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가족성 암 증후군이 의심되는 환자에서 유전 상담의 영향: *TP53* 유전자 돌연변이가 확인된 Li-Fraumeni 유사 증후군 환자를 통한 증례 보고

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Genetic Counseling Can Influence the Course of a Suspected Familial Cancer Syndrome Patient: From a Case of Li-Fraumeni Like Syndrome with a Germline Mutation in the *TP53* Gene

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We report a 26-yr-old female patient with bilateral breast cancer who was clinically diagnosed with Li-Fraumeni like syndrome (LFL) and subsequently found to have a germline mutation of the *TP53* gene. The patient was initially diagnosed with right breast cancer at age 24 yr and then with left breast cancer at age 25 yr. Surgery and radiotherapy were performed accordingly. The patient had a family history of various types of early onset cancers and was referred to a genetic counseling clinic. She was clinically diagnosed with LFL. Genetic analysis of the *TP53* tumor suppressor gene was performed with the patient's consent. Direct sequencing of *TP53* gene exons 5, 6, 8, 9, and 11 revealed a germline missense mutation, resulting in an amino acid change from an arginine to a histidine (g.13203G>A, p.R175H). Considering the family history, individualized cancer surveillance was performed including a gastroscopy and a brain MRI. Even though the patient had not shown any neurological symptoms, a huge mass on the temporal lobe was incidentally found and the patient received surgery and radiotherapy. Although the residual mass required further treatment, the patient decided on supportive care alone and was discharged. We report a case of LFL, with a germline *TP53* mutation, which was confirmed by gene sequencing in Korea. This case shows how genetic predisposition screening and counseling in patients, suspected of having a familial cancer syndrome, can influence the course of the patient. *(Korean J Lab Med 2008;28:493-7)*

Key Words : *Li-Fraumeni syndrome, Li-Fraumeni like syndrome, TP53 gene, Genetic counseling*

INTRODUCTION

Li-Fraumeni syndrome (LFS; MIM #151623), which was first described in 1969 by Li and Fraumeni [1], is a cancer predisposition syndrome associated with sarcoma and a wide spectrum of tumors. Birch et al. [2] and Eeles [3] identified several families exhibiting traits similar to LFS, but

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as these families did not exactly satisfy LFS criteria, they were designated as having Li-Fraumeni like syndrome (LFL). Classic LFS is defined as having a proband with sarcoma before the age of 45 yr, and a first-degree relative with any cancer under the age of 45 yr, and also a first- or second-degree relative with any cancer under the age of 45 yr or sarcoma at any age [1]. LFL is defined as two first- or second-degree relatives with LFS related malignancies including sarcoma, breast cancer, brain tumor, adrenal cortical tumor, or leukemia at any age [3].

TP53 is the main tumor suppressor gene associated with LFS and LFL, and over 50% of the LFS families have an identifiable *TP53* germline mutation [4]. In LFL families, *TP53* germline mutation rates vary from 8% to 22% according to the definition used [5]. LFS is a highly penetrant cancer syndrome, and identification of a germline mutation can confirm its diagnosis.

Here, we report a case of LFL with a germline mutation in the *TP53* gene. With the patient's consent, genetic counseling and germline mutation analysis of the *TP53* gene were performed, which subsequently led to the diagnosis of another malignancy.

CASE REPORT

A 26-yr-old female patient with bilateral breast cancer was initially diagnosed with right breast cancer at age 24 yr, and received a right quadrantectomy with lymph node dissection and postoperative radiotherapy. One year later, a left breast mass was detected and excision of the left tumor and radiotherapy were performed. She was being treated with tamoxifen and no evidence of recurrence was detected on ultrasonography or breast MRI. Given her family history of various types of early onset cancers, she was referred to a genetic counseling clinic. Thorough review of her family history revealed multiple early onset cancers, including brain tumors, gastric cancer, and lung cancer (Fig. 1). The patient's younger brother was diagnosed with, and died from, a brain tumor at the age of 4 yr. Her younger sister was diagnosed with gastric cancer at the age of 20 yr and died at the age of 22 yr. The patient's older sister died of an undefined cause with ascites at the age of 11 yr. Her father died of lung cancer at the age of 36 yr, and one of her paternal uncles died of a brain tumor at the age of 10 yr. Therefore, she was clinically diagnosed with LFL. Genetic analysis of the *TP53* tumor suppressor gene was performed with the patient's consent. Leukocyte DNA was extracted using the Puregene DNA Purification kit (Gentra System, Minneapolis, MN, USA) according to the manufacturer's instructions. Genetic analysis of the *TP53* tumor suppressor gene was performed using direct sequencing, covering exons and exon-intron borders of exons 5, 6, 8, 9, and 11. The PCR reaction mixture (10 μL) contained 1.0 μL 10× PCR buffer (Takara, Tokyo, Japan), 0.7 μL 2.5 mM each dNTP (Takara), 0.3 μM each primer (Bioneer Corp., Cheongwon, Korea), 0.5 U Taq DNA polymerase (Takara), and 1 μL (0.5 μg) genomic DNA. The thermal cycler (Biometra T Gradient PCR, Gottingen, Germany) amplification profile used was as follows: 35 cycles of 30-sec denaturation at 95℃, 30-sec annealing at 60℃ (up to 65℃), and 30-sec extension at 72℃. Amplified DNA (1.5 μL) was incubated with 2 U shrimp alkaline phosphatase and 5 U exonuclease I (USB Corp., Cleveland, OH, USA) at 37℃ for 15 min. The enzymes were inactivated by incubation at 80℃ for 15 min, after which the DNA was denatured at 95℃ for 15 min. The presence of a PCR product was determined using agarose gel

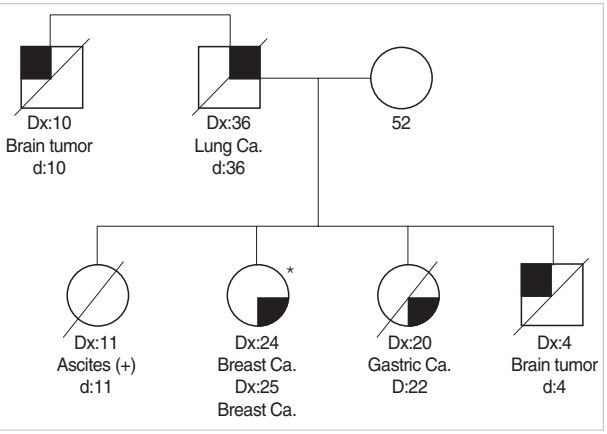

Fig. 1. Pedigree of the Li-Fraumeni like syndrome family. Solid symbols represent individual with tumors. Types of tumors are indicated, with ages (yr) at the time of diagnosis and death (if applicable). Asterisk (*) marks the individual analyzed and found to carry the germline *TP53* mutation.
Abbreviations: Dx, diagnosis; d, death; Ca, cancer.

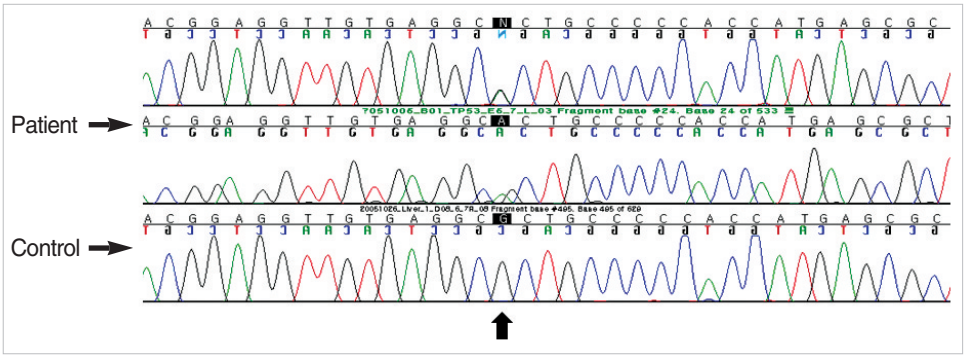

Fig. 2. *TP53* gene sequencing analysis representing missense mutation of codon 175 (p.R175H).

electrophoresis. Cycle sequencing was performed using a BigDye Terminator Cycle Sequencing Ready Reaction kit v3.0 (Applied Biosystems, Foster City, CA, USA) and an automated ABI Prism 3100 Genetic Analyzer (Applied Biosystems).

Sequencing revealed that the patient had a mutation in codon 175, resulting in an amino acid change from arginine to histidine (g.13203G>A, p.R175H) (Fig. 2). Considering the family history, an individualized cancer surveillance program, including gastroscopy and brain MRI, was recommended. The patient agreed to preventive cancer surveillance. Although she had no neurological symptoms, a huge mass on the temporal lobe was found on a brain MRI, and pleomorphic xanthoastrocytoma was pathologically diagnosed. The tumor was removed, and this procedure was followed by a radiotherapy. Despite this treatment, the residual brain tumor mass required further treatment; however, the patient decided on supportive care alone and was discharged.

DISCUSSION

This study revealed a 26-yr-old LFL patient with a germline *TP53* missense mutation in codon 175, resulting in an amino acid change of arginine to histidine. This is the third report of LFS/LFL in Korea. The first case was reported in 1995 by Bang et al. [6] with a family history of early onset breast cancer with p53 germline mutation confirmed in the proband and her sister. The second report was in a hereditary diffuse gastric cancer family with LFS related phenotype [7]. The mutations found in all three cases were different and the mutation of this case is in the L2 loop that binds the minor groove of DNA and has been found to be deleterious in *in silico* analysis using both Align-GVGD and SIFT methods [8]. It is one of the most common mutations reported, and is found in 20 Li-Fraumeni families according to the International Agency for Research on Cancer *TP53* database R12 release [8]. The present case is also consistent with a study by Olivier et al. [9], in which brain tumors were associated with missense *TP53* mutations located in the DNA-binding loop.

Although the mutation in the case was one of the most common mutations found in LFL/LFS, this case proposes many key issues in genetic testing for familial cancer syndrome patients. As can be seen in this case, the timing of genetic screening is crucial. Despite the family history of multiple early onset cancers, the patient was offered testing only after the diagnosis of bilateral cancers. There is evidence to suggest that a prophylactic mastectomy in women with a high risk of breast cancer is beneficial, and may have been of benefit to the patient in this report [10]. Several cases of radiation-induced secondary malignancies have been reported, and there are many reviews on the controversial effects of radiation in LFS patients [11-13]. If the mutation status had been revealed, the patient and the medical staff may have chosen an alternative to radiotherapy after the diagnosis of the right breast cancer.

Another issue is the genetic testing of family members of a known familial cancer syndrome patient. Testing of other family members who are at risk of carrying the same mutation may be helpful in early diagnosis and treatment. This report shows a family of index case with two members

with brain tumors and one member with breast cancer at the time of genetic counseling. Since gastric cancer and lung cancer have not yet been defined as LFL malignancies, only three individuals, including the proband, are included in LFL-defining criteria. However, other cancers that have not yet been classified as typical LFL malignancies might still be considered as a phenotype in LFL or LFS. This is supported by the discovery of a *TP53* germline mutation in a family with hereditary diffuse gastric cancer in Korea [7]. Although the genetic testing was not performed in other family members, genetic counseling was done informing the possibilities of their mutation status and their options following the testing.

The last is the issue of surveillance after genetic screening. Due to the lack of effective screening, and the complex nature of the disease, Chompret et al. [14] suggested that individuals be offered *TP53* analysis if they fulfill certain criteria. Although the effectiveness of genetic testing and surveillance have not yet been proven with respect to LFS or LFL, many clinicians conduct surveillance strategies according to their policy [15]. In this case, a huge brain tumor was found incidentally after the surveillance measure, which enabled surgery and treatment.

Our experiences with the patient described herein, which highlight the impact of genetic testing in patients suspected of familial cancer syndrome, indicate that such patients should be given a genetic counseling by professionals before and after a genetic testing.

요 약

26세 양측성 유방암 여성 환자가 임상적으로 Li-Fraumeni 유사 증후군 환자로 진단받았고 이후에 *TP53* 유전자의 배자계열 변이가 발견되었다. 환자는 초진 시 24세에 우측 유방암을, 25세에 좌측 유방암을 진단받았다. 이에 따라 수술 및 방사선 치료를 받았다. 가족력상 다양한 암의 조기 발병력이 드러나 유전상담 클리닉에 의뢰되어 임상적으로 Li-Fraumeni 유사 증후군으로 진단되었다. 유전자 검사에 대한 설명 후 환자 동의하에 종양억제 유전자인 *TP53* 유전자 분석이 시행되었다. 염기서열 분석법으로 엑손 5, 6, 8, 9, 11에 대한 검사 결과 아미노산이 아르기닌에서 히스티딘으로 변하는 *TP53* 유전자의 배자계열 변이가 발견되었다(g.13203G>A, p.R175H). 가족력을 고려하여 위내시경과 뇌 자기공명영상을 포함한 맞춤 검진이 시행되었다. 신경학적 증상을 호소하지 않았음에도 우측 관자엽의 큰 종양이 우연히 발견되었고 수술과 방사선 치료를 받았다. 치료 후 잔존암이 남았으나 환자는 지지 치료만 받기 원해 퇴원하였다. 한국에서 염기서열 분석으로 *TP53* 유전자 돌연변이가 확진된 Li-Fraumeni 유사 증후군 환자를 보고하는 바이다. 본 증례는 가족성 유전암이 의심되는 환자에서 유전자 검사 및 유전 상담의 영향을 보여주는 예이다.

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