information of value. This communication attempts to summarize present findings.

Eukaryotic chromosomes are composed of DNA, histones and non-histone ('acidic', 'residual') proteins. DNA and histones occur in relatively equivalent and, at least for DNA, in constant amounts per nucleus. Histones in all species examined vary little from the five types: lysine-rich histone F1, slightly lysine-rich histone F2b and arginine-rich histones F2a1, F2a2 and F3 (Butler, Johns & Phillips, 1968). Amino acid sequences are known for histones F2b (Iwai, Ishikawa & Hayashi, 1970) and F2a1 (DeLange, Fambrough, Smith & Bonner, 1969; Ogawa *et al.* 1969) and part sequences for histone F1 (Rall & Cole, 1970).

On the basis of knowledge about proteins for which complete structures are known (Prothero, 1966; Schiffer & Edmundson, 1967), the sites of α-helix in histones F2b and F2a1 have been predicted and suggestions for their structural arrangement in the deoxyribonucleohistone complex have been made (Richards & Pardon, 1970; Boublik, Bradbury & Crane-Robinson, 1970).

X-ray-diffraction studies of fibre and gel specimens of deoxyribonucleoprotein indicated that the DNA in the complex is coiled (deoxyribonucleoprotein supercoil) into a regular structure with pitch 120 Å and diameter 100 Å. At very low concentrations in solutions lacking bivalent ions the deoxyribonucleoprotein supercoil appears to be absent. As the deoxyribonucleoprotein concentration increases, or on the addition of Mg^{2+} ions (concentration above 1 mM), the supercoil appears, indicating that some of the forces responsible for generation of this configuration may be intermolecular. Intramolecular forces are very important in maintaining the supercoil, since partial removal of histone by dissociation of the complex in salt (sodium chloride at above 1.2 M concentration or magnesium chloride at above 0.3 M concentration) results in the loss of supercoiling and successful reconstitution of the complex from histones is dependent on the method by which the protein was prepared. The supercoil configuration is, however, the form in which most and possibly all the DNA of the eukaryotic chromosome is to be found.

Molecular models constructed from space-filling CPK (Ealing Corporation, Cambridge, Mass., U.S.A.) components can be built to investigate the likely arrangement of the histone on the supercoiled DNA. The model for histone F2a1 gives a value of 0.88:1 for the mass ratio protein/DNA, which agrees with the calculated value for native nucleohistone lacking histone F1, when histone F2a1 is attached only in the large groove of the DNA. This implies that access for sequence-recognizing molecules might be via the small groove in the DNA. Further implications of the model for the mechanics of genetic de-repression will be discussed.

Electron microscopy of cell nuclei and metaphase chromosomes shows that chromatin consists of 'unit' threads having a diameter of approx. 170–250 Å in sectioned (Davies, 1968) or thread preparations in the presence of bivalent ions, and approx. 100 Å in preparations treated with chelating agents (Ris, 1969).

In the electron microscope the absence of water prevents the deoxyribonucleohistone complex from assuming the supercoil configuration. Thus direct comparison between the results of electron microscopy and X-ray diffraction of wet specimens is not possible.

Boublik, M., Bradbury, E. M. & Crane-Robinson, C. (1970). *Eur. J. Biochem.* **12**, 258.
Butler, J. A. V., Johns, E. W. & Phillips, D. M. P. (1968). *Prog. Biophys. molec. Biol.* **18**, 211.
Davies, H. G. (1968). *Proc. 4th Eur. Regional Conf. Electron Microscopy, Rome*, p. 175.
DeLange, D. J., Fambrough, D. M., Smith, E. L. & Bonner, J. (1969). *J. biol. Chem.* **244**, 319.
Iwai, K., Ishikawa, K. & Hayashi, H. (1970). *Nature, Lond.*, **226**, 1056.
Ogawa, Y., Quagliarotti, G., Jordan, J., Taylor, C. W., Stanbuck, W. C. & Busch, H. (1969). *J. biol. Chem.* **244**, 4387.
Prothero, J. W. (1966). *Biophys. J.* **6**, 367.
Rall, S. C. & Cole, R. D. (1970). *Fedn Proc. Fedn Am. Socs exp. Biol.* **29**, 535 abs.
Richards, B. M. & Pardon, J. F. (1970). *Expl Cell Res.* **62**, 184.
Ris, H. (1969). In *The Handbook of Molecular Cytology*, p. 222. Ed. by Lima-de-Faria, A. Amsterdam: North-Holland Publishing Co.
Schiffer, M. & Edmundson, A. B. (1967). *Biophys. J.* **7**, 121.

Control of Transcription of Chromatin Deoxyribonucleic Acid

By A. J. MacGillivray and J. Paul. (*Beatson Institute for Cancer Research, Royal Beatson Memorial Hospital, 132 Hill Street, Glasgow C.3, U.K.*)

The mechanism of protein synthesis appears to be basically similar in many forms of life. However, the composition of DNA and the control of the expression of its genetic information for protein synthesis varies considerably between eukaryocytes and prokaryotes. Apart from being present in large amounts, the DNA of mammalian cells is heterogeneous in its composition, since it contains sequences that are highly repetitious and those that are not (Britten & Kohne, 1968). In addition, in eukaryotes the DNA is complexed with protein to form chromosomes that are housed in the nucleus separate from the protein-synthesizing machinery of the cytoplasm. The differentiated cells of an

animal appear to contain the same genetic information (Gurdon & Laskey, 1970), but the populations of RNA transcribed vary from tissue to tissue. In prokaryote cells we know that transcription can be regulated through positive controls (RNA polymerase factors) and by negative means (protein repressors). These mechanisms may be present in eukaryotes, but the situation in higher organisms suggests that other types of transcriptional control may be necessary.

Much effort has recently been applied to the study of the biochemical properties of chromatin, the deoxyribonucleoprotein complex isolated from nuclei. Work carried out in this and other laboratories has shown that chromatin is less efficient than DNA as a primer for RNA synthesis *in vitro*, and moreover the sequences of DNA available for transcription in chromatin are restricted in an organ-specific manner. Several approaches have been used in attempts to resolve the mechanism of these effects. The addition of histones to DNA or their removal from chromatin indicate that these proteins can inhibit DNA as a primer for RNA synthesis *in vitro*. DNA–RNA hybridization studies show that the non-histone fraction of chromatin also restricts the DNA available for transcription. Further evidence has been obtained from experiments in which chromatin has been reassembled from its major components, i.e. DNA, histones and the non-histone fraction. From DNA–RNA hybridization results it appears that histones prevent the transcription of DNA, whereas the presence of the non-histone fraction is required in order to regain a template specificity similar to that of native chromatin (Paul & Gilmour, 1968; Gilmour & Paul, 1969). Further, the non-histone fraction is responsible for the organ-specific properties of the chromatin template (Gilmour & Paul, 1970; Spelsberg & Hnilica, 1970). Although the hybridization techniques are limited to dealing with only the repetitious sequences of DNA, it does appear that the non-histone fraction is associated with certain aspects of the control of transcription.

Analysis of the non-histone fraction shows it to consist largely of acidic protein together with some RNA. Bekhor, Kung & Bonner (1969) and Huang & Huang (1969) have presented evidence that the recognition of the DNA sequences available for transcription in chromatin is associated with a small RNA (chromosomal RNA) bound to acidic protein. In our laboratory non-histone protein fractions containing approx. 1% of RNA have been found to give reconstituted chromatins with templates similar to that of native chromatin. Hyden & Zachau (1971) have shown that chromosomal RNA may be derived from tRNA. The importance of the non-histone proteins themselves in regulating chromatin template activity is indicated by the synthesis of specific non-histone proteins after steroid treatment (Shelton & Allfrey, 1970; Teng & Hamilton, 1970). Non-histone proteins, when added to chromatin *in vitro*, alter the template, indicating that they have made new DNA sequences available for transcription (Kamiyama & Wang, 1971).

There appears to be little tissue or species specificity of the major non-histone proteins of chromatin (Elgin & Bonner, 1970; MacGillivray, Carroll & Paul, 1971), this probably being associated with the fact that many proteins, e.g. RNA polymerase, are common to all chromatins. Hence, to detect specificity-determining proteins, DNA-binding studies have been initiated. These indicate that species-specific proteins are detectable in chromatin non-histone protein preparations (Kleinsmith, Heidema & Carroll, 1970; Teng, Teng & Allfrey, 1970).

A number of investigations have shown that chemical modification of chromatin proteins, e.g. acetylation, methylation, phosphorylation and oxidation of thiol groups, could be associated with de-repression mechanisms. In general the systems studied have been those in which large changes in nuclear activity would be expected to take place, e.g. after hepatectomy. In more discrete examples of gene activation, e.g. during 'puff' formation in polytene chromosomes, such modifications appear unlikely to occur (Clever & Ellgaard, 1970).

Bekhor, I., Kung, G. M. & Bonner, J. (1969). *J. molec. Biol.* **39**, 351.
Britten, R. J. & Kohne, D. E. (1968). *Science, N.Y.*, **161**, 529.
Clever, U. & Ellgaard, E. G. (1970). *Science, N.Y.*, **169**, 373.
Elgin, S. C. R. & Bonner, J. (1970). *Biochemistry, Easton*, **9**, 4440.
Gilmour, R. S. & Paul, J. (1969). *J. molec. Biol.* **40**, 137.
Gilmour, R. S. & Paul, J. (1970). *FEBS Lett.* **9**, 242.
Gurdon, J. B. & Laskey, R. A. (1970). *J. Embryol. exp. Morph.* **24**, 227.
Huang, R. C. & Huang, P. C. (1969). *J. molec. Biol.* **39**, 365.
Hyden, H. W. & Zachau, H. G. (1971). *Biochim. biophys. Acta*, **232**, 651.
Kamiyama, M. & Wang, T. Y. (1971). *Biochim. biophys. Acta*, **228**, 563.
Kleinsmith, L. J., Heidema, J. & Carroll, A. (1970). *Nature, Lond.*, **226**, 1025.
MacGillivray, A. J., Carroll, D. & Paul, J. (1971). *FEBS Lett.* **13**, 204.
Paul, J. & Gilmour, R. S. (1968). *J. molec. Biol.* **34**, 305.
Shelton, K. R. & Allfrey, V. G. (1970). *Nature, Lond.*, **228**, 132.
Spelsberg, T. C. & Hnilica, L. S. (1970). *Biochem. J.* **120**, 435.
Teng, C. S. & Hamilton, T. H. (1970). *Biochem. biophys. Res. Commun.* **40**, 1231.
Teng, G. T., Teng, G. S. & Allfrey, V. G. (1970). *Biochem. biophys. Res. Commun.* **41**, 690.