

Histopathological and Clinical Features of Methotrexate-Associated Lymphoproliferative Disorders and Post-Transplant Lymphoproliferative Disorders

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ABSTRACT

Objective: To study histopathological and clinical features of methotrexate-associated lymphoproliferative disorders (MTX-LPD) and post-transplant lymphoproliferative disorders (PTLD).

Material and Methods: A retrospective study on 30 cases of MTX-LPD and 2 cases of PTLD from 2006 to 2021.

Results: By histopathology, the MTX-LPD group had 21 cases of lymphoma (MTX-Lymphoma) and 9 cases of reactive changes (MTX-Reactive). The PTLD group included diffuse large B-cell lymphoma and polymorphic PTLD (1 case each). The distinctive findings in MTX-Lymphoma and PTLD were association with Epstein-Barr virus (EBV) (8/12 cases, 66.7%) and CD30 positivity (13/18 cases, 72.2%). The MTX-LPD group had median MTX dosage of 10 mg/week, median MTX cumulative dosage of 2,613.75 mg, and median duration of MTX usage of 2,186 days. The 14 MTX-LPD and PTLD patients has median duration to response after the varied interventions of 47 days and the time to the first complete remission (CR) of 126 days. The MTX-Reactive patients had a significantly higher absolute lymphocyte count, younger median age, fewer B symptoms, higher rate of single site involvement, less extranodal involvement, shorter duration to response, less time to enter CR, and higher CR rate than the MTX-Lymphoma patients ($p < 0.05$).

Conclusion: Histopathology in MTX-LPD and PTLD patients can vary from reactive changes to lymphoma. EBV study and CD30 immunostaining help identify MTX-LPD and PTLD. History of MTX usage and other causes of immunodeficiency should be considered before diagnosing lymphoma. MTX discontinuation or reduction of immunosuppressant dosage are recommended before administering combined chemotherapeutic agents in unresponsive cases.

Keywords: Methotrexate-associated lymphoproliferative disorders; post-transplant lymphoproliferative disorders; other iatrogenic immunodeficiency-associated lymphoproliferative disorders (Siriraj Med J 2022; 74: 575-589)

INTRODUCTION

Methotrexate (MTX) is an anti-metabolite of folic acid, used as an anti-neoplastic drug in many neoplasms (including malignant lymphoma) and as an immunosuppressive drug for autoimmune diseases.¹

But MTX has been associated with lymphoproliferative disorders (LPD) secondary to immunosuppression.²⁻⁴ The revised 4th edition of the World Health Organization (WHO) Classification of Tumours of Haematopoietic and Lymphoid Tissues (WHO-HAEM4R) recognizes 4 types

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of immunodeficiency-associated LPD: post-transplant lymphoproliferative disorder (PTLD), lymphomas associated with HIV infection, lymphoproliferations associated with primary immune disorders, and other iatrogenic immunodeficiency-associated LPD that includes MTX-associated LPD (MTX-LPD).⁵ Recently, the 5th edition of the WHO classification of lymphoid tumours (WHO-HAEM5) has been introduced as a review article since 22 June 2022 proposing a three-part nomenclature for LPD and lymphomas arising in the setting of immune deficiency/dysregulation: 1) histological diagnosis (hyperplasia, polymorphic LPD, mucocutaneous ulcer, or lymphoma classified as for immunocompetent patients); 2) viral association (EBV or KSHV/HHV8); and 3) immune deficiency/dysregulation setting (inborn error or immunity, HIV infection, posttransplant, autoimmune disease, iatrogenic/therapy-related, or immune senescence).⁶

The risk and type of MTX-LPD are varied upon the type of immunosuppressive agents used in addition to MTX, the degree of immune deficiency, and the nature of the underlying disorder being treated. Even though the disease is being more recognized now, there are no definitive diagnostic criteria and histologic categories. The diagnosis is based solely on LPD found in patients with a history of MTX usage who recover spontaneously after MTX discontinuation.^{2,3} On the contrary, MTX-LPD patients with aggressive clinical courses at times fail to regress after MTX discontinuation.⁷ It is unclear when MTX-LPD patients should receive chemotherapy after MTX discontinuation and the response may differ depending on the histopathology of MTX-LPD. Recently, The EBV real-time PCR test value in the peripheral blood⁸ and programmed cell death-ligand 1 (PD-L1) expression in the lymphoma cells⁹ were associated with spontaneous regression in MTX-LPD. Moreover, MTX-LPD patients previously treated with tumor necrosis factor-alpha or Janus kinase (JAK) inhibitors prior to MTX-LPD onset had more aggressive disease.¹⁰

This study aimed to investigate the histopathological and clinical features of MTX-LPD and PTLD, specifically of any distinct features to help pathologists identify these cases without knowing any clinical data, to clarify the differences in characteristics between MTX-LPD subcategories, and to identify MTX-LPD requiring chemotherapy upfront.

MATERIALS AND METHODS

Patients and tissue samples

This study was approved by the Institutional Review Board, Faculty of Medicine Siriraj Hospital, Mahidol University (SIRB) (Si 408/2021). For the MTX-LPD

group, we compared the list of 14,139 patients with MTX treatment at Siriraj Hospital during January 2006 and May 2021 from the Department of Pharmacy and the Siriraj Informatics and Data Innovation Center to the archive list in the pathology laboratory information system (LIS) to identify the patients with a history of MTX usage and the histopathology of LPD. For the PTLD group, we searched for the cases diagnosed in the pathology LIS during the same period. We found MTX-LPD patients with lymphoma diagnosis (MTX-Lymphoma) 32 cases, MTX-LPD patients with reactive changes (MTX-Reactive) 21 cases, and PTLD patients 7 cases. The cases that lacked hematoxylin and eosin (H&E) staining and/or immunostained slides were excluded. Finally, we identified MTX-Lymphoma patients 21 cases, MTX-Reactive patients 9 cases, and PTLD patients 2 cases.

Patients were defined as having primary autoimmune disease by the information in the medical records. We defined MTX-LPD in patients receiving MTX at the time of diagnosis of LPD. In patients with underlying rheumatoid arthritis (RA), we evaluated the disease activity score in 28 joints count with 3 variables (DAS28-3) based on clinical and laboratory data 6 months prior to LPD diagnosis, which included the number of tender joints and number of swollen joints out of 28 joints, erythrocyte sedimentation rate (ESR), or C-reactive protein (CRP) calculated by <http://www.das-score.nl>.¹¹ All slides were re-evaluated by two pathologists (PN and SS) to record histopathologic features as listed in Table 1 and the disease diagnosis was made according to the WHO-HAEM4R.⁵

Immunohistochemical study (IHC) and Epstein-Barr virus-encoded small RNA (EBER) in situ hybridization (ISH)

The immunohistochemical study and EBV ISH were performed in an automated staining machine (The Ventana BenchMark XT automated slide-staining system, Tucson, AZ, USA). The IHC panels were listed in Table 2. Immunostained slides and the EBER ISH slides were re-evaluated by two pathologists (PN and SS).

Statistical analysis

Continuous demographic data were presented as the median and range. Categorical demographic data were presented as a percentage. Statistical analysis was performed to investigate the associations between the clinical and laboratory variables and the diagnosis of MTX-LPD by using Fisher's exact test or Mann-Whitney tests, as appropriate (SPSS Statistics, v. 18.0; SPSS, Inc.,

TABLE 1. List of histopathologic features recorded for evaluation in the study.

Histopathologic features	How to record
Residual lymphoid follicle	presence or absence
Tissue necrosis	presence or absence
Vascular invasion	presence or absence
Sclerosis	partial fibrous septa or absence
Histiocytic aggregation	presence or absence
Polymorphic reaction:	
Neutrophil	presence of ≥ 1 cell/HPF*
Eosinophil	presence of ≥ 1 cell/HPF
Plasma cells	presence of ≥ 2 cell/HPF
Hodgkin-Reed-Sternberg-like (HRS-like) cells	presence or absence

* HPF: high power microscopic field images were obtained under microscopy using a 40x objective lens with a field area of 0.159 mm².⁵

Chicago, IL, USA). A p-value < 0.05 was considered to be statistically significant. The cumulative 1-year and 5-year overall survival (OS) rates were analyzed using survival curves and were estimated by the Kaplan-Meier method and compared using log-rank tests.

RESULTS

Patient characteristics

In the 21 cases of the MTX-Lymphoma group, there were RA 17 cases, systemic lupus erythematosus 1 case, idiopathic exfoliative dermatitis 1 case, juvenile idiopathic arthritis 1 case, and idiopathic orbital inflammation 1 case. In the 9 cases of the MTX- Reactive group, there were RA 5 cases, antiphospholipid syndrome 1 case, ankylosing spondylitis 1 case, psoriasis 1 case, and Behçet disease 1 case.

Comparison of the clinical characteristics of the MTX-Lymphoma and MTX-Reactive patients (Table 3) showed that the MTX-Reactive patients had a significantly higher absolute lymphocyte count, younger median age, fewer B symptoms, a higher rate of single site involvement, less extranodal involvement, shorter duration to response, less time to CR, and a higher CR rate than the MTX-Lymphoma patients (p < 0.05).

Among the RA patients were DAS28-3 ESR available in 18 patients (13 MTX-Lymphoma and 5 MTX-Reactive) and DAS28-3 CRP available in 2 MTX-Lymphoma.⁷ Interestingly, 5 MTX-Lymphoma patients had low RA

disease activity (DAS28-3 CRP < 3.2) while 8 MTX-Lymphoma patients and 5 MTX-Reactive patients had moderate RA disease activity (DAS28-3 CRP > 3.2 but < 5.1) (Table 3).

The median MTX dosage at diagnosis of MTX-Lymphoma (7.5 mg/week) was lower than that of MTX-reactive (10 mg/week) (p = 0.071) but the median MTX cumulative dosage at LPD diagnosis of MTX-Lymphoma (2,880 mg) was higher than that of MTX-Reactive (665 mg) (p = 0.167). The median MTX usage duration at LPD diagnosis of MTX-Lymphoma (2,440 days) was longer than that of MTX-Reactive (410 days) (p = 0.288) (Table 3).

Of the 2 PTLD patients, one was a 59-year-old man when PTLD first developed. He had received a living-related kidney transplant for his idiopathic end-stage renal disease 8 years before PTLD diagnosis. He had received cyclosporin (125 mg/day), prednisolone (5 mg/day), and mycophenolic acid (720 mg/day) as immunosuppressants. His EBV PCR titer was less than 5,000 copies/ μ L and his anti-EBV IgG was more than 1:320. The other case was a 16-year-old man when PTLD first developed. He had received matched-unrelated donor hematopoietic stem cell transplantation for his aplastic anemia 90 days before PTLD diagnosis. He only received mycophenolate mofetil as an immunosuppressant at 40 mg/kg/day. His EBV PCR titer was less than 5,000 copies/ μ L and his anti-EBV IgG was 1:80.

TABLE 2. Immunohistochemical study panels used in the study.

Marker	Positive cell/other remark	Source	Antibody clone	Dilution
CD2	T-cell	BioGenex	AB75	1:200
CD3	T-cell	Novocastra	LN10	1:600
CD4	T-cell, helper	Ventana	SP35	RTU
CD5	T-cell	Cell Marque	4C7	1:100
CD7	T-cell	DAKO	CBC.37	1:100
CD8	T-cell, cytotoxic	Cell Marque	C8/144B	RTU
CD10	GCB	Novocastra	56C6	1:300
CD19	B-cell	Genova	LE-CD19	1:300
CD20	B-cell	DAKO	L26	1:2,000
CD21	Follicular dendritic cell	Cell Marque	2G9	1:100
CD23	Follicular dendritic cell	Cell Marque	MRQ-57	1:300
CD30	Activated lymphoid cell	Cell Marque	Ber-H2	1:100
CD56	NK cell	Cell Marque	123C3.D5	1:50
CD79a	B-cell	DAKO	JCB117	1:150
CD138	Plasma cell	DAKO	MI15	1:300
PD1	T-cell, follicular helper	Cell Marque	NAT105	1:50
CXCL13	T-cell, follicular helper	R&D	53610	1:300
βF1	T-cell receptor-beta	Thermo Scientific	8A3	1:20
γ-TCR	T-cell receptor-gamma	Thermo Scientific	γ3.20	1:20
TIA1	CGAP	Biocare	TIA-1	1:500
PAX5	B-cell	Cell Marque	SP34	1:200
c-myc	MYC transcription protein	Biocare	Y69	1:200
BCL2	Anti-apoptotic protein	DAKO	124	1:200
BCL6	GCB	Novocastra	LN22	1:200
MUM1	Plasma cell; non-GBC	DAKO	MUM1p	1:300
cyclin D1	MCL cell	Thermo Scientific	SP4	1:200
SOX11	MCL cell	Cell Marque	MRQ58	1:100
kappa	Plasma cell	Cell Marque	L1C1	1:1000
lambda	Plasma cell	Cell Marque	Lamb14	1:2000
TdT	Lymphoblast	Novocastra	SEN28	1:50
Ki-67	Proliferation index	DAKO	MIB1	1:300
LMP-1	EBV LMP	DAKO	CS1-4	1:150

Abbreviations: CGAP, Cytotoxic granule-associated protein; EBV LMP, EBV latent membrane protein; GCB, Germinal center B-cell; MCL, Mantle cell lymphoma; RTU, Ready to use

TABLE 3. Comparison of the clinical characteristics of MTX-Lymphoma and MTX-Reactive patients.

Parameter	MTX-Reactive (n = 9)	MTX-Lymphoma (n = 21)	P-value
Age in years, median (range)	47 (19–72)	62 (10–86)	0.02
Sex, n (%)			
Male	4 (44.4)	7 (33.3)	0.687
Female	5 (55.6)	14 (66.7)	
Presenting symptoms ^a , n (%)			
Lymphadenopathy	7 (77.8)	12 (57.1)	0.419
Mass or nodule	0 (0)	6 (28.6)	0.141
Fever	0 (0)	3 (14.3)	0.534
Incidental findings	2 (22.2)	1 (4.8)	0.207
Others	0 (0)	4 (19)	0.287
B symptoms, n (%)	0 (0)	14 (66.7)	0.001
Focality			
Single	6 (66.7)	2 (9.5)	0.003
Multiple	3 (33.3)	19 (90.5)	
DAS28-3 ESR, median (range), n	3.64 (3.27–4.78), 5/18	3.59 (2.53–4.69), 13/18	0.424
DAS28-3 CRP, median (range), n	NA	3.245 (2.09–4.40), 2/2	NA
DAS28-3 ESR & CRP, median (range)	3.64 (3.27–4.78)	3.59 (2.09–4.69)	0.424
Absolute lymphocyte count in cells/ μl, median (range)	1,938.30 (573.40–4,757.85)	780.80 (207.48–8,472.70)	0.028
Duration from primary autoimmune disease to LPD in days, median (range)	3,962 (79–4,787)	3,068 (385–7,421)	0.572
Extranodal involvement ^b , n (%)	0 (0)	11 (52.4)	0.011
Bone marrow involvement, n (%)	(n = 2) 0 (0)	(n = 20) 4 (20)	1.000
MTX dosage at Dx in mg/ week, median (range)	10.0 (7.5–20.0)	7.5 (2.5–17.5)	0.071
Duration of MTX usage in days, median (range)	410 (7–4,681)	2,440 (43–6,742)	0.288
MTX cumulative dosage at LPD Dx in mg, median (range)	665 (10–6,802.5)	2,880 (110–14,650)	0.167
Duration to response in days, median (range)	0 (0–329)	(n = 17) 65 (0–196)	0.024
Time to CR in days, median (range)	0 (0–485)	(n = 10) 177 (64–774)	0.013
Treatment response, n (%)		(n = 18)	
Complete remission	8 (100)	10 (55.6)	0.031
Not CR	0 (0)	8 (44.4)	
Relapse, n (%)	0 (0)	1 (4.8)	
1-year overall survival, n (%)	9 (100)	(n = 16) 10 (62.5)	0.057
5-year overall survival, n (%)	1 (100)	(n = 8) 2 (25.0)	0.333

DAS28-3, disease activity score 28-joint count–3 variables.

^asome patients had more than one presenting symptoms.^blung, liver, spleen, kidney, salivary gland, lacrimal gland, and brain.

Histopathology and immunohistochemistry

The histopathology findings and immunohistochemistry results are demonstrated in [Table 4](#). The histopathology in the MTX-Lymphoma group (21 cases) included B-cell lymphoma 16 cases (76.2%), T-cell lymphoma 3 cases (14.3%), classic Hodgkin lymphoma (CHL) 1 case (4.8%) and EBV-associated LPD (EBV+ LPD) 1 case (4.8%). The most common histological pattern was DLBCL (8 cases, 38.1%). The other types included peripheral T-cell lymphoma, not otherwise specified (PTCL, NOS) (3 cases, 14.3%), marginal zone lymphoma (MZL)/mucosa-associated lymphoid tissue lymphoma (2 cases, 9.5%), and one each (4.8%) for CHL, lymphomatoid granulomatosis (LYG), intravascular large B-cell lymphoma (IVL), mantle cell lymphoma (MCL), B-lymphoblastic lymphoma (B-LBL) and EBV+ LPD. The last 2 cases could be diagnosed as unclassifiable B-cell lymphoma (case no. 15 & 16) because they lacked comprehensive immunostaining for a definite diagnosis.

Other histopathologic findings found in the lesions were as follows: residual follicles in 4 of 21 cases (19%), tissue necrosis in 8 (38.1%), vascular invasion in 4 (19%), sclerosis in 6 (28.6%), histiocytic aggregation in 3 (14.3%), HRS-like cell in 9 (42.9%), plasma cell reaction in 6 (28.6%), eosinophil reaction in 3 (14.3%), and neutrophil reaction in 1 (4.8%). The majority of lesional cells were large atypical lymphoid cells with varying numbers of admixed inflammatory cells among the lesional cells. The distinctive ancillary findings in MTX-Lymphoma and PTLD were EBV study and CD30 positivity. We found positive EBV LMP-1 (3 of 5 cases, 60%), positive EBER ISH (8 of 12 cases, 66.7%) and CD30+ cells (13 of 18 cases, 72.2%).

The histopathology in the MTX-Reactive group (9 cases) included mixed paracortical and follicular hyperplasia 5 cases (55.6%), follicular hyperplasia 2 cases (22.2%), paracortical hyperplasia 1 case (11.1%) and florid follicular hyperplasia 1 case (11.1%).

The histopathology in the 2 cases of PTLD included DLBCL 1 case and polymorphic PTLD 1 case.

Bone marrow status

Twenty MTX-LPD patients underwent bone marrow biopsy for staging. Two of them (10%) had diffuse involvement, another two (10%) minimal involvement by scattered lesional cells, and the remaining 16 cases (80%) negative marrow staging. One of the 2 PTLD patients had minimal involvement by scattered lesional cells and the other negative marrow staging.

Treatment and survival outcome

Details of the treatments and outcomes of the

MTX-Lymphoma, MTX-Reactive, and PTLD cases are demonstrated in [Table 5](#). The median follow-up was 394 days.

Only 6 of the 21 MTX-Lymphoma patients (28.6%) discontinued MTX as the first-line management (cases no. 1, 13, 15, 17, 20, and 21). Only 2 cases had spontaneous regression (cases no. 1 and 17) while 3 cases died (cases no. 13, 15, and 20; case no. 15 with progressive disease and case no. 20 with septic shock), and the other lost to follow-up (case no. 21). Among the cases that responded to MTX discontinuation alone, case no. 1 with histopathology of DLBCL had complete remission (CR) at 64 days after MTX discontinuation. Case no. 17 with histopathology of PTCL, NOS had partial remission at 20 days after MTX discontinuation; then computed tomography (CT) at 383 days after MTX discontinuation confirmed CR. Case no. 16 with unclassifiable B-cell lymphoma received prednisolone at the time of MTX discontinuation but lost to follow-up for 2 years but upon return to the hospital, physical examination showed no lymphadenopathy.

In this retrospective study, many patients did not have full records for the exact duration of the response after intervention, so the duration of the response shown in [Table 3](#) refers to the evaluation after diagnosis. The median duration to response in 27 combined MTX-LPD and PTLD patients was 47 days (range, 0-329 days). The median duration to first CR in 20 combined MTX-LPD and PTLD patients was 126 days (range, 0-774 days).

Ten of the 21 MTX-Lymphoma patients (47.6%) discontinued MTX and simultaneously received combined chemotherapeutic agents (CMT) as the first-line management (cases no. 2-7, 9, 14, 18, and 19), 6 cases are still alive at the time of the report writing (5 without disease in cases no. 4, 5, 7, 9, and 14; while case no. 2 received only 1 cycle of R-CHOP and developed congestive heart failure but still alive with disease). Three cases died due to septic shock (cases no. 3, 6, and 18) and the other case lost to follow-up at 104 days after receiving the first-line management (case no. 19). There were 6 cases that achieved CR after MTX discontinuation and simultaneous CMT, including DLBCL 4 cases (cases no. 4, 5, 6, and 7), MZL 1 case (case no. 9), and B-LBL 1 case (case no. 14). The time to CR ranged from 110 to 344 days, with a median of 163 days. Two MTX-Lymphoma patients developed relapse of the disease (cases no. 6 and 9) based on examination of the bone marrow and lymph node, respectively. Case no. 6 had progressive disease after relapse and died. Case no. 9, however, could achieve CR after relapse and is still alive without disease.

Two of the 21 MTX-Lymphoma patients (9.5%) received only surgery as the first-line management without discontinuation of MTX or any CMT (cases no. 8 and 10).

TABLE 4. Histopathology and immunohistochemistry of MTX-Lymphoma, MTX-Reactive, and PTLD patients.

Case No.	Histopathology	Residual follicles	Tumor Necrosis	Vascular invasion	Sclerosis	Histiocytes aggregation	Polymorphic reaction	HRS-like cell	Tumor cell size	EBV-LMP-1	EBER	CD30
MTX-Lymphoma												
1	DLBCL	-	-	-	-	-	-	+	L	+	+	+
2	DLBCL	-	-	+	-	-	PC	-	M to L	-	-	+
3	DLBCL	-	-	-	P	-	-	-	M, L	-	-	-
4	DLBCL	+	-	-	P	+	Eo	+	M to L	NA	NA	+
5	DLBCL	-	+	-	-	+	-	+	L	NA	-	+
6	DLBCL	-	-	-	-	-	-	+	M, L	NA	NA	-
7	DLBCL	-	+	-	-	-	-	-	M, L	NA	NA	-
8	DLBCL	-	-	-	-	-	-	+	L	NA	NA	+
9	MZL	+	-	-	P	-	PC, Eo	-	S	NA	NA	NA
10	MALT	+	-	-	P	-	PC	-	S, few L	NA	NA	NA
11	LYG	-	+	+	-	+	Neu	+	L	NA	+	+
12	IVL	-	-	-	-	-	-	-	L	NA	NA	NA
13	MCL	-	-	-	-	-	-	-	S to M	NA	NA	NA
14	B-LBL	-	-	-	P	-	-	-	M	NA	NA	NA
15	B-Lym	-	-	-	-	-	-	-	M to L	NA	NA	+
16	B-Lym	-	+	-	-	+	-	+	M	NA	+	+
17	PTCL, NOS	+	+	-	-	-	PC	-	S to M	NA	+	+
18	PTCL, NOS	-	+	+	-	+	PC	+	S, M, L	NA	+	+
19	PTCL, NOS	-	-	-	-	-	PC	-	S, M, L	NA	NA	-
20	CHL	-	+	-	P	+	Eo	+	S, L	+	+	+
21	EBV+LPD	-	+	+	-	+	-	-	S to M, few L	NA	+	+

TABLE 4. Histopathology and immunohistochemistry of MTX-Lymphoma, MTX-Reactive, and PTLD patients. (Continued)

Case No.	Histopathology	Residual follicles	Tumor Necrosis	Vascular invasion	Sclerosis	Histiocytes aggregation	Polymorphic reaction	HRS-like cell	Tumor cell size	EBV-LMP-1	EBER	CD30
PTLD												
22	DLBCL	-	-	-	-	-	-	-	M to L	NA	-	-
23	Poly-PTLD	-	+	-	-	-	-	-	S to M, few L	+	+	+
MTX-Reactive												
24	F+PH	NA	NA	NA	-	-	-	-	NA	NA	NA	NA
25	F+PH	NA	NA	NA	-	-	-	-	NA	NA	NA	NA
26	F+PH	NA	NA	NA	-	-	-	-	NA	NA	NA	NA
27	F+PH	NA	NA	NA	-	-	-	-	NA	NA	NA	NA
28	F+PH	NA	NA	NA	-	-	-	-	NA	NA	NA	NA
29	F	NA	NA	NA	-	-	-	-	NA	NA	NA	NA
30	F	NA	NA	NA	P	-	-	-	NA	NA	NA	NA
31	PH	NA	NA	NA	-	-	-	-	NA	NA	NA	NA
32	F*	NA	NA	NA	-	-	-	-	NA	NA	NA	NA

*, focal positive; -, negative; +, positive

Abbreviations: B-LBL, B-lymphoblastic lymphoma/leukemia; B-Lym, B-cell lymphoma, unclassifiable; CHL, classic Hodgkin lymphoma; DLBCL, diffuse large B-cell lymphoma; EBER, Epstein–Barr virus-encoded small RNA in situ hybridization; EBV, Epstein–Barr virus; EBV LMP-1, Epstein–Barr virus latent membrane protein-1; Eo, eosinophil; F, follicular hyperplasia; F*, florid follicular hyperplasia; HRS, Hodgkin–Reed–Sternberg; IVL, intravascular large B-cell lymphoma; L, large; LYG, lymphomatoid granulomatosis; M, medium; MALT, mucosa-associated lymphoid tissue lymphoma; MCL, mantle cell lymphoma; MZL, marginal zone lymphoma; NA, not available; Neu, neutrophil; P, partial fibrous septa; PH, paracortical hyperplasia; PC, plasma cell; Poly-PTLD, polymorphic post-transplant lymphoproliferative disorder; PTCL, NOS, peripheral T-cell lymphoma, not otherwise specified; S, small.

TABLE 5. Diagnosis, treatment, and outcome of MTX-Lymphoma, MTX-Reactive, and PTL D patients.

Case no. Age/Sex	Primary immune disease	Immuno- modulator	Biopsy site	Diagnosis	Focality	Extranodal involvement site	Clinical stage	IPI Score	1 st -Line management	Duration to response (days)	Relapse	Outcome	Time to CR (days)	Mortality
1 68/F	RA	MTX, CQ	LN	DLBCL	Multiple	No	II	1	Off MTX	64	No	CR	64	Alive
2 58/F	RA	MTX, CQ	LN	DLBCL	Multiple	Kidney	III	1	Off MTX + R-CHOPx1	65	No	PR	NA	Alive
3 62/M	RA	MTX, Rituximab	Retroperi- toneal mass	DLBCL	Multiple	No	III	4	Off MTX + R-CHOPx6	90	No	PR	NA	Dead, Septic shock
4 70/M	RA	MTX, CQ, SSZ, LFM	LN	DLBCL	Multiple	No	II	2	Off MTX + R-CHOPx6	158	No	CR	158	Alive
5 71/F	RA	MTX, CQ	LN	DLBCL	Multiple	No	II	2	Off MTX + R-CHOPx6	40	No	CR	191	Alive
6 56/F	SLE	MTX, CQ	LN	DLBCL	Multiple	BM Liver	IV	3	Off MTX + R-CHOPx8	110	BM	CR	110	Dead, Septic shock
7 60/F	RA	MTX, CQ, AZA	Tonsil	DLBCL	Multiple	No	III	2	Off MTX + R-CHOPx8	163	No	CR	163	Alive
8 86/F	RA	MTX	Lung	DLBCL	Multiple	Lungs	IV	2	Lobectomy	0	No	CR	402	Alive
9 49/F	RA	MTX, LFM	Salivary gland	MZL	Multiple	Salivary gland	III	2	Off MTX + CVPx6+ CHOPx8	112	No	CR	344	Alive
10 69/M	IOL	MTX	Lacrimal gland	MALT	Single	Lacrimal gland	I	1	Lateral orbitotomy	0	No	Unknown	NA	Unknown
11 58/M	RA	MTX, SSZ, LFM	LN	LYG	Multiple	Lung NP	IV	2	PSL	120	No	PR	NA	Alive
12 75/F	RA	MTX	Skin	IVL	Multiple	Skin	IV	3	BSC	NA	No	Unknown	NA	Unknown

TABLE 5. Diagnosis, treatment, and outcome of MTX-Lymphoma, MTX-Reactive, and PTLT patients. (Continued)

Case no. Age/Sex	Primary immune disease	Immuno- modulator	Biopsy site	Diagnosis	Focality	Extranodal involvement site	Clinical stage	IPI Score	1 st -Line management	Duration to response (days)	Relapse	Outcome	Time to CR (days)	Mortality
13 63/M	RA	MTX, CQ	BM	MCL	Multiple	Spleen	IV	3	Off MTX	NA	No	Unknown	NA	Death
14 10/F	JIA	MTX	Left orbital mass	B-LBL	Multiple	BM	IV	1	Off MTX + ThaiPOG-ALL	0	No	CR	142	Alive
15 54/M	RA	MTX, CQ	NP	B-Lym	Multiple	No	III	2	Off MTX	NA	No	PD	NA	Dead, progress
16 76/F	RA	MTX, CQ	LN	B-Lym	Multiple	No	III	4	Off MTX + PSL	NA	No	CR	774	Alive
17 62/M	RA	MTX, CQ	LN	PTCL, NOS	Multiple	No	III	3	Off MTX	20	No	CR	383	Alive
18 74/F	RA	MTX, CQ	LN	PTCL, NOS	Single	No	I	2	Off MTX + mini-CHOPx1	14	No	PR	NA	Dead, Septic shock
19 55/F	IED	MTX	LN	PTCL, NOS	Multiple	BM Liver	IV	3	Off MTX + CHOPx3	55	No	PR	NA	Unknown
20 69/F	RA	MTX, CQ	Liver	CHL	Multiple	Spleen Liver	IV	4	Off MTX	196	No	PR	NA	Dead, Septic shock
21 62/F	RA	MTX	Lung	EBV+LPD	Multiple	Lung Pleura	IV	1	Off MTX	78	No	PR	NA	Unknown
22 59/M	LRKT	CS, PSL, MA	Tonsil	DLBCL	Multiple	BM Spleen	IV	2	RI + CHOPx6	56	Tonsil	CR	71	Alive
23 16/M	MUD- HSCT	MM	LN	Poly-LPD	Single	No	I	NA	RI + Rituximabx4	NA	No	Unknown	NA	Dead, Septic shock
24 32/F	APS	MTX, CQ	LN	RL	Multiple	No	NA	NA	Off MTX	329	No	CR	329	Alive

TABLE 5. Diagnosis, treatment, and outcome of MTX-Lymphoma, MTX-Reactive, and PTLD patients. (Continued)

Case no. Age/Sex	Primary immune disease	Immuno- modulator	Biopsy site	Diagnosis	Focality	Extranodal involvement site	Clinical stage	IPI Score	1 st -Line management	Duration to response (days)	Relapse	Outcome	Time to CR (days)	Mortality
25 19/M	Behçet's disease	MTX, PSL, HCQ	LN	RL	Single	No	NA	NA	F/U	0	No	CR	0	Alive
26 47/F	RA	MTX, CQ	LN	RL	Single	No	NA	NA	F/U	0	No	CR	0	Unknown
27 67/M	RA	MTX, CQ	LN	RL	Multiple	No	NA	NA	F/U	47	No	CR	47	Alive
28 72/F	RA	MTX, CQ, LFM	LN	RL	Single	No	NA	NA	F/U	0	No	CR	0	Alive
29 47/M	RA	MTX, CQ, LFM	LN	RL	Multiple	No	NA	NA	Off MTX	14	No	CR	485	Alive
30 32/F	Psoriasis	MTX	LN	RL	Single	No	NA	NA	F/U	0	No	CR	0	Unknown
31 56/F	RA	MTX, PSL, SSZ, LFM	LN	RL	Single	No	NA	NA	F/U	0	No	CR	0	Alive
32 56/M	SpA	MTX, SSZ	LN	RL	Single	No	NA	NA	F/U	0	No	CR	0	Alive

Abbreviations: APS, anti-phospholipid syndrome; AZA, azathioprine; B-LBL, B-lymphoblastic lymphoma/leukemia; B-Lym, B-cell lymphoma, unclassifiable; BM, bone marrow; BSC, best supportive care; CHL, classic Hodgkin lymphoma; CQ, chloroquine; CR, complete remission; CS, cyclosporin; F/U, follow-up; DLBCL, diffuse large B-cell lymphoma; HCQ, hydroxychloroquine; IED, idiopathic exfoliative dermatitis; IOL, idiopathic orbital inflammation; IPI, international prognostic index; IVL, intravascular large B-cell lymphoma; JIA, juvenile idiopathic arthritis; LFM, Leflunomide; LN, lymph node; LRKT, living-related kidney transplant; LYG, lymphomatoid granulomatosis; MALT, mucosa-associated lymphoid tissue lymphoma; MCL, mantle cell lymphoma; MA, mycophenolic acid; MM, mycophenolate mofetil; MTX, methotrexate; MUD-HSCT, match-unrelated hematopoietic stem cell transplant; MZL, marginal zone lymphoma; NA, not available; Not CR = not in complete remission (PD, PR or SD); NP, nasopharynx; PD, progressive disease; Poly-PTLD, polymorphic posttransplant lymphoproliferative disorder; PR, partial remission; PSL, prednisolone; PTCL, NOS, peripheral T-cell lymphoma, not otherwise specified; RA, rheumatoid arthritis; RI, reduction of immunosuppressants; RL, reactive lymphadenitis; SD, stable disease; SLE, systemic lupus erythematosus; SpA, ankylosing spondylitis; SSZ, sulfasalazine; ThaiPOG-ALL, Thai pediatric oncology group treatment protocol for acute lymphoblastic leukemia.

Case no. 8 (aged 86 years) refused to receive any further treatment after surgery due to concerns about the side effects from CMT. The patient achieved CR 402 days after surgery as evaluated by CT and is still alive without detectable disease at 1,638 days after surgery. Case no. 10 lost to follow-up at 151 days after surgery before receiving any CMT according to the plan of treatment.

Of the 2 PTLD patients, the patient with DLBCL (case no. 22) achieved CR after immunosuppressants discontinuation and the completion of 6 cycles of CHOP. One year later, the patient had relapsed disease at the tonsils and received DA-EPOCH regimen for 6 cycles with rituximab for 4 courses. The patient achieved CR and is still alive. The patient with polymorphic PTLD (case no. 23) received a decreased dose of immunosuppressants and 4 courses of rituximab as the first-line management. Unfortunately, the patient died of a superimposed infection.

Of the 9 MTX-Reactive patients, only 2 cases discontinued MTX as the first-line management (cases no. 24 and 29); the others continued receiving MTX, but

2 of these 7 cases lost to follow-up. The 2 cases with MTX discontinuation and the other 5 cases with continued MTX had spontaneous regressions of the lymph node. The MTX-Reactive group tended to have a better survival rate when compared with the MTX-Lymphoma group, but without statistical significance ($p = 0.065$), and the PTLD group, with statistical significance ($p = 0.034$). The MTX-Lymphoma group tended to have better 1-year and 5-year survival rates than the PTLD group, but without statistical significance ($p = 0.685$) (Fig 1).

DISCUSSION

MTX-LPD is still not very well understood in terms of its definition, pathogenesis, prognosis, and treatment, but there are studies available in the literature to help us better understand and handle the disease. MTX-LPD is highly heterogeneous with various clinical presentations, histopathology, and disease progression. Our retrospective study categorized MTX-LPD according to histopathology and found that DLBCL was the most common pattern

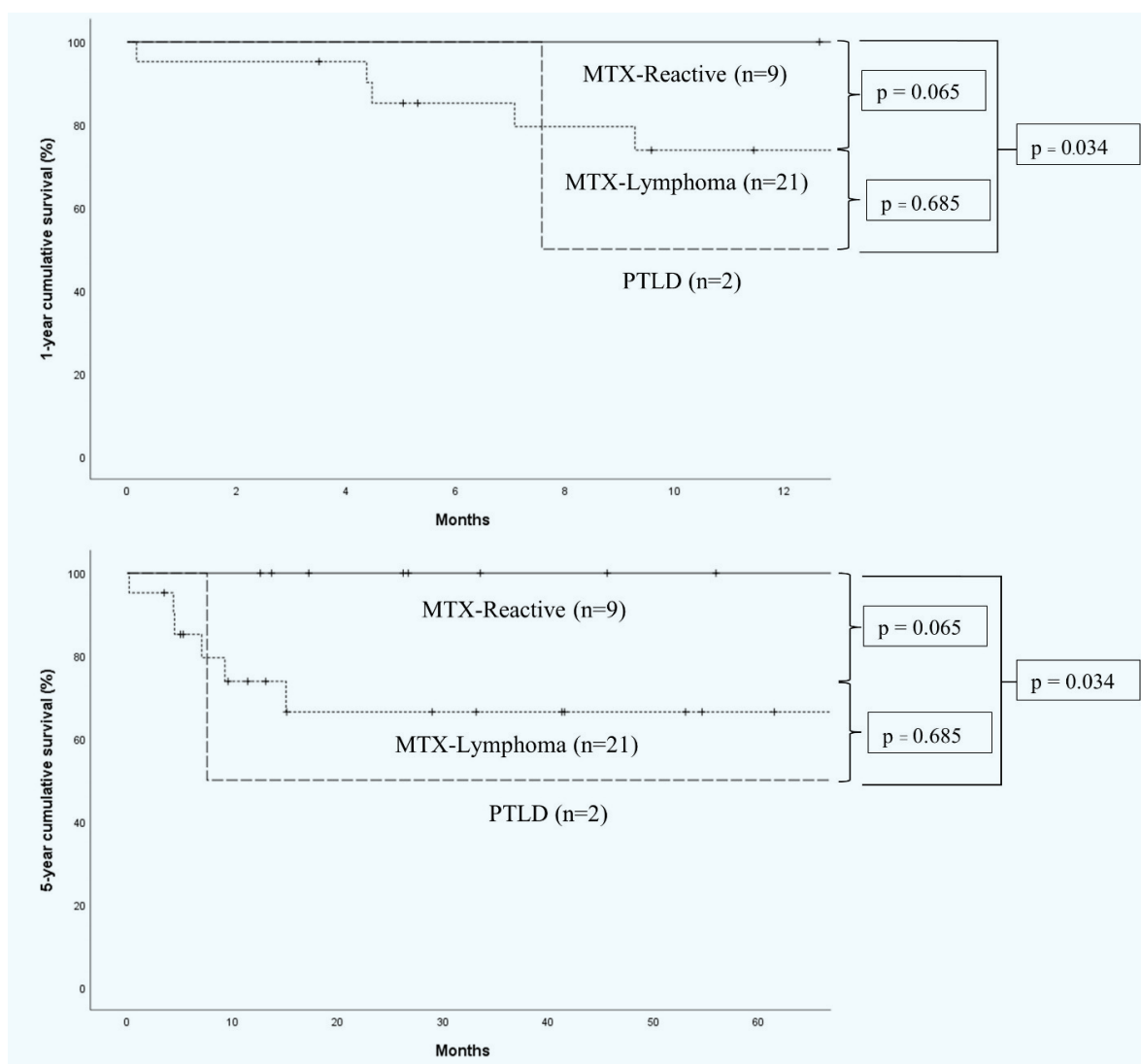


Fig 1. One-year and five-year cumulative survivals between patients of MTX-Reactive, MTX-Lymphoma, and PTLD.

in the MTX-Lymphoma group in concordance with previous reports,^{2-4,12-16} but PTCL, NOS in our study was higher than in other reports; the reason is not known. Moreover, we encountered various kinds of the MTX-Lymphoma group, not only the common entities like DLBCL, PTCL, NOS, or CHL, but also MZL, MCL, and uncommon entities, like LYG and IVL.

Residual lymphoid follicles and vascular invasion were not common in our cases. However, tissue necrosis was frequently found in our MTX-Lymphoma cases; albeit, it can also be found in other lymphomas with aggressive behavior.¹² Histiocyte aggregation and plasma cell reaction were also frequently found in MTX-Lymphoma cases, but the significance and association with MTX have not been yet established.¹² Interestingly, Hodgkin-Reed-Sternberg-like cells were surprisingly common among the MTX-Lymphoma cases, but the significance and association with MTX have not been established yet either.¹²

A panel of immunostaining may give some additional information in these cases. In addition to the positivity of EBER ISH and/or EBV LMP-1 immunostaining, CD30 positivity in scattered large cells was found in many cases in our MTX-Lymphoma group, including DLBCL and PTCL, NOS. The 2 cases in our study (case no. 1 DLBCL and case no. 17 PTCL, NOS) showed regression of the lymph nodes after MTX discontinuation as the first-line management, corresponding to the expected clinical course of MTX-LPD.^{2-4,12-16} Both cases showed EBV+ tumor cells by EBER and/or EBV LMP-1 as well as scattered CD30+ large cells, in agreement with previous reports.¹³ These EBV association and CD30 positivity may help identify MTX-LPD,^{12,13,17-19} but the distinction from de novo lymphoma with EBV association and/or CD30 positivity has not been studied.

Among the 9 MTX-Lymphoma cases in our study who had MTX discontinuation and who received CMT as first-line management, 3 of them died of septic shock that might or might not have been a consequence of the chemotherapy. One may wonder what would have happened if these patients had only MTX discontinuation so that they could have avoided the risk of superimposed infection following chemotherapy. As MTX-LPD is not a well-known entity, this may be the reason why MTX-Lymphoma patients often received a diagnosis of lymphoma instead of MTX-LPD, so that most patients received chemotherapy. This is problematic because the recommended management of MTX-LPD is to discontinue MTX and to wait for a response,²⁰ but there is no consensus about how long to wait for the response or how to assess the response. The response after MTX discontinuation

reported in the literature varied from spontaneous to more than 8 weeks in some studies.^{7,20} Our study had only 2 cases who responded to MTX discontinuation alone; one with histopathology of DLBCL that showed a response at 63 days after discontinuation. The other with histopathology of PTCL, NOS that showed a response at 20 days after discontinuation. These observations were in agreement with previous studies.^{7,20} So, the administration of CMT in these patients may expose them to unnecessary risks from the chemotherapeutic agents. The MTX-Reactive group may not have even needed MTX discontinuation, since 5 of these cases showed a spontaneous regression despite continuing MTX.

The PTLN cases in our series did not have EBV serology tests, so we cannot be certain whether there was an EBV donor/recipient serology mismatch or not. Our polymorphic PTLN case died of superimposed infection despite the dosage of immunosuppressants being reduced and the patient receiving rituximab simultaneously according to the standard of care for LPD after hematopoietic stem cell transplant;²¹ the superimposed infection seemed not to be related to the LPD treatment. The other PTLN case with histopathology of DLBCL was not EBV-associated; the patient had immunosuppressants discontinued and received R-CHOP according to the standard of care for LPD after solid organ transplant,²¹ leading to CR. Despite relapse, the patient achieved second CR by salvage R-DA-EPOCH.

More than half of this group of patients had RA and interestingly about one-third of them had low disease activity index (DAS28-3) at LPD diagnosis. Thus, disease activity may not be useful to distinguish any group of patients. However, this study had a small sample size and may not be able to give adequate results.

Among the findings in the MTX-Reactive patients significantly different from MTX-Lymphoma patients, most of them held true for any reactive versus lymphoma comparisons. However, one of the interesting findings in our study was the absolute lymphocyte count. We found that the MTX-Reactive patients had a significantly higher median absolute lymphocyte count than the MTX-Lymphoma patients (1,938.3 vs. 780.8 cells/ μ l) ($p = 0.028$). This was in agreement with the report by Kurita et al. of peripheral blood lymphocyte counts of less than 1,000 cells/ μ l being found in their MTX-Lymphoma group more than in their MTX-Reactive group.¹² Saito et al. reported that the median lymphocyte count in persistent MTX-LPD was lower than in regressive MTX-LPD, with statistical significance,²² but Nakano et al. reported that the median lymphocyte count in persistent MTX-LPD was higher than in regressive MTX-LPD,

though without statistical significance.¹⁶ The absolute lymphocyte count may need further validation in a larger cohort to determine whether it can be used as one of the predictor for the regression of MTX-LPD.

Due to the small sample size of the study, we could not construct any link between the histopathology and the clinical features, the response after the discontinuation of MTX or immunosuppressants, or other clinical courses among the cases between the MTX-Lymphoma and PTLN groups. For the same reason, we could not find the pathognomonic histopathology for the pathologists to identify MTX-LPD without reviewing the medical records. We could not find a helpful clue to predict which case should respond to MTX discontinuation alone and which case would need upfront CMT unless we wait for a certain time to see the response of MTX discontinuation.²⁰ Further study should emphasize a comparison between MTX-Lymphoma patients with the same histopathology and, if applicable, a determination of clonality and genetic landscapes for any prominent features that would be helpful for pathologists to reach a diagnosis of MTX-LPD.

Based on this retrospective study, it is clear that we need in the future a comprehensive database to identify patients who have been receiving MTX so that this important piece of information can be linked to the laboratory information system for pathologists at the time of making a diagnosis. It could be very problematic if pathologists diagnose lymphoma without knowing that the patient is under MTX treatment, because the patient may be treated with chemotherapy for lymphoma without trying MTX discontinuation first. This could be harmful to the patient, because the side effects of chemotherapy may outweigh the benefit and can lead to death in some cases.

In conclusion, MTX-LPD and PTLN are highly heterogeneous diseases with a wide variety clinical spectrum and histopathology. Definitive diagnostic criteria and histopathologic categories for MTX-LPDs are yet to be established. Identifying MTX, other immunosuppressive drugs, and the immunodeficiency status of the patient are crucial in establishing the diagnosis. Immunostaining for CD30, EBV LMP-1, and EBER ISH study may be helpful to alert pathologists to consider MTX-LPD prior to making a diagnosis of lymphoma. MTX discontinuation is still the first-line management before the administration of chemotherapy in unresponsive cases of MTX-lymphoma, and similarly, lowering immunosuppressive drugs used in PTLN needs to consider with the hope of spontaneous regression of PTLN.

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REFERENCES

1. Koźmiński P, Halik PK, Chesori R, Gniazdowska E. Overview of dual-acting drug methotrexate in different neurological diseases, autoimmune pathologies and cancers. *Int J Mol Sci.* 2020;21:3483.
2. Salloum E, Cooper DL, Howe G, Lacy J, Tallini G, Crouch J, et al. Spontaneous regression of lymphoproliferative disorders in patients treated with methotrexate for rheumatoid arthritis and other rheumatic diseases. *J Clin Oncol.* 1996;14:1943-49.
3. Mariette X, Cazals-Hatem D, Warszawski J, Liote F, Balandraud N, Sibilia J, et al. Lymphomas in rheumatoid arthritis patients treated with methotrexate: a 3-year prospective study in France. *Blood.* 2002;99:3909-15.
4. Hoshida Y, Xu J-X, Fujita S, Nakamichi I, Ikeda J-I, Tomita Y, et al. Lymphoproliferative disorders in rheumatoid arthritis: clinicopathological analysis of 76 cases in relation to methotrexate medication. *J Rheumatol.* 2007;34:322-31.
5. Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J. WHO Classification of Tumors of Haematopoietic and Lymphoid Tissues (Revised 4th edition). Lyon: IARC; 2017.
6. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBO, Berti E, et al. The 5th edition of the World Health Organization Classification of Haematolymphoid Tumours: Lymphoid Neoplasms. *Leukemia.* 2022;36:1720-48.
7. Inui Y, Matsuoka H, Yakushiji K, Okamura A, Shimada T, Yano S, et al. Methotrexate-associated lymphoproliferative disorders: management by watchful waiting and observation of early lymphocyte recovery after methotrexate withdrawal. *Leuk Lymphoma.* 2015;56:3045-51.
8. Kitamura N, Sugiyama K, Nagasawa Y, Hamaguchi M, Kobayashi H, Takei M. Involvement of Epstein-Barr virus in the development and spontaneous regression of methotrexate-associated lymphoproliferative disorder in patients with rheumatoid arthritis. *Clin Exp Rheumatol* 2022;40:1330-5.
9. Gion Y, Doi M, Nishimura Y, Ikeda T, Filiz Nishimura M, Sakamoto M, et al. PD-L1 expression is associated with the spontaneous regression of patients with methotrexate-associated lymphoproliferative disorders. *Cancer Med* 2022;11:417-32.
10. Harada T, Iwasaki H, Muta T, Urata S, Sakamoto A, Kohno K, et al. Outcomes of methotrexate-associated lymphoproliferative disorders in rheumatoid arthritis patients treated with disease-modifying anti-rheumatic drugs. *Br J Haematol* 2021;194:101-10.
11. Fransen J, Stucki G, van Riel PLCM. Rheumatoid arthritis

- measures: Disease Activity Score (DAS), Disease Activity Score-28 (DAS28), Rapid Assessment of Disease Activity in Rheumatology (RADAR), and Rheumatoid Arthritis Disease Activity Index (RADAI). *Arthritis Care & Rheumatism* 2003;49:S214-24.
12. Kurita D, Miyoshi H, Ichikawa A, Kato K, Imaizumi Y, Seki R, et al. Methotrexate-associated lymphoproliferative disorders in patients with rheumatoid arthritis: clinicopathologic features and prognostic factors. *Am J Surg Pathol*. 2019 Jul;43:869-84.
 13. Ichikawa A, Arakawa F, Kiyasu J, Sato K, Miyoshi H, Niino D, et al. Methotrexate/iatrogenic lymphoproliferative disorders in rheumatoid arthritis: histology, Epstein-Barr virus, and clonality are important predictors of disease progression and regression. *Eur J Haematol*. 2013;91:20-8.
 14. Yamakawa N, Fujimoto M, Kawabata D, Terao C, Nishikori M, Nakashima R, et al. A clinical, pathological, and genetic characterization of methotrexate-associated lymphoproliferative disorders. *J Rheumatol*. 2014;41:293-9.
 15. Tokuhira M, Saito S, Okuyama A, Suzuki K, Higashi M, Momose S, et al. Clinicopathologic investigation of methotrexate-induced lymphoproliferative disorders, with a focus on regression. *Leuk Lymphoma*. 2018;59:1143-52.
 16. Nakano K, Saito K, Nawata A, Hanami K, Kubo S, Miyagawa I, et al. Clinical aspects in patients with rheumatoid arthritis complicated with lymphoproliferative disorders without regression after methotrexate withdrawal and treatment for arthritis after regression of lymphoproliferative disorders. *Mod Rheumatol*. 2021;31:94-100.
 17. Satou A, Tabata T, Miyoshi H, Kohno K, Suzuki Y, Yamashita D, et al. Methotrexate-associated lymphoproliferative disorders of T-cell phenotype: clinicopathological analysis of 28 cases. *Mod Pathol*. 2019;32:1135-46.
 18. Yamada K, Oshiro Y, Okamura S, Fujisaki T, Kondo S, Nakayama Y, et al. Clinicopathological characteristics and rituximab addition to cytotoxic therapies in patients with rheumatoid arthritis and methotrexate-associated large B lymphoproliferative disorders. *Histopathology*. 2015;67:70-80.
 19. Koens L, Senff NJ, Vermeer MH, Willemze R, Jansen PM. Methotrexate-associated B-cell lymphoproliferative disorders presenting in the skin: A clinicopathologic and immunophenotypic study of 10 cases *Am J Surg Pathol*. 2014;38:999-1006.
 20. Tokuhira M, Tamaru J-I, Kizaki M. Clinical management for other iatrogenic immunodeficiency-associated lymphoproliferative disorders. *J Clin Exp Hematop*. 2019;59:72-92.
 21. Dierickx D, Vergote V. Management of Post-transplant Lymphoproliferative Disorders. *Hema Sphere*, 2019;3:74-7.
 22. Saito S, Kaneko Y, Yamaoka K, Tokuhira M, Takeuchi T. Distinct patterns of lymphocyte count transition in lymphoproliferative disorder in patients with rheumatoid arthritis treated with methotrexate. *Rheumatology (Oxford)*. 2017;56:940-6.