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mooth muscle cell (SMC) accumulation in the arterial
intima is a key event in restenosis after angioplasty and
by pass surgery and in the development of atheroscle-
rotic lesions.1 Restenosis, which is defined as “the arterial
healing response after injury incurred during transluminal
coronary revascularization,” has been the principal drawback
of percutaneous coronary interventions (PCI) since their
introduction. The accumulation of arterial SMCs is caused by
a combination of proliferation and directed migration of
arterial SMCs from the media into the intima. Both these
activities can be induced by cytokines and growth factors
produced within the arterial wall and circulating cells in
response to the vascular injury. A commonly accepted model
of the response to arterial injury suggests that growth factors
are released after injury, thereby changing the composition of
the extracellular matrix and triggering a proliferation and
migration program. SMCs undergo a phenotypic modulation
from a contractile to a synthetic phenotype (dedifferentia-
tion), proliferate into the media, migrate from the media into
the intima, and subsequently form the neointima.

SMC transition from G1 to Gs cell phase induces neointi-
ma formation and modulates the atherosclerotic growth. The
cell cycle is regulated by the interaction of multiple proteins,
including cyclins, cyclin-dependent kinases, and phospho-
tases. Molecular complexes containing CDKs, cyclins, pro-
iferating cell nuclear antigen (PCNA), and several other
proteins regulate the major cell cycle transition points at the
G1/S and G2/M boundaries. In addition to p21 and PCNA,
the CDK2/cyclin A kinase complex includes a 19-kDa
protein (p19, or SKP1) and a 45-kDa protein (p45, or
SKP2).2,3

SKP2 contains a binding motif of \( \approx 40 \) residues, termed the F
box associated with leucine-rich regions (LRRs). Essential for
S-phase entry, Skp2, component of the SCFskp2 ubiquitin
ligase, is responsible for polyubiquitylation of cell-cycle regula-
tors.4–9 Its major physiological target is cdk inhibitor
p27kip1.6,10 SKP2 specifically recognizes phosphorylated p27
showing that Skp2 is an important factor in VSMC
proliferation.11–13 Furthermore, it has been shown that
the antiproliferative effects mediated by increase in cAMP
and cGMP levels are mediated by the upregulation of
p27kip1 levels and inhibition of CDK2.14–16 In addition,
recent data show that phosphodiesterase 1A inhibition attenu-
ates SMC proliferation through elevation of cGMP levels,
p27kip1 upregulation, and cyclin D1 downregulation.16 To
date it is poorly understood how cyclic nucleotides regulate
intracellular cyclins trafficking and particularly p27kip1
levels.

Cyclic Nucleotides Modulates Skp2 Levels and
SMC Proliferation

In this issue of *Circulation Research*, Wu et al show that
S-phase kinase-associated protein-2 (Skp2) levels are upregu-
lated in proliferating vascular smooth muscle cells (VSMCs)
in vitro and after balloon injury of the rat carotid artery in
vivo.17 Skp2 siRNA results in increased levels of p27Kip1
and reduced proliferation. cAMP is shown to suppress Skp2
levels by inhibiting transcription and by decreasing protein
stability. The authors show that Skp2 overexpression over-
came cAMP-induced cell cycle arrest and upregulation of
p27Kip1 showing that Skp2 is an important factor in VSMC
proliferation and its inhibition of cyclic nucleotides in vitro
and in vivo.17

A novel aspect of this work is that for the first time it has
been shown that activating signaling downstream to cyclic
nucleotides regulates Skp2 expression via inhibition of FAK.
Interestingly, exogenous expression of a constitutively active
mutant of FAK was able to rescue Skp2 expression and
markers of G1–S phase progression after forskolin treatment,
showing the role of FAK as mediator of cyclic nucleotides of
SMCs proliferation (see Figure).

FAK activation is required to integrate integrin signals
with those from receptor tyrosine kinases and G protein–
coupled receptors through downstream activation of Rac1.18
FAK activation by the mechanical stress of the arterial wall
triggers a molecular cascade involving the mitogen activated
protein kinases (MAPKs)19,20 leading to cell proliferation.
Several biological processes occurring in the arterial wall
from angiogenesis to inflammation involve the focal adhesion
molecules.21,22 FAK is also involved in controlling cell
motility, an important step in neointima formation.23 Alter-
native signal pathways can modulate the molecular com-
plexes of the adhesion system regulating a variety of cellular
processes. Further investigation of the mechanisms that
underlie Skp2 controlling from the PKA–FAK axis is needed.
A crucial step that also needs to be investigated is p27kip1
breakdown control. Indeed, recent studies suggest that alter-
native molecular pathways can be involved in p27kip1 degradation.

Perspectives

The ability to modulate SMC proliferation in vivo has given the possibility to generate new therapeutic tools for cardiovascular disorders. Drug eluting stents are changing the outcome of patients undergoing percutaneous coronary interventions for the treatment of atherosclerotic lesions. The present study improves in a significant manner our understanding of cAMP signaling, a key pathway in the regulation of cardiovascular homeostasis. This work suggests new critical steps in the regulation of SMC proliferation. Further studies are needed to address the therapeutic potential of manipulating these pathways for vasculoproliferative disorders.

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References


