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Case Report

Tuberous sclerosis complex case series with novel duplication mutations of *TSC2*

Jong-Mok Lee[a], Ju-Hyun Kim[b], Seock Hwan Choi[c,*]

[a] Department of Neurology, Kyungpook National University Hospital, Daegu, Korea
[b] Department of Neurology, Gyeongsan Joongang Hospital, Gyeongsan, Korea
[c] Department of Urology, School of Medicine, Kyungpook National University, Daegu, Korea

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1. Introduction

Tuberous sclerosis complex (TSC) is a disorder that affects multiple organ systems. It is characterized by the tumors of the skin, heart, lung, kidneys, and brain, leading to health problems like seizures, autism, cognitive impairment, and developmental delay [1]. TSC protein complex consists of harmatin and tuberin, encoded *TSC1* and *TSC2*, respectively. It acts as an inhibitor of the mechanistic target of rapamycin (mTOR) signaling pathway [1,2]. Inactivating mutations of *TSC1* or *TSC2* initiate mTOR complex 1 signaling networks, leading to cell growth, decreased autophagy, or apoptosis [1]. To date, 800 mutations in *TSC1* and 2200 mutations in *TSC2* have been reported in the Leiden Open Variation Database (LOVD, http://www.lovd.nl/TSC1, and http://www.lovd.nl/TSC2). Here, we report two cases of tuberous sclerosis with novel *TSC2* duplication mutations.

2. Case report

2.1. Case 1

A 24-year-old female patient visited the emergency room because of a squeezing pain in the left flank for four days. Multiple tuberous lesions were first observed on her face when she was three years old. She was diagnosed with angiofibroma at the age of thirteen. Her conditions were managed by laser treatment. She denied having seizures, intellectual disabilities, or dyspnea in childhood. Family members did not exhibit any symptom associated with TSC and refused to have genetic analysis.

Physical examination at the emergency room identified tenderness on the bilateral costovertebral angle and the epigastric region. Laboratory tests revealed elevated levels of aspartate transaminase (115 U/L, normal < 32 U/L), alanine aminotransferase (49 U/L, normal < 32 U/L), and C-reactive protein (22.56 mg/dL, normal < 0.5 mg/dL). Computerized tomography (CT) of the abdomen revealed multiple variable-sized fat-containing masses on bilateral kidneys. The largest such mass, located on the left kidney, was approximately 8 cm in size and exhibited a high density in the non-enhanced image, suggesting an intra-tumor hemorrhage as the source of pain. From the CT findings, we diagnosed angiomyolipoma (AML) with spontaneous rupture (Fig. 1A). We successfully performed an immediate angio-embolization (Fig. 1B).

After the treatment, we investigated systemic involvement based on the presentations of angiofibroma (Fig. 1D) and AML. Patient exhibited shagreen patches on the lower back, subungual fibroma on the toes, and dental enamel pits (Fig. 1E–G). Retinal examination revealed multiple retinal hamartomas in both eyes. High-resolution CT scan of the lung revealed multiple cystic lesions less than 5 mm, strongly suggesting lymphangioleiomyomatosis (LAM, Fig. 1H). Brain magnetic resonance imaging (MRI) revealed multiple nodular lesions in the left lateral ventricular ependymal lining, in addition to multiple lesions with high signal intensity in the left temporal and bilateral parietal lobe (FLAIR, Fig. 1I). Results of the pulmonary function test, electroencephalography, and the Wechsler adult intelligence test were all

* Corresponding author at: Department of Urology, School of Medicine Kyungpook National University, 130 Dongdeok-ro, Jung-gu, Daegu, 41944, Republic of Korea.
E-mail address: skhwan.script@gmail.com (S.H. Choi).

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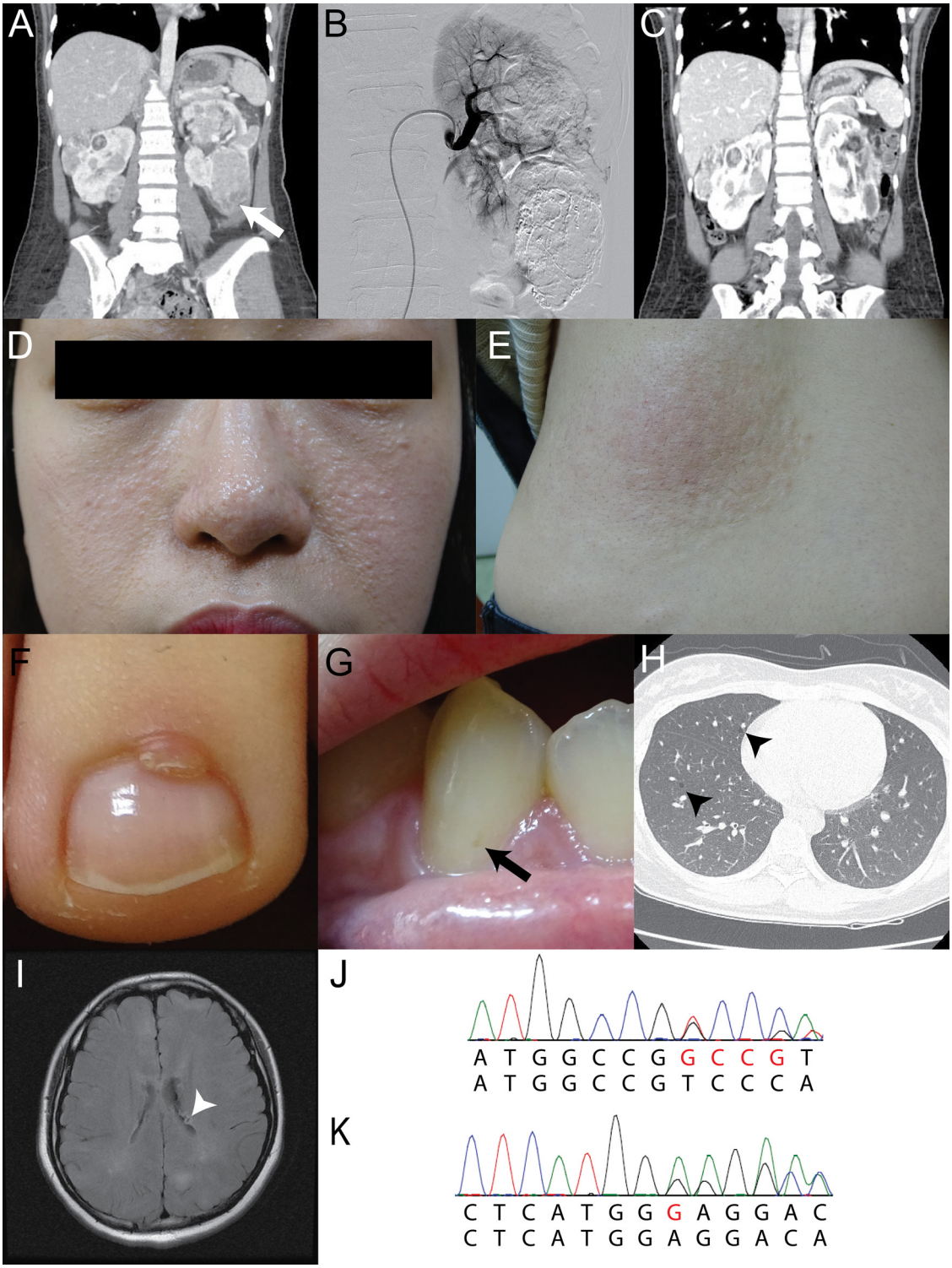

Fig. 1. Systemic involvement of tuberous sclerosis complex. Abdomen computerized tomography (CT) reveals angiomyolipoma (AML) with rupture (A, white arrow). Conventional angiogram reveals the defect of contrast media filling on left kidney after embolization (B). Shrinkage of ruptured AML on abdomen CT after 4 months of the procedure (C). Clinical photograph of the patient includes Angiofibroma, shagreen patch, subungual fibroma, and dental pits (D, E, F, and G, black arrow). High-resolution CT scan of lung reveals multiple cystic lesions suggesting lymphangioleiomyomatosis (H, black arrowheads). The subependymal nodule is noted in the FLAIR image of brain magnetic resonance imaging (I, white arrowhead). Chromatogram of Sanger confirmation indicates c.2518_2521dupGCCG (p.Val841 Glyfs*43, NM_000548.3) and c.841dupG (p.Glu281 Glyfs*57) mutations in *TSC2*, respectively (J and K).

normal.

We performed genetic analysis to confirm a final diagnosis based on the clinical findings. Genomic DNA was isolated from peripheral blood leukocytes and direct sequencing was performed on it. A novel heterozygous duplication mutation c.2518_2521dupGCCG (p.Val841 Glyfs*43, NM_000548.3) in *TSC2* was detected, which is not listed in the LOVD (http://www.lovd.nl/TCS2). It was assessed as pathogenic according to 2015 American College of Medical Genetics (ACMG) and Association for Molecular Pathology (AMP) guideline (Fig. 1J).

After 4 months of diagnostic procedures, a follow-up CT revealed that the ruptured AML shrunk to approximately 3 cm, without any complications. (Fig. 1C)

2.2. Case 2

A 27-year-old female patient with uncontrolled epilepsy and multiple facial tuberous lesions visited the department of neurology for TSC evaluation. Because the patient was orphaned at the age of seven, childhoodd history and family members' clinical information could not be obtained. She suffered from mental retardation. Physical examination at the age of 27 discovered angiofibroma on the face, shagreen patch on the left shoulder and lower back, enamel pits, and periungual fibromas, suggesting TSC. Brain MRI revealed multiple subependymal hamartomas in bilateral lateral ventricles. CT identified LAM in both lungs and AML in both kidneys. The target direct sequencing of TSC2 revealed heterozygotic c.841dupG (p.Glu281 Glyfs*57), which is not listed in the LOVD (Fig. 1K).

3. Discussion

TSC is an autosomal dominant disorder affecting multiple organs, resulting from loss-of-function germline mutations in *TSC1* or *TSC2*. The heterozygous c.2518_2521dupGCCG and c.841dupG variants of the *TSC2* gene, resulting in premature truncation, are pathogenic variants and could influence the clinical phenotype of the patient.

Two-thirds of patients with TSC harbor *de novo* germline mutations, whereas one-third of those harbor inherited mutations [1]. Seventy percent (70%) of TSC patients have *TSC2* mutations. Twenty percent (20%) of TSC patients have *TSC1* mutations [1]. The remaining 10% of TSC patients have intronic splicing variants or low-level somatic mosaicism affecting *TSC1* or *TSC2*. Missense mutations (30% of *TSC2* associated cases) and large deletions (5%) were identified in *TSC2*. In contrast, most *TSC1* mutations include small nonsense insertion and deletion mutation [1].

Our cases confirm with a previous study in genotype-phenotype comparison [2]. Both of our cases show phenotypical characteristics, namely: angiofibroma, enamel pits, periungual fibromas, subependymal hamartomas, LAM, and AML. Symptoms in TSC2 patients are more severe than those in TSC1 patients. In our study, the patient harboring truncated mutation close to the N-terminal had more severe phenotype than the patient with truncated mutation close to the C-terminal. However, the explanation of phenotypical variability owing to *TSC2* has not been well-established.

Renal lesions are commonly observed among patients with TSC. Among renal lesions, AML is the most common manifestation of TSC. AML has been reported in up to 85% of TSC patients with renal lesions and in 49%–60% of all TSC patients [3]. The size of renal AML progresses with age. It can result in painful hemorrhaging in the tumor [3]. The prevalence, number, and size of renal AML also increase with age [3]. Many TSC patients with renal AML exhibit only a few renal AML symptoms. The majority of TSC patients with renal AML come to attention via surveillance imaging [4]. Some TSC patients with renal AML can experience hemorrhage, hematuria, and chronic kidney disease [5].

Everolimus—a rapamycin analog—was approved for the treatment of subependymal giant cell astrocytoma and renal AML [1]. Sirolimus (rapamycin) exhibited improvements of forced vital capacity and forced expiratory volume in 1 s in patients with LAM [1], leading to its FDA approval. If the renal lesion is small and stable, surveillance is recommended. For asymptomatic, growing AML measuring larger than 3 cm in diameter, treatment with an mTOR inhibitor is currently recommended as the most effective first-line short-term therapy [1]. However, available evidence does not sufficiently support the administration of an mTOR inhibitor as an anti-epileptic therapy [1].

4. Conclusion

We report novel mutations in *TSC2*. These mutations can expand the genetic spectrum of TSC. The administration of an mTOR inhibitor is noteworthy and can be another challenge in treating our patient.

Conflict of interest

The authors state that there is no conflict of interest.

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References

[1] E.P. Henske, S. Jozwiak, J.C. Kingswood, J.R. Sampson, E.A. Thiele, Tuberous sclerosis complex, Nat. Rev. Dis. Primers 2 (2016) 16035.
[2] S. Avgeris, F. Fostira, A. Vagena, Y. Ninios, A. Delimitsou, R. Vodicka, R. Vrtel, S. Youroukos, D.J. Stravopodis, M. Vlassi, A. Astrinidis, D. Yannoukakos, G.E. Voutsinas, Mutational analysis of TSC1 and TSC2 genes in Tuberous Sclerosis Complex patients from Greece, Sci. Rep. 7 (1) (2017) 16697.
[3] S.K. Rakowski, E.B. Winterkorn, E. Paul, D.J. Steele, E.F. Halpern, E.A. Thiele, Renal manifestations of tuberous sclerosis complex: incidence, prognosis, and predictive factors, Kidney Int. 70 (10) (2006) 1777–1782.
[4] F.J. O'Callaghan, M.J. Noakes, C.N. Martyn, J.P. Osborne, An epidemiological study of renal pathology in tuberous sclerosis complex, BJU Int. 94 (6) (2004) 853–857.
[5] C.P. Nelson, M.G. Sanda, Contemporary diagnosis and management of renal angiomyolipoma, J. Urol. 168 (4 Pt 1) (2002) 1315–1325.