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Letters

Pulmonary fibrosis in dyskeratosis congenita with *TINF2* gene mutation

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Running head: Pulmonary fibrosis in DC with *TINF2* mutation

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To the Editor:

Dyskeratosis congenita (DC) is a rare inherited disorder of ectodermal dysplasia characterized by the classical mucocutaneous triad of abnormal skin pigmentation, nail dystrophy, and leukoplakia[1-3], at least one of which is present in around 80-90% of DC cases. Bone marrow failure is another common feature, and a variety of other abnormalities (*e.g.* dental, gastrointestinal, neurological, ophthalmic pulmonary, and skeletal) have been also described[1-3]. The main causes of mortality in DC are bone marrow failure, pulmonary disease and malignancy[1]. Three modes of inheritance have been recognized: X-linked recessive, autosomal dominant, and autosomal recessive[1, 3]. Eight DC genes (*DKC1*, *TERC*, *TERT*, *NOP10*, *NHP2*, *TINF2*, *TCAB1*, and *RTEL1*) have already been identified, and their mutations account for approximately 60% of all DC cases[1]. In the DC genes, mutations in *TERC*, *TERT* and *DKC1* have recently been reported to be associated with familial pulmonary fibrosis and idiopathic pulmonary fibrosis, and pulmonary fibrosis is recognized as one of the features of DC. However, the relationship between mutations in the other DC genes and pulmonary fibrosis has not yet been clarified. To the best of our knowledge, this is the first case report describing a DC patient with pulmonary fibrosis who had a *TINF2* mutation.

A 43-year-old woman visited our hospital with cough and progressive dyspnea. She had never smoked and had a history of aplastic anemia, ocular pemphigoid, erythroplasia of Queyrat, and infertility. Her father had been diagnosed as having aplastic anemia, and his whole body was pigmented. About two years ago, she complained of cough and consulted her personal doctor. Her chest radiographs showed diffuse reticular shadows in the bilateral lung fields. She was referred to a general hospital and was diagnosed with idiopathic interstitial pneumonia. Because her general condition was stable at that time, she was followed up without any specific therapy for one year. She was referred to our hospital due to gradual worsening of dyspnea, and admitted for further examinations. Her physical examination was remarkable for skin pigmentation on her whole body, ocular pemphigoid in the left eye, and fine crackles in both lung fields. Her fingertip skin was rough but the nails were not dystrophic. Although no leukoplakia was found in the oral mucosa, she had erythroplasia of Queyrat of the vulva. Laboratory data showed elevated lactate dehydrogenase, transaminases, erythrocyte sedimentation rate, and sialylated carbohydrate antigen KL-6 with thrombocytopenia. Chest roentgenograms demonstrated consolidation and reticular shadows in the bilateral lung fields. Furthermore, chest computed tomography (CT) revealed consolidation and reticular shadows in both lung fields, as well as bronchiectasis and cystic shadows in the left lung.

At this point, we strongly suspected that she had DC. To make a definite diagnosis, we first examined the two DC genes *TERC* and *TERT* by direct sequencing. However, no mutations were found in either gene. Southern blot analysis showed short telomere length (Fig. 1A), therefore mutations in *TINF2* were next explored. As shown in Fig. 1B, because direct sequencing showed a n871-874 tetra-nucleotide AGGA deletion in *TINF2*, she was diagnosed as having DC with pulmonary fibrosis associated with *TINF2* mutation. As her respiratory condition progressed, steroid pulse therapy followed by oral prednisolone was conducted. However, no improvement of her symptoms was observed, and bilateral pneumothorax with mediastinal and subcutaneous emphysemas developed. She died of respiratory failure one year after starting the treatment.

DC is a rare genetic ectodermal disorder characterized by skin hyperpigmentation, nail dystrophy, and leukoplakia of the mucous membranes. Bone marrow failure is a frequent finding, and a predisposition to malignancy has been noted. Although pulmonary manifestations of DC were believed to be uncommon, Dokal reported that abnormal pulmonary features may be seen in as many as 10-15% of patients[1].

Genetically, DC is heterogeneous, with 3 forms having been identified: X-linked recessive, autosomal dominant, and autosomal recessive. In the present case, the patient's father had suffered from the same disease, therefore we suspected that the form of DC of this

patient was autosomal dominant. Autosomal dominant form of DC is caused by heterozygous mutations in the core components of telomerase, *TERC*[4, 5] and *TERT*[6, 7] as well as in the component of the shelterin telomere protection complex, *TINF2*[3]. In this patient, the *TINF2* mutation, but not *TERC* and *TERT*, was confirmed by gene mutation analysis. It has previously been reported that mutations in *DKC1*[8], *TERC*[5], and *TERT*[6] were associated with pulmonary fibrosis in DC patients. *DKC1* was not analyzed in this patient, because mutation in *DKC* causes the X-linked form of DC. Regarding the relationship between pulmonary fibrosis and *TINF2* mutation in DC, Walne *et al.* have reported that only one patient had pulmonary fibrosis among other clinical features in 33 DC patients with *TINF2* mutations[3]. However, they did not describe the patient in detail. To the best of our knowledge, this is the first case report showing pulmonary fibrosis in DC with *TINF2* mutation.

TINF2 mutations were reported to be heterozygous mutations in a sixth-found DC gene by Savage *et al.* in 2008[9]. *TINF2* encodes TIN2, and is a component of the shelterin telomere protection complex. The shelterin complex has at least 3 effects on telomeres, which determines the structure of the telomeric terminus, is implicated in the generation of t-loops, and controls the synthesis of telomeric DNA by telomerase[1, 10]. Without the protective activity of shelterin, telomeres are no longer hidden from DNA damage-repair

mechanisms, and chromosome ends are therefore incorrectly processed by the DNA-repair pathways. Approximately 11% of all DC has been reported to be accounted for by *TINF2* mutations, and patients with *TINF2* mutations have significantly shorter telomeres than other DC subtypes[3]. It has also been reported that most patients with DC with *TINF2* mutations have severe disease, and, compared with other DC genes, DC patients with *TINF2* mutations have a high incidence of aplastic anemia before the age of 10 years[3].

Aberrant repair process by enhanced apoptosis of alveolar epithelial cells plays a critical role in the pathogenesis of pulmonary fibrosis such as idiopathic pulmonary fibrosis, although the precise mechanism of developing pulmonary fibrosis is still unclear. The mechanism(s) of pulmonary fibrosis in DC has also not yet been clarified. However, because mutations in DC genes cause short telomere length with functional deficits in telomere maintenance, telomere in alveolar epithelial cells may be short. In patients with DC, we speculate that aberrant lung repair by enhanced cell death causes pulmonary fibrosis, although the short telomere length in alveolar epithelial cells has not been directly demonstrated.

Herein, we describe the first case report of DC with pulmonary fibrosis associated with *TINF2* mutation. This report proved that mutations not only in *TERC*, *TERT* and *DKC1*, but also *TINF2* cause pulmonary fibrosis in DC. However, we do not know why mutations in

TERC, *TERT* and *DKC1* are frequently found in DC patients with pulmonary fibrosis in contrast to the other five genes. In addition, sex hormones which can increase telomerase activity are potential therapeutic drugs, however, no standard treatment has been established for pulmonary fibrosis in DC patients. Because the clinical characteristics and pathogenesis of pulmonary fibrosis in DC is not clear, the accumulation of case-based reports sheds light on the understanding of this devastating disease.

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FIGURE LEGENDS

Fig. 1: (A) Southern blot analysis showed shorter telomere length of the patient (P) compared to age-matched healthy controls. MWM: molecular weight marker. (B) Gene mutation analysis by direct sequencing showed n871-874 tetra-nucleotide AGGA deletion in *TINF2* gene.

Fig. 1