The BMPR2, ALK1 and ENG Genes Mutation in Congenital Heart Disease-Associated Pulmonary Artery Hypertension

Anggoro Budi Hartopo[1,*], Dyah Wulan Anggrahini[1], Noriaki Emoto[2], Lucia Kris Dinarti[1]

[1] Department of Cardiology and Vascular Medicine, Faculty of Medicine, Public Health and Nursing, Universitas Gadjah Mada / Dr. Sardjito Hospital, Yogyakarta, Indonesia
[2] Laboratory of Clinical Pharmaceutical Science, Kobe Pharmaceutical University, Kobe, Japan

*Corresponding author:
Anggoro Budi Hartopo MD, PhD. –email: a_bhartopo@ugm.ac.id
Address: Department of Cardiology and Vascular Medicine, Faculty of Medicine, Public Health and Nursing, Universitas Gadjah Mada / Dr. Sardjito Hospital, Yogyakarta, Indonesia, Jalan Farmako Sekip Utara, Yogyakarta, 55281, Indonesia

Manuscript submitted: September 11, 2019; Revised and accepted: September 18, 2019

ABSTRACT

The gene mutation is one of the background underlie the pathogenesis of pulmonary artery hypertension (PAH). Several genes have been recognized to be responsible for the development of PAH. The mutation in transforming growth factor-β (TGF-β) pathway is considered to be major genotype background in heritable PAH. The genetic mutation in bone morphogenetic protein receptor-2 (BMPR2), activin receptor-like kinase 1 (ALK-1) and endoglin (ENG) are known to cause heritable PAH. In congenital heart disease–associated PAH (CHD-APAH), their mutation are also presence.

Keywords: gene mutation; pulmonary artery hypertension; congenital heart disease–associated PAH

INTISARI

Mutasi genetik merupakan salah satu kondisi yang mendasari patogenesis hipertensi arteri paru (HAP). Beberapa gene telah dikenal bertanggungjawab terhadap terjadinya HAP. Mutasi pada gen jalur *transforming growth factor-β* (TGF-β) diduga merupakan melatarbelakangi HAP yang diwariskan. Mutasi gen pada *bone morphogenetic protein receptor-2* (BMPR2), *activin receptor-like kinase 1* (ALK-1) dan *endoglin* (ENG) diketahui menyebabkan HAP yang diwariskan. Pada HAP yang terkait penyakit jantung kongenital, mutasi pada gen tersebut juga terjadi.

INTRODUCTION

Pulmonary artery hypertension (PAH) is a disease of pulmonary artery obstruction, due to complex plexyform lesions in the distal artery, with subsequent elevation in pulmonary vascular resistance.[1] Right ventricular failure will develop in the eventual process of PAH.[1] The PAH is characterized by a persistent increase in mean pulmonary artery pressure (≥ 25 mm Hg at rest), a normal value of pulmonary capillary wedge pressure (≤ 15 mmHg) and an increased level of pulmonary vascular resistance (> 3 Wood units).[2] Clinically, the PAH can be divided into idiopathic PAH, heritable PAH, or associated with other conditions (APAH).

Several diseases can be associated with PAH, such as such as drug-induced, toxin exposures, connective tissue diseases, human immunodeficiency virus infection, portal hypertension, congenital heart disease and schistosomiasis.[2] Heritable PAH is recognized by the presence of genetic mutation.

The genetic mutation in bone morphogenetic protein receptor-2 (BMPR2) gene has been reported in about 75% of heritable PAH and nearly 25% of sporadic PAH.[3] Other mutations of gene encoding activin receptor-like kinase 1 (ALK-1) and endoglin (ENG) have been recognized in PAH with a history of hereditary haemorrhagic telangiectasia.[3]

Pathophysiology of PAH is multifactorials and can be attributed to various pathologies. One of the important aspects of pathophysiology of PAH is the role of genetic factors such as mutations in BMPR2, ALK1 and ENG genes. The BMPR2, ALK1 and ENG gene mutations have been determined to have a high level of evidence as a causal role in PAH.[4] The congenital heart disease–associated PAH (CHD-APAH) most commonly occurs with septal defect and systemic-to-pulmonary shunt lesions. It accounts for around 10% of all patients with PAH.[5] Pulmonary hypertension in this condition is due to increasing blood flow in pulmonary vascular systems and subsequent endothelial damage and pulmonary vascular remodeling. The pulmonary vascular lesions in CHD-APAH are similar with lesions in idiopathic PAH and heritable PAH.[5] In CHD-APAH), the role of genetics mutations has been reported, however the result until currently cannot be completely concluded yet.

THE BMPR2 GENE MUTATION IN CHD-APAH

Mutation in BMPR2 genes has been recognized in 70-80% of familial PAH and 10-20% of idiopathic PAH. BMPR2 is gene that encode BMPR2 protein, a member of the transforming growth factor-β (TGF-β) receptor superfamily.[4] The BMPR2 is an active serine-threonine kinase receptor which is expressed constitutively in the lung.[1] Ligation of BMPR2 by its ligands (BMP2, BMP4, BMP6, BMP7, BMP9 and BMP10), triggers BMPR2 phosphorylation and Smad protein-signaling activation.[1]

The BMPR2 mutations in PAH result in derangement of Smad signaling pathways, which increases the tendency of endothelial and artery smooth muscle cells to proliferate and reduce their apoptotic capacity.[4] BMPR2 receptor-activated Smads forms a complex with Smad4 (canonical pathway) and translocatesinto the nucleus, where it regulates gene transcription.[1] Other Smad4-independent pathways are related to Smad8 pathway, in which mutation of SMAD9 gene (gene encodes Smad8) leads to significant loss of function in microRNA maturation and is implicated in PAH pathogenesis.[4] SMAD9 mutation demonstrates a cellular hyperproliferative state, blunted BMP signaling, and dysregulated Smad-mediated microRNA activity.[6] Drake et al. reported the case of dyregulated SMAD9 pathway in CHD-APAH.[6] The majority of the patients with heritable PAH have mutations in BMPR2 which causes increased cellular proliferation.[7] Furthermore, BMPR2 mutation patients with PAH have more severe clinical condition and low response to acute vasodilator testing.[7]

The prevalence of PAH-associated mutations in the CHD-APAH population indicate that up to 10% have a germline BMPR2 mutation.[8] Robert et al. reported six missense BMPR2 mutations in exons 2 and 3 (an extracellular domain of BMPR2), exon 5 (kinase domain for phosporylation) and exon 11 (cytoplasmic tail with unknown function), which associated with altered protein sequence at evolutionary conserved amino acids and deranged BMPR2 function.[8] Patients with BMPR2 mutations were identified as having atrioventricular septal defects (AVSD) and multiple defects (ASD+PDA, ASD+PDA and PAPVD, and AP window+VSD).[8] A study in 11 children with CHD-APAH (10 ASD and 1 VSD) indicated 1 subject (female, VSD) with BMPR2 mutation (deletion in exon 1).[9] Study in Chinese CHD-APAH population indicated mutation rate of 7.5% in BMPR2 gene among CHD-APAH, whereas in CHD

with no PAH the mutation rate of BMPR2 is much lower (1.2 %).[10] Females have increased BMPR2 mutation rate and the dominant mutation type is missense mutation.[10] The type of defects were not associated with BMPR2 mutation.[10]

BMPR2 is highly expressed on the pulmonary vascular endothelial cells.[4] It forms a complex with the type I receptors, ALK1.[4] The complex of ALK1/BMPR2 receptor signaling pathway requires the utilization of ENG as a co-receptor.[4] This membrane bound receptor complex regulates various cellular processes including cell differentiation, proliferation, and apoptosis in embryonic and mature cells.[4] Similar with BMPR2 expression, the ALK1 and ENG are also abundantly expressed in the pulmonary endothelial cells.[4,5] Figure 1 shows the diagram of BMPR2/ALK1/ENG complex receptors in endothelial cells. The mutation of ALK1 and ENG also deranges the BMPR2 downstream signaling and is implicated in the presence of PVD in CHD-APAH.

THE ALK1 GENE MUTATION IN CHD-APAH

Mutation in ALK1, which is also a member of the TGF-β receptor superfamily, is currently reported in PAH patients, although the mutation rate is not as high as BMPR2 gene mutation.[11] In a European cohort of adult-onset PAH, the mutation rate of ALK1 gene was 0.9%.[4] However this collaborative research did not include CHD-APAH. The ALK1 is located on chromosome 12q13 and plays a role in various tissues such as cell proliferation and differentiation, cell migration, cell survival and angiogenesis.[4] Its missense mutations are located in the serine-threonine kinase domain, which is responsible for PAH development in patients with hereditary hemorrhagic telangiectasia (HHT) as well as familial and idiopathic PAH.[12]

The HHT and heritable PAH represent inherited diseases caused by mutation of the TGF-β receptor-mediated signalling, particularly involving ALK1, in the vasculatures.

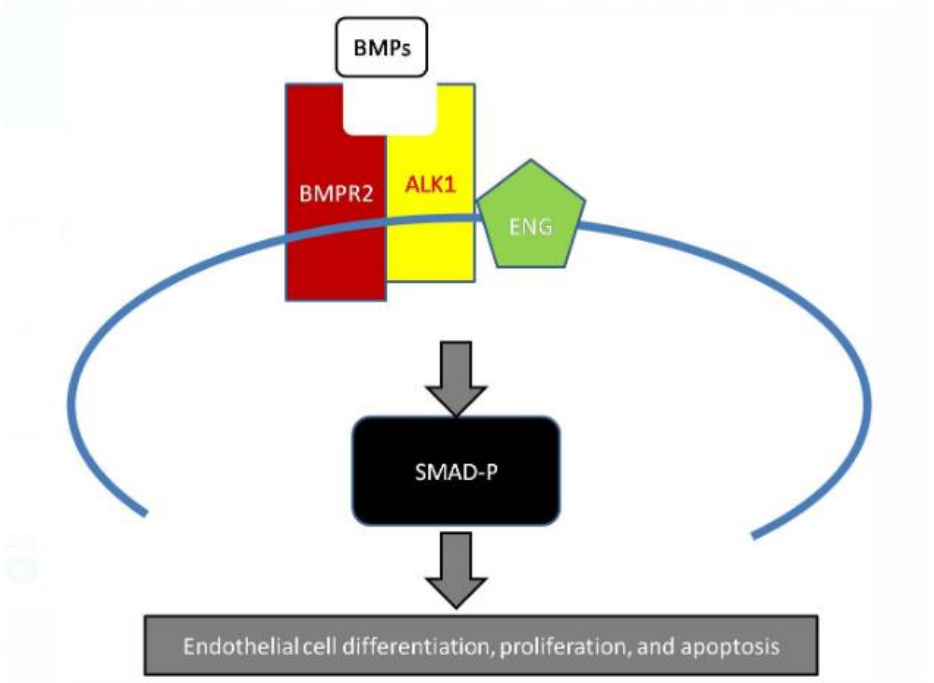

Figure 1. The diagram of BMPR2/ALK1/ENG complex receptor in surface membrane of endothelial cells which exhibit its role on endothelial cell differentiation, proliferation, and apoptosis upon ligation by BMPs.

The TGF-β signaling in endothelial cells modulates vascular remodeling, therefore mutation in ALK-1 gene may induce derangement of remodeling of the vascular bed.[13] A mutation analysis in 18 children with PAH (16 idiopathic PAH and 2 CHD-APAH), detected a missence mutation in idiopatic PAH subject but not in CHD-APAH (BMPR2 mutation instead).[14] Another study involved 11 children with CHD-APAH (10 ASD and 1 VSD) and performed PCR amplification of the entire coding regions and the exon/intron of the ALK1 gene did not find ALK1 mutation.[9]

THE ENG GENE MUTATION IN CHD-APAH

Endoglin is a homodimeric glycoprotein ubiquitously located in the vascular endothelial cells of human body.[4] It is an integral membrane glycoprotein in the celular membranes.[4] The ENG gene encodes a type I integral membrane glycoprotein receptor.[4] It belongs to the TGF-β signaling superfamily.[4] This receptor is expressed on proliferating vascular endothelial cells and other cell types associated with cardiovascular system, in which indicate its important of endothelial and vascular function. It promotes cell differentiation, proliferation, angiogenesis, inflammation, and also induce wound healings.[15] The ENG gene is located on chromosome 9q33-34 and the encoded protein demonstrates an extracellular domain, hydrophobic transmembrane domain and a cytosolic domain.[15]

Similar with the mutation on ALK1 gene, the mutation on ENG gene is associated with HHT, in which the mutation involved are missense, splicing mutations, and novel protein-truncating mutations.[13] In a European cohort of adult-onset PAH, the mutation rate of ALK1 gene was 0.6%.[4] Unfortunately, this collaborative research did not include CHD-APAH. The study by Harrison et al. in 18 children with PAH, found no ENG mutation in CHD-APAH patients (only two subjects in this study), but only found one branch-site ENG mutation in a subject with HHT and IPAH.[14] The study by Pfarr et al. described a small number of pathogenic mutations in patients with CHD-APAH.[9] In 11 children with CHD-APAH without history of HHT, 1 patient with secundum ASD had mutation in the ENG gene (missense mutation in exon 12).[9]

CONCLUSION

In CHD-APAH patients, mutation in the TGF-β pathway genes involving BMPR2/ALK1/ENG complex receptors is needed to be explored. Unlike idiopathic PAH and heritable PAH, the genetic mutation in CHD-APAH is quite rare and its clinical implication need further corroboration.

ACKNOWLEDGEMENT

Authors acknowledge the research grant from Universitas Gadjah Mada. This review article is part of the outcome of research grant Penelitian Dasar 2019 from Universitas Gadjah Mada (No: 2798/UN1.DITLIT/DIT-LIT/LT/2019) to Principal Investigator Anggoro Budi Hartopo.

REFERENCES

1. Archer S.L., Weir E.K., Wilkins M.R. 2010. Basic science of pulmonary arterial hypertension for clinicians: new concepts and experimental therapies. Circulation, 121:2045-2066.
2. Galiè N., Humbert M., Vachiery J.L., Gibbs S., Lang I., Torbicki A., et al. 2015. 2015 ESC/ERS Guidelines for the diagnosis and treatment of pulmonary hypertension: The Joint Task Force for the Diagnosis and Treatment of Pulmonary Hypertension of the European Society of Cardiology (ESC) and the European Respiratory Society (ERS): Endorsed by: Association for

European Paediatric and Congenital Cardiology (AEPC), International Society for Heart and Lung Transplantation (ISHLT). Eur Heart J, 37:67-119.
3. Soubrier F., Chung W.K., Machado R., Grunig E., Aldred M., Geraci M., et al. 2013. Genetics and genomics ofpulmonary arterial hypertension. J Am Coll Cardiol, 62(Suppl):D13–D21.
4. Morrell N.W., Aldred M.A., Chung W.K., Elliott C.G., Nichols W.C., Soubrier F., et al. 2019. Genetics and genomics of pulmonary arterial hypertension. Eur Respir J, 53(1): 1801899.
5. Badesch D.B., Raskob G.E., Elliott C.G., Krichman A.M., Farber H.W., Frost A.E., et al. 2010. Pulmonary arterial hypertension: baseline characteristics from the REVEAL Registry. Chest, 137:376–387.
6. Drake K.M., Comhair S.A., Erzurum S.C., Tuder R.M., Aldred M.A. 2015. Endothelial chromosome 13 deletion in congenital heart disease-associated pulmonary arterial hypertension dysregulates SMAD9 signaling. Am J Respir Crit Care Med, 191:850-854.
7. Rosenzweig E.B., Morse J.H., Knowles J.A., Chada K.K., Khan A.M., Roberts K.E., et al. 2008. Clinical implications of determining BMPR2 mutation status in a large cohort of children and adults with pulmonary arterial hypertension. J Heart Lung Transplant, 27:668-674.
8. Roberts K.E., McElroy J.J., Wong W.P., Yen E., Widlitz A., Barst R.J., et al. 2004. BMPR2 mutations in pulmonary arterial hypertension with congenital heart disease. Eur Respir J, 24:371-374.
9. Pfarr N., Fischer C., Ehlken N., Becker-Grünig T., López-González V., Gorenflo M., et al. 2013. Hemodynamic and genetic analysis in children with idiopathic, heritable, and congenital heart disease associated pulmonary arterial hypertension. Respir Res, 9;14:3.
10. Liu D., Liu Q.Q., Guan L.H., Jiang X., Zhou D.X., Beghetti M., et al. 2016. BMPR2 mutation is a potential predisposing genetic risk factor for congenital heart disease associated pulmonary vascular disease.Int J Cardiol, 15;211:132-136.
11. Pousada G., Baloira A., Vilariño C., Cifrian J.M., Valverde D. 2014. Novel mutations in BMPR2, ACVRL1 and KCNA5 genes and hemodynamic parameters in patients with pulmonary arterial hypertension. PLoS One, 17;9:e100261.
12. Fujiwara M., Yagi H., Matsuoka R., Akimoto K., Furutani M., Imamura S., et al. 2008. Implications of mutations of activin receptor-like kinase 1 gene (ALK1) in addition to bone morphogenetic protein receptor II gene (BMPR2) in children with pulmonary arterial hypertension. Circ J, 72:127-133.
13. Berg J.N., Gallione C.J., Stenzel T.T., Johnson D.W., Allen W.P., Schwartz C.E., et al. 1997. The activin receptor-like kinase 1 gene: genomic structure and mutations in hereditary hemorrhagic telangiectasia type 2. Am J Hum Genet, 61:60-67.
14. Harrison R.E., Berger R., Haworth S.G., Tulloh R., Mache C.J., Morrell N.W., et al. 2005. Transforming growth factor-beta receptor mutations and pulmonary arterial hypertension in childhood.Circulation, 111:435-441.
15. Pousada G., Baloira A., Fontán D., Núñez M., Valverde D. 2016. Mutational and clinical analysis of the ENG gene in patients with pulmonary arterial hypertension. BMC Genet, 4;17:72.