CD31: beyond a marker for endothelial cells

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This editorial refers to ‘A CD31-derived peptide prevents angiotensin II-induced atherosclerosis progression and aneurysm formation’ by G. Fornasa et al., pp. 30–37, this issue.

CD31 is a 130 kDa platelet–endothelial cell (EC) adhesion molecule that was initially identified from ECs and platelets\textsuperscript{1} and later from blood leucocytes.\textsuperscript{2} Mature CD31 contains a short (22-amino acid (aa)) NH\textsubscript{2}-terminal peptide followed by six C2-type immunoglobulin (Ig) domains, each flanked by two conserved cysteine residues outside the cells,\textsuperscript{3} a 19 aa transmembrane domain, and a 118 aa cytoplasmic tail containing two immunotyrosine-based inhibitory motifs (ITIM)\textsuperscript{4} (Figure 1) that mediate intracellular signalling. Although CD31 was initially classified as a cell adhesion molecule,\textsuperscript{3} later studies suggested that CD31 triggers downstream inhibitory signalling\textsuperscript{5} upon transhomophilic CD31 engagement during cell–cell interaction.\textsuperscript{5} CD31 signalling participates in the regulation of leucocyte detachment, T-cell activation, platelet activation, and angiogenesis, all of which are critical to the pathogenesis of atherosclerosis and abdominal aortic aneurysms (AAAs).

Fornasa et al.\textsuperscript{6} use atherosclerosis-prone apolipoprotein E-deficient (Apoe\textsuperscript{-/-}) mice to demonstrate that aa551–574—a synthetic peptide located to the carboxyl-terminal of the Ig domain 6 (Figure 1)—suppressed angiotensin II (Ang-II) perfusion-induced AAAs and atherosclerosis. This peptide reduced atherosclerotic lesion and peri-atherosclerotic and AAA lesions, its expression in other cells makes its pathobiology complicated. In certain environments, cells may express different forms of CD31 that exert different functions. Each domain on CD31 (Figure 1) plays distinct roles in the cells as well as in the development of vascular and other inflammatory diseases. For example, CD31 loses its intercellular junction expression pattern after ECs are exposed to inflammatory cytokines. Neutrophils lose CD31 surface expression after in vitro transendothelial migration or extravasation in human skin transplanted to severe combined immunodeficiency mice.\textsuperscript{7,8} Prior studies showed that CD31 is cleaved between domains 5 and 6 and secreted to the media in primarily cultured human T cells or T-cell lines, thereby enhancing T-cell activation.\textsuperscript{9} A lack of CD31 signalling enhances T-cell activation\textsuperscript{10} and increases T-cell infiltration to atherosclerotic arteries.\textsuperscript{11} Therefore, at least some of the extracellular domains of CD31 are required for CD31 signalling and T-cell activation suppression, which may get lost during the development of atherosclerosis and AAAs. Indeed, Fornasa et al.\textsuperscript{6} demonstrate that CD4\textsuperscript{+} T cells from mouse atherosclerotic lesions, or from peripheral blood from mice with atherosclerosis or AAAs, lack extracellular CD31. Exogenous CD31 peptide aa551–574 from humans or mice (Figure 1) showed a dose-dependent suppression of human T-cell immune responses.\textsuperscript{9} In vitro, aa551–574 suppressed CD3 and CD28 cross-linking-induced mouse splenocyte intracellular Ca\textsuperscript{2+} mobilization. In a mouse model of delayed hypersensitivity, this peptide reduced 2-chloro-3-nitrobenzene-elicited ear thickness.\textsuperscript{9} The reduced atherosclerosis and AAA in the study by Fornasa et al. therefore may result from aa551–574 participation in suppressed T-cell immune responses. Although aa551–574 did not affect the total number of circulating blood cells, it reduced CD69\textsuperscript{+}-activated T cells and inflammatory cytokine secretion and reciprocally increased peripheral CD4\textsuperscript{+} CD25\textsuperscript{+}FoxP3\textsuperscript{+} regulatory T cells.\textsuperscript{6}

The study by Fornasa et al. focuses on the role of CD31 in atherosclerosis and AAAs, but CD31 also may contribute to other T-cell-associated human diseases and may affect any CD31-expressing cells. Fornasa et al. used CD31 peptide aa551–574 to reduce CD4\textsuperscript{+}-specific immune responses against oxidized low-density lipoprotein (oxLDL)—an autoantigen that reduces extracellular CD31 with a concomitant increase in soluble CD31 in the supernatant in cultured CD4\textsuperscript{+} T cells,\textsuperscript{9} suggesting that CD31 participates in T-cell autoimmunity. In atherosclerotic plaques and in the aneurysmal peri-aorta, CD31 peptide also targets macrophages, leading to reduced intracellular protease expression and reduced activity and release of macrophage cytokines and chemokines (e.g. IL6, MCP-1, MIP-1α, and MIP-1β).\textsuperscript{6} Macrophages are probably the most abundant inflammatory cells in atherosclerotic and AAA lesions. Although whether CD31 expression on macrophages\textsuperscript{12} changed upon Ang-II perfusion and atherosclerosis and AAA initiation was not tested, aa551–574 in situ targeting on macrophages in vivo and effective inhibition of macrophage activity in vitro extend CD31 pathobiology to this common inflammatory cell type that is implicated in most, if not all, human inflammatory diseases.

The role of CD31 in T-cell activation has been implicated previously in thrombosis, atherosclerosis, AAAs, and many other inflammatory...
were immunized with minimally modified LDL, CD4+ with ageing, and thrombosis risk increases concomitantly.13 In older females, in multiple sclerosis and AAAs, in humans and in mice. Immunosuppressive activities, and with increased incidence of atherosclerosis, arthritis, and experimental autoimmune encephalomyelitis. In human blood, both CD4+ and CD8+ T cells lose CD31 with ageing, and thrombosis risk increases concomitantly.13 In older Apoe−/− mice (both sexes), CD4+CD31+ cell numbers in aortic root lesions or in the circulation are three-fold lower in mice with thrombus than in those without thrombus. When Apoe−/− mice were immunized with minimally modified LDL, CD4+CD31+ cells from the spleen proliferated twice as fast as CD4−CD31+ cells did.11 These observations suggest that thrombosis reduces CD31 expression, thereby increasing T-cell proliferation. In humans, peripheral CD4+CD31+ and CD8+CD31+ cell numbers decrease significantly in patients with AAAs, with reciprocal increases in CD4−CD31− and CD8−CD31− cells. Peripheral CD4+CD31− cell numbers correlate positively (R = 0.324, P < 0.01), whereas CD8+CD31− cell numbers correlate negatively (R = 0.244, P < 0.05) with AAA cross-section surface area.14 Fornasa et al.6 presented the same observations in experimental AAAs and atherosclerosis as those made in humans.14 Lack of intact CD31 expression associates with loss of immunosuppressive activities, and with increased incidence of atherosclerosis and AAAs, in humans and in mice.

Studies from the same group revealed some controversial observations. In female Apoe−/− mice, intramuscular gene transfer of the entire CD31 extracellular portion, or those missing Ig domains 1–2, developed different phenotypes. After 6 months, mice that received the DNA construct containing the entire CD31 extracellular portion demonstrated significantly reduced atherosclerotic lesion size, intraplaque fibrin deposition, and Th1.2+ T-cell infiltration, peripheral-activated T-cell (CD3−CD4+CD69+) numbers, and spleen T-cell proliferation compared with those treated with the DNA construct missing the Ig domains 1–2 or with vehicle alone.15 These observations indirectly suggest that Ig domains 1–2 help regulate T-cell activation and atherosclerosis, while the presence of Ig domain 6, which contains the peptide aa551–574, is not sufficient to suppress T-cell activities. It is possible that Ig domains 1–2 are required to mediate CD31 transhomophilic interaction on cells expressing intact CD31.5 In contrast, on cells expressing truncated CD31—such as T cells and macrophages from atherosclerotic and AAA lesions—the remaining juxtamembrane extracellular fragment of Ig domain 6 is exposed on leucocytes after activation-induced shedding of CD31. With undefined molecular mechanisms, peptide aa551–574 may restore the CD31/SHP2 inhibitory pathway that was otherwise invalidated by the loss of the transhomophilic Ig domains 1–2 with CD31 shedding. The study by Fornasa et al. proposes a therapeutic application of this peptide in experimental atherosclerosis and AAAs.

Conflict of interest: none declared.

References

Figure 1 CD31 protein domains and their corresponding cellular functions. Ig, immunoglobulin; aa, amino acid; ITIM, immunotyrosine-based inhibitory motif; SHP2, Src homology-2 phosphatase; Y, tyrosine.


