

Résumé. Nous avons enregistré 88 afférences respiratoires du nerf vague chez le poulet. Quelques unes se déclenchaient au cours de l'inspiration eupnéique, d'autres au cours de l'expiration. Jusqu'à présent, on a l'impression qu'il existe plus de types d'activité afférente respiratoire chez l'oiseau que chez le mammifère, mais de nouvelles analyses seront nécessaires pour mieux les préciser. Nous croyons que c'est ici la première preuve évidente de l'existence d'un contrôle afférent vagal de la respiration chez l'oiseau.

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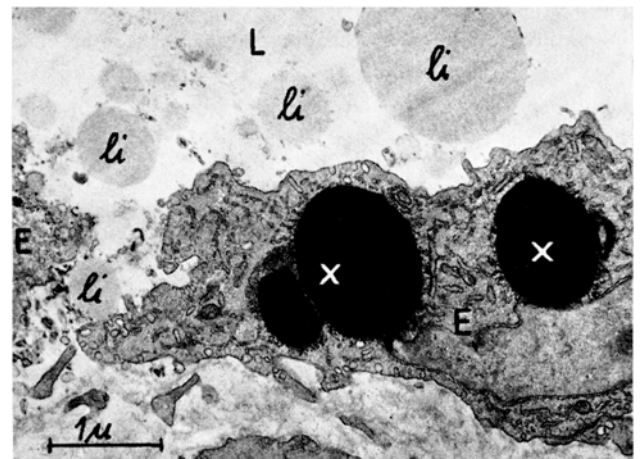
An Electron Microscopy Study of Fat Uptake by Endothelial Cells of Doubly-Ligated Carotid Artery Segments

Earlier light microscopic studies by FRIEDMAN et al.¹ demonstrated large accumulations of lipid in the form of droplets within the endothelial cells of the doubly-ligated rabbit carotid artery, into whose lumen had been injected suspensions of rat thoracic lymph chylomicrons. We felt that a similar study with this model system, in which fat injections into the vessel lumen simulated hyperlipemia while double-ligation induced hypoxia, but using the electron microscope, might give additional information on the question of lipid uptake and transport into and across the endothelial lining of arteries under normal and hypoxic conditions, a problem possibly related to the pathogenesis of atherosclerosis.

The right external carotid artery of ether anaesthetized rabbits was doubly-ligated and the lumen of the ligated segment filled with a suspension of the artificial fat emulsion intralipid (20%, Vitrum, Stockholm) using a fine hypodermic needle. After 3 days animals were sacrificed and the ligated segment removed and prepared for electron microscopy.

No intimal thickening was observed 3 days following double-ligation. The condition of the ligated segment's endothelial lining varied from experiment to experiment. Some areas were necrotic and even denuded while others were quite intact. The latter probably occurred around areas in which deeper penetration of vasa vasorum into the inner media of the occluded segment had commenced enabling oxygen to diffuse to the lining. These intact areas were investigated since we were mainly interested in the effects of hyperlipemia and hypoxia rather than anoxia on the arterial endothelia. As is illustrated in the micrograph, the lumen is filled with the round, grey-toned intralipid particles. Some are seen passing into a gap between the endothelial cell illustrated and a necrotic neighbour on the left. Striking is the high electron-density of the lipid droplets found in all endothelial cells observed. Such massive accumulation of intracellular lipid droplets was not found in previous studies² nor in our controls. No evidence for the mechanism of lipid uptake into the endothelial cells could be found such as via pinocytotic vesicles, although the intralipid spheres often formed smaller particles when in contact with the endothelial surface. They might then be more easily attacked by lipases and transported into the cell interior in the form of smaller metabolites. The increase in electron-density of the intracellular droplets relative to the intralipid particles might be due to a metabolic transformation of the intracellular lipid to a more unsaturated and hence more electron-dense (osmiophilic) form. This would appear more likely than a preferential uptake of phospholipids, found on the surface of the intralipid particles³ and functioning as an emulsifier for the hydrophobic triglyceride core. The phospholipids would probably form myelin figures intracellularly rather

than the dark diffuse droplets seen in these experiments. Why the electron-dense rim of the intralipid particles is seen *in vitro*³ but not *in vivo*, as in this and other studies⁴, remains unclear.



Electron micrograph: endothelial lining of the rabbit carotid artery 3 days following double ligation and intralipid injection. Note light lipid droplets (li) in lumen (L), and dark lipid droplets (x) in endothelial cells (E).

Zusammenfassung. Die Endothelschicht von doppelt unterbundenen Abschnitten der A. carotis des Kaninchens wird 3 Tage nach der Unterbindung und Injektion von Intralipid (künstliche Fett-Emulsion) in das Lumen elektronenmikroskopisch untersucht. Die künstliche Hyperlipämie zusammen mit Hypoxie erzeugte eine starke Aufnahme von Fett in Form von intrazellulären Tröpfchen, die viel elektronendichter waren als die ursprünglichen Intralipid-Partikel. Ein morphologischer Hinweis auf den Mechanismus der Fettaufnahme wurde nicht gefunden.

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¹ M. FRIEDMAN, S. O. BYERS and S. ST.-GEORGE, *Am. J. clin. Path.* 45, 238 (1966).

² H. A. HACKENSELLNER, H. DAVID and I. UERLINGS, *Acta biol. med. germ.* 14, 34 (1965).

³ G. I. SCHOEFL, *Proc. R. Soc. B* 169, 147 (1968).

⁴ G. I. SCHOEFL and J. E. FRENCH, *Proc. R. Soc. B* 169, 153 (1968).