

Comments and Opinions

A Call for Rigorous Study of Statins in Resolution of Cerebral Cavernous Malformation Pathology

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Cerebral cavernous malformations (CCMs) are cerebrovascular disorders with an approximate prevalence of 1 in 200. The CCM pathology is typified by abnormally dilated clusters of blood vessels with defective endothelial cell–cell junctions, sluggish blood flow, and almost always associated with hemosiderin deposition in the surrounding parenchyma.^{1–3} CCM can arise sporadically or be inherited in an autosomal dominant pattern.^{4–6} In some cases, CCMs cause hemorrhagic stroke that elicits neurological defects and rarely death. Extravasation of blood components and hematoma expansion can often occur asymptotically. Three structurally unrelated genes, *CCM1* (*KRIT1*), *CCM2* (*MGC4607*), and *CCM3* (*PDCD10*), have been implicated in CCM pathobiology.^{7–9} It is now thought that the CCM proteins form a ternary complex near the plasma membrane of endothelial cells and act as scaffolds linking the junctional proteins, integrins and vascular endothelial–cadherin, with intracellular signaling components.¹⁰ However, the specific mechanisms through which reductions in the expression of the structurally diverse genes associated with CCM induce their formation and pathobiology are not well explained. Studies using mouse and zebrafish models have been instrumental in functionally characterizing and, to an extent, faithfully recapitulating the molecular and ultrastructural underpinnings of CCM pathology.^{11–15}

Results from these studies have led to the suggestion that pharmacological inhibition of 3-hydroxy-3-methyl-glutaryl-coenzyme A reductase (HMGCR) by statins may be an effective approach to prevent CCM-induced vascular instability and cerebral hemorrhage.^{12,16} The basis for this proposition stems from in vivo and in vitro evidence suggesting that loss of function of *CCM1*, *CCM2*, or *CCM3* genes are associated with RhoA hyperactivation and downstream signaling via Rho kinase and increased stress fiber assembly, which leads to disrupted endothelial cell–cell junctions and loss of vascular stability.^{17,18} More specifically, activated RhoA, through its effector, Rho kinase, mediates actin stress fiber formation by increasing myosin light chain phosphorylation, as well as inhibiting

myosin phosphatase, which is associated with vascular hyperpermeability.¹⁹ Consistently, pharmacological curtailment of RhoA activity, using fasudil, a relatively selective RhoA/Rho kinase inhibitor,²⁰ has been shown to enhance vascular stability effectively in vivo and in vitro and significantly reduce the prevalence of CCM lesions in a in *Krit1^{+/-}* and *CCM2^{+/-}* mice.^{17,21}

Alongside, it has been reported, in ≥ 1 animal study, that in mice with a heterozygous mutation of *CCM2*, treatment with statins (simvastatin) effectively restores the endothelial barrier function by inhibiting Rho GTPase activity,¹² presumably through abrogating the prenylation process. This experimental finding has led the authors to the conclusion that statin treatment could be an effective alternative to neurosurgical intervention to improve CCM outcome.^{12,16} However, there are several outstanding questions about the efficacy and specificity of statins as therapeutic agents to improve CCM outcome. First, unlike fasudil, which is a somewhat selective inhibitor of RhoA/Rho kinase,²⁰ statins are competitive inhibitors of HMGCR, the rate-limiting enzyme in the biosynthesis of isoprenoid pyrophosphates, a highly conserved metabolic pathway (Figure).²² These mevalonate derivatives in turn serve as substrates for post-translational modification (prenylation) of a variety of cell signaling proteins (>100) that harbor the C-terminal CaaX motif, the most well characterized of which include the small GTPase family of molecular switch proteins, RhoA, Rac1, and Cdc42 (Figure).^{23,24} The CaaX proteins interact with prenyltransferases that modify the CaaX cysteine residue by forming a thioether linkage with either a farnesyl or a geranylgeranyl lipid moiety, which ensures membrane localization.²⁵ Hence, inhibition of HMGCR not only reduces the availability of prenyl-based metabolites but also curtails the activity of key cell signaling molecules that require prenylation for activation and downstream signaling.

Therefore, a point of contention about statin therapy is whether inhibition of RhoA hyperactivity is outweighed by any potential pathological outcomes associated with general or indiscriminate depletion of all prenylation-dependent

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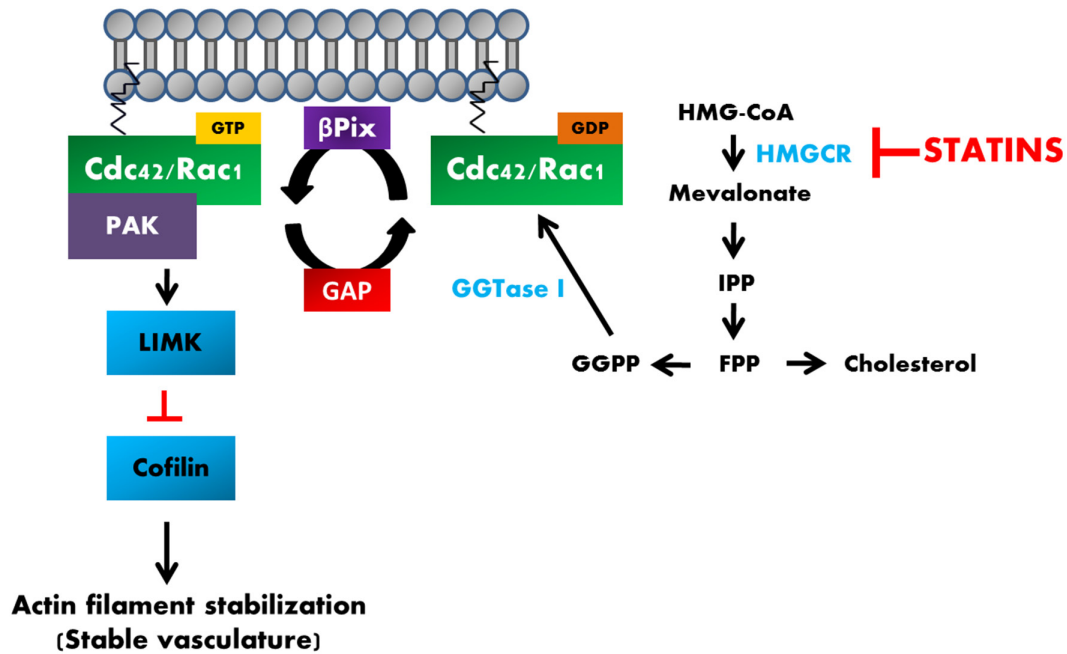


Figure. 3-Hydroxy-3-methyl-glutaryl-CoA reductase (HMGCR)–dependent regulation of Cdc42/Rac1 GTPase prenylation and β Pix/PAK–mediated signal transduction pathway involved in actin filament stabilization. FPP indicates farnesyl pyrophosphate; GAP, GTPase activating protein; GGPP, geranylgeranyl pyrophosphate; IPP, isopentenyl diphosphate; LIMK, LIM kinase; and PAK, p21 activated kinase.

cellular processes. Although statins are generally well tolerated in patients, with rare medically significant side effects, little is known about the possible complications of impaired HMGCR function on the endothelium. For example, prenylated and GTP-bound cdc42/Rac1 plays vital roles in the regulation of vacuole formation, lumenization of vessels, and mediation of endothelial barrier function through regulating vascular endothelial–cadherin dynamics.^{26–31} Consistently, studies in vivo in zebrafish have shown that genetic depletion of β Pix, a guanine nucleotide exchange factor involved in activating Cdc42/Rac1 (by increasing affinity for GTP), as well as p21-activated kinase, a kinase acting downstream of Cdc42/Rac1 (Figure), disrupt vascular integrity and is associated with cerebral hemorrhages and defective vascular stabilization.^{32,33} Waterborne exposure to statins (atorvastatin and cerivastatin) and morpholino-mediated depletion of embryonic HMGCR transcripts in zebrafish have been shown to induce intracerebral hemorrhage in both embryos and larvae due likely to impaired prenylation-dependent processes.^{34–36} Surprisingly, though, there exist no experimental or anecdotal studies to suggest statin-induced enhancement of vascular permeability in mice or other mammalian studies, which could suggest species-specific physiological differences. There have also been no studies, to date, to suggest that statins disrupt vascular stability in murine models of aging, hypertensive and amyloid angiopathies. Nonetheless, in light of the theoretical/mechanistic considerations derived from in vitro and in vivo studies (Figure), we acknowledge that there is a need for a more rigorous set of mammalian studies to investigate the putative molecular mechanisms or the risk benefit of statins in CCM pathology.

Disclosures

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