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ENDOTHELIAL INJURY IN HUMAN ATHEROSCLEROSIS

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Abstract

A light and electron microscopic investigation (scanning and transmission electron microscopy) was performed on 51 human atherosclerotic carotid lesions. The purpose of this study was to establish whether features of endothelial injury such as those described in animals occur in man and whether these features can be related to specific stages of human atherosclerosis.

Irrespective of their histological appearance the atherosclerotic lesions were covered with endothelium which showed non-specific changes in cell shape and size. However, all complicated lesions appeared denuded. Moreover, a peculiar interaction of endothelium with monocytes and lymphocytes as well as blood components (e.g., fibrin and lipoproteins) was observed in intimal thickenings, fatty streaks and uncomplicated plaques. The surface exposure of macrophage-derived foam cells was seen on florid fatty lesions. Large areas of the arterial surface lacking any endothelial coverage were characteristic of complicated plaques. They appeared to be a consequence of the arterial wall degeneration with an associated failure in endothelial repair.

KEY WORDS: Endothelial injury, Scanning electron microscope, Transmission electron microscope, Human atherosclerosis.

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Introduction

During the past 10 years, the response-to-injury hypothesis, proposed by Ross and Glomset in 1976 (61), has gained popularity among pathologists interested in atherosclerosis. This was mainly due to its ability to integrate, in a synthetical and logical manner, the information coming from experiments performed in different labs. The authors' view was that the earliest detectable event in the disease was an undefined injury to the endothelium which led to cell loss and subsequent exposure of the thrombogenic subendothelial matrix (denuding injury). In animal models, in fact, the endothelial denudation induced by mechanical, chemical, viral, immunologic and toxic agents appeared to be initially followed by platelet adherence and degranulation and later by smooth muscle cell migration and proliferation resulting in lesions very close to the human plaque (62, 63).
The discovery of the platelet-derived growth factor (PDGF) as well as the identification of its biological activities (PDGF strongly chemoattracts and stimulates arterial smooth muscle cells to proliferate) strengthened the conviction that endothelial loss was correlated with intimal smooth muscle cell (SMC) accumulation (49, 51, 64).

In 1985, Reidy (53) critically stated: "a major defect in almost all of these studies is the lack of any clear definition of what actually constitutes endothelial injury and, perhaps more importantly, how do we recognize it?". Therefore, the precise role of endothelial injury in the disease remains a debated issue, particularly in man.

In the early 1970s techniques for culturing endothelial cells (EC) were developed, thus demonstrating how complex EC biology is (39). ECs present numerous functions, most of which can be relevant to the physiology of the vessel wall. In particular, the local EC modulation of

various biologic systems such as the coagulative system, vascular tone, inflammation and immunity has been recently demonstrated (12, 14, 24, 25, 34). Moreover, there are also a few interesting papers on the potential implications of such endothelial functions in the onset and subsequent development of the atherosclerotic plaque (16, 48, 64, 67). Therefore, a dynamic participation of EC in the disease is gradually being defined.

Some recent morphological investigations have questioned the presence of denuding endothelial injury both in animals (53) and in man (5, 31). We observed foci of frank endothelial denudation only in the presence of advanced and complicated human plaques (45). Richardson and Parbtani (58) demonstrated that minimal endothelial changes (non-denuding injury) can be induced by a variety of stimuli in animals.

However, a complete and detailed documentation of such endothelial injuries has not yet been provided in human specimens. Scanning electron microscopy (SEM) has been demonstrated to be a reliable tool for detecting the surface endothelial responses to vascular grafting (9, 11, 29, 32, 47, 50, 55), balloon catheter injury (37, 43, 52, 71) as well as experimentally induced and human atherosclerosis (44, 45, 53, 63, 70). Accordingly, we mainly undertook an SEM investigation on endothelium overlying the diseased wall in different stages of human atherosclerosis. Moreover, transmission electron microscopy (TEM) was also used for correlating the surface with internal endothelial morphology. The purpose of this study was to verify whether features of endothelial injury such as those described in animals occur in man and whether they can be correlated with specific stages of the human disease.

Materials and Methods

51 atherosclerotic carotid lesions were surgically treated. 74.5% of these lesions were revealed by an episode of cerebro-vascular insufficiency whereas 25.5% had no clinical symptoms. The asymptomatic lesions were submitted to surgery due to the high risk of cerebral ischemia. In fact, they were either hemodynamically significant lesions (stenosis over 50%) or lesions that with echotomography and angiography showed a complicated atheromatous plaque with thromboembolic risk.

As far as risk factors are concerned, 64.7% of the treated lesions occurred in smokers, 49% in patients with arterial hypertension, 31.4% were associated with hyperlipidemia (8 with hypercholesterolemias, 4 with hypertriglyceridemias, 4 with hypercholesterolemia plus hypertriglyceridemia), and 11.7% with diabetes. Moreover, the carotid lesion in 56.9% was associated with chronic peripheral obstructive arteriopathy and in 35.3% with ischemic heart disease.

In all cases, a carotid thromboendarterectomy (T.E.A.) was performed. T.E.A. was initiated in the common carotid artery at 4 cm. from the main atheromatous lesion. This operation, involving the removal of the intima and media from the arterial wall, allowed us to obtain an arterial segment composed of the terminal part of the common carotid, the atherosclerotic plaque at the carotid bifurcation, and the initial portion of the internal carotid.

Two arterial segments were selected: the carotid bifurcation (segment 1) and the apparently unaffected common carotid artery (segment 2). The samples were briefly rinsed in chilled phosphate buffer saline (PBS) and then fixed by submersion in 2.5% buffered glutaraldehyde for 10 min. in the operating theatre. Each segment was divided into small pieces for light microscopy (LM), transmission and scanning electron microscopy (TEM/SEM) investigation.

Samples for TEM were additionally fixed with 2.5% glutaraldehyde in 0.1 M cacodylate buffer, pH 7.3, for 3 h, postfixed with 1% OsO_4 in the same buffer for 1 h, dehydrated in a graded series of alcohol and embedded in araldite.

Semithin and thin sections were obtained with a Reichert OMU3 ultramicrotome. Semithin sections for LM observation were stained with toluidine blue. Thin sections were counterstained with uranyl acetate and lead citrate and examined in a JEOL 100B transmission electron microscope.

Samples for SEM were fixed as above and after ethanol dehydration they were critical point dried, coated with gold and observed in a Philips 505 scanning electron microscope. In addition, after glutaraldehyde fixation some specimens were subjected to the methenamine silver reduction staining. The staining procedure was performed according to the method described by Becker and Sogard in 1979 (1). The specimens were coated with evaporated carbon and observed in the scanning electron microscope by means of backscattered electron detectors.

Additional specimens were fixed in 2% glutaraldehyde containing 0.02% mg/ml of filipin (Sigma), postfixed in 1% OsO_4 and submitted to a tannic acid incubation according to the method of Simionescu et al. (69) and then processed

for TEM examination.

Results

Light microscopy

The histological appearance of segment 1 was that of a fibrolipidic plaque in 41.2% (21 cases) and of a complicated plaque (calcium salts deposition -13, hemorrage -10, ulcer -18) in 58.8% (30 cases).

In contrast, LM investigation of segment 2 provided the following results: diffuse intimal thickening (3.9%), fatty streak (15.6%), gelatinous lesion (1.9%), eccentric intimal thickening (9.8%), fibrolipidic plaque (43.1%), fibrous plaque (5.8%), and complicated plaque (19.6%).

Electron microscopy

Normal surface endothelial morphology. Normal arterial endothelium appears as a sheet of regular, rather uniform cells evenly aligned along the flow direction. They are narrow, flat and lacking in surface projections (Fig. 1). At low magnification the cell margins are scarcely visible while nuclei slightly bulge into the lumen. The identification of endothelial lining can be made easier by the use of silver methenamine staining. In this case, subsurface nuclei are superimposed on EC surface morphology (Fig. 1, insert). Moreover, there is no evidence of fibrin formation. Blood cells, mainly erythrocytes, are observed over the endothelial layer.

Changes in endothelial cell shape and size. Endothelium continuously covered atherosclerotic lesions irrespective of their histological appearance except for the complicated lesions. ECs were mainly arranged in a polygonal pattern. Polygonal ECs (Fig. 2) showed occasional surface microvillous projections, marginal folds and slightly bulging nuclei thus resulting in a cobblestone appearance. This pattern was predominant even though areas of spindle-shaped ECs as well as of irregularly aligned ECs were also observed. In addition, ECs showed a variable change in size. Anisocytosis was particularly evident when small-sized ECs were seen in conjunction with large ECs.

Endothelial non-denuding injury. TEM of filipin/tannic acid treated samples shows lipid vesicles (35-70 nm) within endothelial transport channels and/or vacuoles in the intimal thickenings associated with raised lesions. Lipid material, most likely transported along a concentration gradient, appears to be segregated from any lysosomal influence. Moreover, lipid vesicles are seen next to the endothelial adluminal and abluminal membrane, thus suggesting a probable endothelial discharge. In the subendothelial space, lipids, undergoing a chemical-physical remodelling appear as vesicular lakes (Fig. 3) in close contact with matrix components (proteoglycans, microfibrils, collagen fibres). The vesicles appear tightly packed with electron-lucent core and lamellar periphery ranging 35-700 nm in diameter (Fig. 4).

A regional adhesion of leukocytes was

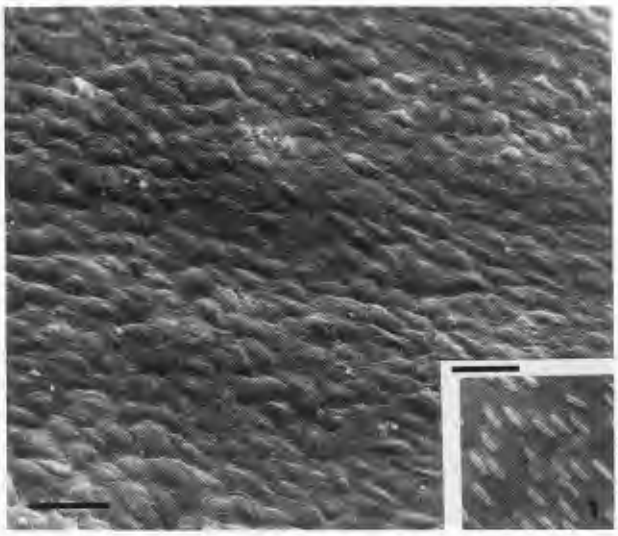


Fig. 1. Normal arterial endothelium. SEM shows sheets of flat ECs evenly aligned along the flow direction. Subsurface nuclei superimposed to endothelial surface morphology are illustrated in the insert (silver methenamine staining). SEM. Bar = 50 µm; insert, Bar = 50 µm.

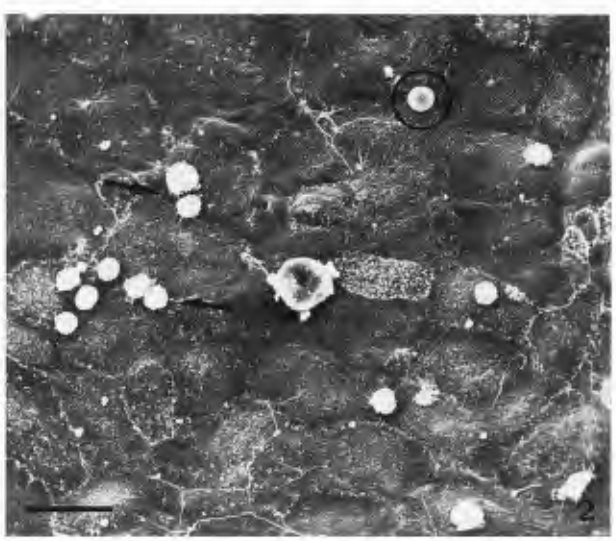


Fig. 2. Changes in EC shape and size. SEM shows ECs arranged in a polygonal pattern. A moderate anisocytosis is also evident. Leukocytes (arrows) adhere to the EC surface. The circle outlines a red blood cell. SEM. Bar = 20 µm.

clearly disclosed on the endothelium lining fatty streaks and uncomplicated plaques. In these areas, the polygonal ECs occasionally showed a contraction of the cell body with thin peripheral intercellular bridges and leaky junctions (Fig. 5). Foci of degenerating ECs and true endothelial necrosis could be observed by TEM (Fig. 6). Monocytes were mainly detected in fatty streaks, whereas clusters of lymphocytes predominated on the proliferative lesions (Fig. 7). TEM revealed that a large amount of the cells within fatty lesions were monocytes/macrophages in origin. Scattered lymphocytes can be observed in close association with monocytes/macrophages and cell debris resulting, in part, from necrosis of foam cells. The frequency of lymphocyte adhesion was unexpectedly high in the uncomplicated plaques. With TEM, the fibrous cap contained newly formed capillaries, containing lymphocytes, as well as clusters of subendothelial lymphocytes (Fig. 8). Fibrin, one of the hallmarks of inflammation, was frequently observed along with the surface inflammatory cells. Fibrin deposition can take the appearance of strands, threads and fibrous tangles (Fig. 7). Its presence may contribute to EC injury and to subendothelial oedema (66).

Borderline injury. The surface exposure of subendothelial macrophage-derived foam cells has been demonstrated in swine (21, 22), nonhuman primates (18, 19), Watanabe heritable hyperlipemic and hypercholesterolemic rabbits (59, 60). We observed this injury in man and particularly in well-developed fatty streaks. Basically, the lesion consists of a focal or a diffuse EC retraction (Fig. 9) with consequent exposure to the blood flow of underlying foam cells. The separation of this entity from other endothelial injuries (i.e., denuding and non-denuding) seems to be valid since the lesion has proved to play a crucial role in the conversion of the fatty streak to a proliferative lesion (18, 59, 63). Earlier in the disease, the EC retraction allows the blood access to the subendothelial space. A dramatic increase in wall permeability can thus be expected. Massive platelet deposition is also observed (Fig. 10). In particular, close platelet-to-foam cell contacts are clearly demonstrated. This may enhance macrophage cholesteryl ester accumulation (13).

Endothelial denuding injury. With SEM, the endothelial denuding injury appears as a large area of the arterial surface lacking any endothelial coverage. The surface consists of fibrin strands, blood cells, extracellular matrix components and cell debris (Fig. 11). Endothelium, with occasional features of regeneration, is observed at the edge of the lesion.

The denuding injury should be accurately

Fig. 3. EC non-denuding injury. Filipin/tannic acid treated specimens show numerous lipid vesicles in the subendothelial space. The arrows indicate the apparently unaffected endothelium. TEM. Bar = 2 μm.

Fig. 4. EC non-denuding injury. Filipin/tannic acid treated specimen. The subendothelial lipid vesicles are shown at higher magnification. TEM. Bar = 600 nm.

Fig. 5. EC non-denuding injury. The asterisk marks a polygonal EC showing a contraction of the cell body. Note the thin peripheral intercellular bridges and the centrally placed microvilli. The arrow indicates a desquamating EC. SEM. Bar = 10 μm.

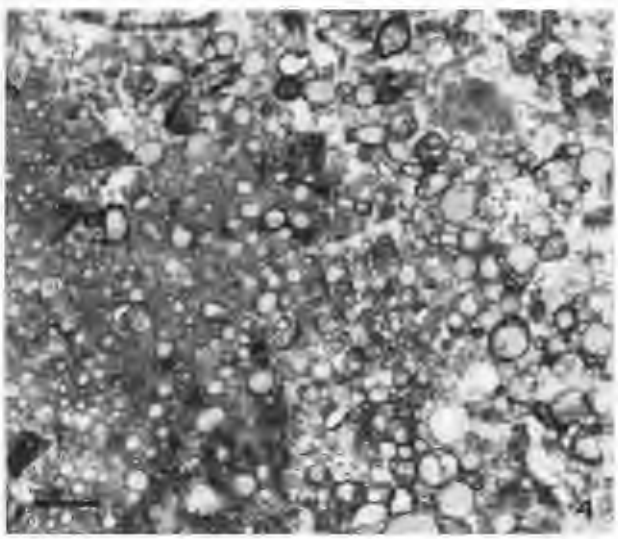
Fig. 6. EC non-denuding injury. Focal necrosis of ECs. Activated platelets as well as fibrin strands (arrows) are close to the subendothelial matrix. TEM. Bar = 2 μm.

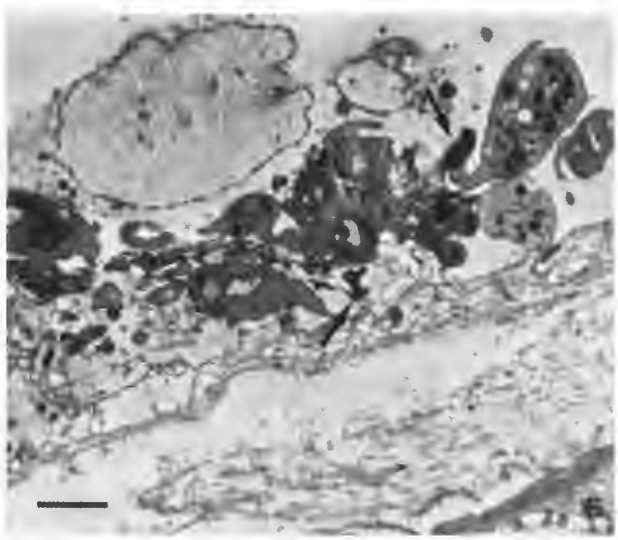
Fig.7. EC non-denuding injury. Surface inflammatory responses. Numerous leukocytes take tenacious contact with the endothelial surface. The surface infiltrate is mainly composed of lymphocytes. Monocytes are also present (arrows). Fibrin deposition takes form of threads (arrowheads) and tangles (*). SEM. Bar = 20 μm.

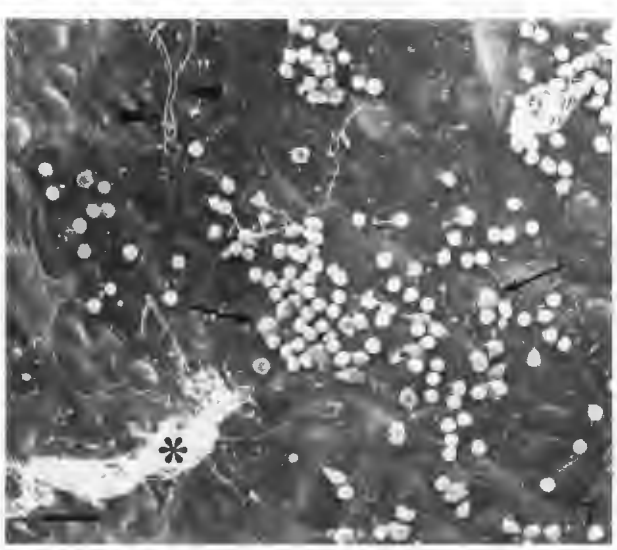
Fig. 8. EC non-denuding injury. The electron micrograph shows some subendothelial lymphocytes (arrows). TEM. Bar = 5 μm.

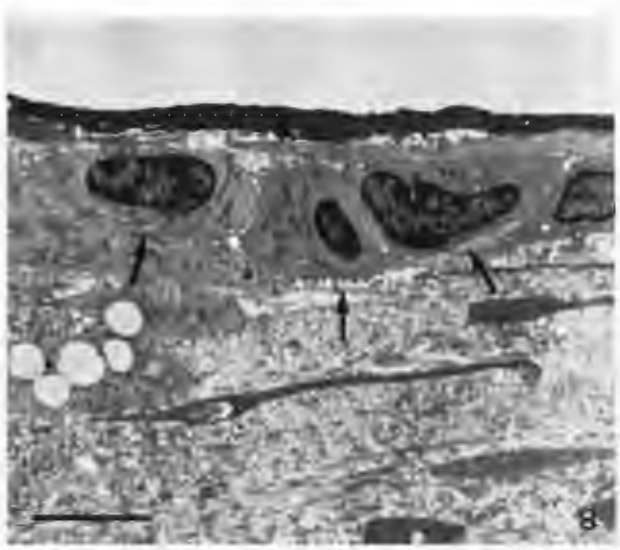
differentiated from endothelial defects caused either by the C.P.D. procedure or by improper handling of the specimen as well as from simple endothelial exfoliation. In the former case, there are many foci of denuded surface with a resultant "moth-eaten" appearance. In addition, the residual endothelium is detached from the subendothelial matrix. Handling artifacts may be suspected when "scraped" areas are observed. As to endothelial exfoliation, only individual cell loss is observed (Fig. 12). Interestingly, no platelet deposition can be detected on the exposed subendothelial matrix. This is in agreement with the recent observation that subendothelial matrix immediately beneath endothelium is not thrombogenic (4). In addition, desquamating cells can also be observed while protruding into the lumen (Fig. 5). They appear as swollen, rounded cells with smooth surfaces. In this case, no evident subendothelial defect is present.

In our series, the endothelial denuding injury is a consistent feature of advanced and complicated plaques. We never found it over









fatty streaks. Fibrolipidic plaques occasionally and gelatinous lesions at all times showed a denuded appearance.

Endothelial regeneration. In experimental models, endothelial injury is associated with varying degrees of endothelial regeneration. Basically, the residual ECs are stimulated to move toward the wound and, after a variable lapse of time, are able to repair the defect. The structural integrity of the endothelial lining is, therefore, maintained. The different phases of endothelial regeneration have been extensively investigated by Reidy's group (10, 54, 55, 72). These studies have proved that small endothelial defects (10-15 cells wide) are followed by rapid endothelial repair (72 h). In contrast, wounds 90-120 cells wide are repopulated by ECs only after 7-8 days. Endothelial repair involves at least four separate processes which include EC activation, spreading, migration, and proliferation. As well assessed by Gotlieb et al. (26), these processes occur sequentially in time and depend on the wound extension. Moreover, complex cytoskeletal rearrangement are involved.

In human disease, two forms of endothelial repair are recognized. At the edge of focal surface defects, we observed a continuous sheet of cells with features of spreading and migration (Fig. 13). ECs were particularly flat and irregular in shape. They showed characteristic long, peripheral lamellipodia. Tenuous cell-to-cell contacts were also shown. Migration was further confirmed by the presence of isolated cells at some distance from the advancing edge. More severe defects involving both surface and subsurface wall components are due to plaque splitting and ulceration. In this case, the endothelium failed to repair completely. With TEM regenerating ECs are characterized by the cytoplasmic presence of many intermediate filaments and of stress fibres. The stress fibre assembly has been related to the inability of injured ECs to repopulate intimal defects (38). Moreover, ECs are frequently located on a reduplicated basal lamina. The most striking finding was the monocyte/macrophage and the smooth muscle cell recruitment. Aspects of thrombus incorporation were also observed. However, we never noted any suggestive features of complete endothelial repair.

Conclusions

ECs are very sensitive to the different procedures of processing material, and artifacts can occur. It is well known that pre-perfusion and fixation protocols are critical for a proper evaluation of endothelial surface morphology (35). Wrinkling, collapse, foci of denudation and

Fig. 9. Borderline injury. EC retraction producing the surface exposure of macrophage-derived foam cells (arrows). SEM. Bar = 50 μm.

Fig. 10. Borderline injury. Exposed macrophages (M) and aggregating platelets (arrow) are evident. The arrowheads indicate the endothelial layer. SEM. Bar = 10 μm.

Fig. 11. Denuding endothelial injury. The arterial surface lacks any endothelial coverage. The subendothelial matrix is focally covered by activated platelets (arrowheads), fibrin threads (arrows) and cell debris (large arrow). SEM. Bar = 10 μm.

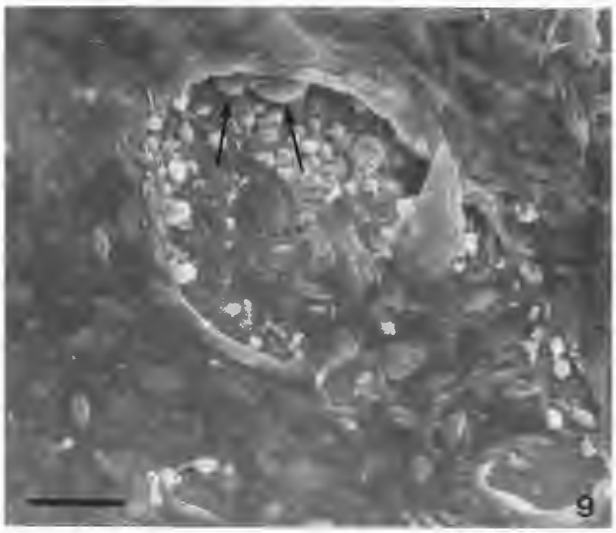
Fig. 12. Endothelial desquamation. Individual cell loss is evident. No platelet deposition is observed on the exposed matrix. SEM. Bar = 5 μm.

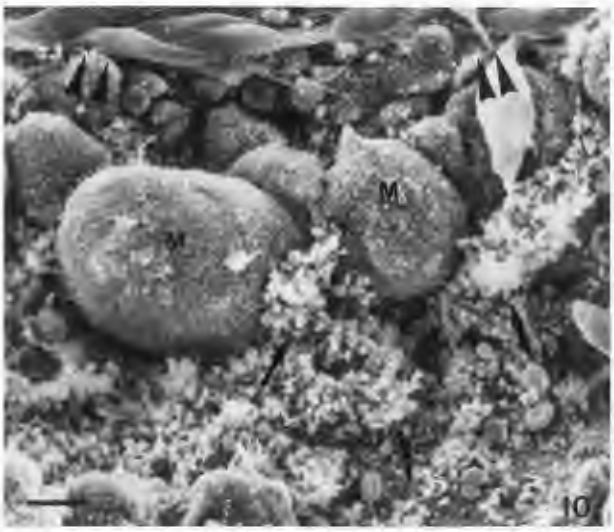
Fig. 13. Endothelial regeneration. Regenerating cells present features of spreading and migration. Peripheral lamellipodia are visible. SEM. Bar = 30 μm.

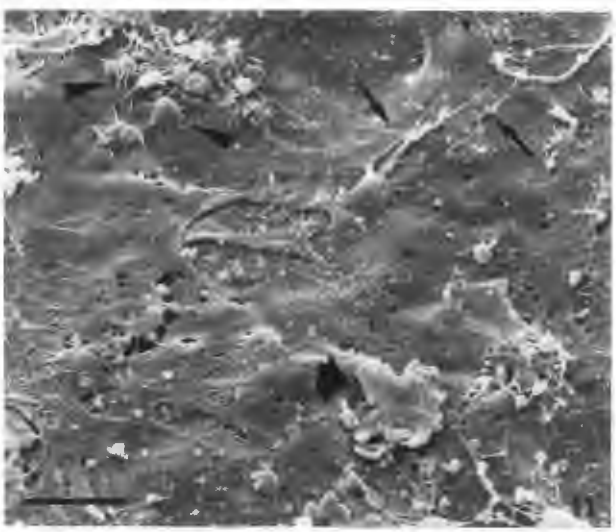
membrane defects have been described (57). This may greatly limit the assessment of eventual endothelial injury, particularly in man. Optimal fixation procedures have been developed for animals and excellent results have therefore been achieved (36, 57, 58). As to human specimens, a correct approach is virtually impossible. A perfusion fixation design can be attempted only when entire arterial segments are attainable. Thus, autopsy and transplant are the potential sources for this kind of tissue collection. Unfortunately, autopsy provides badly preserved material and legal and ethical consideration limit the access to a better maintained morphology.

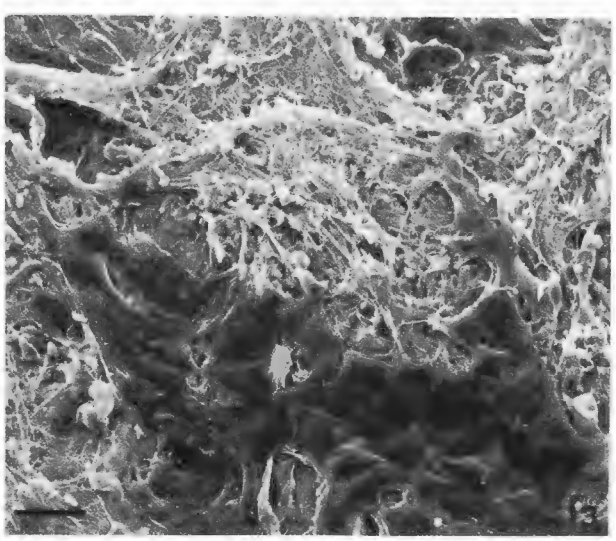
In order to partially overcome such limitations, we utilized the surgical source. In fact, sufficiently good results can be obtained from arterial segments taken at by-pass surgery and at endoarteriectomy, exspecially when the specimens are processed immediately after removal. Fully conscious of its intrinsic limitations, we used an immersion fixation procedure after a short rinse in chilled phosphate buffer saline. This schedule appeared to us an acceptable compromise to evaluate endothelial surface morphology in man.

SEM investigation showed that a continuous endothelial layer is usually present over atherosclerotic lesions irrespective of their histological appearance. Endothelial cells are mainly arranged in a polygonal fashion and present changes in cell size.









In the past, these changes have been considered by some authors as an expression of subtle endothelial injury (8, 56). Other investigators have questioned the true endothelial nature of the polygonal lining. In fact, medial smooth muscle cells can migrate onto the vessel surface, giving

origin to a pseudo-endothelial lining very similar to the polygonal endothelium (28, 53, 68). In our study, these changes appeared to be almost non-specific. TEM demonstrated that the layer was endothelial in origin. Whether these changes should be considered a sign of endothelial injury remains an open question. However, inferences from animal studies suggest a relation with local changes in fluid haemodynamic, including flow patterns, shear stress, and stagnation points (6, 15, 74).

Features which may be considered an expression of non-denuding endothelial injury have been observed in intimal thickenings, fatty lesions and uncomplicated plaques. Basically, they are characterized by: i) lack of any extensive endothelial loss; ii) interaction of endothelium with blood cells and components most likely due to the acquisition of an activated endothelial state in selective fields of the endothelial layer.

Intimal thickenings close to raised lesions showed extensive, mainly extracellular lipid collections immediately beneath an apparently intact endothelium. The subendothelial deposits are composed of many tightly packed lamellar vesicles which are believed to derive from an increased transendothelial passage of LDL (7,30, 69). Our TEM observation of lipid vesicles within transendothelial channels and in close association with both the adluminal and abluminal aspect of the endothelial cell membrane may support this view. Lipid deposits have been demonstrated to chemoattract blood monocytes in animals (23). Modification of LDL by endothelium may render it recognizable by the scavenger receptor on macrophages (3, 20, 73). It may therefore be hypothesized that the ECs may induce the generation of the subendothelial deposits of monocyte-derived foam cells which are virtually characteristic of human fatty streaks.

Focal adhesion of leukocytes to the endothelium has been appreciated in fatty streaks and uncomplicated plaques. This may be correlated to the endothelium activation induced by specific cytokines, including γ-interferon, interleukin-1, and tumor necrosis factor (12, 33, 48). Activated ECs may, therefore, express specific binding sites for monocytes and lymphocytes such as ELAM-1 (2) and ICAM-1 (17, 65).

As described in the early stages of hypercholesterolemia-induced atherosclerosis in animals, including the pig (21, 22), monkey (18, 19), rat (42), pigeon (40) and rabbit (59, 60), monocytes are specifically involved in the composition of human fatty streak. In contrast, lymphocytes predominate in the uncomplicated plaques. This is in accordance with previous histological and immunohistochemical reports (27, 31, 41). Lymphocytes were observed while adhering to the endothelium as well as in the plaque fibrous cap. Diapedesis through the endothelial layer may be the source of the lymphocytes contained in the fibrous cap. In addition, newly formed vascular channels containing lymphocytes may provide an alternative lymphocyte source. In a previous paper (46) we also demonstrated that lymphocytes are of the T-subset. However, we were not able to detect any difference in the T4/T8 ratio. Moreover, T lymphocytes and perhaps also monocyte/macrophage displayed features of activation as demonstrated by focal positivity for the monoclonal antibody BER-H2 which recognizes the CD-30 antigen strictly related to cell activation. Interestingly, many of these cells were also activated while still adhering to the endothelium. This may be indirect evidence that endothelial cells, in particular conditions, may take on inducible functions (class II histocompatibility antigens, release of interleukin-1).

The surface exposure of macrophage-derived foam cells was observed mainly over fatty streaks for the first time in man. However, its potential role in the conversion of the fatty lesion to the proliferative plaque (18, 19, 59, 60, 63) remains to be established in man.

Finally, all complicated lesions lack endothelial coverage. This applies both to usual atherosclerotic plaques and to debatable lesions, e.g., gelatinous lesions. Therefore, it may be suspected that the typical endothelial denuding injury is specifically related to the presence of complications of the diseased arterial wall, such as thrombosis, haemorrhage and ulceration. It appears to be a consequence of the arterial wall degeneration with an associated failure in endothelial repair.

In summary, this paper has concentrated upon EC morphological changes which can be observed during the evolution of atherosclerosis in man. The presence of changes in endothelial shape and size, features of endothelial injury, including non-denuding, borderline and denuding injuries as well as of endothelial regeneration, has been documented and discussed. Each type of EC morphological changes was mainly observed in specific stages of the evolution of the disease. Precise correlations between morphological and functional information as well as inferences from animal studies are needed to draw proper conclusions. However, our data support the view that endothelial cells are actively involved in the evolution of atherosclerorosis in man.

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Discussion with Reviewers

B.E. Spurlock: Were the lesions reported as denuded and non-denuded at the SEM level previously observed grossly ?

Authors: No, they were not. The sample collection was irrespective of the gross surface appearance. In each case, the symptomatic plaque at the carotid bifurcation and the common carotid artery at 2 cm. from the main lesion were selected.

Reviewer II: The cellular mechanisms regulating transendothelial transport of solutes is controversial and at best, poorly understood. The authors have stated they observe leaky junctions, vesicular components and leaky channels. Scanning electron micrographs are used to substantiate these conclusions. Can the authors truly establish these cellular structures as the site of vascular permeability ?

Authors: On the basis of scanning electron information we cannot substantiate these conclusions. However, TEM of filipin/tannic acid treated samples showed the presence of lipid vesicles in

correspondence of both the abluminal and adluminal aspects of the endothelial cell membrane as well as within cytoplasmic transport channels. We defined as transport channels those vacuoles or cystic spaces which most probably derive by invagination of the plasma membrane and are characterized by a serpentine appearance continuously stained by tannic acid. However, dynamic studies using permeability markers are needed before drawing definitive conclusions.

Reviewer II: One possible explanation for the observation of activated endothelium on carotid arteries from these patients could be trauma related to surgical procedures or presurgical procedures such as angiograms. Do the authors have information they can provide concerning these possible effects ?

Authors: No, we do not. We agree that the relationship between trauma due to invasive procedures and perturbation of the endothelial cell membrane deserves particular attention and needs further investigation.

Reviewer II: The authors state several times that the luminal lining of cells are endothelium and not smooth muscle. The major proof of this claim rests on two low power electron micrographs. What is the morphological criteria by which the endothelial nature of these cells is established ?

Authors: Basically by identification of Weibel-Palade bodies. In addition, the presence of marginal folds, micropinocytotic vesicles, intermediate filaments 10 nm thick, tight junctions, and basal lamina were also taken into account.

M. Richardson: What is the relationship of areas showing "polygonal" or irregular endothelial cells to the bifurcation of the carotid artery ?

Authors: There was no particular relationship. Unlike experimental atherosclerosis, in human disease the local variations in blood hemodynamics are very complex and only partially predictable. This may explain the lack of a clear association between endothelial surface morphology and specific topographical areas of the carotid district.