cally effective dose of chlorambucil (Leukeran) (8mg/kg body wt.) an increased incorporation of thymidine into the DNA of sensitive cells was observed. The rates of RNA and total protein biosynthesis were not affected by drug treatment (Hill & Harrap, 1970).

In the present studies the effects of chlorambucil on the nuclear proteins of Yoshida cells have been investigated. Cells were harvested from animals at various time-intervals after treatment with chlorambucil (8mg/kg body wt.). Histone was extracted in 0.25M-hydrochloric acid from chromatin obtained after saline washing of isolated nuclei. The remaining protein, after histone extraction, is referred to as non-histone protein. Changes in the histone/DNA and non-histone protein/DNA ratios were seen only in the drug-sensitive cells: the histone/DNA ratio fell, whereas the non-histone protein/DNA ratio increased. Electron micrographs of sensitive treated cells have shown that these changes were accompanied by 'giant-cell' formation and an altered appearance of the chromatin. Dense material (heterochromatin), seen in untreated sensitive cells, disappeared after treatment, becoming more diffuse (probably euchromatin). No alteration in the chromatin of resistant cells was observed after treatment.

The rate of incorporation of [^{14}C]leucine into histone and non-histone protein has shown an inhibition of synthesis of both proteins, after treatment, in the drug-sensitive cells. This observation, together with a concurrent increase in DNA synthesis, would account for the low histone/DNA ratio. The increase in non-histone protein/DNA ratio, however, cannot be accounted for in this way, and might therefore suggest a reorganization of previously synthesized material.

Since total protein synthesis was not affected under these conditions, chlorambucil appears to have a specific effect in inhibiting nuclear protein synthesis.

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Isolation of Messenger-Like Ribonucleic Acid from Immunochemically Precipitated Polyribosomes

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Specific antibodies to a given protein have been used to precipitate polyribosomes synthesizing that protein. For example, the technique was used for the identification of polyribosomes synthesizing the heavy chains and light chains of immunoglobulin G (Williamson & Askonas, 1967) and for the isolation of mRNA for α -glucosidase (Hartlief & Koningsberger, 1968). However, this study showed that precipitation of intact polyribosomes by whole antibodies was not specific, owing to binding of immunoglobulin G through the Fc portion to ribosomes.

Polyribosomes of MOPC 149 plasmacytoma, which synthesized an immunoglobulin light chain, were labelled *in vivo* with sodium [^{32}P]phosphate and their nascent peptides incorporated ^{14}C -labelled amino acids during incubation of the tumour cells *in vitro*. Cell disruption and polyribosome fractionation occurred in one step on a sucrose density gradient (Davis, Delovitch & Sehon, 1969). Fractions from the gradient, or suspensions of the total polyribosomes, were treated with either rabbit antiserum to MOPC 149 light chain (specific) or normal rabbit serum (control). The optimum amount of sheep antiserum to rabbit immunoglobulin G was then added to cause complete precipitation.

Specific and control antisera precipitated similar amounts of ^{32}P -labelled components from each polyribosome fraction when precipitation occurred in TMK buffer [10mM-tris-HCl buffer (pH 7.3)–3mM-magnesium chloride–15mM-potassium chloride). Removal of nascent peptides with puromycin had no effect on the precipitation of ^{32}P -labelled components. Both antisera precipitated similar amounts of ^{32}P - and ^{14}C -labelled components from a suspension of total polyribosomes in TMK buffer. In contrast, in phosphate-buffered saline [20mM-phosphate buffer (pH 7.4)–0.14M-sodium chloride] the ^{14}C -labelled components in the control precipitates were decreased. However, in phosphate-buffered saline ^{32}P -labelled components were absent from both precipitates, indicating degradation of ribosomes.

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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