Inactivation of catalytic and non-catalytic PI3-kinase gamma function enhances hypoxia-induced pulmonary hypertension in mice

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Rationale: Pulmonary hypertension (PH) is a pulmonary vascular disease characterised by chronically elevated pulmonary arterial mean pressure, increased pulmonary vascular resistance and right ventricular (RV) dysfunction and hypertrophy. The pathogenesis is characterised by vasoconstriction of pulmonary vessels and pulmonary vascular remodelling. PI 3-kinase γ (PI3Kγ) is activated via G protein-coupled receptors and abundantly expressed in all cell types involved in the pathogenesis of PH including leukocytes, cardiomyocytes, and endothelial cells (EC). The catalytic function of PI3Kγ is involved in numerous processes that are important for both vascular remodelling and maladaptive cardiac hypertrophy, including leukocyte recruitment and EC proliferation and survival. On the other hand, its non-catalytic function affects cyclic adenosine monophosphate (cAMP) production and thus cardiac contractility. Furthermore, inactivation of PI3Kγ impairs nitric oxide (NO) production by endothelial NO synthase (eNOS), which may lead to increased vascular resistance. Therefore, the aim of our study was to investigate the role of both catalytic and non-catalytic functions of PI3Kγ in the pathogenesis of PH.

Methods: PI3Kγ knockout mice (PI3Kγ−/−), as well as mice with a catalytically inactive form of PI3Kγ (PI3KγKD/KD) were analysed in vivo using the hypoxia-induced mouse model of PH (21 days at 10% O2 hypoxia (HOX)). To specifically inhibit the non-catalytic function of PI3Kγ, C57BL/6N mice were treated with a mimetic peptide (MP) via intratracheal instillation and studied in the same model. Systolic right ventricular pressure (RVSP) was measured using a Millar pressure catheter inserted via the jugular vein. RV hypertrophy was determined by Fulton’s index (RV/LV+S).

Results: PI3Kγ−/− mice showed significantly increased RVSP after three weeks of hypoxia compared to WT littermate controls (HOX WT 35.64 ±3.21mmHg vs HOX PI3Kγ−/− 37.04±2.31mmHg;p=0.0049). A significant increase in RVSP was also detected in PI3KγKD/KD and MP-treated mice compared to respective WT controls (HOX WT 34.67±3.83mmHg vs. HOX PI3KγKD/KD 37.9±2.19mmHg;p=0.0404 and HOX vehicle 34.19±2.74mmHg vs. HOX MP 37.61±2.46mmHg;p=0.0111). Under normoxic conditions (NOX), an increased RVSP was already measured in PI3Kγ−/− mice (NOX WT 26.13±1.2mmHg vs. NOX PI3Kγ−/− 28.07±0.88;p=0.0157). Heart rate and systemic blood pressure remained unchanged. A significant increase in RV hypertrophy was only evident for PI3KγKD/KD compared to WT controls (HOX WT 0.38±0.06 vs. HOX PI3KγKD/KD 0.46±0.09mmHg;p=0.0037).

Conclusion: The results show that both catalytic and non-catalytic inactivation of PI3Kγ in vivo do not counteract the pathogenesis of PH, but conversely enhance it. In this context, reduced phosphorylation of eNOS may play a crucial role, leading to increased vasoconstriction, whereas increased cAMP levels may help the RV to adapt to the increased pressure. The exact mechanisms will be analysed in the future.