition to the culture medium of the drug toyocamycin blocks the maturation in mammalian cells of the 45s ribosomal precursor RNA, but 5s RNA still appears in the cytoplasm (E. J. Smillie & R. H. Burdon, unpublished work).

A low-molecular-weight component from ribosomal RNA. When 28s RNA from the larger mammalian-cell ribosomal subunit is heated in the presence of detergent a small RNA molecule about 130 nucleotides in length is released (Pene, Knight & Darnell, 1968; Burdon & Clason, 1969). This small RNA can also be released by other treatments designed to break hydrogen bonds. Evidence has been obtained (Pene, Knight & Darnell, 1968) that this RNA arises from the same ribosomal RNA precursor molecules as does the 28s RNA. The function of this small RNA is unknown but it may play a role in maintaining the conformation of the RNA of the larger animal-cell ribosomal subunit. So far no equivalent RNA has been detected in bacteria.

Small monodisperse nuclear RNA from animal cells. Polyacrylamide-gel electrophoresis of RNA isolated from chromatin and nucleolar fractions from animal cells has revealed the presence of several distinct low-molecular-weight RNA species (Prestayko & Busch, 1968; Weinberg & Penman, 1968; Burdon & Clason, 1969). Their size ranges from 100 to 180 nucleotides, and they appear quite stable metabolically and are confined to the cell nucleus. It has been suggested (Prestayko & Busch, 1968) that these RNA molecules may be important in the regulation of genetic expression of chromosomes. Experiments with synchronized cultures of mammalian cells indicate that they are only synthesized at about the time of DNA synthesis in the S phase of the cell cycle (A. E. Clason & R. H. Burdon, unpublished work).

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* Abbreviation- *t*-RNA, transfer RNA.

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Biological Role of Transfer Ribonucleic Acid Molecules

By U. Z. LITTAUER. (*Weizmann Institute of Science, Rehovoth, Israel*)

The different known reactions in which transfer RNA is involved and the different enzymic alterations that may take place on this macromolecule will be summarized. Some aspects of the secondary and tertiary structure of transfer RNA and their relationship to transfer RNA function will also be examined. Finally, evidence for the induction of transfer RNA synthesis after T₄ bacteriophage infection will be presented.

The Effect on Transfer Ribonucleic Acid of Modification *in vitro*

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There have been many studies on the chemical modification of t-RNA* bases, and the resulting changes in the biological activity of these t-RNA species has been correlated with alterations in configuration or base sequence at specific sites in the molecule (Miura, 1967).

In this context, modification of t-RNA by chemical or enzymic overmethylation is of particular interest, since it has been shown that, in tumour tissue, t-RNA has an abnormal methylated base content (Borek, 1969).

By using conditions under which methylguanine is formed predominantly, *Escherichia coli* B t-RNA has been treated with dimethyl sulphate. Increasing levels of overmethylation, up to 13 moles % nucleotide excess, were used to study the physical behaviour of the t-RNA: in particular, the effects of over-methylation on melting curves and on optical rotatory dispersion have been examined.

Amino acid acceptance by overmethylated t-RNA was decreased by an amount characteristic for each of 18 of the common amino acids.

* Abbreviation: t-RNA, transfer RNA.

The formation of cysteinyl-t-RNA is an exception, which will be discussed. No evidence of mis-recognition at the amino acid acceptance level by overmethylated purified phenylalanyl-t-RNA was found.

The ability of purified phenylalanyl-t-RNA to bind to ribosomes (Nirenberg & Leder, 1964) was unimpaired by levels of methylation that inhibited its accepting ability by 55%. The formation *in vitro* of polyphenylalanine, polylysine and polypyrroline was considerably more sensitive to over-methylation of t-RNA than the corresponding amino acid-acceptance function. This suggests that the presence of additional methyl groups interferes with an activity of t-RNA other than amino acid acceptance or ribosome binding.

The use of a modifying method with considerable specificity for a particular base allows an attempt at interpretation of the biological effects of modification in terms of the involvement of defined areas in t-RNA. On this basis, some correlation has been attempted between modification of the anticodon and inhibition of amino acid acceptance: Weil, Befort, Rether & Ebel (1964) found no such correlation in yeast t-RNA. There is considerable evidence for and against the involvement of the anticodon region in aminoacyl-t-RNA synthetase recognition (e.g. Miura, 1967; Carbon & Curry, 1968) but recent work shows that such involvement is not a universal feature of the recognition system (Thiebe & Zachau, 1968; Geffer & Russell, 1969). This aspect will be fully discussed.

We are grateful to Dr D. Novelli, Oak Ridge National Laboratory, for a gift of phenylalanyl-t-RNA. The work outlined here was carried out with Dr D. J. Fillinger and Professor Ernest Borek.

We acknowledge support from the Damon Runyon Foundation.

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