



높은 침투도를 보인 *BMPR2* Intron 3 Splice Site Pathogenic Variant와 연관된 폐동맥고혈압 한국인 가족 증례 보고

Clinical Presentation with High Penetrance in a Korean Family with Pulmonary Arterial Hypertension Associated with a *BMPR2* Intron 3 Splice Site Pathogenic Variant

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Pathogenic variants of bone morphogenetic protein receptor type 2 gene (*BMPR2*) are related to the majority of cases of heritable pulmonary arterial hypertension (PAH). Over 400 pathogenic variants have been identified. However, clinical characterization of PAH is still incomplete. We present a case of heritable PAH in a Korean family showing serious clinical presentation with high penetrance. Genetic sequencing revealed a known heterozygous *BMPR2* pathogenic variant, c.418+5G>A, at a splice site of intron 3. Serious clinical presentation with high penetrance suggested that the interplay of other factors with pathologic variants might be in genotype-phenotype correlation. Further studies are needed to clarify these issues for the development of personalized medicine approaches for PAH.

Key Words: Pulmonary hypertension, Bone morphogenetic protein receptor type 2, Pulmonary artery, Heritable pulmonary arterial hypertension

INTRODUCTION

Pulmonary arterial hypertension (PAH) is a devastating disease characterized by progressive elevation of pulmonary vascular re-

sistance leading to right ventricular failure and death [1]. It is classified as heritable when offspring also suffer from the disease because of genetic susceptibility. Autosomal dominant inheritance, reduced penetrance, presentation at late age, and female predominance are features of heritable PAH. As many as 75% of cases of heritable PAH are attributable to pathogenic variants of the bone morphogenetic protein type 2 receptor gene (*BMPR2*) (MIM# 600799) [2, 3]. The lifetime risk of PAH among pathogenic variant carriers is reported to be approximately 20%; however, the clinical phenotype is variable and much higher penetrance has been reported in some families [4]. We present a family case showing serious clinical presentation of *BMPR2*-related heritable PAH with high penetrance.

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CASE

The proband case (Fig. 1, II:3) was a 58-year-old woman with exertional dyspnea of WHO functional class III at presentation [5]. She was diagnosed with essential hypertension one year prior and prescribed 5 mg amlodipine once daily. Her two daughters (Fig. 1, III:4, III:5) had been diagnosed with PAH during their third decade and had died during treatment. Chest X-ray showed

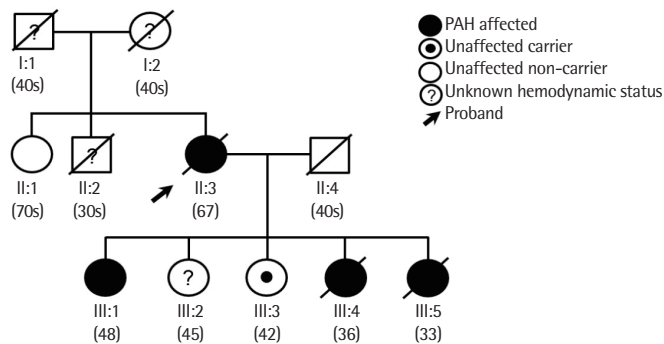


Fig. 1. Pedigree of the case family. The proband (II:3) was diagnosed with PAH later than her fourth (III:4) and fifth (III:5) daughters. Numbers in parentheses are age at death or current living age.

cardiomegaly and bilateral pulmonary arterial enlargement (Fig. 2A). Electrocardiography showed atrial fibrillation, right axis deviation, and right ventricular hypertrophy (Fig. 2B). Her six-minute walking distance was 230 m. Echocardiography showed severe pulmonary hypertension, right ventricular dysfunction, and small amount of pericardial effusion (Fig. 2C-F). Cardiac catheterization indicated a mean pulmonary arterial pressure (PAP) of 64 mmHg. There was no significant hemodynamic response to an acute vasoreactivity test using adenosine. A treatment plan of endothelin receptor blocker (ETRB), 62.5 mg bosentan twice a day, was started. Sildenafil was added 3 years later. She suffered from progressive right heart failure and died suddenly at age 67.

The proband's fourth daughter (Fig. 1, III:4) was the first case diagnosed with PAH in this family. She was diagnosed with idiopathic PAH at 29 years of age based on exertional dyspnea. Chest X-ray showed bilateral hilar enlargement (Fig. 3A). Severe pulmonary hypertension and right ventricular dysfunction were observed in echocardiography (Fig. 3B-D). In cardiac catheterization, mean PAP was 41 mmHg with pulmonary vascular resistance of 12 Wood units. She was treated with a prostacyclin in-

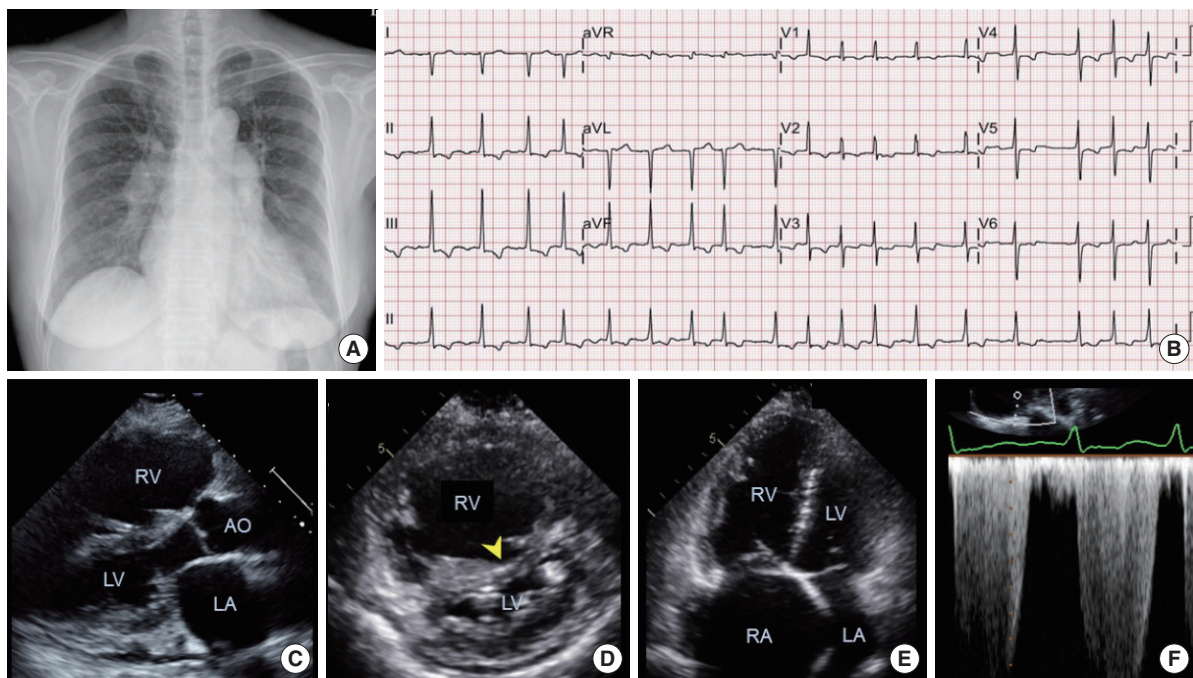


Fig. 2. Chest radiograph, electrocardiogram, and echocardiogram of the proband at the time of PAH diagnosis. Cardiomegaly with bilateral hilar enlargement in chest radiograph (A), atrial fibrillation and right ventricular hypertrophy in electrocardiogram (B), right ventricular dilation and hypertrophy, interventricular septal bowing (arrowhead), resultant left ventricular compression, and small amount of pericardial effusion in echocardiography (C-E). Estimated systolic pulmonary arterial pressure from peak tricuspid regurgitation flow velocity is around 110 mmHg (F). Abbreviations: AO, aorta; LA, left atrium; LV, left ventricle; RA, right atrium; RV, right ventricle.

haler, which was later combined with oral ETRB. She died at the age of 36 from progressive right heart failure.

The proband's youngest daughter (Fig. 1, III:5) was diagnosed with PAH at 28 years of age. Laboratory data were not available, as she was treated at another tertiary hospital. She was treated

with a combination of a prostacyclin inhaler and ETRB; however, she died at 33 years of age from right heart failure.

The proband's first daughter (Fig. 1, III:1) was diagnosed with PAH at 45 years of age based on exertional dyspnea. Her six-minute walking distance was 450 m. Chest X-ray showed cardiomeg-

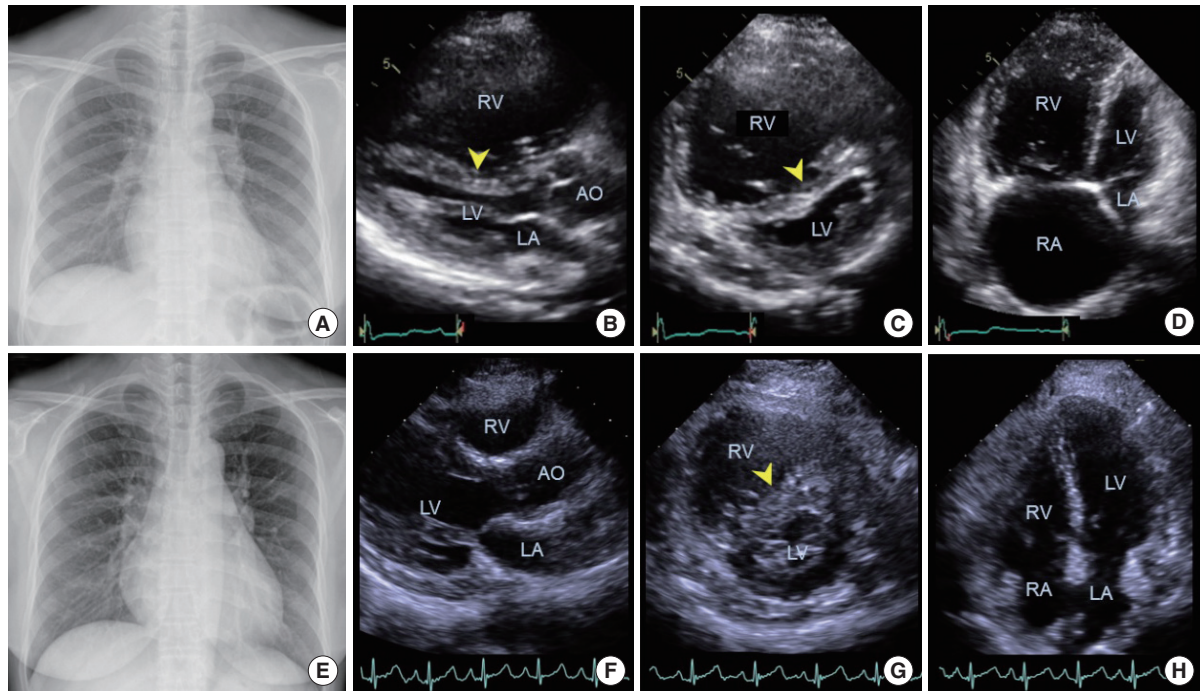


Fig. 3. Chest radiograph and echocardiogram of the proband's fourth daughter (III:4) and first daughter (III:1) at the time of PAH diagnosis. Cardiomegaly and hilar enlargement in X-ray (A, E), right ventricular dilation and hypertrophy, interventricular septal bowing to left ventricle (arrow head) in echocardiogram (B-D, F-H).

Abbreviations: AO, aorta; LA, left atrium; LV, left ventricle; RA, right atrium; RV, right ventricle.

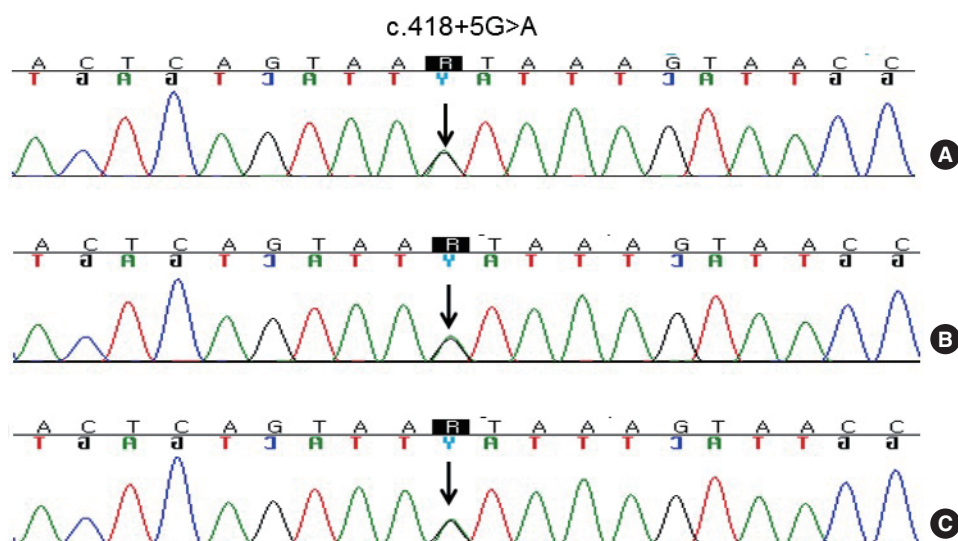


Fig. 4. Sequencing chromatogram of *BMPR2* gene from proband (A), proband's first daughter (III:1, B), and unaffected third daughter 5 (III:3, C) showed identical heterozygous pathogenic variants c.418+5G>A in the splice site of intron 3.

aly (Fig. 3E). Echocardiography showed marked pulmonary hypertension and right ventricular dysfunction (Fig. 3F-H). Her echocardiography results, obtained two years prior, were normal. In cardiac catheterization, the mean PAH was 46 mmHg with pulmonary vascular resistance of 9 Wood units, and right atrial pressure was 14 mmHg. With combination treatment using ETRB and sildenafil, she survives at WHO functional class of II.

The proband's third daughter (Fig. 1, III:3) was 42 years of age and showed normal cardiac function without symptoms at the last follow-up.

After informed consent was obtained from the proband (Fig. 1, II:3) and available family members of her two daughters (Fig. 1, III:1 and III:3), analysis of the *BMPR2* gene was performed. Sanger sequencing chromatogram revealed that all of them were heterozygous for a variant of the *BMPR2* gene (NM_001204.6) at nucleotide position 418 (c.418+5G>A), which is a splice site of intron 3 and causes splicing defect (Fig. 4).

DISCUSSION

The present family case involved heritable PAH resulting from a pathogenic variant of the *BMPR2* splice site located in intron 3. Although the c.418+5G>A variant of *BMPR2* has been reported previously [2, 6], there was no characteristic description of its genotype-phenotype association. Here we newly describe the detailed clinical presentation of this pathogenic variant. The case is remarkable in that several prototypical features of heritable PAH were apparent, such as high penetrance. Each case in the family had a varying degree of clinical presentation, including variability in the age of disease onset of PAH, even though they all expressed the same pathogenic variant.

PAH syndrome is a heterogeneous genetic disease. To date, over 400 *BMPR2* variants have been identified [7]. As each variant exhibited a different functional impact on the bone morphogenic protein signaling pathway, each pathogenic variant of *BMPR2* might have a different clinical phenotype [8]. Alteration of the BMP pathway is considered a central process in the pathogenesis of PAH. However, the precise sort of genetic change required to cause critical bone morphogenic protein dysfunction is not well known. Clinical presentation is complex and highly variable.

The characteristic feature of the present family case is a high disease penetrance, which is defined as the proportion of individ-

uals who exhibit the phenotype among the individuals who have the phenotype-associated genotype [9]. Although this study has limitations in that a complete survey has not been conducted for all family members, the authors could predict that cases 2 and 3 carried the pathogenic variant inherited from case 1. Therefore, the penetrance might be predicted at 80% in this case. The average penetrance has been known to be relatively low, around 20% among pathogenic variant carriers of familial PAH, though the penetrance among affected families could be variable [4]. Such high penetrance might be a characteristic of the c.418+5G>A pathogenic variant in this case, which is located in one of the extracellular ligand-binding domains of *BMPR2* [2, 10]. It might cause an extensive loss of conformational integrity of bone morphogenic protein receptors, type 2 (BMPR2), of the transforming growth factor β (TGF- β) superfamily of cell-signaling molecules, resulting in the development of PAH [2, 10]. We could not adduce experimental evidence on c.418+5G>A pathogenic variant's genotype-phenotype correlation in the current situation. However, further experimental studies are needed to clarify this issue.

In addition, all members of the present family case were women, although that seems to be incidental. Female predominance in heritable and idiopathic PAH has been noted since the earliest studies [8]. A gender skew was prominent among younger patients, with a female-to-male ratio of 2.3:1, which weakened among the elderly to 1.2:1 [11]. In a previous study, which enrolled 24 families with PAH, the female-to-male ratio at birth was 160:122 among pathogenic variant carriers. As father-to-son transmission was observed, despite low frequency, X-linked inheritance was excluded [12]. Such gender skewness might suggest the possible selective loss of male fetuses before birth [12]. Gender-dependent penetrance suggests that such pathogenic variants would affect even the early stages of embryonic development.

Furthermore, the phenomenon of genetic anticipation, that is, earlier age of diagnosis in the subsequent generation, has still been reported in cases of heritable PAH [12]. The prototypical example of genetic anticipation is Huntington's disease and myotonic dystrophy, in which trinucleotide repeat amplification is the responsible molecular mechanism [2, 12]. However, anticipation is controversial in heritable PAH because incomplete penetrance and highly variable clinical presentation complicate observation of anticipation phenomena [2, 8]. Lack of a biological explanation also drives controversy, because *BMPR2* does not include a trinucleotide repeat.

cleotide repeat [2, 10]. In the present case, however, a kind of phenomenon suggesting genetic anticipation was observed in which two daughters of the proband were diagnosed with PAH ahead of their mother. The daughters' PAH was also more severe at presentation and progressed rapidly than that of their mother.

Several hypotheses have been suggested to fit the complicated clinical phenotype of heritable PAH. A germline pathogenic variant of *BMPT2* would predispose the carriers to pulmonary vascular remodeling and PAH [10]. Considering the low penetrance and late presentation among carriers, "the second hit," such as modification of genes or environmental factors, is required for the development of PAH in genetically predisposed individuals [13]. Anticipation might be explained by cumulative genetic damage or by some other mechanism besides a trinucleotide repeat. Recent work suggests a role for epigenetic, such as DNA methylation, in the pathogenesis of PAH [14]. Inflammatory and hypoxic milieu might inflict oxidative damage to DNA. Such epigenetic change can be heritable and accumulate sufficiently to have a stronger impact on subsequent generation.

Genetic counselling for PAH seems to be complex because of incomplete penetrance and variable expression [4, 15]. Genetic counselling and genetic testing should be considered not only for patients with idiopathic PAH or patients with a family history of PAH according to local regulations and ethnic principles [16], but also for family members of individuals expressing heritable PAH, including those who are ostensibly healthy [4, 15]. In particular, considering the characteristic incomplete penetrance and variability in age of disease onset of PAH [4, 15], for asymptomatic carriers of PAH, such as the proband's third daughter (III:3) in this case who had a pathogenic variant of PAH without symptoms at the age of 42, yearly echocardiography for pulmonary arterial pressure should be recommended for early diagnosis and treatment [15, 16].

In conclusion, a family with heritable PAH caused by a *BMPT2* intron 3 splice site pathogenic variant showed serious clinical presentation with high penetrance. Variable *BMPT2*-related PAH might be a heterogeneous group of diseases with variable phenotypes and prognosis according to pathogenic variant loci, which might affect drug response. Characterization of variable *BMPT2* pathogenic variants and their genotype-phenotype correlations would facilitate future individualized treatment for PAH.

요 약

대부분의 유전성 폐동맥 고혈압은 bone morphogenic protein receptor type 2 (*BMPT2*) 유전자의 pathogenic variants와 관련되어 있으며 현재까지 400개가 넘는 pathogenic variants가 발견되어 왔다. 그러나, 폐동맥고혈압의 임상적 특징은 여전히 완벽하지 않다. 저자들은 높은 침투도와 중증의 임상 양상을 보인 유전성 폐동맥고혈압 한국인 가족 증례를 보고하는 바이다. 염기순서검사에서 알려진 *BMPT2* 유전자의 intron 3의 splice site에서 c.418+5G>A, heterozygous pathogenic variant가 관찰되었다. 높은 침투도와 중증의 임상 양상은 pathogenic variant와 다른 인자들과의 상호작용이 유전형-표현형 연관성에 영향을 줄 수 있음을 의미하였다. 폐동맥고혈압에 대한 정밀의료 접근법 개발을 위해 이러한 문제를 명확히 하기 위해서는 더 많은 연구가 필요할 것으로 사료된다.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

No potential conflicts of interest relevant to this article were reported.

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