(4) The DNA is assumed to be in the super-coil configuration (Pardon, Wilkins & Richards, 1967; J. F. Pardon & M. H. F. Wilkins, unpublished work).

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The Structure of Deoxyribonucleohistone and the Role of Individual Histone Proteins

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In studies of the reconstruction of the nucleoprotein complex from separated histones and DNA the characteristic X-ray-diffraction pattern can be used as the criterion for the recovery of the correct molecular configuration. In this way we can test for the nativity of mixtures and individual histone fractions isolated by both physical and chemical means.

Histones produced by salt dissociation of nucleohistone are native by the stated criterion, but fractionation is virtually non-existent except for the separation of lysine-rich histone F1 (Ohlenbusch, Olivera, Tuan & Davidson, 1967). Similarly attempted fractionation by acid titration (Murray, Vidali & Neelin, 1968) is only partially successful, but nevertheless yields mixtures of native histones provided that they are sulphates precipitated with ethanol.

Individual histones fractionated by selective chemical separation or separation by column chromatography are being investigated for their ability to produce native material. Thus far the original mixture of five histones has been reduced to a mixture of three fractions that have undiminished ability to regenerate the super-coil configuration of nucleohistone (Pardon, Wilkins & Richards, 1967).

Progress towards a two-component nucleohistone is essential for the full elucidation of the interaction between any given histone molecule and DNA at the atomic level. Until this is achieved the application of crystallographic procedures remains severely limited.

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The Isolation of Rapidly Labelled Deoxyribonucleic Acid Fragments of Nuclear Origin from the Polyribosome Fraction of Cultured Embryonic Liver Cells

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During experiments designed to isolate low-molecular-weight RNA species, it was found that a large amount of DNA accumulates in the cytoplasm of cultured embryonic liver cells. Mouse 14½-day embryonic livers were disaggregated and cultured in the presence of [³²P]phosphate for 16 h at 37°C. The cells that do not stick to glass, predominantly erythroid in type, were isolated and lysed with a hypo-osmotic buffer. Total nucleic acid was isolated either from the postmitochondrial supernatant or from isolated polyribosomes; the results were apparently similar with regard to DNA in both cases.

Up to 20% of the total DNA was found in the cytoplasm. The base compositions of nuclear and cytoplasmic DNA were identical with each other and with established values for mouse cells. The nuclear and cytoplasmic DNA both had a buoyant density indistinguishable from that of native total embryonic DNA, demonstrating not only the identity of base composition but also that the cytoplasmic DNA is double-stranded.

Electrophoresis of the DNA on 2.4% polyacrylamide gels gave a regular series of bands roughly from the position of 5S to the top of the gel. At least ten bands can be detected by the eye. The four smallest DNA components were isolated by preparative polyacrylamide-gel electrophoresis, and their sedimentation coefficients were determined and molecular weights calculated. The DNA fragments appear to be multiples of a unit molecular weight approx. 150 000.

The specific radioactivity of the DNA from the cytoplasm was 1.3–2-fold higher than that of the nucleus, and the radioactivity was non-terminal, thus ruling out the introduction of [³²P]phosphate via a phosphokinase mechanism during degradation. The DNA of one band (molecular weight approx. 300 000) has been purified, depurinated and 'finger-printed' and is definitely not of unique base sequence.

It is thought that this DNA may be similar to that reported from a cultured embryonic muscle-cell system by Bell (1969). However, certain of the experimental results outlined here do not fully accord with his conclusions.