Introduction

Mixed connective tissue disease (MCTD) is an autoimmune disease containing elements of three rheumatic diseases: systemic lupus erythematosus, systemic sclerosis, and polymyositis/dermatomyositis. Proteinase 3 (PR3)-antineutrophil cytoplasmic antibody (ANCA) is an autoantibody found in granulomatosis with polyangiitis (GPA) and is not considered as a specific autoantibody for MCTD. However, it has been suggested that MCTD positive for PR3-ANCA complicates systemic atherosclerosis\(^1\). Moreover, diabetes mellitus (DM) is classified into two categories: type 1 and type 2, with type 1 DM being considered as the autoimmune form of diabetes\(^2\). There are currently no reports of MCTD positive for PR3-ANCA in patients with chronic thyroiditis and slowly progressive type 1 DM (SPT1DM) positive for anti-glutamic acid decarboxylase (GAD) antibodies.

Here we report a case of MCTD positive for PR3-ANCA in a patient with SPT1DM and chronic thyroiditis.

Case report

A woman in her sixties was referred to the rheumatology department in our hospital with a 3-month history of swelling and numbness in her fingers. A prominent atherosclerotic lesion was revealed upon brain magnetic resonance imaging, and she was found to have mixed connective tissue disease (MCTD) positive for proteinase 3 (PR3)-antineutrophil cytoplasmic antibody (ANCA). This rare case of MCTD accompanying SPT1DM and PR3-ANCA suggested that a synergy between MCTD and PR3-ANCA triggers atherosclerosis.
1,968 cm/sec; left, 2,090 cm/sec. Both values were more than the mean value for her age plus two standard deviations). Chronic thyroiditis was diagnosed based on the diffuse swelling of the thyroid, anti-thyroglobulin and thyroid peroxidase antibody positivity, and fine needle aspiration specimen findings. The cause of her double vision was not clarified. Brain magnetic resonance imaging (MRI) did not reveal any findings of an enlargement of extraocular muscles or orbital tumors. However, fluid-attenuated inversion recovery imaging showed multiple high signal intensities in the white matter, which were compatible with chronic ischemic changes (Fig. 1).

The results of blood examinations undertaken upon referral to the rheumatology department were as follows: white blood cell count, 3,700/µL; hemoglobin, 10.8 g/dL; platelet count, 203,000/µL; C-reactive protein, < 0.14 mg/dL; blood urea nitrogen, 15.8 mg/dL; serum creatinine, 0.49 mg/dL; glucose, 215 mg/dL; hemoglobin A1c, 6.4% (National Glycohemoglobin Standardization); thyroid stimulating hormone, 3.632 µIU/ml; thyroid hormone (T4), 0.81 ng/dL; total cholesterol, 177 mg/dL; triglyceride, 146 mg/dL, high-density lipoprotein cholesterol, 56 mg/dL; and low-density lipoprotein cholesterol, 109.0 mg/dL. Her serum antinuclear antibody (ANA) titer was elevated (×160, speckled pattern). A serum specific autoantibody screen was positive for anti-U1 ribonucleoprotein (RNP) antibody (concentration, 21.7 U/mL) and PR3-ANCA (17.9 U/mL), but negative for anti-double-stranded DNA antibody, anti-Sm antibody, anti-SS-A-antibody, anti-Scl-70 antibody, myeloperoxidase (MPO)-ANCA, anti-β2-glycoprotein-I antibody, lupus anticoagulant (measured by dilute Russell’s viper venom time), rheumatoid factor, and anti-citrullinated protein antibody. Urine tests for protein and occult blood were negative. Neither white blood cell levels nor interleukin-6 levels (1.4 pg/mL) were increased in the cerebrospinal fluid. Conventional radiography of the hand revealed neither bony erosion nor joint space narrowing. Computed tomography images did not show lung mass lesions or interstitial pneumonia. A pulmonary function test did not reveal an obstructive or restrictive pattern.

An echocardiogram revealed neither pulmonary artery hypertension (estimated pulmonary artery pressure, 26.5 mmHg) nor pericardial effusion. Neck MRI did not show apparent abnormalities in the alignment of vertebrae or spinal cord. A nerve conduction velocity test revealed mild carpal tunnel syndrome in the right arm. Carotid ultrasonography showed thickness in the right bifurcation (intima media thickness, 1.4 mm).

MCTD was diagnosed based on the presence of Raynaud’s phenomenon and swelling in her fingers, detection of anti-U1-RNP antibody, leukocytopenia, and sclerodactyly, which were compatible with the Kasukawa’s criteria. Since neither granulomatous lesion nor vasculitis in organs such as lungs and kidney was found, a diagnosis of GPA was not established. No organ symptoms warranting the initiation of immunosuppressive therapy were found. Therefore, we did not initiate the administration of immunosuppressants, but instead continued careful observation at the outpatient clinic.

Discussion

Our patient was positive for not only anti-U1-RNP antibody but also PR3-ANCA. There have been several reports analyzing the associations among anti-U1-RNP antibody, ANCA, MCTD, and ANCA-associated vasculitis (AAV). Tubery et al have analyzed 11,921 samples in ten European hospitals, out of which 18 were positive for both anti-U1-RNP antibody and PR3-ANCA, and only one patient showed clinical symptoms for both MCTD and GPA. In addition, they also described that 11 of the 18 patients suffered from various autoimmune diseases.
such as rheumatoid arthritis and lupus\textsuperscript{4}. Their findings revealed that although double positivity for anti-U1-RNP antibody and PR3-ANCA is rare, such patients may show various clinical symptoms of not only MCTD and GPA but also other immunological diseases.

In our patient, brain MRI findings were not considered as a symptom of MCTD because the results of cerebrospinal fluid analysis were normal and tests for autoantibodies for antiphospholipid syndrome were negative. However, small-vessel fibrinoid necrosis was found in an autopsied MCTD case\textsuperscript{5}. Kanazawa et al have reported a case of a patient with MCTD positive for PR3-ANCA, which revealed systemic atherosclerosis, including an old infarction in the periventricular white matter, as well as multiple lacunae without traditional risk factors, such as hypertension, hyperlipidemia, DM, and smoking. Although their patient was not compatible with the WG criteria, they did not deny a possibility that multiple small cerebral infarctions were due to vasculitis\textsuperscript{5}. Based on these and our report, we should be aware of the pathophysiology of AAV which might increase damage on vessel walls when cases of MCTD affecting brain blood vessels with neither cerebrospinal inflammation nor thrombus are encountered.

Another interesting feature of our case is that the patient was complicated by SPT1DM. Schlaffke et al have analyzed the prevalence of ANCA in patients with type 1 DM. They found that 11 (12\%) out of 94 patients were positive for ANCA (nine for MPO-ANCA and two for PR3-ANCA)\textsuperscript{6}. They could not detect differences in controls and in the complications of DM between ANCA-positive and ANCA-negative patients and concluded that ANCA was not related to diabetic microangiopathy. However, the high prevalence of ANCA in type 1 DM would suggest that pathophysiology associated with the production of ANCA exists in type 1 DM. In the presented case, it is undeniable that her relatively high age and SPT1DM affected the findings of the brain lesion. Incidence of ischemic lesion on brain MRI has been reported to increase in an aged population\textsuperscript{7}, and DM is related to be an increased risk of brain infarction\textsuperscript{8} and atherosclerosis in carotid arteries\textsuperscript{9}. However, it is not clear how SPT1DM affects brain vessels. Future studies analyzing an association between atherosclerosis and SPT1DM are needed.

Chronic thyroiditis does not seem to be a rare complication of MCTD. Tomsic et al reported 7 out of 18 MCTD patients were confirmed as having chronic thyroiditis\textsuperscript{10}. Another study revealed chronic thyroiditis was found 21\% of MCTD patients\textsuperscript{11}. These data suggests physicians should be alert about chronic thyroiditis when they encounter patients with MCTD. On the other hand, the prevalence of PR3-ANCA in chronic thyroiditis is yet to be clarified and chronic thyroiditis complicating GPA is very rare: we could only one Japanese case positive for PR3-ANCA\textsuperscript{12}. Considering the relatively high prevalence of hypothyroidism in Japanese general population (1–2\%)\textsuperscript{13}, it is presumable not a few patients with chronic thyroiditis are latently positive for PR3-ANCA. Further studies are needed to clarify the precise association between PR3-ANCA and immunological thyroid abnormality.

This is the first report of MCTD positive for PR3-ANCA in a patient with SPT1DM and chronic thyroiditis. Based on a previous report\textsuperscript{1} and this report, it is suggested that synergy between MCTD and PR3-ANCA triggers atherosclerosis, which is exacerbated by SPT1DM. This is an interesting case to consider the role of autoantibodies and autoimmune diseases in the pathogenesis of atherosclerosis.

**Conflict of interests** The authors declare that they have no conflict of interests.

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