Göteborg, 2018

Filamin A in Cardiovascular Remodeling

Akademisk avhandling

Som för avläggande av medicine doktorsexamen vid Sahlgrenska akademin, Göteborgs universitet kommer att offentligen försvaras i hörsal Arvid Carlsson, Academicum, Medicinaregatan 3, Göteborg.

Torsdagen den 29 November 2018, kl 09:00

Av

Sashidhar Bandaru

Fakultetsopponent:

Pontus Aspenström

Institutionen för immunologi, genetik och patologi

Uppsala universitet, Sweden

Avhandlingen baseras på följande delarbeten

I. Deficiency of filamin A in endothelial cells impairs left ventricular remodeling after myocardial infarction. **Bandaru S**, Grönros J, Redfors B, Çil Ç, Pazooki D, Salimi R, Larsson E, Zhou A-X, Ömerovic E, Akyürek LM.
Cardiovascular Research 2014, 105:151-9.

II. Targeting filamin A reduces macrophage activity and atherosclerosis in mice. **Bandaru S**, Salimi R, Ala C, Akula MK, Ekstrand M, Devarakonda S, Karlsson J, Levin M, Borén J, Bergo MO, Akyürek LM.
Under revision 2018.

SAHLGRENSKA AKADEMIN
INSTITUTIONEN FÖR BIOMEDICIN

GÖTEBORGS UNIVERSITET

Göteborg, 2018

Filamin A in Cardiovascular Remodeling

Sashidhar Bandaru

Department of Medical Biochemistry and Cell Biology, Institute of Biomedicine Sahlgrenska Academy, University of Gothenburg, Sweden

ABSTRACT

Filamin A (FLNA) is a large actin-binding cytoskeletal protein that stabilizes actin networks to integrate them with cellular membranes. FLNA is involved in scaffolding and cell signaling that are vital for cell motility and organ development. Recently, we discovered that the C-terminal fragment of FLNA ($FLNA^{CT}$) cleaved by calpain is important for the nuclear transport of transcriptional factors to regulate angiogenesis. As we considered this finding unique, we hypothesized about whether the absence of vascular endothelial cell-specific FLNA impairs angiogenesis after myocardial infarction (MI) and the absence of macrophage-specific FLNA reduces the formation of atherosclerotic plaques.

In Study I, we induced MI by the permanent ligation of the left descending coronary artery on both *wt*-controls ($Flna^{o/fl}$) and mice that are deficient in *Flna* in endothelial cells ($Flna^{o/fl}$/*VECadCre+*). Mice without endothelial FLNA developed an increased size of MI, larger and thinner left ventricles, impaired cardiac pump function, elevated levels of NT-proBNP and reduced VEGF-A secretion. $Flna^{o/fl}$/*VECadCre+* hearts exhibited reduced capillary structures within the infarcted regions. *FLNA*-deficient endothelial cells also showed impaired migration and tubular formation, along with reduced levels of p-ERK and p-AKT and GTPase RAC1.

In Study II, we observed an increase in the expression of FLNA in human carotid arteries with advanced atherosclerotic plaques compared with intermediate plaques. Macrophages that are deficient in *Flna* ($Flna^{o/fl}$/*LC*) proliferated and migrated less compared with controls ($Flna^{o/fl}$). We also observed reduced p-ERK and p-AKT, along with reduced lipid uptake and increased cholesterol efflux, in $Flna^{o/fl}$/*LC* macrophages. To induce atherosclerosis, we transplanted bone marrow with either *wt*- or *Flna*-deficient macrophages into $Ldlr^{-/-}$ mice or overexpressed PCSK9 in $Flna^{o/fl}$ and $Flna^{o/fl}$/*LC* mice by an adenoviral vector. After 20–25 weeks on a Western diet, we observed a reduction in the size of atherosclerotic plaques and a decrease in the number of CD68-positive macrophages within the atherosclerotic plaques from mice that are deficient in *Flna* in macrophages. Interestingly, we discovered that the calpain-cleaved fragment of $FLNA^{CT}$ interacts with STAT3. The inhibition of this cleavage by calpeptin reduced nuclear p-STAT3 levels and then IL-6 secretion. Furthermore, foam cell formation, cell proliferation and migration were reduced in the absence of FLNA in cultured macrophages. Finally, calpeptin treatment of atherogenic mice overexpressing PCSK9 reduced the size of atherosclerotic plaques.

Our results indicate novel functions for FLNA during MI and atherogenesis by interacting with transcriptional proteins in the regulation of angiogenesis and cytokines, which are important events in the progression of MI and atherosclerosis. These novel findings identify FLNA as an important mediator in cardiovascular remodeling. Targeting FLNA in endothelial cells or the cleavage of $FLNA^{CT}$ in macrophages might have the potential to reduce the size of MI or slow the formation of atherosclerotic plaques respectively.

Keywords: Cytoskeleton, Myocardial infarction, Atherosclerosis

ISBN: 978-91-7833-229-8 (Print)
ISBN: 978-91-7833-230-4 (PDF)

http://hdl.handle.net/2077/56922