CONTENT

Chapter 1
ATHEROSCLEROSIS

Atherosclerosis is the process that alters the artery wall so that in some of its sections **atherosclerotic plaques** — local thickening caused by deposition of cholesterol (C), fat and some other blood components, are formed, resulting in reduction of the internal arterial lumen.

As atherosclerosis progresses, the reduction reaches such a degree that the blood, rich with oxygen and other nutrients, hardly reaches the organ supplied by this vessel such as the heart or brain. In severe cases, the artery lumen can be completely closed. Process of artery closing is called occlusion. It occurs due to overlap of the vessel lumen by growing deposits, as well as as a result of blocking of the narrowed lumen by a blood clot. The latter case is referred to as formation of the so-called **complicated plaque** (Fig. 1.1), the surface of which is damaged as a result of rupture or erosion caused by a combination of several factors (loose lipid core, weakened plaque cover — due to a small amount of collagen and calcium, inflammation, unfavorable hemodynamic conditions, and other causes). Due to the disruption of the continuous plaque surface ("cracked" endothelium and adjacent vascular wall structures), the blood begins to contact the internal contents of the plaque, which contains substances able to trigger the processes of blood clot formation. The result of plaque rupture is thrombosis of various severity (mural or occlusive, i.e. completely closing the vessel lumen). To describe the latter condition, the term **"atherothrombosis"** is used, which is the most dangerous complication of atherosclerosis.

Atherothrombosis is considered the main cause of death and disability associated with atherosclerosis. The clinical consequences of atherothrombosis are acute ischemia, necrosis of part of an organ or tissue (myocardium, brain, limb, kidney, abdominal organs, etc.) that receives blood through this artery, or death of the patient (if a significant part of a vital organ has been damaged by ischemia or necrosis). The most frequent clinical symptoms of atherothrombosis are acute coronary syndromes (ACS; myocardial infarction — MI) and ischemic acute cerebrovascular events (ACVE — strokes). If the process of atherothrombosis occurs in the blood vessels of other organs (kidneys, limbs, abdominal organs), this leads to similar severe impairments of their functions.

Pathogenesis

Atherosclerotic damage to the arteries starts with the accumulation of lipoprotein particles rich with cholesterol in the arteries' intima. The possibility of lipoproteins ingress and retention in the intima structures is, on the one hand, a result of increased endothelial permeability, on the other hand, — of binding the lipoprotein particles by components of the extracellular matrix, primarily with proteoglycan molecules.

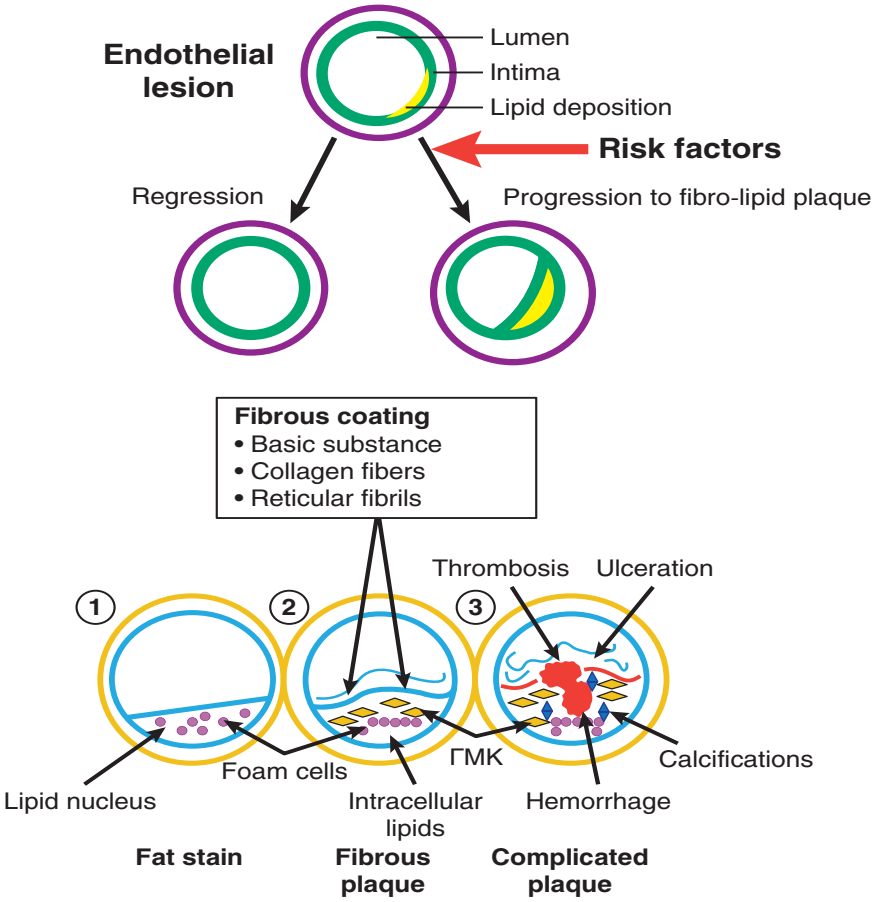

Fig. 1.1. Atherosclerosis development. Schematic representation of the cross-section of the artery at different stages of the atherosclerotic process

Currently, the most substantiated point of view is that the initial stages of atherosclerosis are considered as a result of endothelial damage. The term "damage" in this situation suggests rather not mechanical injury of the endothelium but an impairment of its function manifested by increased permeability and adhesion. Normally, the interendothelial spaces are quite narrow and impenetrable for lipoproteins. Under the influence of certain substances (for example, catecholamines, angiotensin II, serotonin, endothelin, etc.), as well as under the influence of hypercholesterolemia, the interendothelial spaces widen, and low-density lipoprotein (LDL) particles penetrate the arterial intima (Fig. 1.2, part 1). Increase in blood cholesterol-rich lipoproteins content, arterial hypertension (AH), local hemodynamic changes, smoking, inflammatory diseases are considered among the principal factors causing arterial endothelium damage. It is believed that these, and, possibly, other unknown factors lead to loosening and thinning of the protective glycocalyx on the surface of endothelial cells, expansion of interendothelial gaps, loosening of endothelial fibrous structures and edema of the subendothelial intima layer.

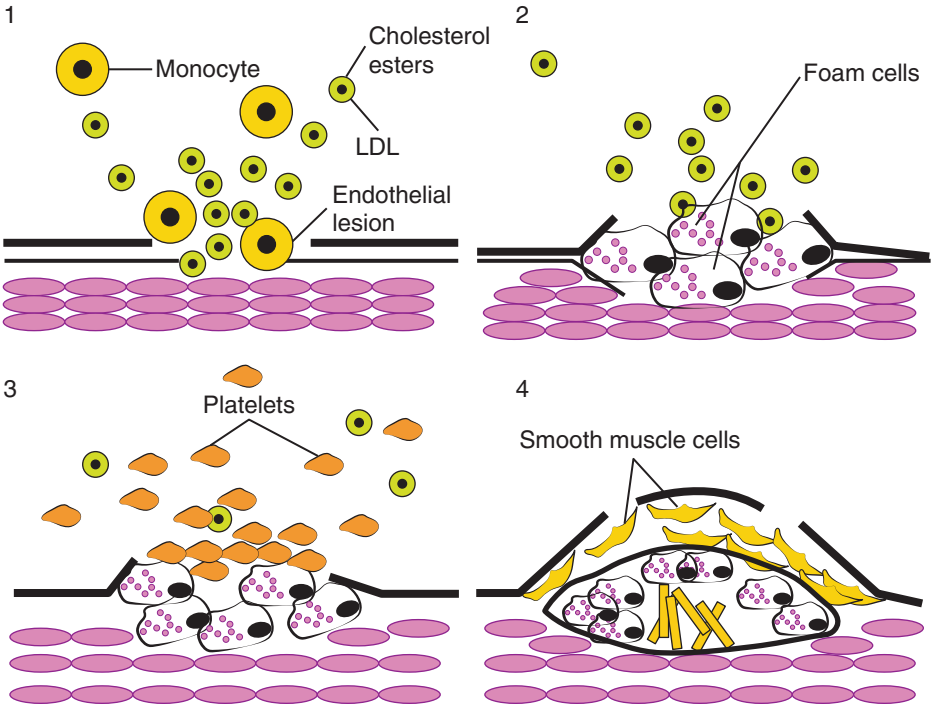

Fig. 1.2. Atherosclerotic plague formation

Changes in the artery intima lipids are the next stage in atherosclerosis development. Lipoprotein particles that have penetrated the extracellular space of intima, bound and retained there by proteoglycans, are modified. Modified lipoproteins include peroxide-modified and glycosylated lipoproteins, autoimmune "lipoprotein-antibody"complexes, products of limited lipoprotein proteolysis, desialized and aggregated lipoproteins, and the above-mentioned complexes of lipoproteins with glycosaminoglycans. Modification of lipoproteins occurs not only in artery intima but also in the blood. The greatest significance in the development of atherosclerosis is currently attached to the chemical modification of lipoproteins occurring in intima: peroxidation and glycosylation.

Intima infiltration by circulating white blood cells and monocytes, which then transform into macrophages and, capturing modified LDL, turn into foam cells (see Fig. 1.2, part 2) is the next stage in atherosclerosis development. Before the interaction of monocytes and T-lymphocytes with endothelial cells and their penetration into the subendothelial space occurs, these cells adhere to the endothelium. This process involves special adhesive molecules and certain cytokines. White blood cells migration to the subendothelial space occurs not only under the influence of chemoattractant cytokines (chemokines) but also with the participation of modified LDL. By loading with lipids, macrophages participate in the removal of lipoproteins accumulated in the focus of developing atherosclerotic lesion. But in case of hyperlipidemia and a significant lipid accumulation in the artery wall, this function of macrophages is

disrupted. As a result, foam cells, i.e. macrophages overloaded with lipids, mostly remain in arteries intima and die, undergoing apoptosis. At this time cholesterol esters, non-esterified cholesterol and cholesterol monohydrate crystals that were accumulated in the foam cells are released. These processes lead to focal cholesterol accumulation in arteries intima and create preconditions for the development of lipid spots, then lipid strips, and subsequently — atherosclerotic plaques. Foam cells also serve as a source of a number of cytokines and effector molecules such as oxygen superoxide anion and matrix metalloproteinases involved in the development and progression of atherosclerotic lesions.

On the whole, the process of inflammation has a great importance in the development of an atherosclerotic plaque. There is a theory, according to which the development of atherosclerosis is based on a chronic inflammatory process and local endothelial reactions in response to various metabolic, mechanical damage and exposure to infectious agents, immune complexes (IC), and various toxins. The role of inflammation in the pathogenesis of atherosclerosis was proven in a placebo-controlled trial of kanakinumab, a monoclonal antibody (AB) to interleukin-1β (IL-1β), which reduced the incidence of cardiovascular events in patients with the history of MI, when added to standard pharmacological therapy, but was not registered for this indication due to an increased likelihood of septic complications.

Normally, smooth muscle cells are located in the tunica media (the middle membrane of the arteries) and perform a contractile function. Smooth muscle cells migrate to the intima under the influence of chemoattractants, which are produced by macrophages, endotheliocytes, and fibroblasts of arteries intima in response to the appearance of modified LDL in it. The platelet growth factor, which is secreted not only by platelets (see Fig. 1.2, part 3) but also by endothelial cells, smooth muscle cells themselves, and macrophages, has a great significance for relocation of smooth muscle cells into arteries intima. Smooth muscle cells that have migrated to intima (see Fig. 1.2, part 4) grow intensively under the influence of fibroblast growth factor and, possibly, tumor necrosis factor alpha (TNF-α) and IL-1. After ingress into the intima and proliferation, smooth muscle cells undergo a number of changes and acquire new features. Firstly, they begin to produce collagen, elastin, and glycosaminoglycans, i.e. the connective tissue basis of the future atherosclerotic plaque. Secondly, smooth muscle cells acquire the ability for unregulated capture of modified LDL without the participation of aporeceptors (these receptors are present on the surface of smooth muscle cells) by direct endocytosis which leads to the accumulation of cholesterol esters in them. During the process of atherogenesis, in developing atheroma, along with the proliferation of smooth muscle cells, their death (apoptosis) is observed which is stimulated by pro-inflammatory cytokines and occurs with the participation of cytotoxic T-lymphocytes (T-killers). In its turn, another subpopulation of T-lymphocytes — T-helper-1 secrete pro-inflammatory cytokines (interferon γ, IL-1, TNF-α). These cytokines contribute to the development of inflammation by activation of endotheliocytes, macrophages, stimulation of the production of free radicals, proteolytic enzymes, and significant increase in coagulant activity. Another subpopulation of T-lymphocytes — T-helper-2, on the contrary, produce cytokines

that have an anti-inflammatory effect, and tissue growth factor. These substances contribute to the proliferation of smooth muscle cells, development of fibrosis, and enhance healing processes.

As the atherosclerotic lesion progresses, an increased blood supply to the newly formed structures evolves, in the form of an abundant microvessel plexus in the atherosclerotic plaque. A new vascular network formation is caused by the effect of angiogenesis factors which include fibroblast growth factors expressed in the atheroma, the endothelial growth factor and other molecules. The evolving new vessels network is largely responsible for the development of atherosclerosis complications. The vessels of this network contribute to hemorrhages in the internal structures of the plaque (see Fig. 1.1), create abundant accumulations of white blood cells ("leukocyte plugs") on its surface which, in its turn, contributes to the penetration of white blood cells including monocytes into the formed plaque and subsequent occurrence of aseptic inflammation in it. In addition, new vessels are characterized by acutely increased permeability and increased capacity for microthrombi formation in them. The formed thrombin can stimulate the proliferation of smooth muscle cells and release of cytokines and growth factors from them.

Pathomorphology

Lipid spots and lipid strips, which are based on the appearance of foam cells, are considered the earliest verified morphological signs of atherosclerosis.

Lipid spots are yellowish points with a diameter of up to 1.5 mm of a soft consistency which do not rise above the surface of the endothelium and do not create obstacles to blood flow. Lipid spots can appear in any part of the arterial system, but, as a rule, appear at first in the aorta. Their further fate is very variable. It is believed that in some cases lipid spots undergo reverse development and disappear without a trace. Sometimes lipid spots do not undergo further development, and the process of atherosclerosis freezes at the initial stage. But in a significant number of cases lipid spots evolve towards further development of the atherosclerotic process.

Lipid strips are the next early stage of atherosclerotic lesion development. They are formed from lipid spots that enlarge, become elongated and wider. In contrast to lipid spots, lipid strips can rise above the surface of the endothelium. Lipid strips are formed beginning from the second decade of life, and consist, as well as lipid spots, of foam cells of macrophage and myocytic origin, loaded with lipids in the form of cholesterol esters and T-lymphocytes. In the lipid strips, extracellularly located cholesterol is almost absent. The next stage in atherosclerosis development is characterized by lipid deposition in the extracellular space in the form of cholesterol esters and free cholesterol. Thus, the preconditions for the formation of atherosclerotic plaque lipid core are created.

Atheroma ("glob of porridge", from the Greek athera — "porridge") is the next stage in the evolution of an atherosclerotic lesion. It is characterized by a large number of extracellular lipids and true lipid core formation. From the pathomorphology point of view, an atheromatous plaque can be divided into three separate components.

- Atheroma — a nodular cluster of soft, flaky, yellowish material in the center of large plaques, consisting of macrophages, closer to the artery lumen.

- Deeper located areas with cholesterol crystals.
- Calcinosis located evev more deeper — at the external basis (in long-existing plaques).

Further progression of the atherosclerotic process naturally leads to the development of **fibroateroma** (Fig. 1.3). The atherosclerotic plaque at this stage has a lipid core and a fibrous capsule. The degree of fibrous changes severity in the atherosclerotic plaque depends on the type of foam cells. If in the atherosclerotic plaque foam cells of macrophage origin dominate, then the number of extracellular lipids in it is large, the lipid core is fairly well expressed, and the fibrous capsule is relatively thin. These plaques are termed "yellow". It is considered that they develop at an earlier stage, in contrast to fibrotic. Yellow plaques are softer and therefore more vulnerable, but they are more elastic and reduce the artery lumen to a small extent. It is supposed that the thin connective tissue capsule of an atherosclerotic plaque can be relatively easily damaged under the influence of high pressure in the artery, compression of the artery from outside, and other factors. The rupture of the connective tissue capsule of the atherosclerotic plaque is facilitated by proteolytic metalloproteinase enzymes produced by macrophages and mast cells that destroy the extracellular matrix. Thinning and rupture of a peculiar fibrous capsule by banal stretching can contribute to an increase in the size of the plaque core. If the foam cells are of myocytic origin, the lipid core is smaller (although it can protrude into the lumen of the vessel to a greater extent), fibrotic changes clearly predominate, the fibrous capsule is well expressed, dense — such a plaque is termed fibrotic, or "white". These plaques contribute to the occurrence of hemodynamically significant reduction of the artery. The size of plaques varies from a few millimeters to several centimeters in diameter, fibrous plaques can merge with each other, they grow gradually, increase in size due to the accumulation of lipids, the fibrous capsule and parietal thrombosis, which is formed as a result of cracks, ulceration, small tears of the fibrous covering, damage and death of endothelial cells above the plaque.

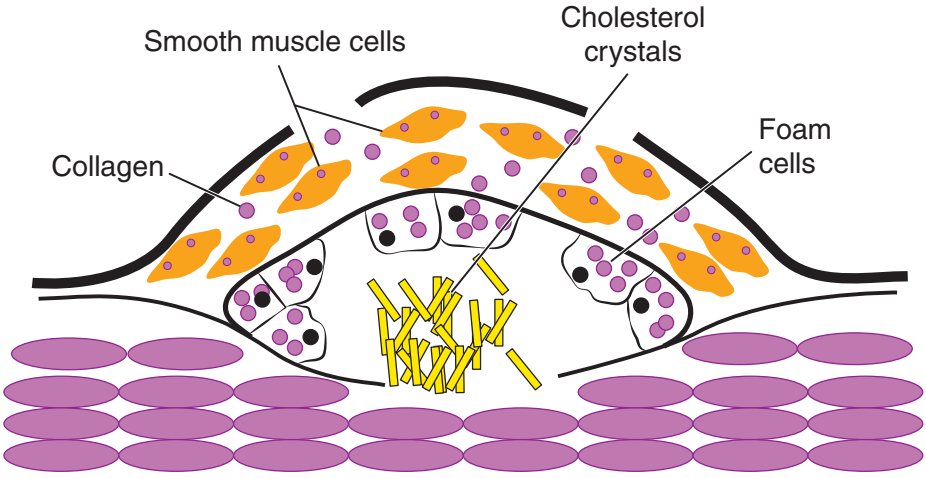

Fig. 1.3. Atherosclerotic plaque structure

In later stages, the plaques **are calcified**. The amount of calcium in the plaque is inversely proportional to the probability of its rupture and can be estimated with the use of modern methods of visualization (intravascular and tomographic). Abdominal aorta, coronary arteries, pelvic arteries, and femoral arteries are calcified most often. One of the stages in the atherosclerotic plaque evolution is critical stenosis of the affected artery and, consequently, ischemia of the corresponding organ. But ischemia is not the most dangerous manifestation of the late stages of atherosclerotic process, because with the slow growth of plaque, collateral blood circulation has time to form. The fact is that the course of atherosclerosis is characterized by multiple alternations of stable and unstable phases. It is the destabilization of the atherosclerotic plaque that is always of a great danger for the patient.

The principal pathomorphological characteristics of an unstable atherosclerotic plaque are considered erosions, cracks, tears, ruptures, and thrombosis. It is suggested that instability of a plaque is based on an increased activity of enzymes — metalloproteinases released by activated macrophages and damaging the fibrous membrane of the plaque. Plaque instability, especially in case of occlusive thrombosis, clinically leads to an acute event such as MI, stroke, or limb gangrene.

Clinical Pathologic Correlations in Atherosclerosis

Atherosclerosis is responsible for two main problems.

- Atheromatous plaques, despite long-term compensation due to distention of the artery lumen, finally break (erode) and/or lead to reduction of the artery lumen, and therefore to insufficient blood supply of the organs and tissues that they supply, i.e. contribute to their ischemia.
- Excessive compensatory artery distention leads to the formation of aneurysms.

Atherosclerosis tends to develop locally, in certain predisposed places. For example, in the coronary vessels system such a target is the proximal section of the anterior interventricular branch of the left coronary artery. Similarly, the preferred places for the development of atherosclerosis are considered the proximal sections of the renal arteries and the place of division (bifurcation) of the common carotid artery into internal and external arteries. This is associated with the fact that atherosclerotic lesions in the arteries are most often formed in the places of branches, i.e. where conditions for a disturbed (“twisted”, turbulent) blood flow exist. It is believed that the impaired laminar blood flow in the arteries causes impaired formation of nitric oxide by endotheliocytes.

Despite the systemic effect of known risk factors that predispose to the development of atherosclerosis, it selectively affects various pools of the arterial vascular bed and causes special clinical manifestations that depend on localization of arteries involved in the pathological process. Coronary arteries atherosclerosis most often leads to MI and angina pectoris. Atherosclerosis of the arteries that supply the central nervous system (CNS) most often leads to stroke and transient ischemic attacks. Peripheral vascular atherosclerosis causes intermittent claudication and gangrene. Visceral arteries atherosclerosis is responsible for mesenteric ischemia and necrosis, and renal arteries lesion can damage the kidneys due to ischemia caused by vessel reduction, and cause the development of renovascular hypertension.