FIB-SEM: an in-depth study of atherosclerotic tissue

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During atherogenesis changes take place in the ultrastructure of the vasculature and in the localisation of proteins within the vasculature. Some of these changes will initially only arise in a relatively small number of cells, positioned in a 'sea' of unaffected cells. To localise these affected cells and to study these cells at high resolution, there is a need to navigate through a large area of the sample and subsequently zoom in onto the area of interest. In this way specific cells within their *in vivo* environment can be studied.

The focussed ion beam-scanning electron microscope was used to study aorta of ApoE-/- mice in a correlative way. Analysis with this microscope enables one to scan a large surface area of interest at low magnification, in this case an atherosclerotic plaque, and subsequently zoom in, for further analysis at an ultrastructural level of, in this case, the endothelial cells and the vessel wall. Three different approaches were used: 1. large cross section through the artery; 2. small cross section specific through the endothelial cells and 3. a slice and view series through the endothelial cells positioned on the lesion towards the healthy site, thereby rendering valuable and detailed 2D and 3D information of the vasculature (Figure 1). Moreover, in combination with pre-embedment labelling of surface exposed antigens, these approaches allow insight into the 3D distribution of these markers.

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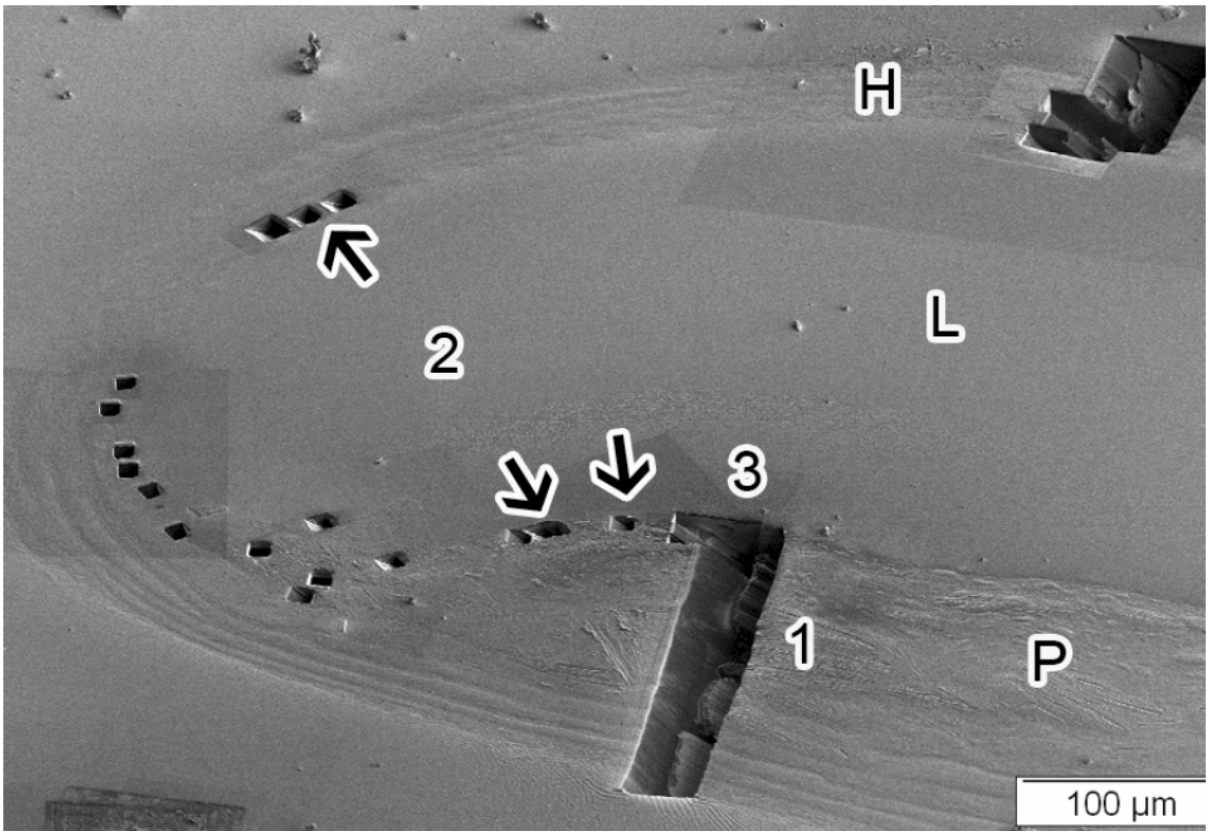

Figure 1. FIB-SEM examination of atherosclerotic tissue of ApoE-/- mice. Overview of the artery and the three different approaches used to study the vasculature. 1. Large cross section; 2. small cross section (white arrows) at various places along the endothelial lining; 3. slice and view series. L=Lumen; P = plaque; H=healthy site.

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