〈Case Report〉

Long-term survival with *RAS*-associated autoimmune leukoproliferative disorder with somatic *KRAS* mutation:A case report

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ABSTRACT *RAS*-associated autoimmune leukoproliferative disorder (RALD) is a recently reported rare nonmalignant autoimmune disorder. The characteristic clinical findings of RALD include monocytosis, leukocytosis, lymphoproliferation, and autoimmune phenomena. RALD is defined by somatic mutations in *KRAS* or *NRAS*. It is a new disease that was reported by Niemela and Takagi in 2011. The prognosis and incidence are currently unknown and the treatment strategy has not yet been established.

Here we describe the long-term survival of a patient with who displayed a somatic *KRAS* G12D mutation. His clinical features and labolatory data were overlapped with juvenile myelomonocytic leukemia and chronic myelomonocytic leukemia. Mercaptopurine hydrate, hydroxycarbamide and azacitizine were administered to control white blood cell count and improve clinical symptoms. He had a long survival time without hematopoietic stem cell transplantation.

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Key words : *RAS*-associated autoimmune leukoproliferative disorder, azacitizine

INTRODUCTION

RAS-associated autoimmune leukoproliferative disorder (RALD) is a chronic, non malignant condition that characterized by monocytosis, leukocytosis, lymphoproliferation, and autoimmune phenomena. RALD is a new class of monogenic autoimmune diseases[1, 2].

Patients with RALD show overlap of clinical features and laboratory data with patients with juvenile myelomonocytic leukemia (JMML), chronic myelomonocytic leukemia (CMML), and autoimmune lymphoproliferative syndrome (ALPS)[3－5].

RALD is caused by activation of *RAS*-family proteins, including *NRAS* and *KRAS*. *KRAS* is a gene that acts as an on/off switch for cell signaling. When it functions normally, it controls cell proliferation. When it is mutated, negative signaling is disrupted. Therefore, cells can continuously proliferate and often develop into cancers. The prognosis of RALD is unknown because there are very few cases[3－5].

We observed the long-term survival of a patient

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with RALD who displayed a somatic *KRAS* G12D mutation.

CASE REPORT

A 6 month-old boy presented with bilateral cervical lymphoadenopathy, splenomegaly, and moderate anemia. Bone marrow (BM) aspiration and a trephine biopsy were performed. His BM revealed normocellular marrow with evident erythrodysplasia and 2% blasts involving trisomy 8. The patient was diagnosed with myelodysplastic syndrome (MDS). His parent refused hematopoietic stem cell transplantation (HSCT), and he remained under observation with red blood cell concentrate transfusion. At the age of 13 years, he had a remarkably short stature of -5.3 SD and a low weight of -3.0 SD (Fig. 1).

He had taken mercaptopurine hydrate to control his splenomegaly since the age of 6 years, but his splenomegaly gradually worsened, and put pressure on his gastrointestinal tract. By the age of 13 years, he was unable to consume food because of the

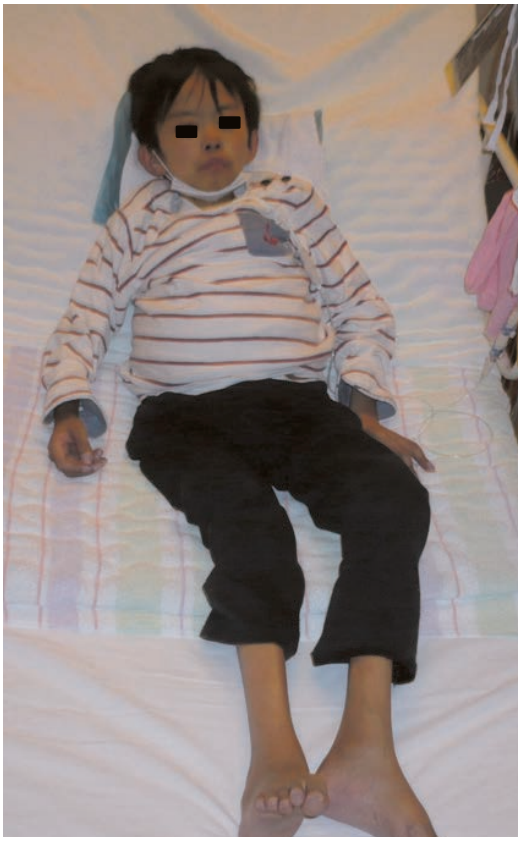

Fig. 1. Physical examination at the age of 18 years

Table 1. Laboratory findings on admission

Peripheral blood	
WBC	34,540 /μl
Neut	73 %
Lym	2 %
Mon	25 %
Eos	0 %
Baso	0 %
RBC	264×10^4 /μl
Hb	9.0 g/dl
HbF	1.0 g/dl
Ht	27.6 %
MCV	104.5 fl
MCH	34.1 pg
Plt	24.7×10^4 /μl

Coaglation	
PT	9.3 sec
APTT	33.0 sec
Fibrinogen	520 mg/dl

Blood chemistry	
TP	8.2 g/dl
Alb	2.4 g/dl
T.Bil	1.3 mg/dl
AST	39 U/l
ALT	17 U/l
LDH	470 U/l
ALP	1116 U/l
γ-GTP	333 U/l
BUN	17 mg/dl
Cr	0.21 mg/dl
UA	4.7 mg/dl
Glu	96 mg/dl
CRP	6.41 mg/dl
Na	128 mEq/l
K	2.9 mEq/l
Cl	90 mEq/l
Ca	7.8 mg/dl
P	3.2 mg/dl
IgG	2,780 mg/dl
IgA	1,398.3 mg/dl
IgM	481.1 mg/dl
ANA	28.3 (+)

Bone marrow	
NCC	28.2×10^4 /μl
Megk	69.4 /μl
Blast	0.2 %
Eo	0.6 %
Baso	0.2 %
Myeloid series	47.4 %
Mono	22.2 %
Erythroid series	15.8

Chromosome G-banding
47, XY, +8 [20/20]
Spontaneous colony (-)
GM-CSF hypersensitivity (-)

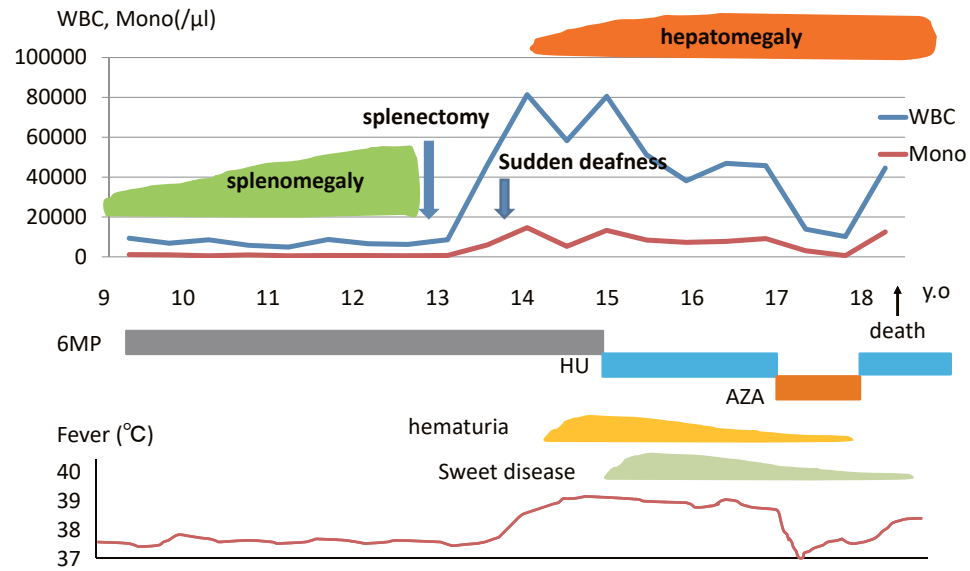

Fig. 2. Clinical course
The fever describes the peak temperature.
6MP: mercaptopurine hydrate, HU: hydroxycarbamide, AZA: azacitizine

splenomegaly. He underwent a splenectomy, and the pressure on the intestinal tract was reduced. However, a hematological examination showed a white blood cell (WBC) count of 31,300 - 121,800/μL with monocytosis (2,308 - 29,841/μL) .

His serum levels of immunoglobulin (Ig) G, IgA, and IgM were 2,780, 1,398 and 481 mg/dL, respectively. His antinuclear antibodies were mildly positive, and his HbF did not increase (Table 1).

He presented with fever, rash, and arthralgia because of the WBC count increase; hydroxycarbamide was used to control his WBC count. Rashes appeared as small red bumps on his arms, legs, neck, and trunk were associated with pain; therefore we diagnosed Sweet's syndrome. Treatment with a corticosteroid was effective, but the rashes recurred frequently. He also had deafness in his right ear at the age of 14 years, and his hearing loss was irreversible (Fig. 2). A peripheral blood smear evaluation showed absolute monocytosis with 1% circulating blast. An assessment of his BM revealed severe hyperplasia, monocytosis, and dysplasia with megaloids, micromegakaryocytes, ringed neutrophils, and hypersegmented neutrophils (Fig. 3). However, BM contained only 1% blasts involving trisomy 8. The Spontaneous colony and GM-CSF hypersensitivity assessments were negative. Therefore, we diagnosed CMML transformed from MDS.

To reduce leukocytosis and improve clinical symptoms, he was administered six cycles of azacitizine (AZA) (75 mg/m2, day1-5, each cycle every 4 weeks) at the age of 17 years. The percentage of bone marrow blasts did not change from 1% to 2% before and after AZA. All cell line did not reach normal maturation. Complete remission was not achieved, but his clinical data and symptoms improved. The white blood cell count decreased and hepatomegaly was reduced. The fever was relieved and the rash with pain disappeared (Fig. 2). Following this, a somatic *KRAS* c.35 G>A mutation was found, and he was diagnosed with

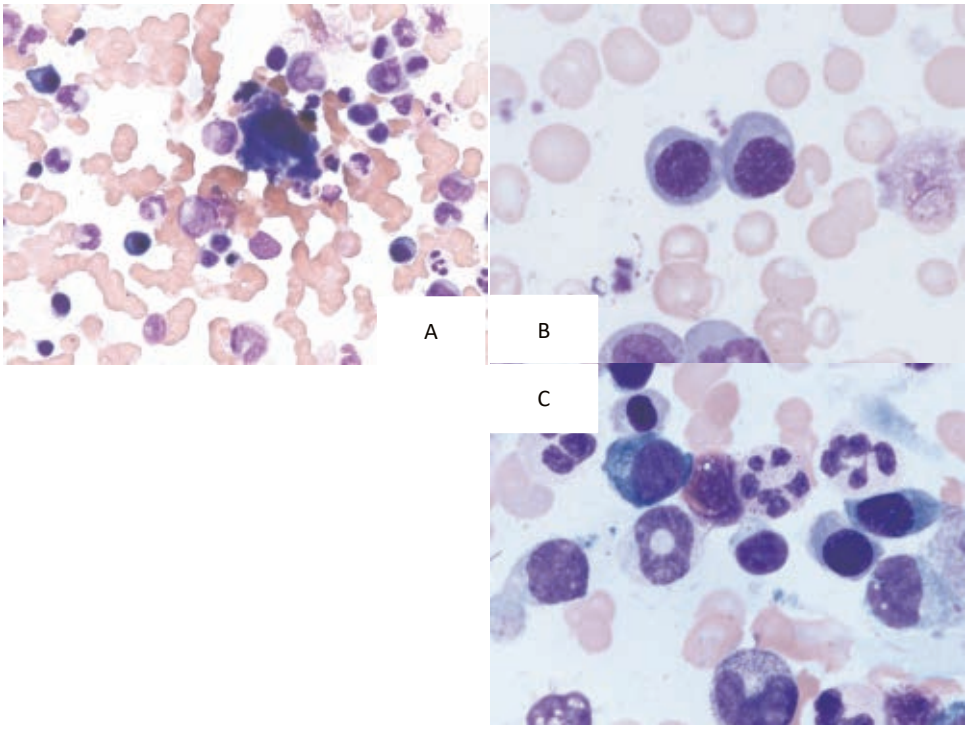

Fig. 3. Electropherogram of the *KRAS* G12D mutation in the bone marrow-CD34- positive cells

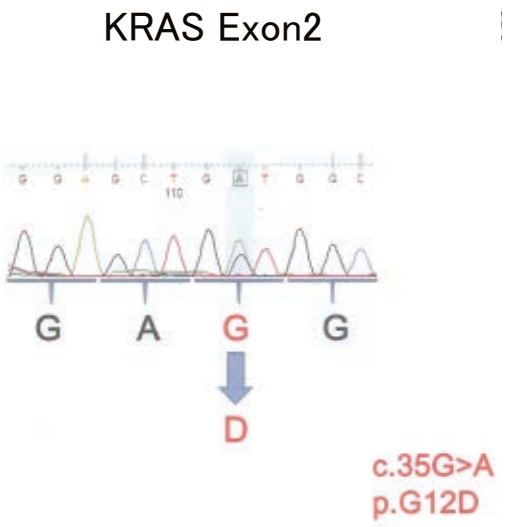

Fig. 4. (A)Bone marrow from the patient showing micromegakaryocytes (× 400)
(B) Bone marrow from the patient showing megaloids (× 1,000)
(C) Bone marrow from the patient showing hypersegmented neutrophils and ringed neutrophils (×1,000)

RALD (Fig. 4). Other JMML and ALPS related gene mutations were not detected.

The patient died due to liver failure at the age of 19 years.

DISCUSSION

RALD is a rare disease caused by a *RAS* pathway mutation. It is characterized by lymphoproliferation with an increase of CD4-CD8- double negative T cells. The clinical features includes monocytosis, leukocytosis, splenomegaly, and hypergammaglobulinemia[1, 2]. Furthermore, in some patients, this disease occurs in combination with autoimmune diseases such as systemic lupus erythematosus, Rosai-Dorfman syndrome, and Sweet's syndrome[6, 7]. In some patients, an increase in HbF and GM-CSF hypersensitivity has been confirmed. RALD is caused by the activation of *RAS*-family proteins, including *NRAS* and *KRAS*. RALD is considered a disease that shares some

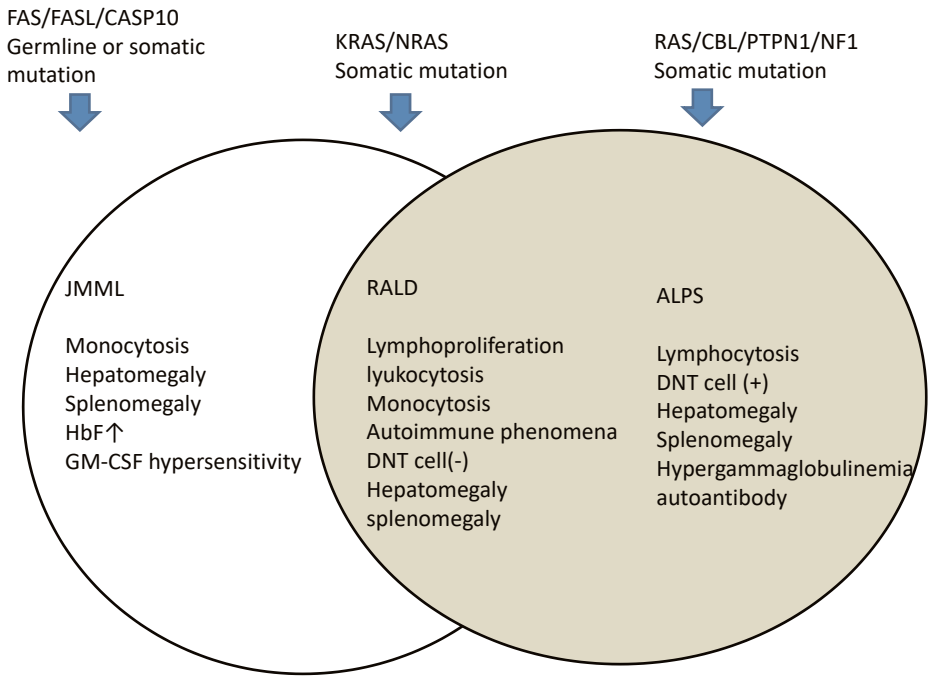

Fig. 5. Comparison of pathologies of JMML, RALD and ALPS

clinical features and molecular genetic abnormalities with other leukoproliferative diseases, such as ALPS, JMML, and CMML (Fig. 5). Several cases have been reported, but the prognosis and standard treatment remain unclear. RALD is a slow disease, but some cases have been reported with rapid progression and pulmonary infiltration[3-5].

Allogenic HSCT is the only treatment for patients with RALD. However, one opinion recommends avoiding aggressive interventions, such as HSCT, in patients with RALD without clear evidence of malignancy[5]. Toyoda et al reported that rituximab could improve the clinical response and quality of life of patients with RALD[8].

For our patient, we administered mercaptopurine hydrate, hydroxycarbamide, and AZA for improving the clinical symptoms and suppressing the WBC count before RALD was diagnosed.

Hypomethylating drugs are useful for the management of MDS and CMML[9,10]. Costa et al reported an overall response rate of 39% (14 of 36) when AZA was administered to patients with CMML[9]. The disorder did not reach remission with AZA treatment, but the condition remained stable, and some improvements in clinical symptoms were observed. Accumulation of the use experience for children is expected in the future.

Our report has several limitations. We did not estimate the TCR $\alpha\beta$+CD4-CD8- double negative T cell and conduct an apoptosis assay. Therefore, differentiation from other autoimmune diseases such as ALPS and JMML, may be difficult.

Transformation from MDS to CMML is rarely observed. After being diagnosed with MDS at the age of 6 months based on BM examination, the patient did not undergo a splenectomy until the age of 13 years because his parents did not want invasive treatment. Therefore, the evaluation of disease activity may have been insufficient.

In summary, we report the long-term survival of a patient with RALD with a somatic *KRAS* mutation. The pathological condition and prognosis of RALD

have not yet been elucidated, and further study of cases is necessary.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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