Mouse bulk and n.e.-DNA's were also compared by analytical CsCl density gradient centrifugation. Bulk DNA banded as a sharp zone of peak density 1.701 g/cm$^3$ with satellite at 1.690 g/cm$^3$. 4 samples of n.e.-DNA banded as broader zones with peak density at 1.705 g/cm$^3$ and without a separate satellite. Low M. W. bulk DNA also banded in this way. In these cases it was not possible to accurately estimate the satellite content of mouse n.e.-DNA, but it was obviously not greatly enriched in this DNA, relative to bulk.

The present results reinforce the view that the DNA associated with isolated nuclear envelopes is to some extent a distinct sub-fraction of the total DNA. It is characterized by its close association with the envelope and by moderate enrichments in late-replicating[4,5] and repetitive DNA sequences and presumably also by an enrichment in sequences most intimately associated with the envelope in vivo. These results are consistent with the n.e.-DNA being derived from peripheral heterochromatin, a conclusion also reached by FRANKE et al[12]. Finally it seems that the association of this peripheral heterochromatin with the envelope is mainly mediated by ionic forces.

*Zusammenfassung.* Nachweis, dass aus Ratten- und Mausleberzellen isolierte, an Zellkernmembrane gebundene DNS, im Vergleich zum totalen Zellkern-DNS besonders reich an wiederholenden Sequenzen waren. Diese DNS stammt vermutlich aus peripherem Heterochromatin und ist an die Membran hauptsächlich durch Ionen gebunden.

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## The Localization of Adenosine Triphosphatase Activity in the Alveolar Macrophages of Hamster Lung

The alveolar macrophages of mammalian lung are known to contain a variety of hydrolytic enzymes[1–4] although the precise intracellular location of many of these enzymes has yet to be determined. In this study the adenosine triphosphatase (ATPase) activity in the alveolar macrophages of hamster lung was localized using an electron cytochemical technique.

*Materials and method.* Young adult hamsters were killed by cervical dislocation and small blocks of lung tissue were excised and fixed for 90 min in ice-cold 2.5% glutaraldehyde in cacodylate buffer, pH 7.3. The blocks were washed for 30 min in cacodylate buffer containing 0.15 *M* sucrose and then cut into sections (about 0.5 mm thick) with a razor blade. The sections were incubated for 20–30 min in a lead capture medium at 37 °C. The medium consisted of 11 ml of 0.2 *M Tris*-maleate buffer (pH 7.4), 11 ml of 2.5 m*M* ATP (Sigma London), 2 ml of 0.1 *M* magnesium sulphate, and 1 ml of 10 m*M* lead nitrate. Control sections were incubated in a substrate-free medium or, alternatively, in a medium in which ATP had been replaced by an equimolar quantity of Na $\beta$-glycero-phosphate. After incubation all sections were post-fixed for 45 min in buffered 1% osmium tetroxide, rapidly dehydrated in absolute ethanol, and embedded in Araldite[5]. Ultrathin sections were cut on a Reichert ultramicrotome and viewed without further staining in an AEI 801 electron microscope at an accelerating voltage of 60 kV.

*Observations.* Enzymic hydrolysis of ATP in the presence of free lead ions results in an insoluble electron-opaque deposit of lead phosphate. In tissues incubated for 30 min or longer a fine particulate deposit of lead phosphate was present in the plasma membranes of the alveolar macrophages. Although membranous structures are generally poorly delineated in sections of unstained material, sufficient contrast was present in the electron micrographs to confirm that the reaction product was associated with the outer leaflet of the plasma membrane. The intensity of the reaction was greatest over the surface of pseudopodia (Figure 1). A moderately intense reaction was present in the membranes of phagocytic and micropinocytotic pits. Many of the digestive vacuoles and dense bodies were labelled (Figure 2) but the mitochondria and Golgi elements were unreactive even after prolonged incubation (120 min). Non-specific deposition of lead salts was commonly seen within the nuclei of macrophages. Control sections incubated in a medium lacking the substrate (ATP) or in a medium in which ATP had been replaced by Na $\beta$-glycerophosphate showed no reaction.

*Discussion.* In this study vigorous hydrolysis of ATP was observed on the plasma membranes of alveolar macrophages, especially in areas where pseudopodia or phagocytic and micropinocytotic pits are present. The absence of reactivity in control incubations where Na $\beta$-glycerophosphate was substituted for ATP in the medium suggests that the hydrolysis of ATP was due in normal circumstances to the action of a specific ATPase rather than a non-specific phosphomonoesterase.

In previous cytochemical studies ATPase has been observed on the microvilli of intestinal epithelium[6], the endothelium of capillaries[7], the basal infoldings of cell membranes in kidney tubules[8], and the microvilli of bile canaliculi[9]. In each of these sites extensive transfer of fluid is known to take place. Furthermore, strong ATPase activity has been detected within motile cell processes[10,11]. It is probable therefore that much of the ATPase contain-

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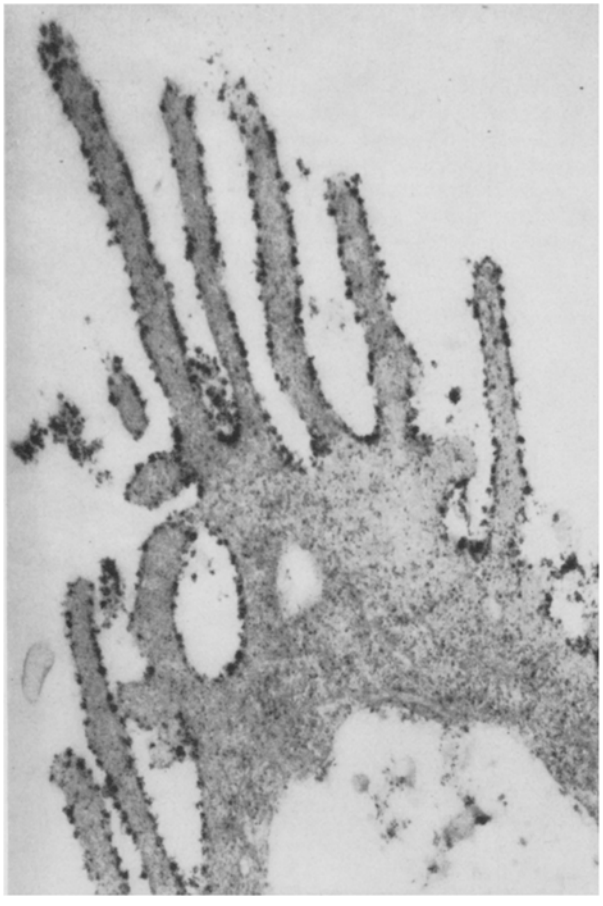

Fig. 1. ATPase reaction in the pseudopodia of an alveolar macrophage. Incubation time 60 min. ×38,000.

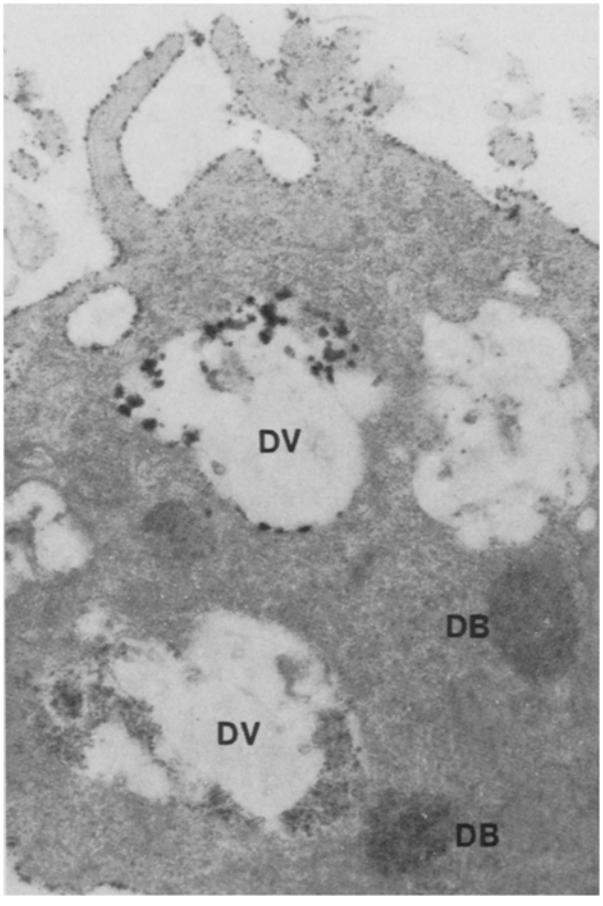


Fig. 2. ATPase reaction in the digestive vacuoles (DV) and dense bodies (DB) of an alveolar macrophage. Incubation time 60 min. ×50,000.

ed within alveolar macrophages is responsible for the active movements of the surface membranes which occur when the cells migrate or engulf foreign material.

*Zusammenfassung.* Durch eine elektronenmikroskopische Technik konnte bei Alveolarmakrophagen der Hamsterlunge die intrazelluläre Lokalisation der ATPase-Aktivität bestimmt werden. Dabei zeigte die Plasmamembran der Makrophagen eine hohe ATPase-Aktivität, während in den Vakuolen und in den elektronendichten Körperchen der Phagozyten eine geringere Reaktion vorgefunden wurde.

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## Presence of the Same Types of Nonhistone Chromosomal Proteins in Different Tissues

It has been proposed that nonhistone chromosomal proteins (NHC proteins) act as the specific regulators of gene expression (for review see [1]). In support of this proposal a considerable tissue specificity in the electrophoretic patterns of NHC proteins has been reported by many authors[2–9]. Since only a small fraction of the genome is active in any given type of cells[10–13], such markedly expressed tissue specificity of NHC proteins was surprising. Therefore, we decided to reexamine the question of the tissue specificity of the total NHC proteins.

*Material and methods.* Chromatin was prepared from isolated pure nuclei and the total chromosomal proteins were analyzed by polyacrylamide gel electrophoresis in a system which separated histones and NHC proteins on the same gel. The nuclei were isolated from liver, cerebral cortex, cerebellum and a mixture of midbrain and brainstem of female albino rats derived from Holtzman strain. In addition nuclei were also prepared from white leghorn chicken embryos in developmental stage 18 and from CHANG's rat ascites hepatoma[14]. From rat organs and