Ac-SDKP suppresses TNF-α-induced ICAM-1 expression in endothelial cells via inhibition of IκB kinase and NF-κB activation

Liping Zhu,1 Xiao-Ping Yang,1 Branislava Janic,1 Nour-Eddine Rhaleb,1 Pamela Harding,1 Pablo Nakagawa,1 Edward L. Peterson,2 and Oscar A. Carretero1

1Hypertension and Vascular Research Division, Department of Internal Medicine, Henry Ford Hospital, Detroit, Michigan; 2Department of Public Health Sciences, Henry Ford Hospital, Detroit, Michigan

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NEW & NOTEWORTHY

TNF-α is one of the major inflammatory cytokines that plays an important function in inflammation and is increased in patients with cardiovascular disease. TNF-α primarily targets vasculature and upregulates adhesion molecules such as ICAM-1. Our study demonstrated that N-acetyl-Ser-Asp-Lys-Pro (Ac-SDKP) diminished TNF-α-induced ICAM-1 expression mainly through inhibition of NF-κB pathway.

INFLAMMATION PLAYS A KEY ROLE in the initiation, development, and progression of cardiovascular diseases, such as hypertension and atherosclerosis (38). One of the critical steps in the inflammatory response is the recruitment of leukocytes to the inflammatory lesion sites (20). This is achieved through processes of leukocyte adhesion and transendothelial migration in which cell adhesion molecules expressed on endothelial cells, such as ICAM-1, play an important role. These molecules bind to different ligands expressed on circulating leukocytes and are upregulated in response to proinflammatory stimuli, such as TNF-α. The increase in endothelial ICAM-1 expression in response to inflammatory stimuli was shown to be in temporal and spatial association with the accumulation of inflammatory cells within the affected tissue (1). Therefore, factors affecting endothelial adhesion molecule expression are important in regulating inflammatory processes.

N-acetyl-Ser-Asp-Lys-Pro (Ac-SDKP) is a naturally occurring tetrapeptide with anti-inflammatory and antifibrotic properties (32). Ac-SDKP is hydrolyzed mainly by angiotensin-converting enzyme (ACE), and it contributes to the anti-inflammatory and antifibrotic effects of ACE inhibitors. It has been shown that Ac-SDKP exerts antifibrotic effects by binding to Ac-SDKP receptors in rat cardiac fibroblasts (43). We previously reported that Ac-SDKP decreased cardiac and renal inflammatory cell infiltration in various models of hypertension, as well as in the model of heart failure after myocardial infarction (27, 28, 33). In addition, Lin et al. (14) demonstrated that Ac-SDKP prevented macrophage infiltration in the aorta in rats with angiotensin II (Ang II)-induced hypertension, and it suppressed ICAM-1 mRNA that is often used as a marker of endothelial cell inflammatory response (15). Furthermore, Nakagawa et al. (22) showed that Ac-SDKP reduced ICAM-1 protein expression in experimental autoimmune myocarditis (EAM) (22). Altogether, these results indicate that, under inflammatory conditions, Ac-SDKP affects ICAM-1 expression. However, the molecular mechanisms involved in Ac-SDKP-mediated anti-inflammatory effects on endothelial cells, including ICAM-1 downregulation, are still largely unclear.

TNF-α is one of the major inflammatory cytokines produced by a number of cells, including vascular endothelial cells (31), that plays an important function in inflammation (35). TNF-α-induced ICAM-1 expression in the endothelium is achieved through the activation of so-called “canonical” or “classical” NF-κB signaling pathway that depends on IκK-β and IκK-γ and induces the transcription of genes that regulate inflammation and cell survival (37). TNF-α initiates ligand-dependent oligomerization of the TNF-α receptor 1 (TNFR1) and recruitment of TNFR-associated factors (TRAFs) to the cytosolic TNFR1 domain (9, 42). These factors activate IκK-β and IκK-γ subunits of IκK complex by phosphorylation and conformational change. Activated (phosphorylated) IκK-β in turn phosphorylates IκB inhibitor on two specific serine residues,
and this causes IkB degradation and the release of NF-κB. Released NF-κB is translocated into the nucleus, where it binds to specific promoter regions of target genes (19, 24). TNF-α signaling was also shown to be transduced via p-38 and ERK MAP kinase pathways (41). The aim of this work was to evaluate the effect of Ac-SDKP on TNF-α-stimulated ICAM-1 expression. We hypothesized that, in human coronary artery endothelial cells (HCAECs), Ac-SDKP suppresses TNF-α-induced ICAM-1 expression by suppressing NF-κB activation via inhibition of IKK-β phosphorylation.

**MATERIALS AND METHODS**

**Materials.** Endothelial cell growth medium-2 (EGM-2) and endothelial cell basal medium (EBM) were purchased from Lonza. Protease inhibitors and phosphatase inhibitors for Western blotting were purchased from Roche. Recombinant human TNF-α was purchased from R&D Systems. IKK inhibitor IMD-0354 (IMD) and the antibody raised against ICAM-1 were purchased from Santa Cruz Biotechnology. The antibodies against phospho-IKK, phospho-p38, total p38, phospho-IκB, IκB, phospho-ERK, total ERK, and GAPDH were from Cell Signaling Technology. Ac-SDKP was purchased from Bachem.

**Cell culture and Ac-SDKP treatment.** HCAECs were purchased from Lonza and maintained in EGM-2 supplemented with 5% FBS. The cells were incubated at 37°C in 5% CO₂-95% air. On the day before experiments, HCAECs were starved in EGM-2 medium containing 0.5% FBS, at 37°C for overnight, and then switched to serum-free EBM medium during the treatment. HCAECs were treated with vehicle (control), TNF-α (0.5 ng/ml), TNF-α plus Ac-SDKP (10 nM), or Ac-SDKP alone. Cells were preincubated with Ac-SDKP for 30 min before the addition of TNF-α. On the basis of our previous experience in performing time-course experiments, TNF-α-induced phospho-IκB reached the highest level at 15 min; therefore, this time point was chosen to stimulate cells with TNF-α to assess phospho-IKK, phospho-IκB, phospho-p38, and phospho-ERK. An ACE inhibitor captopril (10 μM) was added to all groups to prevent breakdown of Ac-SDKP by ACE that is expressed by endothelial cells because the previous study has shown that the addition of captopril to the cell culture media increased the peptide half-life (7).

**Cell lysate preparation and Western blot analysis.** For cell lysate preparation, HCAECs were washed with cold PBS and lysed in lysis buffer (20 mM Tris·HCl, pH 7.5, 150 mM NaCl, 1 mM EDTA, 1 mM EGTA, 2.5 mM sodium pyrophosphate, 1 mM sodium orthovanadate, 1 mM β-glycerophosphate, 1% Triton X-100, 1 mM PMSF, protease, and phosphatase inhibitor cocktail) while being vortexed. Cell lysates were then incubated on ice for 30 min and centrifuged (14,000 rpm, 10 min) at 4°C, and supernatants were transferred to a tube. Protein concentration was determined using the Bradford assay (Bio-Rad). Total protein was separated by electrophoresis on SDS-PAGE, and the proteins were electro-transferred onto PVDF membranes (Millipore). Membranes were then probed with primary antibodies against ICAM-1 (1:1,000), phospho-IκBα (1:1,000), total IκB (1:1,000), phospho-ERK1/2 (1:1,000), phospho-p38 (1:1,000), GAPDH (1:1,000), or phospho-IκK (1:1,000). Horseradish peroxidase (HRP)-conjugated anti-rabbit IgGs or anti-mouse IgGs (Cell Signaling Technology) were used to visualize proteins by a chemiluminescence reaction (GE Healthcare). The expression of proteins was normalized to GAPDH expression.

**RT-PCR for IKK-β mRNA expression.** Total RNA was extracted from HCAECs using an RNaseasy kit (Qiagen). RT-PCR was performed using a two-step protocol. RNA (0.2 μg) was reverse transcribed into cDNA using an Omniscript kit (Qiagen), and mRNA levels of IKK-β were determined by real-time PCR using ABI 7900. Gene expression was quantified and analyzed using the comparative cycle threshold (CT) method as described in the Applied Biosystems user bulletin. All data were normalized to GAPDH as an internal control. Primer sequences are designed by TIB MOLBIOL and are as follows: IKK-β forward 5'-CCCCGATAGGCACTGCACCT, reverse 5'-TTCTCTTGGGCTTGGATAA; GAPDH forward 5'-GGGAAGTTGAAGGTCCGAGTC, reverse 5'-GTTCAATTGAAGGGTGTCATTGATG.

**Extraction of cell nuclear fraction.** Extraction of nuclear fraction of HCAECs was performed by using Active Motif Nuclear extraction kit according to the manufacturer’s protocol. Briefly, HCAECs were washed twice with ice-cold PBS containing phosphatase inhibitors and then detached and incubated in a hypotonic buffer. Cells were then centrifuged, and the supernatants containing cytosolic fraction were removed. The remaining cell pellets were resuspended in lysis buffer and incubated on ice on a rocking platform at 150 rpm for 30 min and then centrifuged. The supernatant was the nuclear fraction. EMSA. EMSA was performed by using EMSA gel shift kit (Panomics) according to the manufacturer’s protocol. Oligonucleotides corresponding to NF-κB binding sequence (5'-AGTTGAGGGGACCTTCCAGGC-3', from Panomics) were used. HCAECs were seeded in a 100-mm culture dish and kept overnight in cell growth medium containing 0.5% FBS. Cells were then preincubated with Ac-SDKP (10 nM) for 30 min in the presence of captopril (10 μM) in serum-free EBM medium (Lonza). On the basis of our previous experience, TNF-α-induced NF-κB DNA binding activity was time dependent (data not shown) and reached the highest level after 1 h of stimulation. Therefore, we used this time point to challenge the cells with TNF-α (0.5 ng/ml for 1 h at 37°C). HCAEC’s nuclear extracts (2 μg) were incubated with Poly D (I-C) at room temperature for 5 min. The nuclear extracts were then incubated with biotin-labeled probes at 15°C for 30 min. After electrophoresis on a 6% polyacrylamide gel, the samples on gel were transferred onto a presoaked Pall Biodyne B nylon membrane (VWR International). The membrane was cross linked using a UV crosslinker for 3 min and then developed by adding the blocking buffer and streptavidin-HRP conjugate. Optical density of the bands was compared as described previously (34).

**Statistical analysis.** Results are expressed as means ± SE. Contrast statements in an analysis of variance routine were used to examine all pairwise comparisons. A Hochberg’s method was used to adjust for multiple comparisons. A difference was considered significant if the adjusted P value was <0.05.

**RESULTS**

**In endothelial cells Ac-SDKP suppresses TNF-α-stimulated ICAM-1 expression.** Effect of Ac-SDKP on TNF-α-stimulated ICAM-1 expression was examined by Western blotting. ICAM-1 expression was undetectable in untreated cells but was greatly increased by TNF-α treatment. Pretreating the cells with Ac-SDKP significantly decreased TNF-α-stimulated ICAM-1 expression in a concentration-dependent manner, without affecting the expression of housekeeping protein GAPDH. The strongest inhibition was achieved with a 10 nM dose (Fig. 1). When concentration of Ac-SDKP was increased to 100 nM, this inhibition was reversed (data not shown). Therefore, the dose of 10 nM Ac-SDKP was used for the rest of the experiments. These results indicate that Ac-SDKP suppresses TNF-α-induced ICAM-1 expression.

**Ac-SDKP suppresses TNF-α-induced NF-κB activation through decreasing the phosphorylation of IKK and IkB.** TNF-α is a potent activator of NF-κB in endothelial cells (35). The TNF-α-induced activation of NF-κB requires activation of the enzymatic complex IKK that is followed by the phosphorylation of IκB protein (8). Therefore, we analyzed the effect of Ac-SDKP on TNF-α-induced IκK expression and phosphorylation.
loration. As shown in Fig. 2A, phosphorylated IKK was undetectable in untreated cells but greatly elevated by TNF-α treatment. Ac-SDKP significantly reduced IKK phosphorylation in TNF-α-stimulated cells. In addition, the data showed that neither TNF-α nor Ac-SDKP treatment affected the protein expression levels of the IKK-α subunit. However, IKK-β subunit protein expression levels were significantly elevated by TNF-α treatment, and this increase was significantly diminished by Ac-SDKP (Fig. 2B). Ac-SDKP treatment alone had no effect on either the basal level of phosphorylated IKK or nonphosphorylated IKK-β. These data indicate that Ac-SDKP inhibits TNF-α-induced IKK phosphorylation and IKK-β protein expression. However, IKK-β mRNA expression was not affected by Ac-SDKP (Fig. 2B).

We next analyzed whether Ac-SDKP affected TNF-α-stimulated IκB phosphorylation. The addition of TNF-α to human endothelial cells caused significant increase in the protein levels of phosphorylated IκB with a concomitant decrease of total IκB protein levels. The levels of phosphorylated IκB were significantly decreased in cells treated with Ac-SDKP (Fig. 3). Phosphorylated IκB protein in non-TNF-α-treated and in cells treated with Ac-SDKP alone was not detectable. These data indicate that Ac-SDKP interferes with TNF-α-induced IκB phosphorylation.

Lastly, we tested whether Ac-SDKP-mediated decrease in TNF-α-induced IKK-β and IκB phosphorylation resulted in inhibited NF-κB activation in human endothelial cells. As shown in Fig. 4, TNF-α significantly increased formation of NF-κB-DNA complex. Conversely, pretreatment with Ac-

![Fig. 1.](http://example.com/f1.png) **Fig. 1.** Effect of N-acetyl-seryl-aspartyl-lysyl-proline (Ac-SDKP) on TNF-α-stimulated ICAM-1 expression. Human coronary artery endothelial cells (HCAECs) were pretreated with Ac-SDKP and then stimulated with TNF-α (0.5 ng/ml) for 6 h. Top: representative Western blots. Bottom: quantitative evaluation of ICAM-1 protein expression normalized to GAPDH, *P < 0.05; **P < 0.01, n = 7.

![Fig. 2.](http://example.com/f2.png) **Fig. 2A:** effect of Ac-SDKP on TNF-α-stimulated IKK phosphorylation (p-IKK). HCAECs were pretreated with Ac-SDKP (10 nM) and then stimulated with TNF-α (0.5 ng/ml) for 15 min. Top: representative Western blots. Bottom: quantitative evaluation of p-IKK expression normalized to total IKK, **P < 0.01, n = 7. B: effect of Ac-SDKP on TNF-α-stimulated IKK-β expression. HCAECs were pretreated with Ac-SDKP (10 nM) and then stimulated with TNF-α (0.5 ng/ml). Top: quantitative evaluation of IKK-β expression normalized to GAPDH. *P < 0.05, n = 7. Bottom: quantitative evaluation of IKK-β mRNA expression, n = 5.

![Fig. 3.](http://example.com/f3.png) **Fig. 3.** Effect of Ac-SDKP on TNF-α-stimulated IκB phosphorylation. HCAECs were pretreated with Ac-SDKP (10 nM) and then stimulated with TNF-α (0.5 ng/ml) for 15 min. Top: representative Western blots. Bottom: quantitative evaluation of p-IκB normalized to total IκB, **P < 0.01, n = 8.
Ac-SDKP significantly reduced TNF-α-stimulated NF-κB-DNA complex formation. Ac-SDKP alone had no effect. These results indicate that Ac-SDKP reduced TNF-α-induced NF-κB translocation from cytosol to nucleus binding to DNA in TNF-α-stimulated cells.

**TNF-α-stimulated ICAM-1 expression is mediated by IKK.** To confirm that TNF-α-stimulated ICAM-1 expression is mediated by IKK in our experimental settings, we used a selective IKK-β inhibitor, IMD (37). As shown in Fig. 5, IMD significantly inhibited TNF-α-induced IKK phosphorylation. In addition, it also reduced IKK-β expression, whereas IMD treatment alone had no effect on the basal phosphorylation of IKK and IKK-β. We further determined whether IMD is able to inhibit TNF-α-mediated ICAM-1 expression. Similar to the data shown in Fig. 1, TNF-α treatment dramatically increased ICAM-1 expression, which was significantly suppressed by IMD. IMD treatment alone had no effect on ICAM-1 expression (Fig. 6). These data indicate that phosphorylation of IKK plays a role in TNF-α-mediated ICAM-1 expression.

**Ac-SDKP does not inhibit TNF-α-induced MAPK activation.** TNF-α-stimulated recruitment of TRAF protein complex to the intracellular domain of TNF-α receptor can activate MAPK pathways as well (18). Therefore, we analyzed the effect of Ac-SDKP on TNF-α-induced ERK and p38 MAP kinase activation. The results in Fig. 7 indicate that TNF-α significantly increased p38 phosphorylation; however, Ac-SDKP failed to inhibit TNF-α-induced activation of this kinase. ERK phosphorylation was also examined, but we found that TNF-α failed to activate ERK under our experimental conditions (Fig. 8).

**DISCUSSION**

Intercellular adhesion molecule ICAM-1 expressed in endothelial cells plays an important role in vascular inflammatory responses in which it regulates leukocyte transendothelial migration to the inflammation sites (10). Previous studies from our group have found that Ac-SDKP reduced cardiac ICAM-1 expression in animals with experimental autoimmune myocarditis.

**Fig. 4.** Effect of Ac-SDKP on TNF-α-stimulated NF-κB activation. HCAECs were pretreated with Ac-SDKP (10 nM) and then stimulated with TNF-α (0.5 ng/ml) for 1 h. **A**: representative NF-κB DNA binding. **B**: quantitative evaluation of NF-κB DNA binding. Negative control is the EMSA probe only without nuclear extract. *P < 0.05, n = 6.

**Fig. 5.** Effect of IKK-β inhibitor IMD-0354 (IMD) on TNF-α-stimulated p-IKK. HCAECs were pretreated with IMD (0.1 μM) and then stimulated with TNF-α (0.5 ng/ml) for 15 min. **Top**: representative Western blots. **Bottom**: quantitative evaluation of p-IKK expression normalized to total IKK. **P < 0.01, n = 6.

**Fig. 6.** Effect of IKK-β inhibitor IMD on TNF-α-stimulated ICAM-1 expression. HCAECs were pretreated with IMD (0.1 μM) and then stimulated with TNF-α (0.5 ng/ml) for 6 h. **Top**: representative Western blots. **Bottom**: quantitative evaluation of ICAM-1 protein expression normalized to GAPDH. **P < 0.01, n = 7.
Ac-SDKP suppresses TNF-α-induced ICAM-1 expression

In vascular endothelial cells, TNF-α-induced ICAM-1 expression is carried out through TNFRI receptor and is mediated by the canonical NF-κB pathway (17, 21, 42). TNF-α-induced endothelial cell inflammatory responses have been associated with various cardiovascular diseases. In particular, long-term responses involving gene upregulation and protein synthesis of TNF-α-dependent molecules such as ICAM-1 affect endothelial cell function and interaction with immune cells (17). Therefore, we hypothesized that in HCAECs Ac-SDKP suppression of TNF-α-induced ICAM-1 expression is achieved through inhibition of NF-κB pathway. We first demonstrated that TNF-α increased ICAM-1 expression in HCAECs, which is in agreement with the previous studies performed on human umbilical vein endothelial cells (21). We then tested whether Ac-SDKP affected TNF-α-stimulated ICAM-1 expression, and we showed that treatment with Ac-SDKP indeed inhibited TNF-α-mediated ICAM-1 expression in a dose-dependent manner. To further dissect the molecular mechanisms involved in the decreased ICAM-1 expression, we analyzed the effect of Ac-SDKP on different components of canonical NF-κB pathway.

We demonstrated that Ac-SDKP inhibited TNF-α-induced cytosol to nuclear translocation of NF-κB that would result in a decreased amount of nuclear NF-κB available to bind to DNA and to initiate the transcription of TNF-α-dependent genes (3). These data are in agreement with our previous in vivo study showing that Ac-SDKP decreased NF-κB activation in Ang II-induced hypertension (6). A key step in NF-κB activation involves phosphorylation of IκB. Therefore, we tested whether Ac-SDKP affects TNF-α-induced IκB phosphorylation. Indeed, we found that Ac-SDKP significantly inhibited TNF-α-stimulated IκB phosphorylation. These data implicated that the inhibitory effect of Ac-SDKP on TNF-α-
induced NF-κB pathway may be exerted on the molecules upstream from IκB such as IKK complex.

It is well established that activation of the IKK that phosphorylates IκB is critical in the activation of NF-κB. The IKK complex consists of two catalytic subunits, IKK-α and IKK-β, and a noncatalytic adaptor subunit, IKK-γ (9). Gene-depletion studies have demonstrated that IKK-β, but not IKK-α, plays an essential role in NF-κB activation mediated by TNF-α (12, 13, 29, 39). Stimulation by TNF-α induces trimerization of TNFR1 that leads to the recruitment of various adaptor proteins, ubiquitin ligases, and kinases to the cytosolic TNFR1 domain that phosphate and activate IKK-β (9). The exact mechanism of this phosphorylation remains unclear, but it could be mediated by both upstream kinases and transautophosphorylation (4). Our data show that in nonstimulated cells IKK phosphorylation was very low, but it dramatically increased in response to TNF-α. Treatment with TNF-α also significantly increased IKK-β protein levels but not IKK-α, which is in agreement with the established role of IκB-κ, but not IKK-α, in IKK activation by TNF-α (4). Pretreatment with Ac-SDKP decreased TNF-α-stimulated IKK phosphorylation and IKK-β protein levels, indicating that the inhibitory effects of Ac-SDKP on TNF-α-stimulated IκB phosphorylation are mediated by a decreased activity of IKK complex. We observed a similar pattern of IKK phosphorylation and IKK-β expression when we used IMD (a specific IKK-β inhibitor that inhibits ATP attachment to IKK-β) (11) to downregulate NF-κB pathway and ICAM-1 expression in response to TNF-α. Therefore, the observed decrease in total IKK phosphorylation in cells treated with Ac-SDKP may be due mainly to decreased IKK-β phosphorylation. There are many IKK kinase candidates implicated in IKK-β phosphorylation, such as TAK1 (36) and MAP kinase kinases (23), and it is possible that some of these kinases may be involved in Ac-SDKP-mediated inhibition of NF-κB signaling pathway. On the other hand, the observed decrease in TNF-α-dependent IKK-β expression in response to IMD was less than the decrease observed in response to Ac-SDKP, indicating that, in addition to altering IKK-β phosphorylation, Ac-SDKP may also affect the levels of nonphosphorylated IKK-β. This effect may involve Ac-SDKP inhibition of IKK-β protein and/or mRNA expression, potentiation of IKK-β degradation, and/or Ac-SDKP inhibition of phosphatases such as PP2A (5) and PP2C-β (30) implicated in IKK-β dephosphorylation. Together our data indicate that Ac-SDKP downregulation of NF-κB pathway and ICAM-1 expression in response to TNF-α is achieved through the inhibition of IKK-β kinase. However, the complexity of the signaling pathway upstream of the IKK-β introduces numerous molecules as possible Ac-SDKP targets. Therefore, further studies are needed to dissect this question.

In addition to activating NF-κB pathway, TNF-α stimulation of TNFR1 and subsequent recruitment of protein complex to TNFR1 cytoplasmic domain activates MAP kinase pathway as well (18). These pathways involve signaling through ERK and p38 MAP kinases. Our data show that TNF-α dramatically increased p38 phosphorylation but did not affect ERK phosphorylation. Ac-SDKP had no effect on TNF-α-stimulated p38 phosphorylation. These data suggest that the inhibitory effect of Ac-SDKP is mainly through inhibition of NF-κB pathway and is independent of p38 MAP kinase or ERK. However, it is unclear at which point Ac-SDKP interacts with NF-κB pathway. Because TNF-α-mediated p38 phosphorylation is not affected by Ac-SDKP, it is not likely that Ac-SDKP directly interacts with the TNF-α receptor or associated proteins (TRAFs).

In summary, in the present study, we demonstrated that Ac-SDKP inhibits TNF-α-induced ICAM-1 expression in HCAECs by targeting NF-κB signaling pathway through the inhibition of IκB kinase (Fig. 9). These data add insight into the molecular mechanism of our previous in vivo evidence that Ac-SDKP downregulated ICAM-1, and this may contribute to further investigation on Ac-SDKP anti-inflammatory activity and cardiovascular protective effect, as well as to the development of Ac-SDKP-based therapies for treating inflammatory diseases.

**Limitation.** We demonstrate that the inhibitory effect of Ac-SDKP on TNF-α-induced ICAM-1 expression is mainly through inhibition of the NF-κB pathway, and that is independent of p38 MAP kinase or ERK. We cannot exclude the possibility that other signaling pathways also contribute to this inhibitory effect of Ac-SDKP, and therefore further investigations are warranted. In addition, TNF-α is one of the stimuli that induce ICAM-1 expression. We have not explored whether Ac-SDKP modulates other cytokines that mediate ICAM-1 expression because this was not the focus of our current study.

**Perspective.** TNF-α is one of the major inflammatory cytokines produced by a number of cells, including vascular endothelial cells, that plays an important function in inflammation. Patients with cardiovascular disease have increased expression of p38 MAP kinase and TNF-α receptor, which primarily target vascular endothelial cells and upregulate specific adhesion molecules such as ICAM-1, contributing to the pathophysiology of cardiovascular disease. Our study
demonstrated that Ac-SDKP diminished TNF-α-induced ICAM-1 expression mainly through inhibition of NF-κB pathway in HCAECs. These data, along with our previous in vivo data, support the possibility that Ac-SDKP may be beneficial for the treatment of cardiovascular diseases with inflammatory and fibrotic components.

DISCLOSURES
No conflicts of interest, financial or otherwise, are declared by the authors.

AUTHOR CONTRIBUTIONS

REFERENCES


