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Tobacco-Related Cardiovascular Diseases in the 21st Century

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Smoking and Cardiovascular Disease

Mechanisms of Endothelial Dysfunction and Early Atherogenesis

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Abstract—Smoking represents one of the most important preventable risk factors for the development of atherosclerosis. The present review aims at providing a comprehensive summary of published data from clinical and animal studies, as well as results of basic research on the proatherogenic effect of smoking. Extensive search and review of literature revealed a vast amount of data on the influence of cigarette smoke and its constituents on early atherogenesis, particularly on endothelial cells. Vascular dysfunction induced by smoking is initiated by reduced nitric oxide (NO) bioavailability and further by the increased expression of adhesion molecules and subsequent endothelial dysfunction. Smoking-induced increased adherence of platelets and macrophages provokes the development of a procoagulant and inflammatory environment. After transendothelial migration and activation, macrophages take up oxidized lipoproteins arising from oxidative modifications and transdifferentiate into foam cells. In addition to direct physical damage to endothelial cells, smoking induces tissue remodeling, and prothrombotic processes together with activation of systemic inflammatory signals, all of which contribute to atherogenic vessel wall changes. There are still great gaps in our knowledge about the effects of smoking on cardiovascular disease. However, we know that smoking cessation is the most effective measure for reversing damage that has already occurred and preventing fatal cardiovascular outcomes. **(*Arterioscler Thromb Vasc Biol*. 2014;34:509-515.)**

Key Words: endothelial damage ■ flow-mediated dilatation ■ inflammation ■ lipid modification ■ nitric oxide ■ oxidative stress

In their pathogenesis, cardiovascular diseases (CVDs) are among the most complex human diseases. The interplay of genes, lifestyle, and the environment defines the time point of onset, mode of initiation, location of onset, sites most severely affected, dynamics of disease progression, and type of cardiovascular outcome. The large number of risk factors, physicochemical interactions, cell types, and biological processes involved add to the complexity of these diseases.

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Cigarette smoking is probably the most complex and the least understood among the risk factors for CVDs. This is because cigarette smoke contains ≈4000 different chemicals (with sizes from atoms to particulate matter).[1,2] These chemicals are further modified in the human body by detoxification systems, such as the CYP family members (cytochrome P450 oxidase family is involved, among others, in metabolization of toxic substances). Individual smoking behavior and intensity and the brand of cigarettes smoked further modulate the amount, number, and type of smoke chemicals to which an individual is exposed.[3] To date, apart from a handful of candidate compounds, the relevance of most compounds in cigarette smoke in CVD initiation, progression, and cardiovascular outcome has not been studied. Importantly, it is likely that it is not just a single compound or a compound class, such as oxidants, that are the CVD-relevant fraction of cigarette smoke but rather a highly complex and changing mixture of compounds that is responsible for disease initiation, progression, and cardiovascular outcome. The interplay of these compounds with the individual's genetic background and the individual environment defines the onset, location, and pace of CVDs.

For the past decades, it has been clear that smoking is an important (and modifiable) risk factor for CVDs; according to World Health Organization data, smoking is responsible for 10% of all CVD cases.[4] However, for a long time it remained unclear how smoking causes CVDs. In 1993, Celermajer et al[5] published a study showing that smoking reduces flow-mediated dilatation (FMD) in systemic arteries in healthy young adults. Because altered FMD is a well-defined and early marker for vascular (endothelial) dysfunction, the above study represented a clinical landmark in the understanding of smoking-induced CVD pathophysiology. In the following years, several experimental studies suggested a link between proatherogenic cellular and molecular effects of cigarette smoke and initiation of CVD.

This review summarizes the current knowledge on how cigarette smoking causes endothelial dysfunction and initiates atherogenesis.

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Clinical Data

Evidence for smoking-induced initial vascular damage and endothelial dysfunction stems from an array of clinical studies analyzing endothelial function using various techniques.[6] As mentioned above, in 1993, Celermajer et al[5] were able to show that continuous smoking impairs FMD of the brachial artery in a dose-dependent manner, shown by the strong association between FMD and pack years smoked. This was also found to be the case with coronary arteries in a study by Zeiher et al.[7] Interestingly, reduction of endothelium-dependent dilatation by smoking was reversible, and significant improvement of FMD 1 year after cessation has been reported.[8] Likewise, a recent study of Amato et al[9] revealed that smoking light cigarettes impairs FMD as much as smoking regular cigarettes, arguing against light cigarettes as a less harmful alternative.

Measurement of FMD represents a useful tool to assess the effects of smoking on the vascular wall. Independent of its FMD reducing activity, smoking was shown to induce other proatherogenic alterations in the vascular wall (eg, by deposition of smoke chemicals). The time needed to restore interrupted functions of the vascular endothelium will depend on the specific process that has caused the endothelial damage; some alterations of the endothelial wall may vanish more rapidly than others, without these being reflected by improved FMD.

Clinical studies assessing the interrelation of secondhand smoking and FMD reported strong correlations. Secondhand smoking was shown to impair endothelium-dependent arterial dilatation in young subjects,[10] which was shown to be reversible 1 year after cessation of exposure.[11]

Because FMD is a sensitive marker for smoke exposure as also for cessation, FMD was also used as a tool to assess the efficacy of treatment options for early smoking-induced proatherogenic changes in the vasculature. Heitzer et al[12] demonstrated that treatment of chronic smokers with tetrahydrobiopterin (BH4, cofactor of endothelial NO-synthase) improved smoking-impaired vasodilatation. Furthermore, the authors hypothesized that reduced NO production in chronic smokers may, in part, be caused by reduced BH4 availability. Furthermore, Papamichael et al[13] showed that acute smoking-induced impairment of FMD (healthy nonsmokers smoked 1 cigarette) can be improved by simultaneous consumption of red wine, probably because of its antioxidant properties.

Several studies in the past 40 years have analyzed the effect of smoking on serum lipids. However, the first clear results stem from 1989; Craig et al[14] showed a statistically significant correlation between smoking and increased total serum cholesterol, very–low-density lipoprotein, low-density lipoprotein (LDL), and triglyceride serum concentrations. Furthermore, they found that high-density lipoprotein and apolipoprotein A1 levels to be decreased in smokers, in a dose-dependent manner. Similar findings were reported by subsequent clinical studies, all demonstrating that smoking modifies serum lipid profiles in a proatherogenic manner.[15,16] Neufeld et al[17] found a significant association between secondhand smoking and impaired serum lipids in children at high risk for early heart disease (based on existing dyslipidemia). These children were exposed to secondhand smoke in the household and had significantly reduced levels of high-density lipoprotein. In contrast to the above-mentioned clear improvement of FMD after smoking cessation, there was only minor improvement in lipid profiles in response to cessation. Cessation led only to an increase in serum high-density lipoprotein levels, whereas the levels of total cholesterol, LDL, and triglyceride remained unchanged.[18,19]

Apart from modulating lipid quantities, smoking was also shown to change lipids qualitatively. Free radicals and oxidants present in cigarette smoke, as well as endogenously produced oxidants and radicals (resulting from the smoke chemical-induced alteration in the cellular redox system), cause a prooxidative environment.[20] This general shift is likely to contribute to lipid oxidation and to a general increase in oxidative modification (and inactivation) of biomolecules. Morrow et al[21] reported increased presence of lipid peroxidation products in the serum of smokers, and Salonen et al[22] found increased levels of circulating autoantibodies against oxidized LDL. Later, Yamaguchi et al[23] suggested that peroxynitrite (generated by a reaction between NO and superoxide anion) is involved in the oxidative modification of LDL in smokers' blood. Further support for these findings comes from studies by Pilz et al,[24] Reilly et al,[25] and Solak et al,[26] showing increased lipid oxidation, signs of oxidative stress, and impairment of antioxidant systems. It is well known that only oxidatively modified LDLs are recognized by macrophage scavenger receptors, taken up by these macrophages and causing their transformation into foam cells, which are essential elements in lipid deposition and plaque formation. Therefore, it is likely that in vivo the oxidation of lipids is another way by which smoking induces and accelerates atherosclerosis.

Inflammation is known to constitute an essential element in atherogenesis.[27] One of the most important risk factors for CVDs, smoking, influences and activates the immune system both systemically and locally. Smokers were shown to have significantly elevated white blood cell counts, which was tightly correlated to the formation of carotid atherosclerotic plaques.[28] Lavi et al[29] reported increased levels of neutrophils, lymphocytes, and monocytes in smokers when compared with nonsmokers. Furthermore, smokers were found to have significantly increased serum levels of proinflammatory cytokines, such as tumor necrosis factor α and interleukin-1β.[30] A widely accepted marker for the occurrence of inflammation is elevated serum C-reactive protein, which was found to be increased in smokers.[31,32] However, in vivo data on the mechanisms involved in smoking-induced inflammation are virtually absent.

Systemic immunologic alterations by smoking were found to correlate with local processes in the vascular wall (viz., in the atherosclerotic plaque), characterized by inflammation and increased expression of matrix metalloproteinases.[33] Interestingly, the systemic proinflammatory effect of smoking is not limited to active smokers. Also patients exposed to secondhand smoke exhibit increased concentrations of inflammatory markers.[31]

Furthermore, leukocyte recruitment mediated by endothelial adhesion molecules as an essential element in the initiation of atherosclerosis is increased by smoking. For example, Cavusoglu et al[34] showed that smoking increases the plasma concentration of soluble vascular cell adhesion molecule-1 in patients with coronary artery disease. Further evidence for the strong association between smoking and the increased expression of adhesion molecules comes from cell culture–based studies discussed below.

Data on the influence of smoking on blood pressure, one of the most important risk factors for the development of CVDs, are conflicting. Early epidemiological studies from the 1970s failed to find elevated blood pressure in smokers,[35,36] whereas data from the Annual Health Survey for England revealed a significant increase in blood pressure in old male smokers when compared with nonsmokers.[37] Other smoking status-related comparisons of blood pressure in this study did not reveal significant differences between smokers and nonsmokers. In summary, there may be an influence of smoking on blood pressure in defined subgroups. However, for the general population, there does not seem to be a close association between smoking status and increased blood pressure.

Finally smoking, whether active or secondhand, has dramatic effects on the coagulation system. Numerous studies have shown that exposure to cigarette smoke activates platelets, stimulates the coagulation cascade, and reduces fibrinolysis. In 2 recent review articles, the association between smoking, the coagulation system, and development of atherothrombosis were extensively discussed.[38,39]

Animal Studies

Animal experiments led to an increase in the understanding of smoking-induced atherogenesis. However, there are significant differences in the techniques to expose animals to cigarette smoke, ranging from intravenous or subcutaneous injection of smoke extracts to inhalation (the most physiological method). Furthermore, cigarette smoke can be generated in different ways, with resultant differences in the composition of smoke extracts (eg, nicotine-containing versus nicotine-free extracts). All these make results of studies difficult to compare; in addition, extrapolation of findings from animal studies to the situation in human smokers may not be feasible.

An animal study using C57/BL6 mice undertaken by Talukder et al[40] supported many results obtained in previous clinical studies. It was found that chronic cigarette smoke exposure causes oxidative stress and impairs endothelium-dependent vasorelaxation by reduction of NO bioavailability. Interestingly, the same study found blood pressure in smoke-exposed animals to be increased, which contradicts clinical findings. Cigarette smoke-mediated impairment of endothelium-dependent relaxation and the role of oxidants, radicals, and reduced NO in animals were previously described by studies in rats and rabbits.[41–44]

As mentioned above, clinical data clearly show the pro-oxidative (and therefore proatherogenic) potential of smoking, which are further supported by a limited number of animal studies. In humans and in animals (mice and rabbits), smoking increases serum levels of oxidative stress markers, oxidative modification of LDL, lipid peroxidation, and lipid deposition in the vessel wall and causes plaque development and progression.[45] In addition, Orosz et al[46] showed in vivo a link between smoking-induced increase in reactive oxygen species and the expression of proinflammatory cytokines (interleukin-1β, interleukin-6, and tumor necrosis factor α) in the vascular wall by the activation of NAD(P)H oxidase.

Of interest are studies addressing the effect of smoke exposure when combined with other CVD risk factors, for example, secondhand smoking combined with infection with Chlamydia pneumonia, all of which show increased atherosclerosis development.[47]

The induction of hyperlipidemia by smoking has been shown in humans, whereas the data from animal experiments are ambiguous. Yamaguchi et al[48] showed that injection of cigarette smoke extract in rabbits leads to an increase in LDL cholesterol levels. In contrast, inhaled smoke was reported by Kunimoto et al[45] to have no effect on serum cholesterol and triglyceride levels in ApoE$^{-/-}$ mice.

In conclusion, results from animal studies confirmed and extended clinical findings; however, few animal studies provide new data on the mechanisms of cigarette smoke-induced endothelial dysfunction, damage, and initiation of atherogenesis.

In Vitro Studies

Smoking reduces FMD in vivo, the earliest marker of atherogenesis and vascular dysfunction. Experimental studies showed that this clinical phenomenon is based on a significant reduction of NO bioavailability in the vasculature (Figure). Cigarette smoke contains ≈10^{17} long-lived radicals/g tar (hydrophobic) fraction and 10^{15} radicals/g of the volatile fraction.[49] Particularly superoxide anion is known to reduce bioactive NO by the formation of peroxynitrite, which itself leads to protein nitration and oxidation of LDL.[50] The shift to a pro-oxidative, hence, NO level-reducing environment in the vascular wall of smokers is further facilitated by the deposition of oxidation catalyzing metals in cigarette smoke, as well as by an alteration in the balance between of oxidant-generating and oxidant-reducing cellular systems, in favor of the former.[51,52] Macrophages, neutrophils, the mitochondrial respiratory chain, and xanthine oxidases are major sources of the increased levels of oxidants in the vascular wall of smokers. Furthermore, stable aldehydes in cigarette smoke increase reactive oxygen species production by the activation of NADPH oxidases. Beyond that, smoking compounds have been proven to increase eNOS acetylation and expression while decreasing and uncoupling eNOS activity.[53–55] Smokers further exhibit reduced levels of selenium, a central element in many antioxidant systems.[51]

The shift to a vascular pro-oxidative state not only reduces NO levels but may also significantly contribute to lipid oxidation,[50,56] foam cell formation, foam cell death, and inflammation. Inflammation and oxidation are central elements in the activation of several CVD-relevant cell types, such as macrophages, endothelial cells, and platelets. Cigarette smoke chemicals were reported to lead to adhesion molecule expression on the surface of endothelial cells and induce the release of proatherogenic cytokines, such as interleukin-6 and interleukin-8.[57] These processes, in combination with cigarette smoke-induced increase in the number and activation of platelets, lead to a significant shift toward a prothrombotic and procoagulative state in the vascular wall of smokers.[38] The core of these processes is the activation of the NFkB cascade. In human lung epithelial cells and fibroblasts, it was recently shown that cigarette smoke activates mitogen- and stress-activated kinase by phosphorylation at Thr581 leading to activation of NFkB and downstream proinflammatory genes, involving a modification of chromatin structure.[58]

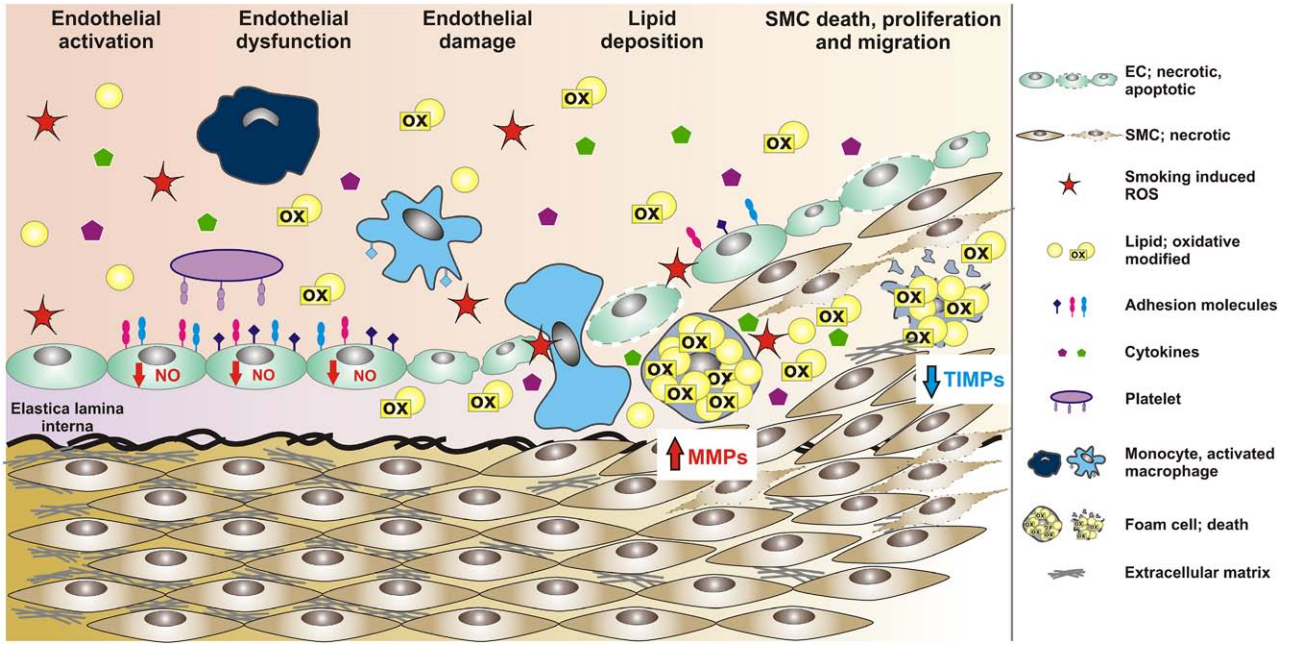

Figure. Schematic representation of smoking-induced signaling pathways in the vessel wall. Cigarette smoke–induced oxidative (OX) stress was shown to activate the endothelium by induction of adhesion molecule expression (eg, intracellular adhesion molecule, vascular cell adhesion molecule), as well as macrophages and platelets. Endothelial activation is characterized by the reduction of NO levels within the cells and resulting in loss of function of smooth muscle cells (SMCs) in the vessel media. In response to smoke exposure, endothelial cells are known to release inflammatory and proatherogenic cytokines. All these processes lead to endothelial dysfunction. Direct physical effects of smoke compounds and produced reactive oxygen species (ROS) lead to endothelial cell loss by apoptosis or necrosis. Besides endothelial cells, also macrophages are activated by the expression of adhesion molecule receptors recognizing adhesion molecules on endothelial cells. After adhesion and transendothelial migration, macrophages take up oxidized lipids produced by oxidative modification through smoke-increased ROS production. Scavenger receptor-mediated uptake of lipids induces the formation of so-called foam cells within the aortic wall, and subsequent death of foam cells induces the release of these lipids and the formation of lipid-rich aortic plaques. Likewise, it is postulated that smoking induces an increase in SMC proliferation and migration provoking intimal thickening and plaque formation. Triggering of SMC death by necrosis is a further consequence of exposure to smoke that triggers inflammatory signals, as well as the release of intracellular proteolytic enzymes inducing cleavage of extracellular matrix proteins. Destruction of extracellular matrix proteins is further enhanced by increased expression of matrix metalloproteinases (MMPs) and reduced expression of tissue inhibitors of MMPs (TIMPs).

Apart from triggering immune system-activating oxidant reactions and damage, cigarette smoke, with its pathogen-associated molecular patterns (microorganisms on tobacco), modulates innate immune system function. Cigarette smoke extract, by causing protein carbonylation, was shown to reduce alveolar macrophage sensitivity to lipopolysaccharides and tumor necrosis factor α.[59] Because chronic infections are well known to constitute a risk factor for CVDs, the above findings may also be relevant for CVDs. However, this aspect has not been studied in detail. The role of Toll-like receptors in smoking-induced lung diseases (and potentially also in CVDs) is, also, still not clear. Some data suggest that Toll-like receptor expression and function in cigarette smoking are causally linked to COX and NADPH activation, oxidative stress, and damage,[60] whereas other results show that the activation of Toll-like receptor-9 by stimulation with an agonist reduces lung inflammation in response to secondhand smoke.[61]

Several experimental studies demonstrated that, apart from causing vascular endothelial dysfunction, cigarette smoke causes physical damage to the vascular endothelium (Figure). In essence, 2 forms of damage were observed: (1) contraction of endothelial cells, mediated by oxidation and collapse of the tubulin system, which is reversible and may be part of the clinical symptoms of smoking-caused reduction of FMD,[57] and (2) endothelial cell death. In general, all forms of cell death, that is, apoptosis, necrosis, programmed necrosis (necroptosis), and autophagy, were found to be induced by cigarette smoke chemicals. Importantly, the use of different preparations of cigarette smoke chemicals by different groups, concentrations, and cell types used may explain some of the reported effect discrepancies. We have shown that the hydrophilic fraction of cigarette smoke activates an autophagic signaling pathway initiated by endoplasmic reticulum stress. At lower concentrations, the hydrophobic fraction induces a programmed, caspase-independent form of cell death. However, at higher concentrations, an apoptosis-inducing factor-mediated form of cell death causes lysosomal membrane permeabiliztion-dependent form of necrosis. In all these processes, reactive oxygen species play a central role.[62–64] Recent data suggest that cadmium present in cigarette smoke may play a significant role in causing endothelial cell death.[65,66]

Clearly, lack of knowledge about the concentrations of cigarette smoke chemicals in smokers' vascular tissue, and even more importantly, their identity still represents a major problem for in vitro cigarette smoke studies.

Apart from the significant and clinically proven relevance of endothelial dysfunction in CVD initiation, loss of endothelial integrity is assumed to play a crucial role. Alterations in endothelial cell structure and the induction of cell death reduce endothelial functions, may contribute to thrombogenic events,

and promote inflammation (adhesion molecule expression, cytokine release, endothelial necrosis, lack of barrier function; Figure). Barbieri et al,[67] for instance, demonstrate that the combination of cigarette smoke extract and interleukin-1β deactivated phosphatase and tensin homolog, which may contribute to endothelial junction disassembly.

The effects of cigarette smoke on other CVD-relevant vascular cell types were much less studied. However, several studies found that cigarette smoke increases vascular smooth muscle cell proliferation and migration. Proliferation of smooth muscle cells seems to be mediated via activation of the platelet-derived growth factor–protein kinase C signaling cascade.[68,69] Furthermore, polycyclic aromatic hydrocarbons (part of the hydrophobic fraction of cigarette smoke) were shown to activate aryl hydrocarbon receptor, which induces iNOS, leading finally to intimal thickening.[70] Likewise, polycyclic aromatic hydrocarbon–activated aryl hydrocarbon receptors can induce the transcription of receptors for chemokines and adhesion molecule receptors for leukocytes, thus amplifying inflammatory signals on the vascular endothelium.[71]

Alterations of vascular extracellular matrix and tissue remodeling processes in general are central elements in atherogenesis (Figure). Several studies showed that cigarette smoke leads to an increased expression and activity of matrix metalloproteinases (MMPs), as well as to decreased expression of MMP inhibitors (eg, tissue inhibitors of MMPs). For instance, cigarette smoke extract and acrolein (a combustion product of the tobacco additive glycerin) were shown to increase MMP-1 expression, an effect that may be mediated by the inhibition of the mammalian target of rapamycin pathway.[72] In the same study, it was also reported that cigarette smoke extract and acrolein reduce tissue inhibitors of MMPs-3 levels in aortic endothelial cells. Moreover, it was shown that cigarette smoke condensate and nicotine increase MMP-1, MMP-8, and MMP-9 expression, increase xenobiotic metabolism genes, and reduce genes responsible for proliferation.[73] Apart from the specific effects of cigarette smoke chemicals on MMP-activating and tissue inhibitors of MMPs–decreasing cellular pathways, necrotic cell death of endothelial cells and smooth muscle cells can lead to proteolysis of extracellular matrix, by the release of, for example, lysosomal proteases. The proteolytic environment is further enhanced by inflammatory cells, such as neutrophils.[74] Apart from contributing to classical atherogenesis, degradation of extracellular matrix induced by chemicals in cigarette smoke is likely to play a significant role in the formation of aneurysms, in which smoking is one of the few known risk factors.

Most studies on the effects of cigarette smoke on macrophages were conducted in a pulmonary setting. Data on vascular macrophages are sparse. However, it is likely that similar observations can also be made in the vascular system and may be involved in atherogenesis. Despite the fact that smoking causes inflammation, at the level of macrophages, it is interesting to note that cigarette smoke chemicals seem to reduce the activity and function of macrophages. Previous studies showed that cigarette smoke extract reduced proinflammatory cytokine expression in response to lipopolysaccharides or tumor necrosis factor α stimulation but did not prevent pseudopodia formation in alveolar macrophages. The central hypothesis of this study was that massive protein carbonylation accounts for the observed inhibition of NFkB signaling.[59] One may hypothesize that reduced macrophage function caused by smoking may lead to the accumulation of otherwise phagocytosed and disposed cell remnants (including cholesterol) in the vasculature. This hypothetical effect would reduce vascular repair and function and may accelerate the formation of a necrotic core within an atherosclerotic plaque.

Cigarette smoke is well known to interfere with the coagulation system dramatically and to cause a shift to a local and systemic prothrombotic state. Importantly, cigarette smoking was shown to have an effect at all stages of atherogenesis, such as plaque formation, plaque stability, etc. Because these aspects were recently discussed in depth by our team and Barua et al, we would herewith like to refer to the following reviews.[38,39]

Summary

A modern cigarette is a high-tech product, designed to deliver nicotine efficiently without an unpleasant taste but with a reduced potential to cause throat irritation and coughing and thus to ultimately maximize pleasure for the smoker. Together with the aggressive merchandising practices and the highly addictive nature of nicotine, cigarettes are currently 1 of the most dangerous drugs freely available to human beings. Smoking kills 6 million humans per year, with ≈10% of these deaths related to secondhand smoking. Smoking is responsible for an alarming but preventable 10% of CVDs.[75]

Smoking plays a strong role not only in CVD initiation but also significantly contributes to and causes disease progression and fatal cardiovascular outcomes. The key processes in smoking-induced atherogenesis initiation are endothelial dysfunction and damage, increase in and oxidation of proatherogenic lipids, as well as decrease of high-density lipoprotein, induction of inflammation, and the shift toward a procoagulant state in the circulation (Figure). Current data clearly show that also secondhand smoking can trigger life-threatening conditions (including in children).

The addictive nature of smoking and the increasing number of smokers worldwide make it imperative to intensify research to (1) enable greater understanding of the health damage caused by smoking, (2) develop markers for smoke exposure (there is not a single marker available to determine long-term exposure), and (3) to design more effective therapies of CVDs specifically caused by smoking.

Smoking cessation is and will remain the most effective measure to prevent smoking-caused CVDs. However, long-term smokers who have quit smoking and secondhand smokers will benefit from increased knowledge of the association between cigarette smoke and CVDs, availability of new exposure markers, and novel therapies.

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Disclosures

None.

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Significance

The present review focuses on the effect of cigarette smoking on the vascular endothelium and the initial events in the process of lesion development in atherosclerosis. On the basis of an extensive literature search, this review offers a comprehensive summary of results of basic science experiments over animal testing to human studies performed for the past 40 years. The major events by which smoking initiates atherogenesis are (1) endothelial activation and dysfunction by reducing vascular NO availability, biomolecule oxidation, and endothelial damage, (2) by the proatherogenic alteration of lipid profiles and lipid oxidation, (3) by the induction of a local and systemic proinflammatory status, and (4) by causing a procoagulative environment. Secondhand smoking causes effects similar to active smoking. Significant lack of knowledge exists in the following fields: proatherogenic smoke chemicals are not well defined. Markers for nonacute smoke exposure and smoking-caused atherosclerotic processes are missing. Not a single smoke-specific therapy is available.