Interaction of carbamylated LDL with LOX-1 in the induction of endothelial dysfunction and atherosclerosis

Jawahar L. Mehta* and Alexei G. Basnakan

Department of Medicine, University of Arkansas for Medical Sciences, and Central Arkansas Veterans Healthcare System, Little Rock, AR, USA

This editorial refers to ‘Carbamylated low-density lipoprotein induces endothelial dysfunction’, by T. Speer et al., on page 3021.

Urea is a normal component of human blood plasma, but it is not inert. Urea-derived cyanate modification of LDL-cholesterol, known as LDL carbamylation, has been discussed for more than a decade. Recent studies have linked LDL carbamylation to atherosclerosis in uremic patients and animals with chronic kidney disease (CKD). More broadly, protein carbamylation has been linked with atherosclerotic cardiovascular disease (CVD) in CKD. Endothelial cell (EC) dysfunction induced by carbamylated LDL (cLDL) seems to be a plausible explanation for the high frequency of CVD in CKD. However, despite considerable efforts in determining the mechanistic basis of atherosclerosis induced by cLDL, the link remains blurry since it is hard to relate the complex events based on in vitro, in vivo, and clinical studies done independently.

An important contribution by Speer and associates is that they were able to combine all these approaches into a unified concept. As opposed to many previous studies which used LDL artificially carbamylated by potassium cyanate, or measured protein carbamylation, or measured plasma cLDL by enzyme-linked immunosorbent assay (ELISA), these investigators isolated LDL from plasma samples and measured its carbamylation directly using HPLC. Surprisingly, this natural cLDL in CKD patients was found to contain more carbamylation sites than artificially ex vivo cLDL. This may be an indication of more than one mechanism of LDL carbamylation in the human body, or the presence of factors in blood that affect the efficiency of LDL carbamylation.

The authors determined the link of LDL-carbamyl-lysine levels with cardiovascular (CV) outcomes in patients with CKD followed for a median duration of 4.7 years. Their remarkable finding was that LDL-carbamyl-lysine levels in these patients were significant predictors for CV events and all-cause mortality. Taken together with recent animal and clinical studies, these data link cLDL with vascular dysfunction and adverse outcome in patients with CKD.

Findings by this team confirmed several previous observations and moved the discovery forward by providing evidence of an interesting new mechanism. The authors examined vascular reactivity in isolated aortic rings, and measured reactive oxygen species (ROS) and nitric oxide (NO) production by electron-spin resonance spectroscopy. While native LDL (nLDL) showed no effect, cLDL impaired EC-dependent relaxation, but not EC-independent relaxation. cLDL increased ROS production in aortic rings by activating NADPH oxidase, and stimulated endothelial nitric oxide synthase (eNOS) uncoupling apparently by promoting S-glutathionylation of eNOS. Importantly, by using aortic rings from lectin-like oxidized LDL (oxLDL) receptor (LOX-1) transgenic mice, Speer et al demonstrated that the impaired EC-dependent relaxation was more pronounced in the LOX-1 transgenic mice.

These authors further confirmed that it is the activation of LOX-1 that mediates cLDL-induced EC dysfunction by examining NO production using small interfering RNA (siRNA) targeting LOX-1. They showed that silencing of LOX-1 abrogated the inhibitory effect of cLDL on EC NO release. Nonetheless, while the effect of cLDL being mediated through LOX-1 activation is clearly suggested, demonstration of LOX-1 gene up-regulation in ECs by cLDL and absence of cLDL’s effect on vasoreactivity in LOX-1 knockout mice might have provided additional evidence of a cLDL–LOX-1 pathway.

These findings are an important addition to previous reports of cLDL-induced dysfunction of ECs shown mainly in vitro studies. Previous studies showed that cLDL is capable of binding to ECs and cLDL may transmigrate through ECs, activate the mitogen-activated protein kinase (MAPK) pathway, and induce injury. It also induces monocyte adhesion and activation and their transformation into macrophages utilizing a unique spectrum of scavenger receptors. cLDL also activates smooth muscle cells (SMCs) and induces
Further support for the critical role of LOX-1 in atherogenous signals, and SMC proliferation, and restoration of EC-dependent oxidation of ROS, reduction in eNOS synthesis, and activation of the inflammatory cascade11 (Figure 1). LOX-1 can itself act as an adhesion molecule since human peripheral blood mononuclear cells and the monocyte cell line THP-1 bind to a recombinant LOX-1-coated surface.12 The importance of LOX-1 became evident in recent studies in LDL receptor null mice on a high fat diet in which abrogation of LOX-1 resulted in reduction in oxidative stress, inflammatory signals, and SMC proliferation, and restoration of EC-dependent vasorelaxation; all of these collectively resulted in decreased atherogenesis.13 Further support for the critical role of LOX-1 in atherogenesis comes from the multitude of in vitro studies implicating oxLDL, but not nLDL, in forcing proatherogenic phenotypes of vascular cells.14 Recently, an elegant study on oxLDL misdirection15 showed that removal of oxLDL from the circulation with overexpression of LOX-1 in the liver drastically inhibited formation of atherosclerotic lesions in ApoE knockout mice without affecting any other parameter of the lipid profile. Speer et al.16 found that cLDL stimulates both whole blood and aortic production of ROS via LOX-1 activation. This indicates that cLDL may cause oxidation of LDL in plasma and vascular walls, and thus may potentially cause oxLDL production or lead to dual modification of LDL. Taken together with the previously demonstrated myeloperoxidase pathway of LDL carbamylation through oxidation,4 it now becomes clear that the two modifications are tightly linked and may, perhaps, coincide on the same LDL particle. It is interesting that such an isoform, carbamylated-oxidized LDL, has recently been demonstrated in another study.16

Newer pathways of oxidative stress and inflammation in atherogenesis are being described with increasing frequency. Some of these pathways are more important in patients with CKD, and others in patients with conditions such as dyslipidaemia and diabetes. Therapeutic interventions may tell us which pathways are critical and which are redundant in different disease states. It is likely that there are connections linking multiple pathways. The study by Speer et al.17 is an important step in the right direction.

Conflict of interest: none declared.

References
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