

POSTER PRESENTATION

Open Access

Identification of protein kinase G I alpha interacting proteins as potential targets to prevent cardiac remodeling

Robert Blanton^{1*}, Angela Lane¹, Mark Aronovitz¹, Guang-Rong Wang¹, Robrecht Thoonen¹, Roger Davis², Michael Mendelsohn^{1,3}, David Kass⁴, Richard Karas¹

From 6th International Conference on cGMP: Generators, Effectors and Therapeutic Implications
Erfurt, Germany. 28-30 June 2013

Background

We recently reported that mutation of the cGMP-dependent Protein Kinase G I alpha (PKG α) N-terminal leucine zipper (LZ) domain (in the PKG α LZ mutant, or Lzm, mouse) accelerates cardiac remodeling and heart failure after left ventricular (LV) pressure overload, and prevents the anti-remodeling effect of sildenafil [1]. We therefore hypothesized that PKG α attenuates remodeling by regulating cardiac signaling pathways that are dependent on substrate interactions mediated by its LZ domain. As a first step to identifying cardiac proteins downstream of PKG α , we screened myocardial lysates for PKG α LZ domain-interacting proteins. Our previous work revealed a requirement for the PKG α LZ domain for the activation of anti-remodeling myocardial JNK activity after LV pressure overload. MLK3 is a MAPKKK that contains an LZ domain and activates JNK.

Results

We now demonstrate, by immunoprecipitation, that MLK3 interacts with the PKG α LZ domain in myocardial lysates. We show further that 8-Br-cGMP induces MLK3 phosphorylation on Thr277 and Ser281 in WT, but not Lzm lysates. And, in 293 cells transfected with FLAG-MLK3, 8Br-cGMP induced PKG α -MLK3 co-precipitation, and increased MLK3 phosphorylation on Thr277/Ser281. Co-transfection of MLK3 and PKG α also induced MLK3 phosphorylation. We next examined the cardiovascular effect of MLK3 deletion in vivo. Male 8 week old MLK3-/mice display basal bi-ventricular hypertrophy compared

with littermate controls (LV/Tibia length 42.8 ± 0.6 mg/cm in WT, 52.9 ± 1.8 in MLK3 -/-; $P<0.01$; RV/TL 10.8 ± 0.1 mg/cm in WT, 13.3 ± 0.3 in MLK3-/-; $P<0.01$; $n= 7$ WT, 5 MLK3-/-). By 14-16 weeks of age, LVH progressed in the MLK3-/- mice (LV/TL 47.7 ± 1.3 mg/cm in WT, 59.8 ± 7.5 in MLK3-/-; $n= 6$ WT, 9 MLK3-/-; $P<0.01$). Arterial blood pressure was modestly increased, though still normal, in MLK3-/- mice (SBP 93 ± 1 in WT, 113 ± 1 in MLK3-/-). And, 14-16 week MLK3-/- mice have impaired LV diastolic function (τ 3.2 ± 0.1 ms WT, 3.7 ± 0.1 MLK3-/-; $P=0.06$).

Conclusion

Our studies reveal a novel function of MLK3 as a myocardial PKG α effector and inhibitor of LVH. These results support the strategy of exploring LZ-dependent PKG α substrates in the myocardium to identify potential therapeutic targets for cardiac remodeling.

Authors' details

¹Molecular Cardiology Research Institute, Tufts Medical Center, Boston, MA, USA. ²Department of Biochemistry, University of Massachusetts School of Medicine, Worcester, MA, USA. ³Merck Pharmaceuticals, Rahway, NJ, USA.

⁴Department of Cardiology, Johns Hopkins Medical Institutions, Baltimore, MD, USA.

Published: 29 August 2013

Reference

1. Blanton R, Takimoto , Lane AM, Aronovitz M, Piotrowski R, Karas RH, Kass DA, Mendelsohn ME: Protein kinase g ia inhibits pressure overload-induced cardiac remodeling and is required for the cardioprotective effect of sildenafil in vivo. *J Am Heart Assoc* 2012, 1:e003731.

doi:10.1186/2050-6511-14-S1-P10

Cite this article as: Blanton et al.: Identification of protein kinase G I alpha interacting proteins as potential targets to prevent cardiac remodeling. *BMC Pharmacology and Toxicology* 2013 14(Suppl 1):P10.

* Correspondence: rblanton@tuftsmedicalcenter.org

¹Molecular Cardiology Research Institute, Tufts Medical Center, Boston, MA, USA

Full list of author information is available at the end of the article