

ORIGINAL RESEARCH

# Arterial Platelet Adhesion in Atherosclerosis-Prone Arteries of Obese, Insulin-Resistant Nonhuman Primates

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**BACKGROUND:** Platelet–endothelial interactions are thought to contribute to early atherogenesis. These interactions are potentiated by oxidative stress. We used *in vivo* molecular imaging to test the hypothesis that platelet–endothelial interactions occur at early stages of plaque development in obese, insulin-resistant nonhuman primates, and are suppressed by NADPH-oxidase-2 inhibition.

**METHODS AND RESULTS:** Six adult rhesus macaques fed a Western-style diet for a median of 4.0 years were studied at baseline and after 8 weeks of therapy with the NADPH-oxidase-2-inhibitor apocynin (50 mg/kg per day). Six lean control animals were also studied. Measurements included intravenous glucose tolerance test, body composition by dual-energy X-ray absorptiometry, carotid intimal medial thickness, carotid artery contrast ultrasound molecular imaging for platelet GPIIb/IIIa (glycoprotein-IIb/IIIa) and vascular cell adhesion molecule-1, and blood oxidative markers on mass spectrometry. Compared with lean controls, animals on a Western-style diet were obese (median body mass: 16.0 versus 8.7 kg,  $P=0.003$ ; median truncal fat: 49% versus 20%,  $P=0.002$ ), were insulin resistant (4-fold higher insulin–glucose area under the curve on intravenous glucose tolerance test,  $P=0.002$ ), had 40% larger carotid intimal medial thickness ( $P=0.004$ ), and exhibited oxidative signatures on proteomics. In obese but not lean animals, signal enhancement on molecular imaging was significantly elevated for GPIIb/IIIa and vascular cell adhesion molecule-1. The signal correlated modestly with intimal medial thickness but not with the degree of insulin resistance. Apocynin significantly ( $P<0.01$ ) reduced median signal for GPIIb/IIIa by >80% and vascular cell adhesion molecule-1 signal by 75%, but did not affect intimal medial thickness, body mass, or intravenous glucose tolerance test results.

**CONCLUSION:** In nonhuman primates, diet-induced obesity and insulin resistance leads to platelet–endothelial adhesion at early atherosclerotic lesion sites, which is associated with the expression of pro-inflammatory adhesion molecules. These responses appear to be mediated, in part, through oxidative pathways.

**Key Words:** atherosclerosis ■ molecular imaging ■ platelets ■ von Willebrand factor

Platelet–endothelial interactions are thought to contribute to early atherogenesis.<sup>1–3</sup> In preclinical models of atherosclerosis, platelet adhesion is mediated largely through interactions with endothelial-associated von Willebrand factor (VWF) in lesion-prone arterial regions.<sup>3–6</sup> Adherent platelets potentially

accelerate atherosclerosis by recruiting leukocytes, serving as a source for pro-inflammatory cytokines and inflammasome activation, and suppressing inflammation resolution.<sup>1,7–10</sup> Most data implicating platelet–endothelial interactions in atherogenesis have come primarily from rabbit and transgenic murine models of

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Supplementary Material for this article is available at <https://www.ahajournals.org/doi/suppl/10.1161/JAHA.120.019413>

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# Atherosclerosis In Primates

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