



Multiplex Ligation-dependent Probe Amplification 방법을 이용한 정신지체와 수면장애를 가진 Smith-Magenis Syndrome 환자의 진단

Diagnosis of Smith-Magenis Syndrome in a Patient with Mental Retardation and Sleep Disturbance Confirmed by Multiplex Ligation-dependent Probe Amplification

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Dear Editor,

Smith-Magenis syndrome (SMS) is characterized by mental retardation and developmental delay caused by an interstitial deletion of chromosome 17p11.2 or a mutation in the *retinoic acid induced 1 (RAI1)* gene [1, 2]. Additional phenotypes include neurobehavioral problems, such as self-injury and attention-seeking, sleep disorder, and distinctive craniofacial anomalies [3]. Most cases of SMS are de novo; in extremely rare cases, inheritance from parents has been reported [4]. While the worldwide incidence of SMS is estimated to be one in 15,000–25,000 people [3, 5], reports of occurrence of SMS are relatively infrequent in Korea [6–9]. We report a Korean SMS case confirmed by multiplex ligation-dependent probe amplification (MLPA) analysis.

The patient was a 25-year-old Korean male, the first child of non-consanguineous parents with no relevant family history of SMS. He visited the outpatient clinical genetics department as he

was suffering from mental retardation and developmental delay. His developmental delay was noticed at the age of 1. Additionally, he exhibited sleep disturbances and abnormal behavior, such as self-injury. He had dysmorphic features including brachycephaly and frontal bossing. For diagnosis of the patient, we conducted a conventional cytogenetic study and a microdeletion assay using MLPA. In the conventional chromosomal study, the karyotype was normal, and it was reported as 46,XY [20]. As we suspected a microdeletion syndrome based on his phenotype, we decided to perform MPLA analysis. We conducted the assay with MLPA P245-B1 microdeletion syndromes-1 probemix (MRC-Holland, Amsterdam, The Netherlands). Analysis of the mutation was performed using GeneMarker 1.70 (Softgenetics, Pennsylvania, USA), and the analysis was based on the relative peak values of each probe amplification divided by the value of the control probes. The threshold values to detect deletion and amplification of the genes were set at 0.65 and 1.3, respectively. A microdeletion screening test revealed deletions of *RAI1*, *dynein regulatory complex subunit 3 (DRC3)*, and *scribble cell polarity complex component (LLGL1)* gene loci (Fig. 1A), which were diagnostic findings of Smith-Magenis syndrome with an interstitial deletion in 17p11.2. To confirm SMS, we conducted MLPA using the P374-A1 probemix covering more loci, including *COP9 signalosome subunit 3 (COPS3)*, *RAI1*, *microRNA 33b (MIR33B)*, *target of myb1 like 2 membrane trafficking protein (TOM1L2)*, *phosphoribosyl pyrophosphate synthetase associated protein 2 (PRPSAP2)*, and *microfibril associ-*

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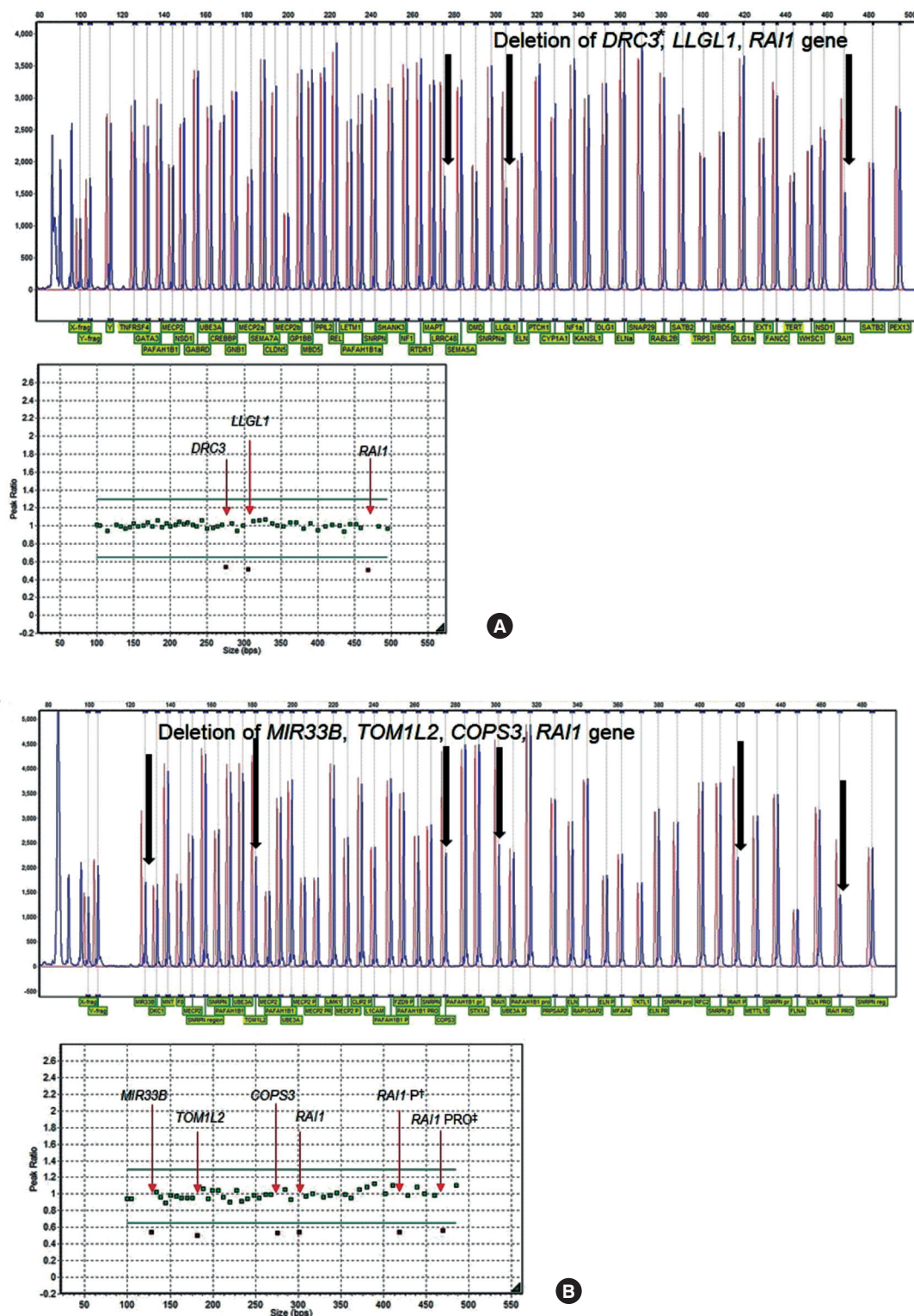


Fig. 1. (A) MLPA analysis (red: control, blue: patient) results reveal heterozygous deletion of *DRC3*, *LLGL1*, *RAI1* gene on 17p11.2. (B) Confirm test covering more loci reveals heterozygous deletion of *COPS3*, *RAI1*, *MIR33B*, *TOM1L2* gene on 17p11.2. Abbreviation: MLPA, Multiplex ligation-dependent probe amplification. *The name of the *Leucine-rich repeat-containing protein 48 (LRRC48)* gene has been changed to *DRC3*; [†]*RAI1* P and *RAI1* PRO are different sets of primers of *RAI1* gene. The names are randomly added with P and PRO as the analysis software requires different names with matching primers.

ated protein 4 (MFAP4). The results revealed deletion of the *COPS3*, *RAI1*, *MIR33B*, and *TOMIL2* genes on 17p11.2 (Fig. 1B). The final diagnosis for the patient was reported as SMS. Further cytogenetic studies of the parents and the sibling were not conducted, as the parents decided to continue treatment of the patient's sleep disorder at another clinic.

SMS exhibits distinctive phenotypes consisting of neurobehavioral problems, developmental delay, and craniofacial anomalies [3]. However, there is a wide variety of phenotypes that overlap with other congenital anomalies and mental retardation-related disease entities. For example, SMS and Down syndrome share several facial phenotypes, including upward slanting palpebral fissures, brachycephaly, flat midface, and a short and broad nose [10]. It can be challenging, especially in early infancy, to suspect SMS solely based on the symptoms and signs of patients, often leading to use of a conventional chromosomal study as a screening test. However, as is often the case, a microdeletion of 17p11.2 is not detectable via conventional karyotyping and diagnostic process can be more difficult. To compensate for the limitations of conventional cytogenetics, further studies such as fluorescent in situ hybridization (FISH), MLPA, quantitative real time PCR, chromosomal microarray (CMA), and targeted sequencing are needed [11].

FISH has been widely used as a first-tier test to detect microdeletion syndromes. The detection rate of cryptic aberrations using FISH is approximately 5–7% [12]. As FISH has a specific limitation, which is it can target only a few loci at a time, MLPA—which can target 40–50 loci in a single test—may be a viable alternative. Detection rates of causative genetic anomalies using MLPA go up to 31.8% when combining several MLPA kits [13]. CMA has a resolution of as much as 400 kb, which is more than a 10-fold improvement compared to G-banded karyotyping [14]. Owing to its clinical effectiveness, CMA is now suggested as the new first-tier screening test for patients with multiple congenital anomalies and mental retardation [14, 15]. However, it can be costly for large-scale use. In Korea, CMA is available only for research, and it is not used for routine diagnostic purposes in clinical laboratories. Considering the large cost and unavailability of CMA, a viable alternative may be MLPA screening. MLPA has a short turn-around time and is easy to conduct; most importantly, MLPA analysis results are highly reliable.

We described an SMS case with microdeletions in the chromo-

somal region 17p11.2. This is the first MLPA-confirmed SMS case in Korea. We suggest the use of MLPA as the choice of screening method for patients with developmental delay and mental retardation, in Korea.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

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

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