Anti-vascular cell adhesion molecule antibody M/K-2.7–conjugated microparticles of iron oxide

VCAM-MPIO

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Chemical name:	Anti-vascular cell adhesion molecule antibody M/K-2.7–conjugated microparticles of iron oxide	
Abbreviated name:	VCAM-MPIO	
Synonym:	M/K-2.7-MPIO	
Agent category:	Antibody	
Target:	Vascular cell adhesion molecule-1 (VCAM-1)	
Target category:	Antigen	
Method of detection:	Magnetic resonance imaging (MRI)	
Source of signal/contrast:	Iron oxide	
Activation:	No	
Studies:	• *In vitro* • Rodents	Click on protein, nucleotide (RefSeq), and gene for more information about VCAM-1.

Background

[PubMed]

Magnetic resonance imaging (MRI) maps information about tissues spatially and functionally. Protons (hydrogen nuclei) are widely used to create images because of their

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abundance in water molecules, which comprise >80% of most soft tissues. The contrast of proton MRI images depends mainly on the nuclear density (proton spins), the relaxation times of the nuclear magnetization (T1, longitudinal; T2, transverse), the magnetic environment of the tissues, and the blood flow to the tissues. However, insufficient contrast between normal and diseased tissues requires the use of contrast agents. Most contrast agents affect the T1 and T2 relaxation times of the surrounding nuclei, mainly the protons of water. T2* is the spin–spin relaxation time composed of variations from molecular interactions and intrinsic magnetic heterogeneities of tissues in the magnetic field (1). Cross-linked iron oxide nanoparticles and other iron oxide formulations affect T2 primarily and lead to a decreased signal. On the other hand, paramagnetic T1 agents, such as gadolinium (Gd^{3+}) and manganese (Mn^{2+}), accelerate T1 relaxation and lead to brighter contrast images.

Endothelial cells have important roles in inflammatory responses (2, 3). Bacterial lipopolysaccharide (LPS), virus, inflammation, and tissue injury increase tumor necrosis factor α (TNFα), interleukin-1 (IL-1), and other cytokine and chemokine secretion. Emigration of leukocytes from blood is dependent on their ability to roll along endothelial cell surfaces and subsequently adhere to endothelial cell surfaces. Inflammatory mediators and cytokines induce chemokine secretion from endothelial cells and other vascular cells and increase their expression of cell-surface adhesion molecules, such as intracellular adhesion molecule-1 (ICAM-1), vascular cell adhesion molecule-1 (VCAM-1), integrins, and selectins. Chemokines are chemotactic toward leukocytes and toward sites of inflammation and tissue injury. The movement of leukocytes through endothelial junctions into the extravascular space are highly orchestrated through various interactions with different adhesion molecules on endothelial cells (4).

VCAM-1 is found in very low levels on the cell surface of resting endothelial cells and other vascular cells, such as smooth muscle cells and fibroblasts (5-9). VCAM-1 binds to very late antigen-4 (VLA-4) integrin on the cell surface of leukocytes. IL-1 and TNFα increase expression of VCAM-1, P-selectin, and other cell adhesion molecules on the vascular endothelial cells, which leads to leukocyte adhesion to the activated endothelium. Furthermore, VCAM-1 expression is induced by oxidized low-density lipoproteins under atherogenic conditions (10). Overexpression of VCAM-1 by atherosclerotic lesions plays an important role in their progression toward vulnerable plaques, which may erode and rupture. Microparticles of iron oxide (MPIO) are composed of iron particles with a diameter of ~4.5 μm. MPIO targeted with anti-VCAM-1 monoclonal antibody (mAb) M/K-2.7 (VCAM-MPIO) is being developed as a non-invasive agent for VCAM-1 expression in vascular endothelial cells during different stages of inflammation in atherosclerosis and renal ischemia (11).

Related Resource Links:

- Chapters in MICAD (VCAM-1)
- Gene information in NCBI (VCAM-1)
- Articles in Online Mendelian Inheritance in Man (OMIM) (VCAM-1)

Synthesis

[PubMed]

The synthesis of VCAM-MPIO was described by McAteer et al. (12). The *p*-toluenesulphonyl-MPIO (diameter, 1 μm) was purchased commercially (Invitrogen). The M/K-2.7 mAb (33 pmol/10^7 MPIO) was incubated with activated MPIO for 20 h at 37°C. IgG control MPIO (IgG-MPIO) was prepared in the same way with 33 pmol M/K-2.7 mAb. MPIO was washed and incubated with 0.1% bovine serum albumin for 4 h at 37°C to block the remaining activated sites. The number of antibodies per MPIO was not reported.

In Vitro Studies: Testing in Cells and Tissues

[PubMed]

McAteer et al. (12) performed *in vitro* cell-binding assays with VCAM-MPIO using mouse sEND-1 endothelial cells. As determined with light microscopy, VCAM-MPIO bound to only TNF-activated sEND-1 cells ($R^2 = 0.94$; $P = 0.03$) in a dose-dependent manner, whereas IgG-MPIO did not. A corresponding decrease in MRI signal intensity of the MPIO was observed on 11.7-T MRI images ($R^2 = 0.98$, $P = 0.01$), indicating the presence of VCAM-MPIO on the cell surface. VCAM-MPIO binding was blocked by preincubation with soluble VCAM-1 and not by ICAM-1. Unconjugated MPIO was phagocytosed by mouse peritoneal macrophages and not by sEND-1 cells.

Animal Studies

Rodents

[PubMed]

Akhtar et al. (11) performed MRI (9.4 T) with VCAM-MPIO in a renal ischemia reperfusion injury (IRI) mouse model. The left renal pedicle was ligated for 30 min with subsequent perfusion for 16–18 h and injection of 4.5 mg iron/kg VCAM-MPIO ($n = 5$ mice) or IgG-MPIO ($n = 3$ mice). MRI was performed at 0, 30, 60, and 90 min after MPIO injection with automated segmentation and quantification of contrast volume. There was a rapid increase in contrast volume at 30 min and maximal contrast by 60 min in the IRI kidneys, with sequestration in the spleen and liver. IRI kidneys ($5{,}991 \pm 354 \times 10^6$ μm^3) showed a 69-fold increase, but a decrease in signal in VCAM-MPIO contrast compared to the control kidneys ($87 \pm 7 \times 10^6$ μm^3; $P < 0.001$). IgG-MPIO showed minimal accumulation (~100×10^6 μm^3) in the IRI and control kidneys. Pretreatment with anti-VCAM-1 antibody (1.32 nmol/kg, 15 min) blocked VCAM-MPIO accumulation by ~90% to $625 \pm 80 \times 10^6$ μm^3. VCAM-1 expression in the IRI kidneys was 65-fold higher than in the control kidneys and in significant ($P < 0.01$) correlation with VCAM-MPIO contrast volume ($R^2 = 0.90$). Histological examination of the IRI kidney sections showed that

VCAM-MPIO adhered to the vessel walls of the peritubular capillaries and was not associated with local infarction and renal tubules.

Other Non-Primate Mammals

[PubMed]

No publication is currently available.

Non-Human Primates

[PubMed]

No publication is currently available.

Human Studies

[PubMed]

No publication is currently available.

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