Multifocal Micronodular Pneumocyte Hyperplasia and Osteosclerotic Lesions in a Patient with Tuberous Sclerosis Complex Misdiagnosed with Lung Adenocarcinoma

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A 45-year-old woman presented with multiple ground-glass nodules (GGNs) in both lungs. Osteosclerotic changes were observed on a thoracoabdominal computed tomography scan. She underwent surgical resection of a single GGN and was diagnosed with lung adenocarcinoma with bone metastases; however, neither lung nor bone lesions progressed. Pathological reexamination of the biopsy confirmed multifocal micronodular pneumocyte hyperplasia (MMPH) diagnosis, and subsequent genetic analysis identified a nonsense mutation in the tuberous sclerosis complex (*TSC*) 1 gene. The case report presented here suggests a potential role for MMPH and osteosclerotic lesions in diagnosing TSC.

Key words: multifocal micronodular pneumocyte hyperplasia, osteosclerotic lesion, tuberous sclerosis complex, ground-glass nodule, genetic analysis

INTRODUCTION

Tuberous sclerosis complex (TSC) is an autosomal dominant genetic disorder characterized by multiple organ hyperplasia [1–3]. Moreover, TSC is caused by mutations in the *TSC1* gene encoding hamartin or *TSC2* gene encoding tuberin [4]. Lymphangioleiomyomatosis (LAM) and multifocal micronodular pneumocyte hyperplasia (MMPH) are well-known lung lesions associated with TSC. MMPH was initially reported in 1991 [5] as a rare pulmonary manifestation of TSC. However, with the development of high-resolution computed tomography (CT), MMPH has been identified as a relatively common lesion, with a prevalence of 40–60% in patients with TSC [6, 7].

MMPH diagnosis is not challenging in cases presenting with typical dermatological and neurological manifestations or a family history of TSC. However, MMPH can remain undiagnosed or misdiagnosed as lung adenocarcinoma in sporadic or subclinical cases of TSC. Furthermore, patients with TSC can present with osteosclerotic lesions resembling bone metastases. Herein, we report a case previously misdiagnosed as lung adenocarcinoma presenting with multiple ground-glass nodules (GGNs) and bone metastases, which were eventually diagnosed as sporadic TSC using genetic analysis.

CASE REPORT

A 45-year-old woman with a smoking history of 40-pack-year presented to her primary care physician with multiple GGNs in both lungs. The lesions were detected on a chest radiograph during an annual health checkup. The patient exhibited no respiratory symptoms or epilepsy. Routine laboratory tests, including blood cell counts and biochemical examinations, revealed no abnormalities. The lung sounds were normal. Additionally, no skin abnormalities were noted. Thoracoabdominal CT revealed multiple GGNs, 1 to 8 mm in size, distributed predominantly in the bilateral upper lobes of the lungs, without spiculations (Fig. 1A). Moreover, multiple osteosclerotic changes were identified on CT (Fig. 1B) with normal bone scintigraphy using metastable technetium 99-labeled methylene diphosphonate (Fig. 1C). Nine months later, a single part-solid GGN with surrounding tiny GGNs in the right middle lobe was surgically removed, resulting in a diagnosis of lung adenocarcinoma in situ (AIS) and multiple atypical adenomatous hyperplasia (AAH).

The patient was referred to our hospital three years after the initial presentation. As the remaining ground-glass nodules remained stable in size with the absence of progressive osteosclerotic changes on CT, MMPH was suspected to be associated with TSC rather than lung cancer. Brain magnetic resonance imaging revealed irregularity in the wall of the lateral ventricle, consistent with subependymal and cortical nodules in the left frontal lobe, which further prompted us to investigate the possibility of TSC. We consulted our pathologist to reexamine the surgical lung biopsy specimens. The specimen demonstrated proliferation of type II alveolar epithelial cells with atypia, homogeneous cells (Figs. 2A

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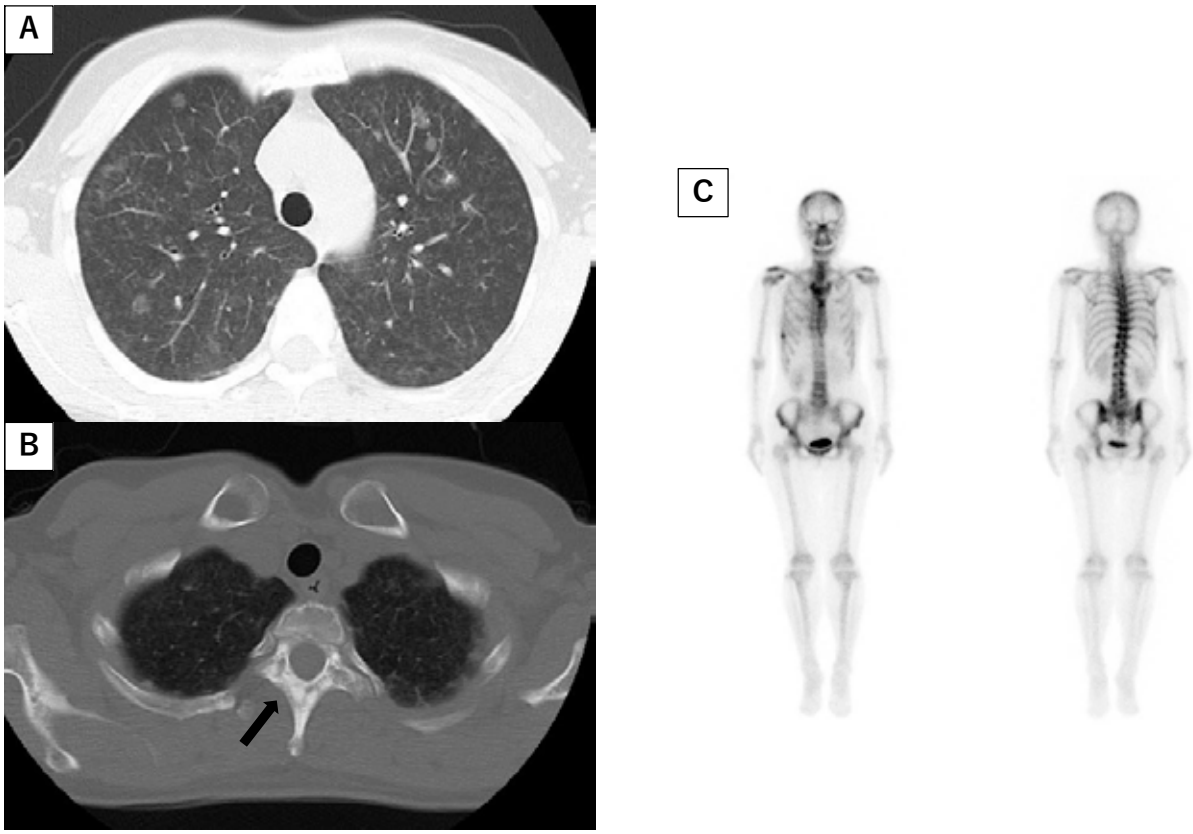

Fig. 1 (A) Multiple ground-glass nodules distributing predominantly in the bilateral upper lobes and (B) multiple osteosclerotic lesions in the vertebral body on thoracic computed tomography scan (arrow). (C) No findings suggestive of bone metastasis on bone scintigraphy using metastable technetium 99-labeled methylene diphosphonate. The traumatic change is identified at the right sixth rib.

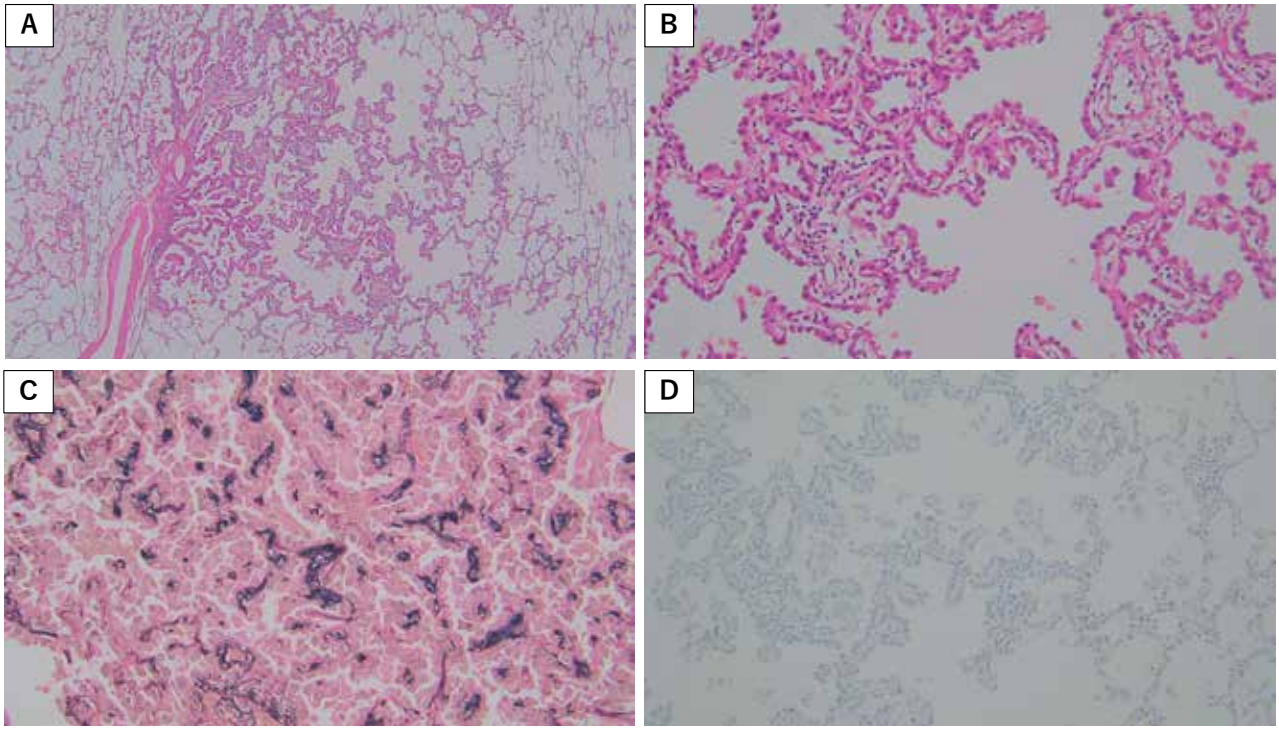

Fig. 2 Pathological findings of surgical specimen of the right middle lobe. (A and B) Proliferation of type II alveolar epithelial cells with atypia homogeneous cells (Hematoxylin and Eosin stain x20 and x100). (C) Aggregation of elastic fibers (Verhoeff's van Gieson stain x400). (D) Negative staining for carcinoembryonic antigen (x100).

Table 1 Cases of multifocal micronodular pneumocyte hyperplasia and multiple sclerotic bone lesions successfully diagnosed with tuberous sclerosis complex by testing gene mutations

References	Age Sex	MMPH	Multiple sclerotic bone lesions	Classical triad for TSC			Other manifestations of TSC	Type of genetic mutation
				Facial angiofibromas	History of epilepsy	Intellectual disability		
Kimura, *et al.* [15]	46 F	diagnosed by TBLB	present	present	absent	absent	Shagreen patch, Ungual fibromas	A frameshift mutation in *TSC1*
Li, *et al.* [16]	53 M	diagnosed by SLB	present	absent	absent	absent	Subependymal calcified nodules, Multiple renal cysts	Nonsense mutation in *TSC1*
Present case	45 F	diagnosed by SLB	present	absent	absent	absent	Subependymal and cortical nodules	Nonsense mutation in *TSC1*

MMPH, multifocal micronodular pneumocyte hyperplasia; TSC, tuberous sclerosis complex; TBLB, transbronchial lung biopsy; SLB, surgical lung biopsy

and B), aggregation of elastic fibers (Fig. 2C), and negative staining for carcinoembryonic antigen (Fig. 2D). Based on these findings, a provisional diagnosis of MMPH secondary to TSC was made. Pathological findings suggestive of LAM were not observed. However, the patient did not meet the clinical diagnostic criteria for TSC [6]. Therefore, she provided written informed consent and underwent testing for *TSC1/TSC2* gene mutations. The whole genome of the *TSC1/TSC2* gene region was amplified using a long-range polymerase chain reaction, followed by next-generation sequencing [8]. The identified mutations were validated by CEL nuclease-mediated heteroduplex incision using polyacrylamide gel electrophoresis, silver staining, and the Sanger sequencing (CHIPS) method [9]. A nonsense mutation in *TSC1*, NM_000368.5:c.2131C > T p.(Gln711Ter) was identified. The absence of a family history of TSC, combined with this *TSC1* mutation, led to a definitive diagnosis of sporadic TSC.

Thereafter, she was followed up at the outpatient clinic. No neurological or dermatological manifestations were observed.

DISCUSSION

MMPH is a pulmonary manifestation commonly observed in TSC, characterized by the proliferation of alveolar type II epithelial cells in small nodules (1–10 mm in diameter) randomly distributed in the bilateral upper lobes or peripheral lung fields [10]. GGNs in MMPH rarely change in number or size [10, 11]. Nonetheless, a report indicated that MMPH-related GGNs can increase over 8 years [12], warranting consideration of potential malignancy.

In subclinical and sporadic cases of TSC, differentiating MMPH from AIS or AAH based on radiological findings or pathological examination is difficult [10]. In the present case, multiple GGNs in both lungs were initially diagnosed as lung adenocarcinoma despite a surgical biopsy. Physicians and pathologists presumably inferred malignant diseases from the concomitant presence of multiple osteosclerotic lesions that mimic bone metastasis. However, considering the possibility of TSC when multiple lung GGNs and osteosclerotic lesions are observed on CT is important. Although osteosclerotic lesions are excluded from the diagnostic criteria for TSC due to their low specificity, a retrospective study examining patients with TSC-LAM and sporadic LAM reported that osteosclerosis was more common than dermatologic manifestations of TSC. These lesions demonstrated a high specificity of 97% in distinguishing TSC-LAM or TSC from sporadic LAM [13]. As the classical triad of TSC, such as facial angiofibroma, a history of epilepsy, and intellectual disability, occurs in only 29% of patients with TSC [14], the presence of osteosclerotic lesions could be a significant indicator of TSC, even in the absence of major dermatologic manifestations.

Genetic analysis plays a key role in the diagnosis of TSC, as the identification of a genetic variant in *TSC1* or *TSC2* is sufficient for the definitive diagnosis of TSC, regardless of the clinical findings [6]. Three cases have been reported, including ours, with multiple GGNs suggestive of MMPH concomitant with multiple osteosclerotic lesions, which contributed to the successful diagnosis of TSC confirmed by genetic analysis (Table 1). Kimura *et al.* reported the case of a 46-year-old woman with pulmonary MMPH that had been present for 10 years along with multiple osteosclerotic lesions. Although her clinical manifestations including shagreen patches and ungual fibromas satisfied the clinical diagnostic criteria for TSC, the presence of the condition was confirmed by genetic analysis, which revealed a *TSC1* mutation [15]. Li *et al.* reported the case of a 53-year-old man who presented with MMPH and multiple osteosclerotic lesions as well as subependymal calcified nodules and multiple renal cysts. Considering his clinical manifestations did not meet the clinical diagnostic criteria, he underwent a genetic analysis, which revealed a *TSC1* mutation, leading to a definitive diagnosis of TSC [16]. Interestingly, Togi *et al.* investigated genotype-phenotype correlations in 283 Japanese patients with TSC and reported that MMPH was frequently detected in patients with *TSC1* mutations, whereas intellectual disability was commonly linked to *TSC2* mutations [17]. These findings are consistent with those of the present case, and the type of *TSC* mutation may be beneficial for predicting organ involvement.

Minimally invasive examinations such as CT can easily identify pulmonary and bone lesions suggestive of TSC. Therefore, patients with multiple GGNs predominantly distributed in the bilateral upper lobes

and osteosclerotic lesions are favorable candidates for further examination of other organ involvement or genetic testing for TSC.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

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