Zoological Studies 34, Supplement I (1995)

Transport Pathways of Lipoproteins across the Arterial Endothelial Cells

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Endothelial injuries in the artery have long been considered as an initial event in the pathogenesis of atherosclerosis. Increasing arterial endothelial permeability can cause low density lipoproteins (LDL) to filtrate and accumulate in the intima (Ross 1993). However, the nature of the initial damage to the arterial wall is not fully understood. Our previous studies showed that the endothelial ultrastructure of the branched regions of aortic arch was quite different from that of the unbranched regions (Kao et al. 1994). In this study, we use fluorescein
(DiL) labeled LDL to compare the distribution of LDL in the different regions of the arteries in the hypercholesterolemic rats. We also use colloidal gold labeled LDL to investigate *in situ* the transport pathways of LDL across the arterial endothelium at the branched regions of the arteries. Rat or human LDL was separated from pooled plasma by differential centrifugation. Then, LDL was coupled to DiI or colloidal gold (diameter =10 nm). Male Sprague-Dawley rats, weighing approximately 250 gm were fed high-cholesterol diet over a period of 1 year. At 1, 3, 6 and 12 months after feeding, three to five animals were anesthetized. The thoracic and the abdominal aorta, the right and the left femoral arteries were cannulated with polyethylene tubings. After the vascular bed was cleared with PBS at 37°C, the tubings between the thoracic aorta and the left ventricle and the tubings between the femoral arteries and the abdominal aorta were connected with peristaltic pump to form two closed circuits. Then, the colloid gold-LDL or DiI-LDL was infused. The animal was then perfused through the left ventricle with the fixative. The aorta, carotid artery, abdominal aorta and their branched regions were collected and prepared for quantitative electron microscopy. The adhesion and penetration of mononuclear cells into the intima occurred after 1-3 months of high cholesterol diet treatment. The fatty streak started to appear in the intima at 6 months, whereas the endothelium still kept intact. More LDL-DiI accumulated in the branched regions than those in the unbranched regions of the arteries. LDL-gold conjugates were observed in the plasmalemmal vesicles, multivesicular bodies and in the subendothelial space in both the branched and the unbranched regions. Quantitative study revealed that the volume densities of plasmalemmal vesicles which contained the LDL-gold particles in the branched regions of aortic arch were significantly ($P<0.05$) higher than the density value in the unbranched regions. The incidence of open junctions was significantly increased in the branched regions of aortic arch whereas no open junctions were observed in the unbranched regions. Moreover, the LDL-gold conjugates were present within most of these open junctions (Fig. 1). These results indicated that the major visible routes for transport of the LDL across the endothelium in the branched regions of the arteries are open junctions as well as plasmalemmal vesicles. The region-associated permeability changes might account for the incidence of atherosclerosis in the branched areas of arteries.

REFERENCES

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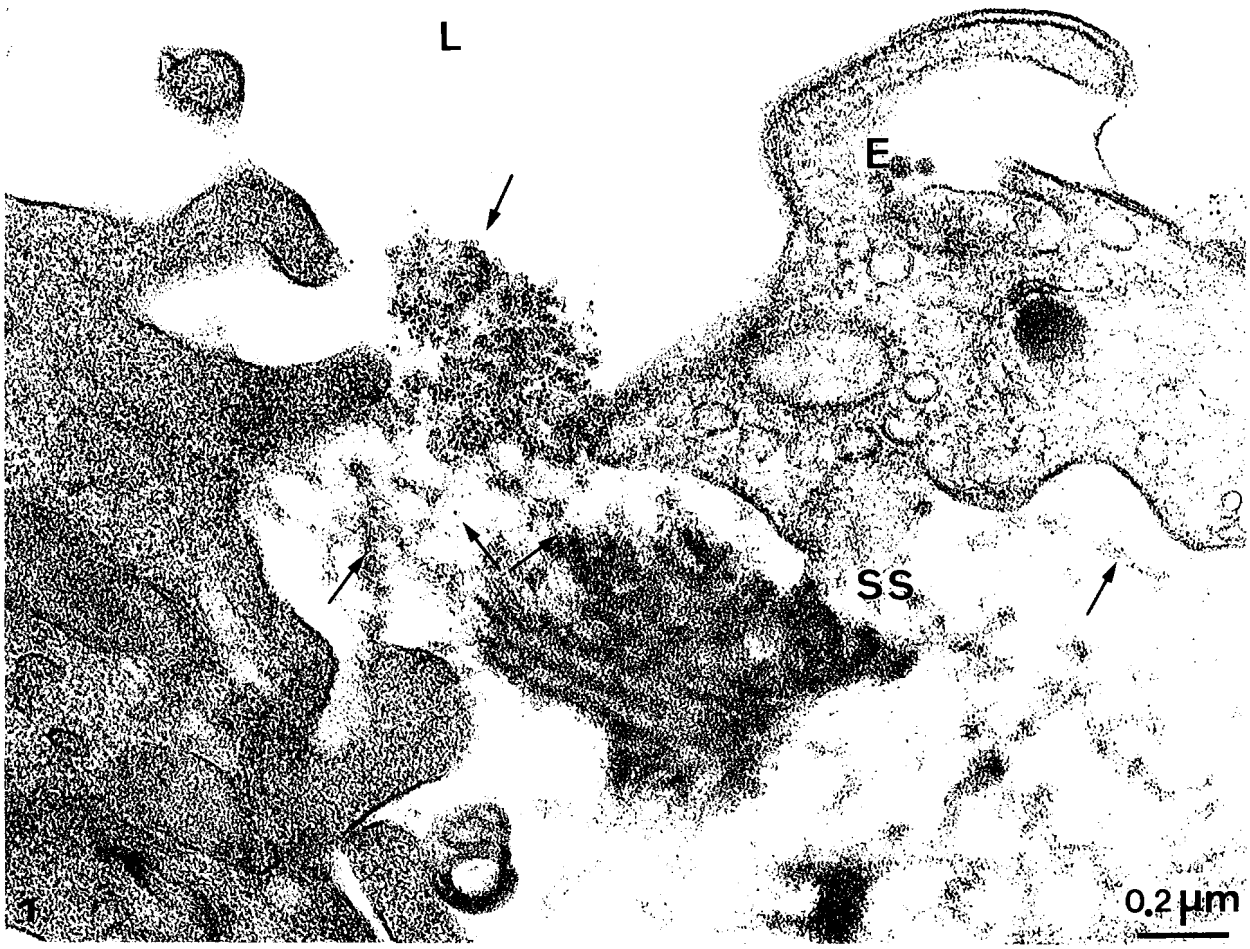

Fig. 1. Endothelial cells (E) of rat aortic arch after 6 months on the high-cholesterol diet. LDL-gold particles (arrow) are seen within the open junctions as well as in the subendothelial space (SS).