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Clinical and Genetic Characteristics of XIAP Deficiency in Japan

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Abstract Deficiency of X-linked inhibitor of apoptosis (XIAP) caused by *XIAP/BIRC4* gene mutations is an inherited immune defect recognized as X-linked lymphoproliferative syndrome type 2. This disease is mainly observed in patients with hemophagocytic lymphohistiocytosis (HLH) often associated with Epstein–Barr virus infection. We described nine Japanese patients from six unrelated families with XIAP deficiency and studied XIAP protein expression, *XIAP* gene analysis, invariant natural killer T (iNKT) cell counts, and the cytotoxic activity of $CD8^+$ alloantigen-specific cytotoxic T lymphocytes. Of the nine patients, eight patients presented with symptoms in infancy or early childhood. Five patients presented with recurrent HLH, one of whom had severe HLH and died after cord blood transplantation. One patient presented with colitis, as did another patient's maternal uncle, who died of colitis at

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4 years of age prior to diagnosis with XIAP deficiency. Interestingly, a 17-year-old patient was asymptomatic, while his younger brother suffered from recurrent HLH and EBV infection. Seven out of eight patients showed decreased XIAP protein expression. iNKT cells from patients with XIAP deficiency were significantly decreased as compared with age-matched healthy controls. These results in our Japanese cohort are compatible with previous studies, confirming the clinical characteristics of XIAP deficiency.

Keywords X-linked lymphoproliferative syndrome · X-linked inhibitor of apoptosis · Epstein–Barr virus · hemophagocytic lymphohistiocytosis · invariant natural killer T cell

Abbreviations

BIR	Baculovirus IAP repeat
CTL	Cytotoxic T lymphocyte
HSCT	Hematopoietic stem cell transplantation
HLH	Hemophagocytic lymphohistiocytosis
IAP	Inhibitor of apoptosis
LCL	Lymphoblastoid cell line
MMC	Mitomycin C
mAb	Monoclonal antibody
MFI	Mean fluorescence intensity
iNKT	Invariant natural killer T
PCR	Polymerase chain reaction
PBMC	Peripheral blood mononuclear cells
TCR	T cell receptor
XIAP	X-linked inhibitor of apoptosis
XLP	X-linked lymphoproliferative syndrome

Introduction

X-linked lymphoproliferative syndrome (XLP) is a rare inherited immunodeficiency estimated to affect approximately one in one million males, although it may be underdiagnosed [1]. XLP is characterized by extreme vulnerability to Epstein–Barr virus (EBV) infection, and the major clinical phenotypes of XLP include fulminant infectious mononucleosis (60%), lymphoproliferative disorder (30%), and dysgammaglobulinemia (30%) [2]. In addition, XLP is associated with a variety of additional clinical phenotypes such as vasculitis, aplastic anemia, and pulmonary lymphoid granulomatosis. Patients with XLP often develop more than one of these phenotypes. The gene responsible for XLP was identified as *SH2D1A*, located on Xq25 and encoding the SLAM-associated protein (SAP) [3–5]. However, gene analysis revealed *SH2D1A* mutations in only 50–60% of presumed XLP patients [6]. Importantly, a mutation in the gene that encodes the X-linked inhibitor of apoptosis (XIAP) called *XIAP* or *BIRC4* was identified as a second causative gene for XLP [7]. *XIAP* is located close to the *SH2D1A* gene on the X chromosome and consists of six coding exons [8–10]. XIAP produces an anti-apoptotic molecule that belongs to the inhibitor of apoptosis (IAP) family proteins. It contains three baculovirus IAP repeat (BIR) domains that, together with flanking residues, bind to caspases 3, 7, and 9, thereby inhibiting their proteolytic activity [11].

The clinical presentations of XIAP-deficient patients have been frequently reported [7,12,13]. More than 90% of patients with XIAP deficiency develop hemophagocytic lymphohistiocytosis (HLH) which is often recurrent. Therefore, it was recently suggested that the phenotype of XIAP deficiency fits better with the definition of familial HLH than with XLP disease [12]. However, familial HLH is characterized by defects in $CD8^+$ T and NK cell cytotoxicity responses, while these responses are normal in XIAP deficiency [7,12]. Other symptoms of XLP, such as splenomegaly, hypogammaglobulinema, and hemorrhagic colitis, have been reported in patients with XIAP deficiency, but lymphoma has never been noted [7,12–15].

We searched for patients with XIAP deficiency in Japan by detection of *XIAP* gene mutations and flow cytometric assessment of lymphoid XIAP expression. We previously reported the first case of XIAP deficiency in Japan [14]. Thereafter, we identified eight additional cases from five families with XIAP deficiency in our country. In this study, we describe the clinical and laboratory findings from nine patients from six unrelated families with XIAP deficiency, including previous cases, to help further the understanding of the pathogenetic features of this disease.

Materials and Methods

Patient and Family Member Samples

Patients without indentified *SH2D1A* mutations but with presumed XLP phenotypes were screened for *XIAP* mutations. Their family members were also screened for the same mutation. Upon identification of *XIAP* mutations, the patients were enrolled in this study. Patient 2.2 passed away before a genetic diagnosis of XIAP deficiency was made, but he was the maternal uncle of patient 2.1 and had presented with a XLP phenotype (Table I). In the end, nine patients from six different families were found to have XIAP deficiencies, three of whom had been reported previously [13,14]. Upon the approval of the Ethics Committee of the University of Toyama and after obtaining informed consent, 5–10 mL heparinized venous blood was collected from the patients, their mothers, and 25 age-matched healthy children (1–13 years of age). All of the samples were

transferred to our laboratory at room temperature within 24 h for analysis.

Mutation Analysis of the *XIAP* Gene

DNA was extracted from peripheral blood using the QuickGene-Mini 80 nucleic acid extraction system (FUJIFILM Co., Tokyo, Japan). The coding regions and the exon–intron boundaries of the *XIAP* gene were amplified by polymerase chain reaction (PCR) using primers flanking each of the six exons by standard methods. PCR products were sequenced using the BigDye Terminator Cycle Sequencing Kit (Applied Biosystems, Foster City, CA, USA) with the same primers used for PCR amplification. Sequencing analysis was performed on an Applied Biosystems Prism 310 Capillary Sequencer (Applied Biosystems).

Flow Cytometric Analysis of XIAP Protein Expression in Lymphocytes

XIAP protein expression was studied by flow cytometric techniques as previously described [16,17]. Peripheral blood mononuclear cells (PBMC) from patients 1, 2.1, 3.1, 3.2, 4, 5, 6.1, 6.2, and 25 age-matched healthy children were prepared by density gradient centrifugation over Histopaque-1077 (Sigma-Aldrich, Inc., St. Louis, MO, USA). The cells were first fixed in 1% paraformaldehyde in PBS for 30 min at room temperature and then permeabilized in 0.5% saponin in washing buffer. The fixed and permeabilized cells were then incubated with an anti-XIAP monoclonal antibody (mAb) (clone 48 (BD Biosciences, Franklin Lakes, NJ, USA) or clone 2 F1 (Abcam, Cambridge, UK)) for 20 min on ice, washed, and then incubated with a FITC-labeled anti-mouse IgG1 antibody (SouthernBiotech, Birmingham, AL, USA) for 20 min on ice. The stained cells were analyzed on the FC500 flow cytometer (Beckman Coulter, Tokyo, Japan).

Western Blot Analysis of XIAP Protein Expression in Lymphocytes

PBMC from normal controls and patients 3.1, 5, and 6.2 were washed and pelleted. The cells were then lysed in 10 μL of lysing solution (l% Triton-X 100; 150 mmol/L NaCl; 10 mmol/L Tris–HCl, pH 7.6; 5 mmol/L EDTA-Na; 2 mmol/L phenylmethylsulfonyl fluoride) per 10^6 cells for 30 min on ice. The lysed cells were centrifuged for 10 min at 15,000*g* to remove nuclei, and the supernatants were diluted in the same volume of Laemmli's sample buffer. Samples were then electrophoresed in sodium dodecyl sulfate–polyarylamide 10% to 20% gradient gel and blotted on nitrocellulose filters. Blots were blocked in 5% skim milk in PBS for 1 h, treated with anti-XIAP mAb (clone 28 or clone 2F1) for 2 h, and then incubated with peroxidase-conjugated anti-mouse IgG antibody (Invitrogen, Grand Island, NY, USA) for 1 h. Immunoblots were developed by the ECL Western blotting detection system (GE Healthcare UK Ltd., Buckinghamshire, England).

Flow Cytometric Identification of Invariant Natural Killer T Cells

PBMC from eight patients (1, 2.1, 3.1, 3.2, 4, 5, 6.1, and 6.2) and 25 controls were incubated with fluorochrome-conjugated anti-CD3 (Dako Japan KK, Kyoto, Japan), anti-TCRVα24, and anti-TCRVβ11 mAbs (Beckman Coulter) to identify invariant natural killer T (iNKT) cells by flow cytometry. After the electronic gating of 100,000 $CD3^+$ T cells, iNKT cell populations were defined by the co-expression of TCRVα24 and TCRVβ11. The iNKT cell counts were evaluated at the diagnosis of XIAP deficiency.

Establishment of Alloantigen-Specific Cytotoxic T Lymphocyte Lines and Analysis of Cytotoxic T Lymphocyte-Mediated Cytotoxity

Alloantigen-specific $CD8^+$ cytotoxic T lymphocyte (CTL) lines were generated as described previously [18,19]. Briefly, PBMC were obtained from patients 1, 2.1, 3.1, and unrelated healthy individuals. These cells were co-cultured with a mitomycin C (MMC)-treated B lymphoblastoid cell line (LCL) established from an HLA-mismatched individual (KI-LCL). Using cell isolation immunomagnetic beads (MACS beads; Miltenyi Biotec, Auburn, CA, USA), $CD8^+$ T lymphocytes were isolated from PBMC that had been stimulated with KI-LCL for 6 days. $CD8^+$ T lymphocytes were cultured in RPMI 1640 medium supplemented with 10% human serum and 10 IU/mL interleukin-2 (Roche, Mannheim, Germany) and stimulated with MMC-treated KI-LCL three times at 1-week intervals. These lymphocytes were then used as $CD8^+$ alloantigen-specific CTL lines. The cytotoxic activity of CTLs was measured by a standard ^{51}Cr-release assay as described previously [20]. Briefly, alloantigen-specific CTLs were incubated with ^{51}Cr-labeled allogeneic KI-LCL or TA-LCL, which did not share HLA antigens with KI-LCL, for 5 h at effector/target cell ratios (E/T) of 2.5:1, 5:1, and 10:1. Target cells were also added to a well containing only medium and to a well containing 0.2% Triton X-100 to determine the spontaneous and maximum levels of ^{51}Cr release, respectively. After 5 h, 0.1 mL of supernatant was collected from each well. The percentage of specific ^{51}Cr release was calculated as follows: (cpm experimental release − cpm spontaneous release) / (cpm maximal release − cpm spontaneous release)×100, where cpm indicates counts per minute.

Table I Summary of our data

	Patient 1 [13]	Patient 2.1 [12]	Patient 2.2 [12]	Patient 3.1	Patient 3.2	Patient 4	Patient 5	Patient 6.1	Patient 6.2
Age at initial presentation	20 months	7 months	3 months	2 months	Asymptomatic	2 months	6 months	17 months	15 months
Current age	4 years	Deceased	Died of colitis	12 years	17 years	15 years	2 years	1 year	12 years
Family history	No	Yes	Yes	Yes	Yes	No	No	Yes	Yes
HLH	+	+	−	+	−	−	+	+	+
Recurrent HLH	+	+	−	+	−	−	+	−	+
Fever	+	+	+	+	−	−	+	+	+
Splenomegaly	+	+	ND	−	−	−	−	+	+
Cytopenia	+	+	ND	+	−	−	+	+	+
EBV	+	−	ND	+	−	−	−	+	+
Hypogammaglobulinemia	−	+	ND	−	−	+	−	−	−
Colitis	−	−	+	−	−	−	+	−	−
Treatment	PSL CsA Dex	PSL CsA Dex	ND	PSL CsA	−	IVIG	PSL, Dex CsA, IVIG Infliximab	IVIG, Dex	PSL
Allogeneic HSCT	−	+	−	−	−	−	−	−	−
Mutation	R238X	R381X	ND	W217CfsX27	W217CfsX27	E349del	Del of exons 1-2	N341YfsX7	N341YfsX7
XIAP protein expression	±	−	ND	−	−	+	±	±	±

HLH hemophagocytic lymphohistiocytosis, *ND* no data, *EBV* Epstein–Barr virus, *PSL* prednisolone, *CsA* cyclosporin A, *Dex* dexamethasone, *IVIG* intravenous immuglobulin, *HSCT* hematopoietic stem cell transplantation, + yes or positive, − no or negative, ± residual expression

Statistical Analysis

Student's *t*-test was used for statistics, with *P*-values <0.05 considered to be statistically significant.

Results

Clinical Manifestations of the Patients

Most of our patients presented with disease symptoms at very early ages; five patients presented in infancy and three patients presented in childhood (Table I). Three of the six families had family history records. Five of the nine patients had recurrent HLH, fever, splenomegaly, and cytopenia. EBV infection and hypogammaglobulinemia were also observed in multiple patients. Most patients with HLH were treated with corticosteroids with or without cyclosporin A to prevent an otherwise rapidly fatal disease course. Patients 2.2 and 5 presented with colitis, whereas patient 2.2 died; patient 5 improved with anti-TNF alpha mAb (infliximab®) treatment. Patient 2.1 underwent cord blood transplantation but died of complications. Patient 4 had a history of recurrent otitis media and pneumonia since 2 months of age, and he was found to have hypogammaglobulinemia. The patient was treated with intravenous immunoglobulin replacement therapy alone, and he is currently doing well. No patient developed lymphoma.

Detection of *XIAP* Mutations

We identified *XIAP* mutations in patients from all six unrelated families (Fig. 1) and analyzed all of the data using the US National Center for Biotechnology Information database (http://www.ncbi.nlm.nih.gov/SNP) to check for single-nucleotide polymorphism in the *XIAP* gene. As previously reported, patient 1 possessed a nonsense mutation, 712 C > T, resulting in an early stop codon R238X [14]. Patient 2.1 had a nonsense mutation in exon 5, 1141 C > T, resulting in R381X [13]. Patient 2.2 might have the same mutation as patient 2.1 because patient 2.2 was the maternal uncle of patient 2.1 [13]. Patients 3.1 and 3.2 were siblings and were found to have a one base pair deletion (650delG) in exon 1, resulting in a frameshift and premature stop codon (W217CfsX27). Patient 4 was found to have one amino acid deletion (1045_1047delGAG; E349del) in exon 3. Patient 5 has a large deletion, spanning exons 1 and 2. Patients 6.1 and 6.2 were brothers and had a two-nucleotide deletion (1021_1022delAA), which resulted in a frameshift and premature stop codon (N341YfsX7). All of the mothers of the patients from families 1–5 were heterozygote carriers of the mutations. Interestingly, we could not find any *XIAP* mutation in the mother of patients 6.1 and 6.2. We identified deleterious *XIAP* mutations in nine patients from six unrelated Japanese families that are likely to underlie their XLP phenotypes.

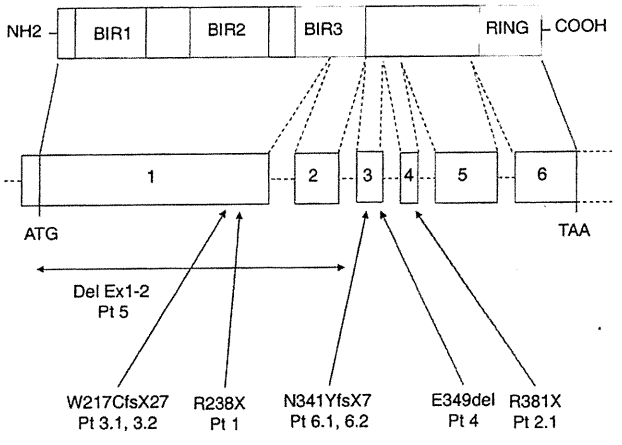

Fig. 1 *XIAP* gene mutations and their consequences for XIAP protein. *XIAP* comprises six exons and encodes the XIAP protein, which consists of 497 amino acids. XIAP contains three BIR domains and one RING domain. Mutations identified in our patients are indicated

XIAP Expression in Lymphocytes from the Patients and Carriers by Flow Cytometry

XIAP expression levels were analyzed in the lymphocytes of patients from all six families (Fig. 2). The lymphocytes of

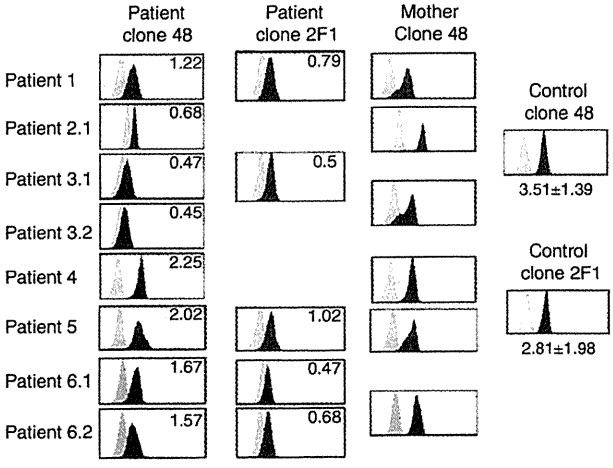

Fig. 2 XIAP protein expression in lymphocytes from the patients and their carriers. Flow cytometric detection of intracellular XIAP in lymphocytes from patients and their maternal carriers. The *gray* and *black areas* indicate the negative control and anti-XIAP staining, respectively. Anti-XIAP staining was performed using the clones 48 and 2 F1 antibodies where indicated. The *number in the box* indicates the log scale difference between the mean fluorescence intensity (ΔMFI) stained by the isotype antibody and that by the anti-XIAP antibodies. XIAP expression in 25 normal controls was also analyzed by the clone 48 and 2 F1 antibodies. The data of mean ± standard deviation of ΔMFI and each representative profile were shown

patients 1, 3.1, 5, 6.1, and 6.2 were examined by two different anti-XIAP mAbs. Using clone 48 antibody, patients 1, 2.1, 3.1, 3.2, 6.1, and 6.2 showed reduced XIAP expression, whereas XIAP was normally expressed in the lymphocytes of patients 4 and 5. In contrast to clone 48, clone 2F1 antibody showed reduced XIAP expression in patient 5. The effects of heterozygous *XIAP* mutations were studied in the lymphocytes of the patients' mothers by anti-XIAP mAb clone 48. The mothers of patients 1, 3.1, and 3.2 showed a bimodal pattern of XIAP protein (Fig. 2). The mothers of patients 2.1, 6.1, and 6.2 did not show a clear mosaic pattern, but all of these patients had reduced XIAP expression levels. Similarly to patients 4 and 5, the mothers of patients 4 and 5 demonstrated a normal XIAP expression pattern.

XIAP Expression in Lymphocytes from the Patients by Western Blot

Western blot analysis was used to evaluate the expression level of XIAP to determine the impact of patient *XIAP* mutations on protein expression and to compare this to the flow cytometric analysis. PBMCs from patients 3.1, 5.1, and 6.2 were available for Western blotting. All of these patients showed a reduction in XIAP protein expression (Fig. 3), fitting with the results obtained by flow cytometric analysis.

iNKT Cell Counts in the Patients

SAP-deficient patients had reduced numbers of NKT cells that expressed an invariantly rearranged T-cell receptor (TCR) consisting of TCRVα24 and TCRVβ11 chains [21,22]. The rare subset of iNKT cells was originally reported to be reduced in XIAP-deficient patients as well [7] but seemed to be present in normal numbers in a later study involving a larger patient cohort [23]. We analyzed the iNKT cell frequencies in 100,000 $CD3^+$ T cells in our XIAP-deficient patients and compared these with healthy controls (Fig. 4). The average frequency of iNKT cells within the $CD3^+$ T cell compartment of our XIAP patients was significantly reduced by twofold when compared with healthy

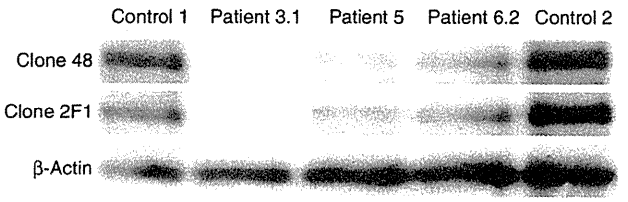

Fig. 3 XIAP expression in lymphocytes from the patients by Western blot. Analysis of XIAP expression in PBMC generated from patients with XIAP deficiency and normal controls using the antibody clone 48 (*upper panel*), the antibody clone 2 F1 (*middle panel*), and the β-actin antibody as an internal control (*lower panel*)

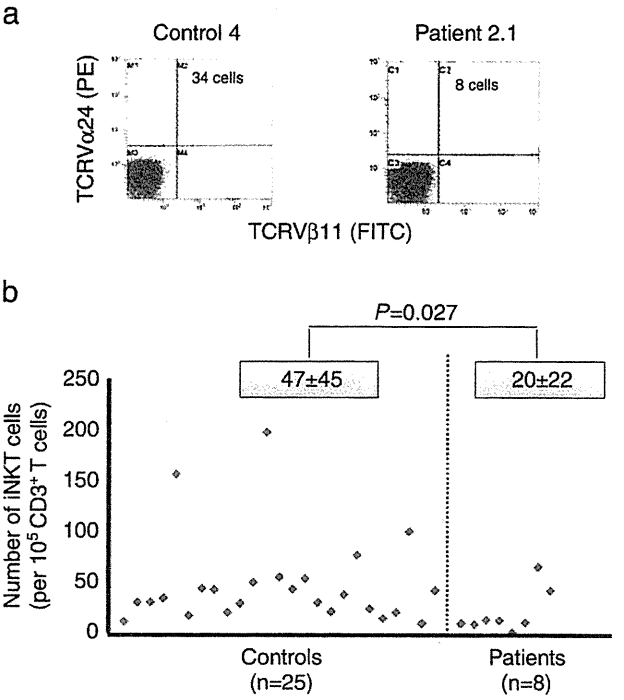

Fig. 4 iNKT cell counts in the patients and healthy controls. **a** Representative flow cytometric analysis of iNKT cells in $CD3^+$ lymphocytes from one XIAP-deficient patient and one healthy control. **b** Comparison of the number of iNKT cells in 100,000 $CD3^+$ lymphocytes between XIAP-deficient patients and control individuals. Statistical significance between patients and controls was determined with the Student's *t*-test (*p*-value=0.027)

controls (20 vs. 47 per 10^5 $CD3^+$ T cells). Therefore, we concluded that the number of iNKT cells was reduced in our patients with XIAP deficiency.

Functional Analysis of CTL Lines Established from the Patients

To test whether our XIAP-deficient patients have similar defects in $CD8^+$ T cell cytotoxicity as described in other subtypes of familial HLH [20,38], we generated $CD8^+$ alloantigen-specific CTL from patients 1, 2.1, 3.1, and three healthy controls (Fig. 5). The cytotoxic activity of the CTL of these patients was similar to that of the healthy controls, indicating that XIAP patients clearly differ from other familial HLH patients in this aspect of the disease.

Discussion

XIAP deficiency is a rare but severe and life-threatening inherited immune deficiency [12,13]. Early diagnosis and life-saving treatment such as hematopoietic stem cell transplantation is especially important. The causative gene for

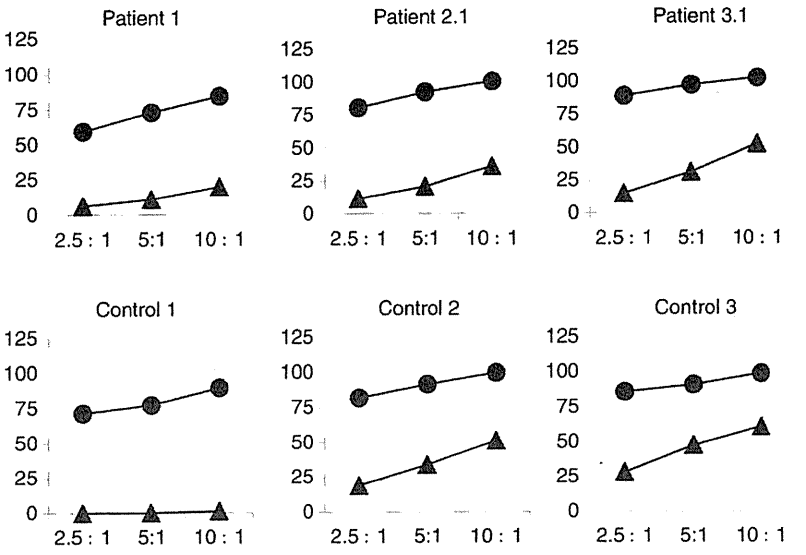

Fig. 5 Cytotoxicity of alloantigen-specific $CD8^+$ T cell lines. $CD8^+$ T cell lines were generated from PBMC of patients with XIAP deficiency and healthy controls by stimulation with allogeneic LCL (KI-LCL). Their cytotoxity was determined against allogeneic KI-LCL (*circles*) and against allogeneic TA-LCL (*triangles*), which does not share alloantigens with KI-LCL

XIAP deficiency was identified to be *XIAP/BIRC4*, and 25 mutations in the *XIAP* gene have been previously reported [7,12–14]. In the present study, we described four novel mutations (W217CfsX27, E349del, deletion of exons 1 and 2 and N341YfsX7) in the *XIAP* genes as well as previously described patients with R381X and R238X mutations [13,14]. The mother of patients 6.1 and 6.2 had no mutation in the *XIAP* gene. Because this is an X-linked inheritance, the failure to identify the same mutation in the mother suggests that the mother had a germline mosaicism for the mutation. Such mosaicism has not yet been described in XIAP deficiency, but it has been reported in Duchenne muscular dystrophy, X-linked severe combined immunodeficiency, X-linked agammaglobulinemia, and many other inherited diseases [24–26]. HLH is common in XIAP-deficient patients, and it is often recurrent [13,14]. In our study, six patients had HLH and five patients presented with recurrent HLH. Therefore, XIAP deficiency should be suspected in certain boys with HLH, especially in those with family history or recurrent HLH. The reason why XIAP deficiency increases susceptibility to HLH remains unclear. Murine studies have also failed to disclose a mechanism for the development of HLH [27]. Interestingly, Xiap-deficient mice possess normal lymphocyte apoptosis induced by a variety of means [28]. Three of our patients presented with EBV-associated HLH. EBV infection has been reported to be a trigger of the first HLH episode in patients with XIAP deficiency [13]. The excess of lymphocyte apoptosis in XIAP deficiency might account for the abnormal immune response to EBV [28]. Splenomegaly is not frequently observed in XLP type 1 or SAP deficiency but might be a common clinical feature in XIAP deficiency [12,13] as four (50%) of eight Japanese patients developed splenomegaly. Pachlopnik Schmid et al. [13] reported that recurrent splenomegaly occurring in the absence of systemic HLH was often associated with fever and cytopenia. XIAP-deficient patients are at risk for chronic colitis, which is possibly a more frequent cause of mortality than HLH [13]. Our study included two patients who developed colitis, and one of the patients died of colitis at 4 years of age. Although we did not have enough clinical information or samples from that patient because of his early death, his symptoms suggest that he had a XIAP deficiency complicated with colitis because he was the maternal uncle of patient 2.1. The other patient was 2 years old and also suffered from chronic hemorrhagic colitis.

In contrast to SAP deficiency, lymphoma has never been reported in XIAP deficiency, including our patients. Some studies indicate that the XIAP protein is a potential target for the treatment of cancer based on the anti-apoptotic function of XIAP [29]. Therefore, the absence of XIAP may protect patients from cancer, explaining why XIAP-deficient patients do not develop lymphoma. We generated a clinical summary to compare XIAP-deficient patients with the previous reports (Table II). Although our study included a relatively small number of patients, our results appear to be consistent with previous large studies [12,13] and confirm the clinical characteristics of XIAP deficiency.

Flow cytometry can be used for the rapid screening of several primary immunodeficiencies including XLP [30]. XIAP protein has been found to be expressed in various human tissues, including all hematopoietic cells [7,10]. Marsh et al. [16] described that XIAP was readily detectable in normal granulocytes, monocytes, and all lymphocyte subsets. Moreover, patients with *XIAP* mutations had decreased or absent expression of XIAP protein by flow cytometry [14,16]. We investigated XIAP expression in lymphocytes from eight patients by flow cytometry as previously described [16,17]. As demonstrated by Marsh et al. [16], clone 48 antibody provided brighter staining compared

Table II Comparison of patients with XIAP deficiency

	Marsh R et al. [12]	Pachlopnik Schmid J et al. [13]	Our study
Number of patients	10	30	9
HLH	9 (90%)	22/29 (76%)	6/9 (67%)
Recurrent HLH	6 (60%)	11/18 (61%))	5/6 (83%)
EBV-associated HLH	3 (30%)	16/19 (84%)	4/6 (67%)
Splenomegaly	9 (90%)	19/21 (90%)	4/8 (50%)
Hypogammaglobulinema	2 (20%)	8/24 (33%)	2/8 (25%)
Lymphoma	0	0	0
Colitis	0	5 (17%)	2 (22%)

to clone 2F1 antibody. In patients 5, 6.1, and 6.2, XIAP protein expression was normal when using clone 48 antibody but decreased when using clone 2F1 antibody. Western blot analysis showed XIAP expression in patients 3.1, ,5 and 6.2, and using clone 48 antibody, we found a discrepancy between flow cytometry and Western blot. Flow cytometric diagnosis may thus result in false positive results, and the gene sequencing of *XIAP* should be performed even when the patient shows normal XIAP expression levels.

All of the mothers examined in this study except for one were carriers of *XIAP* mutations. Analysis of XIAP expression in the mothers of patients 1, 3.1, and 3.2 revealed a bimodal expression pattern of XIAP in lymphocytes with cellular skewing towards expression of the wild-type XIAP allele as previously demonstrated [16]. However, the mother of patients 2.1, 6.1, and 6.2 demonstrated a normal expression pattern, possibly resulting from an extremely skewed pattern of X chromosome inactivation as shown in XIAP deficiency and other primary immunodeficiencies, and de novo mutations in *XIAP* are also observed [16,31]. The mother of patients 6.1 and 6.2 might have a germline mosaicism for the mutation, resulting in normal XIAP protein expression.

iNKT cells represent a specialized T lymphocyte subpopulation with unique features distinct from conventional T cells [32,33]. Human iNKT cells express an invariant TCR that recognizes self and microbacterial glycosphingolipid antigens presented by the major histocompatibility complex class I-like molecule CD1d [28]. The first series of XIAP-deficient patients showed decreased iNKT cell counts similar to SAP deficiency [7]. However, Xiap-deficient mice have normal numbers of iNKT cells and did not show an abnormal response to apoptotic stimuli [34]. Marsh et al. [23] reported a cohort of XIAP-deficient patients with normal numbers of iNKT cells, indicating that XIAP-deficient patients differ from SAP-deficient patients in this respect. In our cohort, we observed significantly decreased iNKT cell numbers in XIAP-deficient patients compared to healthy controls. However, we could not identify a correlation between the number of iNKT cells and the clinical disease features. Flow cytometric evaluation of iNKT cell counts can allow for the discrimination of XLP and other primary immunodeficiency diseases because patients may have normal XIAP protein expression in their lymphocytes.

CTLs kill their targets by one of two mechanisms: granule- or receptor-mediated apoptosis [35]. A recent study showed that the main pathway of cytotoxicity mediated by alloantigen-specific human $CD4^+$ and $CD8^+$ T cells is granule exocytosis and not the FAS/FAS ligand system [18]. Granzyme B is a major effector molecule of granule-mediated killing that rapidly induces cell death after entering the cytoplasm of the target cell [36]. The enzymatic activity of granzyme B is key to its ability to induce cell death. The executioner caspase-3 has been shown to be proteolytically processed and activated by granzyme B [37]. Although XIAP possesses an inhibitory effect for caspases, it is important to study the cytotoxic activities of CTLs in XIAP deficiency. Furthermore, many studies have indicated that some subtypes of patients with familial HLH show a deficiency in their cytotoxic activities [20,38]. To further investigate the function of antigen-specific CTLs, we studied $CD8^+$ alloantigen-specific CTL analysis among three XIAP-deficient patients. XIAP-deficient patients showed a normal level of cytotoxic activity, suggesting that XIAP might not play an important role in the cytotoxic responses of $CD8^+$ T cells as was previously suggested based on the normal NK cell-mediated cytotoxicity found in XIAP-deficient patients [7,12].

In this study, we have described nine Japanese patients with XIAP deficiency with clinical characteristics similar to those of patients in Europe and USA [12,13].

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制御性T細胞に異常を有する原発性免疫不全症

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免疫系には，免疫抑制機能に特化した制御性T細胞(regulatory T cell：Treg)とよばれる少数の細胞集団が存在し，自己免疫やアレルギー，炎症といった過剰な免疫反応を抑制して，免疫恒常性の維持において非常に重要な役割を果たしている．Tregのマスター遺伝子としてforkhead box P3(*FOXP3*)遺伝子が同定されて以来，Tregの発生・分化と抑制機能の分子機構が解明されつつある．Tregの異常，すなわち*FOXP3*遺伝子変異によって，ヒトではimmune dysregulation, polyendocrinopathy, enteropathy, and X-linked(IPEX)症候群を生じる．この疾患は，I型糖尿病(insulin-dependent diabetes mellitus：IDDM)や甲状腺機能低下症などの多腺性内分泌障害，難治性下痢などを主症状とし，自己免疫性と考えられる貧血，血小板減少，腎炎など多彩な症状を呈する．本稿では，IPEX症候群の臨床像ならびに分子学的異常について概説する．

はじめに

自己構成成分に対する免疫不応答，すなわち免疫寛容の維持には，自己反応性T細胞の制御が重要である．T細胞は，T細胞受容体(T cell receptor：TCR)遺伝子V(D)J断片の組換えにより，10^{15}にものぼる多様な抗原特異性をもつが，正常自己抗原に反応して自己組織の破壊を引き起こすことはない．免疫寛容には“中枢性”と“末梢性”があるが，末梢性免疫寛容機構が自己免疫疾患の発症阻止にきわめて重要である．末梢T細胞レパトア中には，自己免疫疾患を引き起こす機能的な自己反応性T細胞が存在するが，健常人においてそれらは特定の細胞集団によって抑制的に制御されている．この特定のT細胞亜集団を，制御性T細胞(regulatory T cells：Treg)と称する(**図1**)．自己免疫を抑制するTregは，インターロイキン(IL)-2受容体α鎖(CD25)陽性CD4陽性サブセット中に局在することがさまざまな実験系で明らかにされ[1]，その後，転写因子forkhead box P3(FOXP3)がその中心的役割を果たすことが明らかにされた[2)3)]．

Tregの異常がある場合には，さまざまな自己免疫疾患をきたしうると考えられるが，ヒトにおいてimmune dysregulation, polyendocrinopathy, enteropathy, and X-linked(IPEX)症候群とよばれる疾患が従来から知られていた[4]．免疫調節障害の結果として，I型糖尿病(insulin-dependent diabetes mellitus：IDDM)や甲状腺機能低下症などの多腺性内分泌障害，難治性下痢などの主症状の他に，湿疹，自己免疫性と考えられる貧血，血小板減少，腎炎など多彩な症状を呈する[5]．その名のとおりX連鎖劣性遺伝形式をとるまれな疾

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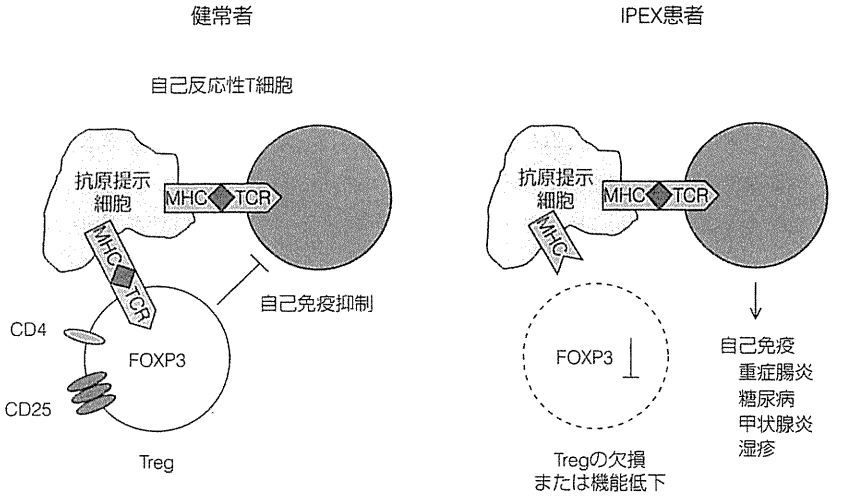

図 1. Tregによる自己反応性T細胞の抑制モデル
健常人では，FOXP3を有するCD4$^+$CD25$^+$T細胞によって自己反応性T細胞の機能が抑制され，自己免疫反応が抑制されているが，IPEX症候群患者では，*FOXP3*遺伝子変異によりTregの欠損または機能低下があり，自己反応性T細胞を抑制することができずに，重症腸炎，糖尿病，甲状腺炎，湿疹などの自己免疫が生じる．

患で，通常，新生児期もしくは乳児期早期に発症し，適切な治療がおこなわなければ，重篤な腸疾患または致死的な感染症のため乳幼児期に死亡することが多い．治療としては，免疫抑制療法が第1選択としておこなわれるが，現時点では造血幹細胞移植(hematopoietic stem cell transplantation：HSCT)が唯一の根本的治療である[6)~9)]．IPEX症候群の責任遺伝子として，染色体Xp11.23に位置する*FOXP3*遺伝子が同定された[10)]．*FOXP3*遺伝子は，ヒトでは11個のエクソンから成り立ち，431個のアミノ酸をコードする．FOXP3はforkheadファミリーに属する転写因子であり，forkhead(FKH)DNA-binding domain, leucine zipper, zinc finger-binding domainを含んでいる．現在まで約20の異なる*FOXP3*遺伝子変異が，同定されているが，多くはFKH DNA-binding domainに位置する[10)]．

ScurfyマウスはIPEX症候群のマウスモデルであり，魚鱗様皮膚病変，発育不良，貧血，血小板減少，リンパ節腫脹，肝脾腫，易感染性，下痢など他臓器にわたる自己免疫性・炎症性疾患を発症して，生後3~4週で死亡する．このscurfyマウスにおいても，*Foxp3*遺伝子変異が同定されている[11)]．

このように，Tregは末梢免疫寛容(免疫調節)の維持のために重要な役割をもつことがわかっており[2)3)]，さまざまな免疫関連疾患に深くかかわっている．数的あるいは機能的異常が自己免疫疾患やアレルギー疾患に関与していると報告があり，数的あるいは機能的な亢進が感染症や担癌患者において報告されている．

本稿では，IPEX症候群を概説することによって，Tregの機能を理解することの助けになれば幸いである．

1．IPEX症候群の臨床像

1982年にPowellら[12)]は，今日のIPEX症候群にあたる新たな疾患“an X-linked syndrome of diarrhea, polyendocrinopathy, and fatal infection in infancy”をはじめて報告した．おもな臨床症

状として，発育不全，難治性重症下痢，IDDMや甲状腺機能低下症，皮疹を認め，その他の症状として，自己免疫性溶血性貧血，血小板減少，リンパ節腫脹，肝脾腫，易感染性などが認められる．大半の患者は，3歳前に低栄養や感染症，その他の合併症によって死亡する[4)5)]．その後，この疾患の報告は散見されるが，きわめてまれな疾患であると考えられている．まとまった症例数を検討した報告は少ないが，Gambineriら[13)]がIPEX患者14例の報告をおこなっており，われわれも，わが国におけるIPEX患者6例(5家系)についてまとめた(**表1**)．

発症年齢は乳児期早期であることが多いが，年長児以降での発症も報告されている[12)]．多彩な症状を呈するため早期診断は非常に困難であると思われるが，Gambineriら[13)]は湿疹の有無にかかわらず，IgE上昇を伴った自己免疫性腸炎(難治性下痢)とIDDMの症状を認めた乳児例では，積極的にIPEX症候群を疑うべきであると述べている．

発育不全は，おもに腸疾患に伴う長期的な吸収障害にもとづくものであるが，IDDMなどの内分泌異常や長期にわたるサイトカインの過剰も影響している可能性がある．過去の報告でも，IPEX症候群患者において最も頻度の高い症状は，乳児期早期に発症する難治性下痢である．これは絨毛萎縮と広範囲にわたる小腸粘膜へのリンパ球浸潤を伴い，結果として水様性下痢や粘血便を呈し，時にイレウスが合併することもある．ミルク変更や食事摂取を中止して，中心静脈栄養にしても下痢は持続することが多い．小腸粘膜生検では，絨

表 1. わが国のIPEX患者における臨床的特徴と免疫学的特徴

患者 no.	1	2	3	4	5	6
発症年齢	日齢5	2ヵ月	日齢19	2ヵ月	4ヵ月	6ヵ月
現在年齢	19歳	10歳	18歳	7歳	5歳	26歳
身長	74.7 cm (−16.5 SD)	116.5 cm (−3.7 SD)	157.1 cm (−2.4 SD)	109 cm (−2.6 SD)	100 cm (−2.2 SD)	157.2 cm (−2.3 SD)
体重	9.15 kg	26.2 kg	44.6 kg	21 kg	14.7 kg	48.95 kg
家族歴	あり	あり	あり	なし	あり	あり
遺伝子変異	227delT (L76fsx53)	Ala384Thr (1150G>A)	Ala384Thr (1150G>A)	Phe373Val (1117T>G)	748delAAG (ΔK250)	IVS1+1A>T
初発症状	自己免疫性 甲状腺炎	アトピー性皮膚炎 哺乳不良	好酸球性胃腸炎	自己免疫性腸炎	糖尿病	糖尿病
その他の 症状	溶血性貧血 腸炎 尿細管障害 骨粗鬆症	気管支喘息 副腎不全 アトピー	アトピー 全身脱毛	なし	ミルクアレルギー アトピー ネフローゼ	ネフローゼ 腸炎
治療	タクロリムス ステロイド	ステロイド ステロイド吸入	ステロイド外用 抗アレルギー剤	免疫グロブリン シクロスポリンA 同種移植	シクロスポリンA ステロイド 同種移植	シクロスポリンA ステロイド

患者2，3は同胞例．患者4，5は同種骨髄移植後．SD：standard deviation(標準偏差)

毛の萎縮，粘膜びらん，粘膜下層・粘膜固有層へのリンパ球浸潤が認められる．血清中に抗 enterocyte 抗体が証明されている症例が多く，その抗原の1つとして，自己免疫性腸症関連 75 kDa 抗原（autoimmune enteropathy-related 75 kDa antigen：AIE-75）が同定されている[14]．その後，抗 AIE-75 抗体は，下痢を呈した IPEX 12 例全例で陽性であったことが確認されている[15]．

多腺性内分泌障害として，IDDM や甲状腺機能低下症が高頻度で認められる．とくに新生児期や乳児期に発症する IDDM は，IPEX 症候群の早期診断には重要な症状であり，インスリンによる血糖コントロールも困難であることが多い．膵臓や甲状腺組織にリンパ球の浸潤が認められ，抗膵島細胞抗体，抗甲状腺ミクロソーム抗体などが検出され，自己免疫の機序によって生じ，臓器の形成不全によるものではない．IPEX 症候群では，副甲状腺や副腎機能異常はまれで，副腎機能低下症が過去に1例報告されているのみである．

IPEX 症候群で上記2つにつづいて多い症状は，皮疹である．落屑性皮疹，アトピー性皮膚炎などを呈するが，最も一般的であるのは，全身性湿疹である．重篤な湿疹患者では，全身脱毛を伴った症例もある．皮膚炎の病理学的な特徴は，リンパ球浸潤である．

IPEX 症候群のその他の症状として，クームス陽性の溶血性貧血，自己免疫性血小板減少や好中球減少を認め，血清中で特異抗体がしばしば検出される．腎疾患は患者の約 1/3 程度に認められ，大半が間質性腎炎であるが，軽微な蛋白尿や血尿を呈する症例や急速進行性糸球体腎炎を合併する症例も存在する．自己免疫性肝炎は約 20％の患者に認められる．その他肝脾腫，リンパ節腫脹や重症感染症を合併する．とくに敗血症，髄膜炎，肺炎，化膿性関節炎などが報告されており，治療に使用される免疫抑制薬が原因と考えられる症例もあるが，免疫抑制薬未使用例にも易感染性がみられる．免疫異常が存在する可能性はあるが，自己免疫性好中球減少症に伴うものや，皮疹や腸炎に関連して病原体が侵入しやすいこと，低栄養状態なども易感染性に関与していると考えられる．病原菌として，*Enterococcus*，*Staphylococcus*，cytomegalovirus，candida が多い[4]．

2．IPEX 症候群の検査所見

IPEX 症候群患者の血液検査では，血清 IgE，IgA の上昇，好酸球増多を認めるが，末梢血リンパ球の CD4/CD8 比は正常で，phytohemagglutinin や pokeweed mitogen，concanavalin A に対するリンパ球幼若化反応も正常である．また *FOXP3* 遺伝子変異を有する大半の患者では，フローサイトメトリーにて FOXP3 を発現している $CD4^+CD25^+$ T 細胞（Treg）は著しく減少しており，診断に有用である（**図 2**）[16]．

3．IPEX 症候群の病態

FOXP3 遺伝子は，Treg の発生・分化や機能をつかさどるマスター遺伝子と考えられている．Treg が自己反応性 T 細胞の活性化，増殖を抑制することで，能動的自己寛容を維持している．この Treg は胸線で分化し，*FOXP3* 遺伝子を発現していることが特徴である．*FOXP3* 遺伝子は，他の $CD4^+$ T 細胞や $CD8^+$ T 細胞にはほとんど発現していない．IPEX 症候群は，*FOXP3* 遺伝子異常のため Treg の機能が獲得できず，能動的自己寛容に破綻をきたし，臓器特異的自己免疫疾患，アレルギー疾患，炎症性疾患など，さまざまな免疫異常による症状を呈する．保因者では X 染色体の不活化はランダムであり，*FOXP3* 遺伝子異常がある T 細胞は，*FOXP3* 遺伝子が正常に機能している Treg によって制御されていると考えられる．また近年では，*FOXP3* 遺伝子変異を認めない IPEX-like syndrome が報告されており，プロモーター領域やエンハンサー領域など翻訳領域以外の変異や *FOXP3* 関連遺伝子による変異の可能性が示唆されている[17]．

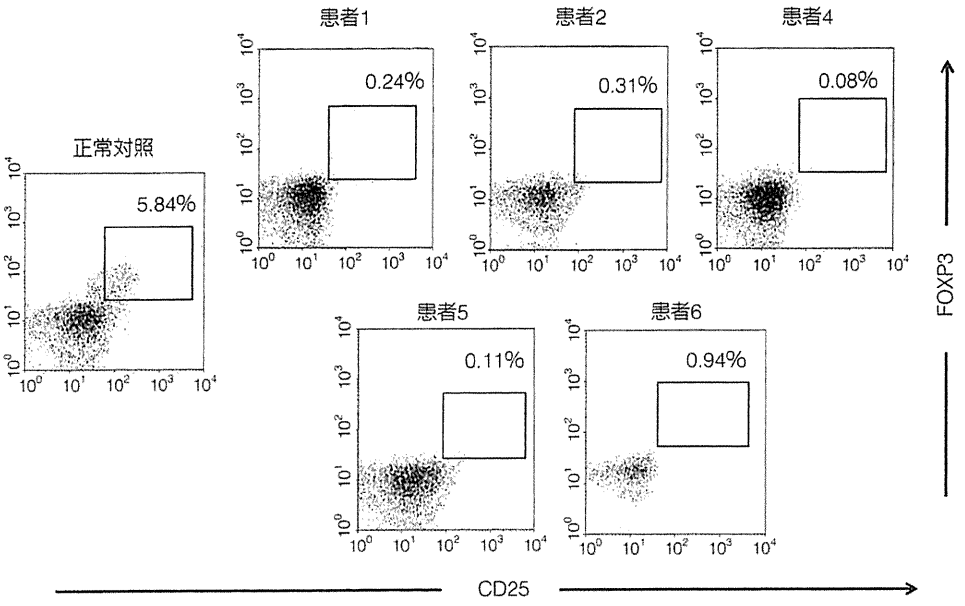

図 2. わが国の IPEX 症候群患者における FOXP3$^+$CD4$^+$CD25$^+$T 細胞
末梢血単核球を固定し，細胞膜透過性を高めた後に，抗 FOXP3，CD4，CD25 抗体による 3 重染色をおこない，CD4$^+$T 細胞における FOXP3 と CD25 の発現を評価した．FOXP3$^+$CD4$^+$CD25$^+$T 細胞は，健常人では約 5%認められるが，IPEX 症候群患者ではほとんど認められない．

4．IPEX 症候群の genotype

IPEX 症候群の詳細な分子機能はいまだ不明である．現在までに約 20 の遺伝子変異が報告されており，ミスセンス，ナンセンス，挿入，欠失，スプライス異常などが報告され，FKH DNA-binding domain に変異が集中しているが，その他の部位にも変異は散在している[5)13)]．現在のところ，遺伝子変異部位による表現型の違いは明らかではなく，またまったく同じ部位の変異であっても，臨床症状や重症度が異なる症例が報告されており，FOXP3 mRNA の発現量の違いやそれ以外の因子が関与している可能性がある．

5．IPEX 症候群の治療

早期診断が，最終的な予後を決定する重要な因子である．症状に応じて，輸血やインスリン投与，経静脈栄養などをおこなうが，IPEX 症候群患者では，種々の抗原に対してアレルギー反応を呈するので注意が必要である．予防接種や分枝鎖アミノ酸補液などによるアレルギー反応に関連した死亡例の報告もある．免疫抑制薬は一定の患者には有効であるが，一般的には効果は限定的であり，長期間の寛解状態を得ることは困難で，重症感染症を引き起こす可能性がある．シクロスポリン A やタクロリムスがステロイドと併用で使用されることが多いが，その他にはメトトレキサート，インフリキシマブ，リツキシマブも使用されている．近年海外では，sirolimus が腎機能障害も少ないため使用されているが，わが国では入手困難である．Sirolimus はエフェクター T 細胞の増加を抑制し，Treg を増加させるという報告もあり，効果に期待がもたれる[18)19)]．しかし IPEX 症候群患者において，唯一有効な治療法は HSCT である．近年では，HSCT が施行され完全寛解に至っている症例報告も認められ，また治療強度を弱めた前処置（reduced-intensity conditioning：RIC）で施行された成功例も散見される[20)21)]．しかし，移植

方法は未確立で，治療成績の詳細も明らかではない．また，IPEX 症候群の移植では生着不全が起こりやすい．この生着不全は，宿主の Treg が機能しないためドナー抗原に対する反応性 T 細胞が除去されないことから，拒絶を受けやすいのかもしれない．IPEX 症候群患者に HSCT が施行されるようになり，移植後の FOXP3$^+$Treg 再構築の報告も散見されるようになった[20)21)]．過去の報告によると，FOXP3$^+$Treg の数が正常化する時期は 1～18ヵ月と幅があり，一定していない．FOXP3$^+$Treg の回復は，過去の治療，移植細胞数，前処置，移植片対宿主病(graft versus host disease：GVHD)予防薬など，さまざまな因子が影響している可能性がある．移植後，長期間経過観察されている症例や，FOXP3 発現維持についての報告はなく，今後も RIC をはじめとする移植方法について，更なる症例の積み重ねが必要と考えられる．

おわりに

FOXP3$^+$Treg は，免疫恒常性の維持にとって本質的な機能を担っており，この Treg のマスター遺伝子である *FOXP3* 遺伝子の発見により，その発生・分化と抑制機能の分子機構に対する理解が急速に進んでいる．そしてこの Treg の解析はまた，IPEX 症候群患者の病態の解明に寄与した．一方で，*FOXP3* 遺伝子異常の見つからない IPEX 症候群様の疾患も存在しており，これらの原因解明や IPEX 症候群に対する HSCT や遺伝子治療などの治療法の確立のためにも，更なる症例の蓄積，検討が望まれる．

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X連鎖リンパ増殖症候群

——SAP欠損症とXIAP欠損症

X-linked lymphoproliferative syndrome

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◎X連鎖リンパ増殖症候群(XLP)は，Epstein-Barrウイルス(EBV)に対する特異的免疫応答の欠陥を有する先天性免疫不全症であり，致死的伝染性単核症，異常γ-グロブリン血症，悪性リンパ腫を臨床的三徴とする．ほとんどの症例(XLPタイプ1)はSLAM-associated protein(SAP)をコードする*SH2D1A*遺伝子変異によって生じるが，XLPタイプ2と称される患者でX-linked inhibitor of apoptosis(XIAP)をコードする*XIAP*遺伝子変異が明らかとなった．XLP患者ならびに*Sap*ノックアウトマウスの観察から，SAPの欠損によりナチュラルキラーおよびCD8^{+}T細胞による細胞障害活性，T細胞からのサイトカイン産生，活性化によって誘発される細胞死，胚中心の形成，ナチュラルキラーT細胞の産生が障害されることが明らかとなったが，XIAPの欠損による分子病態はまだ明らかではない．SAP欠損症とXIAP欠損症の臨床像ならびに分子病態を明らかにすることによって，EBV特異的のみならず普遍的な免疫応答の異常が明らかになると思われる．

Key word：X連鎖リンパ増殖症候群(XLP)，Epstein-Barrウイルス(EBV)，免疫不全症，SLAM-associated protein(SAP)，X-linked inhibitor of apoptosis protein(XIAP)

X連鎖リンパ増殖症候群(X-linked lymphoproliferative syndrome：XLP)は比較的まれな先天性免疫不全症であり，致死的伝染性単核症(fatal infectious mononucleosis：FIM)，異常γ-グロブリン血症，悪性リンパ腫を臨床的三徴とする[1,2]．1970年代にはじめてこの症候群が認識され[3]，1998年にその原因遺伝子*SH2D1A*が報告され[4-6]，2006年に第2の原因遺伝子として*XIAP*が報告された[7]．それぞれの遺伝子はSLAM-associated protein(SAP)ならびにX-linked inhibitor of apoptosis(XIAP)をコードし，XLPはタイプ1(SAP欠損症)とタイプ2(XIAP欠損症)に分けられる．

本稿ではそれぞれの臨床的・分子病態学的特徴の違いについて述べることによって，XLPに対する理解を深めてもらいたい．

臨床的特徴

乳児期にEpstein-Barrウイルス(Epstein-Barr virus：EBV)感染を契機に発症することが多いが，興味深いことにほとんどの患者ではEBV感染前はまったく健康である[1,2]．しかし，EBV感染後にはしばしばTまたはB細胞増殖症をきたし，いわゆる血球貪食性リンパ組織球症(hemophagocytic lymphohistiocytosis：HLH)を発症する．異常γ-グロブリン血症や悪性リンパ腫もよく認められるが，これらはEBV陰性の患者でも認められる(表1)．その他，再生不良性貧血，リンパ性血管炎，肺リンパ様肉芽腫症や腸炎，乾癬などの自己免疫疾患を呈することもある．以前は非常に予後不良な疾患であったが，疾患の認識と診断技術の向上で予後は改善してきている．唯一の根治療法は造血幹細胞移植である．

表 1　SAP欠損症とXIAP欠損症の特徴[2]

特徴	SAP 欠損症	XIAP 欠損症
臨床像		
HLH	あり	あり
低γ-グロブリン血症	あり	あり
悪性リンパ腫	あり	なし
再生不良性貧血	あり	なし
血管炎	あり	なし
遺伝子		
原因遺伝子	*SH2D1A*	*XIAP*
局在	Xq25	Xq25
コードする蛋白	SAP	XIAP
免疫機能		
NK 細胞障害活性	低下	正常
NKT 細胞数	欠損	低下～さまざま
再活性化による細胞死	低下	亢進
メモリー B 細胞数	低下	報告なし
治療		
HLH	免疫抑制療法	免疫抑制療法
	化学療法	化学療法
	rituximab を考慮	rituximab を考慮
液性免疫不全	IgG 補充	IgG 補充
悪性リンパ腫	標準的化学療法	
根治療法	造血幹細胞移植	造血幹細胞移植

HLH：hemophagocytic lymphohistiocytosis(血球貪食性リンパ組織球症).

原因遺伝子

XLP の原因遺伝子は 1998 年に 3 つのグループから独立して報告され[4-6]，*SH2D1A* と同定され，これまで 70 以上のさまざまなタイプの遺伝子変異が報告されている．ほとんどの変異で SAP 蛋白の発現低下または欠損を示すが，遺伝子変異の種類と疾患の重症度には相関関係はない．

SAP 蛋白は 128 個のアミノ酸からなり，1 個の SH2 ドメインを有する(図 1)．SAP は T 細胞，ナチュラルキラー(natural killer：NK)細胞，NKT 細胞に発現するが，B 細胞にはほとんど発現していない．SAP は SLAM に限らず，SLAM ファミリーに属するレセプターにおけるシグナル伝達にかかわっている．

2006 年に *SH2D1A* 変異を認めない XLP を呈する 3 家系 12 例の患者で *XIAP* 変異が同定された[7]．興味深いことに *XIAP* は *SH2D1A* のきわめて近傍に局在する．*XIAP* 変異は *SH2D1A* 変異と同様に機能喪失をきたし，XIAP 蛋白の発現は低下または欠損する．*XIAP* がコードする XIAP は inhibitor of apotosis(IAP)ファミリーに属する．XIAP は 3 つの baculovirus IAP repeat(BIR)ドメインからなり，カスパーゼとの相互作用にかかわる．一方，C 末端に存在する Ring ドメインは E3 ユビキチンリガーゼ活性を有する．XIAP はすべての血液担当細胞に存在する．SAP も XIAP もその欠損により EBV 関連 HLH を発症するが，XIAP 欠損症における臨床的特徴は SAP 欠損症とは完全にオーバーラップしていない[8,9]．

SAPシグナルとXLPの病態

SAP は最初 SLAM レセプターの細胞内ドメインに結合する蛋白として同定され，このレセプターの細胞内シグナルを制御するものと考えられた[6]．その後，構造的に SLAM と相同性を有する SLAM ファミリーレセプターのシグナルにもかかわることが明らかとなってきた[10]．SLAM ファミリーレセプターには 2B4(CD244；SLAMF4)，NTB-A(SLAMF6)，CRACC(CD319；SLAMF7)，CD84(SLAMF5)，Ly9(CD229；SLAMF3)が存在する．これらのレセプターは，基本的には細胞外の 2 個の免疫グロブリン様ドメイン，膜貫通部，すくなくとも 2 個のチロシン残基を含む細胞内テールから構成されている．チロシン残基が SAP と相互