Specific precipitation of polyribosomes, particularly from the 200S region of the gradient, was achieved in TMK buffer with F(ab')₂ fragments of the antibodies. Pretreatment with puromycin resulted in decreased precipitation of polyribosomes by specific F(ab')₂ fragments.

A rapidly labelled RNA was extracted from the precipitated polyribosome-antibody complexes. Its sedimentation coefficient was 11-12S and its base composition was 48mol of G+C/100mol. The size and base composition of this RNA are consistent with its being the mRNA for the immunoglobulin light chain synthesized by MOPC 149 plasmacytoma.

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The Occurrence of 5.8S Ribonucleic Acid in Plants

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A 5.8S RNA species has been shown to be present in several flowering plants, a fern, a club-moss and a green alga, and has several properties in common with the corresponding molecule (also described as 7S, 25S A and 'I' RNA) from animal cells (Pene, Knight & Darnell, 1968; Sy & McCarty, 1970; King & Gould, 1970). First, plant and animal 5.8S RNA species are very similar in size. We found that the plant RNA species varied in molecular weight between 49800 (*Tussilago farfara*) and 51600 (*Selaginella kraussiana*) as determined by polyacrylamide-gel electrophoresis, the mean value being 50400. Secondly, it is associated with the largest (25S) rRNA in equimolar amounts. Thirdly, this complex may be dissociated by conditions that disrupt hydrogen bonds. With the broad-bean RNA we have shown that dissociation by heat occurs within a narrow temperature range (37-52°C in a buffer containing no Mg²⁺ and of low ionic strength). Similar findings have been reported for the animal RNA (Sy & McCarty, 1970; King & Gould, 1970). It is therefore probable that 5.8S RNA is a universal component of cytoplasmic 80S ribosomes.

The 5.8S RNA was recovered in equimolar amounts with 5S RNA from broad-bean roots, but in green leaves it was found in submolar amounts. The relative proportions of 5S and 5.8S RNA in

leaves of dark-grown plants were similar to those in roots, but when such plants were exposed to light, which stimulated the synthesis of chloroplast ribosomes, the proportion of 5.8S RNA progressively decreased until the relative amounts were comparable with those in leaves of light-grown plants. These observations suggested that chloroplast 70S ribosomes do not contain 5.8S RNA, and this finding was confirmed by heating and cooling the RNA fraction from purified chloroplasts prepared from broad-bean and spinach leaves.

As it has also been reported that 5.8S RNA could not be detected in *Escherichia coli* preparations (Pene *et al.* 1968; Sy & McCarty, 1970), we examined two other prokaryotes, the blue-green algae *Nostoc* and *Anabaena flosaquae*, and similarly could not detect this component. The 5.8S RNA is therefore probably not a component of the 70S class of ribosomes, but is restricted to eukaryotic 80S ribosomes. Our findings demonstrate a further similarity in chloroplast and prokaryotic ribosome structure and provide additional support for the endosymbiont theory of the origin of chloroplasts (Margulis, 1970).

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Mutagenicity of 'K-Region' Epoxides of Polycyclic Hydrocarbons towards Bacteriophage T₂

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The microsomal metabolism of polycyclic hydrocarbons is now known to involve the formation of epoxide intermediates that are probably 'K-region' derivatives (Sims, Hewer & Grover, 1971). Synthetically prepared 'K-region' epoxides have been found to be alkylating agents that react with nucleic acid either *in vitro* (Grover & Sims, 1970) or in cells in culture (Grover, Forrester & Sims, 1971). Polycyclic hydrocarbon epoxides are therefore particularly interesting as potential mutagens, and we have now tested a series of these compounds for activity in producing plaque-type and host-range mutations of bacteriophage T₂ (Loveless, 1959).

In the host-range-mutagenesis assay, where buffered suspensions of bacteriophage T₂ were

treated for 2–3 h with dimethyl sulphoxide solutions of the epoxides, 3-methylcholanthrene 11,12-oxide, dibenz[*a,h*]anthracene 5,6-oxide and 7-methylbenz[*a*]anthracene 5,6-oxide were found to be strongly mutagenic. Under these conditions phenanthrene 9,10-oxide gave a very slight mutagenic effect, but chrysene 5,6-oxide and benz[*a*]anthracene 5,6-oxide were inactive. Similar results were obtained when bacteriophage were treated with these epoxides for longer periods and the frequency of plaque-type mutants was determined. The active epoxides were highly effective mutagens when compared with ethyl methanesulphonate, which was used as a positive control (Loveless, 1959). It may also be significant that only those 'K-region' epoxides that are derived from potent carcinogenic hydrocarbons were strongly mutagenic to bacteriophage in these tests. Some possible mechanisms of action of polycyclic hydrocarbon epoxides as mutagens will be discussed in relation to the role that these metabolic intermediates may play in hydrocarbon carcinogenesis.

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Binding of Polylysine to Chromatin Extracted from Various Tissues

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Previous studies on the accessibility of the DNA component of rat thymus chromatin have shown that about 40% of the DNA phosphate groups are available for binding poly-L-lysine hydrobromide of various molecular weights (Itzhaki, 1970, 1971a). This work has now been extended to the DNA-protein complex extracted from a variety of sources.

Mixtures of DNA-protein and polylysine in 0.7 mM-sodium phosphate buffer, pH 6.9, were made over a range of DNA-protein concentrations such that all the DNA-protein was precipitated. Estimates were then made of the excess of unbound polylysine by titration with Methyl Orange.

It has been found that, in chromatin from the livers of normal rats and from rats treated with dimethylnitrosamine, in calf thymus nucleohistone and in avian erythrocyte chromatin, 40–44% of the DNA phosphate groups are available for binding

polylysine, and these are present in stretches sufficiently long to bind most or all of each polylysine molecule. In DNA-protamine from salmon sperm about 24% of the DNA phosphate groups are available for binding, but these free phosphate groups are present only in very small stretches.

Attempts were made to find if there was appreciable binding of polylysine to the chromatin protein carboxyl groups. Two methods were used: first, the binding to rat thymus chromatin was measured at pH 4.5 so as to decrease the dissociation of the protein acidic groups; secondly, the binding to a precipitate consisting mainly of chromatin protein was measured. Both methods showed that the binding of polylysine to the chromatin protein was negligible.

On the basis of the previous studies with polylysine and other studies on the effect of added nucleases on chromatin (Itzhaki, 1971b) it was proposed that the DNA in chromatin is extensively covered by protein but that there are many zones of free DNA beneath the protein. The present results suggest that the structure of chromatin from relatively active cells is similar to that of chromatin from the inactive avian erythrocyte. In fact, calculation of the ratio of protein basic groups to DNA phosphate groups in the extracts shows that the value is lower rather than higher in the DNA-protein complex from the avian erythrocyte and from sperm. It is concluded that any repression of gene activity by histones is unlikely to be due to a simple coverage of the DNA.

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Formation of Polydeoxyadenylylthymidylate after Deoxyribonucleic Acid Synthesis by a Deoxyribonucleic Acid Polymerase from *Mycobacterium smegmatis*

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When the DNA polymerase from *Mycobacterium smegmatis* is incubated with a small amount of primer DNA and an excess of deoxynucleoside triphosphates, synthesis of DNA stops before 50% 'replication' of the primer has taken place. However, this is followed by a phase of exponential incorporation of thymidine, and evidence has been obtained that this is due to synthesis of poly-[d(A-T)] (McNulty & Winder, 1972). The following results support this conclusion.

The polymerase was purified 500-fold from iron-limited *M. smegmatis*, as will be described elsewhere.