



Title	Re-infection of Toxoplasma gondii after HSCT presenting lymphadenopathy resembling recurrence of lymphoma
Author(s)	Hashiguchi, Junichi; Onozawa, Masahiro; Naka, Tomoaki; Hatanaka, Kanako C.; Shiratori, Souichi; Sugita, Junichi; Fujimoto, Katsuya; Matsuno, Yoshihiro; Teshima, Takanori
Citation	Transplant infectious disease, 20(3), e12892 https://doi.org/10.1111/tid.12892
Issue Date	2018-06
Doc URL	http://hdl.handle.net/2115/74504
Rights	This is the peer reviewed version of the following article: Hashiguchi J, Onozawa M, Naka T, et al. Re-infection of Toxoplasma gondii after HSCT presenting lymphadenopathy resembling recurrence of lymphoma. Transpl Infect Dis. 2018;20:e12892., which has been published in final form at https://doi.org/10.1111/tid.12892 . This article may be used for non-commercial purposes in accordance with Wiley Terms and Conditions for Use of Self-Archived Versions.
Type	article (author version)
File Information	TID R1 PDF_proof_hi.pdf



[Instructions for use](#)

Transplant infectious disease

Short communications

Re-infection of *toxoplasma gondii* after HSCT presenting lymphadenopathy resembling recurrence of lymphoma

Junichi Hashiguchi¹⁾, Masahiro Onozawa¹⁾, Tomoaki Naka²⁾, Kanako C. Hatanaka²⁾, Souichi Shiratori¹⁾, Junichi Sugita¹⁾, Katsuya Fujimoto¹⁾, Yoshihiro Matsuno²⁾, Takanori Teshima¹⁾

1) Department of Hematology, Hokkaido University Hospital, Sapporo, Japan

2) Department of Surgical Pathology, Hokkaido University Hospital, Sapporo, Japan

Words counts of text: 1238 words

Table: 0, Figure: 4

Key words: (3-10) *Toxoplasma gondii*, lymphadenopathy, re-infection, HSCT, FDG PET-CT

Running Head: Re-infection of *T. gondii* after HSCT

Correspondence to: Masahiro Onozawa

Department of Hematology, Hokkaido University Hospital, Kita-15, Nishi-7, Kita-ku, Sapporo 060-8638, JAPAN

TEL: +81-11-716-7214

FAX: +81-11-706-7823

E-mail address: onozawa@med.hokudai.ac.jp

Abstract (<75 words)

Toxoplasma gondii (*T. gondii*) reactivation is one of the fatal complications after hematopoietic stem cell transplantation (HSCT); however, re-infection has not been reported. Here we report a case of mycosis fungoides in which cervical lymphadenopathy developed after HSCT. Initially, recurrent lymphoma was suspected. However, biopsy of the lymph node showed typical histology of toxoplasmosis and serology showed re-infection of *T. gondii*. *Toxoplasmosis* needs to be differentiated for cases with lymphadenopathy after HSCT.

Introduction

Toxoplasma gondii (*T. gondii*) is an opportunistic protozoan, and reactivation of latent disease is believed to be the main cause of symptomatic disease after allogeneic hematopoietic stem cell transplantation (HSCT) (1). Disseminated disease is associated with considerable morbidity and mortality (2, 3). Here we report a case of mycosis fungoides in which cervical lymphadenopathy developed due to re-infection of *T. gondii* after HSCT. Re-biopsy of the lymph node was important to rule out recurrent lymphoma.

Case Report

A 58-year-old woman was followed up for diagnosis of parapsoriasis at a regional dermatology clinic for 9 years. She was referred to our hospital because of rapidly growing painful left inguinal lymphadenopathy and masses in her left thigh. Skin biopsy revealed mycosis fungoides and lymph node biopsy showed infiltration of mature T cell lymphoma. She received 4 courses of the CHOP regimen, which resulted in refractory disease, and the EPOCH regimen was therefore given as salvage therapy. Since she still had a refractory skin lesion even after 4 courses of the EPOCH regimen, allogeneic HSCT was performed. The conditioning regimen consisted of Fludarabine, Melphalan and low-dose TBI. Short-term MTX and Tacrolimus were administered as prophylaxis against GVHD. Engraftment was achieved on day 17 after HSCT. A prophylactic oral sulfamethoxazole-trimethoprim (SMX-TMP) mixture against pneumocystis pneumonia was not administered because of sustained thrombocytopenia. She was discharged on day 107 after HSCT without any significant complication or sign of GVHD and was followed at the

out-patient ward monthly. ¹⁸F-fluorodeoxyglucose positron emission tomography-computed tomography (¹⁸F-FDG PET-CT) at 6 month after HSCT showed complete remission without any abnormal FDG uptake. Administration of tacrolimus was ceased at six months after HSCT. A periodical checkup by ¹⁸F-FDG PET-CT at 13 months after HSCT revealed right cervical lymphadenopathy with FDG uptake (Fig.1). Maximum standardized uptake value (SUVmax) of the lesion was 15.4. The lymph node was palpable as 2 cm in diameter without tenderness. Because the patient had history of inguinal lymph node infiltration of cutaneous T-cell lymphoma, recurrence of the initial lymphoma was strongly suspected, and re-biopsy of the lymph node was done. Microscopic examination of the lymph node revealed lymphofollicular enlargement with hyperplastic germinal centers, paracortical aggregation of monocytoid B cells, and epithelioid cell microgranulomas, which all suggested a diagnosis of toxoplasmic lymphadenitis (Fig. 2). There was no histological or immunohistochemical evidence of recurrent disease of cutaneous T-cell lymphoma. To confirm the diagnosis, the *T. Gondii* B1 gene was PCR-amplified using a genome template purified from the lymph node sample. The lymph node biopsy before HSCT, which was histologically shown to be involved by infiltration of mycosis fungoides, was used as a control. Toxoplasma B1 gene was detected by nested PCR in the lymph node sample after HSCT but not in the lymph node sample before HSCT (Fig. 3). Her serology was positive for both anti-toxoplasma IgM and IgG antibodies. Because anti-toxoplasma IgM could persist year-long since initial infection, we retrospectively tested the anti-toxoplasma IgM and IgG antibodies using available cryopreserved serum specimens. The samples before HSCT and 9 months after HSCT were positive for anti-toxoplasma IgG and negative for anti-toxoplasma IgM, indicating that she had previous *T. Gondii* infection (Fig. 4). A follow-up sample at 16 months after HSCT (2 months after lymphadenitis) showed increased IgG and decreased IgM, suggesting re-infection occurred at 14 month after HSCT. Brain MRI was normal without typical toxoplasmosis found in an immunocompromised host. From these findings, we made a diagnosis of re-infection of *T. Gondii* in a patient with previous toxoplasma infection. An oral SMX-TMP mixture was prescribed and no further lymphadenopathy was observed.

Toxoplasmosis is one of the most common parasitic zoonoses worldwide. Its causative agent, *T. gondii*, is a facultatively heteroxenous, polyxenous protozoon that has developed several potential routes of transmission within and between different host species. Transmission to humans may occur via ingestion of infectious oocysts from the environment (usually via soil contaminated with feline feces), via organ transplants, or via tissue cysts in meat from an infected animal or from contaminated fruits, vegetables, or water. However, it is not known which of these routes is more important epidemiologically. It is likely that the major routes of transmission are different in human populations with differences in culture and eating habits (4). The seroprevalence of *T. gondii* varies markedly among different regions, ranging from 10% to 50-80%. In Japan, the seropositivity for *T. gondii* in the general population has been reported to be approximately 10-15% (5). In a chart review in our department, seropositivity of *T. gondii* was found to be 10.8% (33 seropositive patients in 306 patients), which is almost the same as that in the United States and lower than that in other countries (6). When immunocompetent adults are primarily infected with *T. gondii*, the majority of them are usually asymptomatic. Approximately 10% of patients show symptoms like those of infectious mononucleosis including fever, chills, muscular pain, prominent cervical lymphadenopathy, hepatitis. The most common manifestation is bilateral, symmetrical, and non-tender cervical lymphadenopathy. The lymph nodes are usually smaller than 3 cm in size and non-fluctuant (7).

In HSCT recipients, toxoplasmosis is a relatively rare complication. The incidence of toxoplasmosis after HSCT was reported to be 0.1-6.0% (4, 5, 8-11), but it is a potentially fatal opportunistic parasitic infection with an estimated mortality rate of 60-90% (12-15). In Japan, definite diagnosis of toxoplasmosis after allo-HSCT was reported to be very rare (0.22%) (5). Most cases of toxoplasmosis occur due to reactivation with organ symptoms, particularly in patients with GVHD and in umbilical cord blood transplant recipients (16, 17). The largest scale review showed 356 cases of toxoplasmosis following allo-HSCT (6). Among patients with known pretransplant toxoplasma serology, 90% were seropositive and likely had reactivation of toxoplasmosis following HSCT. Central nervous system and

disseminated diseases were the most common manifestations, accounting for 87% of toxoplasma disease cases (6, 18). In most cases (89%), toxoplasmosis developed within 180 days after HSCT (6). Our case was a rare case of re-infection of toxoplasma after HSCT. Pre-HSCT anti-toxoplasma IgG positivity clearly showed that the patient had previous infection of toxoplasma. Toxoplasma transmission via the stem cell graft from the donor was unlikely because lymphadenopathy and anti-toxoplasma IgM were not detected until the episode at 14 months after HSCT. Anti-toxoplasma IgM after HSCT would be produced by naive donor immune cells. Unfortunately, we could not verify toxoplasma serology of the donor because the unrelated donor bone marrow graft was donated via a public registry and anti-toxoplasma IgG is not included in routine screening items. Re-infection of toxoplasma after HSCT could have been derived from latent infection within her body or new infection from the environment. She had swelling of a single cervical lymph node without other organ involvement or systemic infectious symptoms. The lymph node began to swell after she was discharged and went back to her home, where she had a cat as a pet. When she developed lymphadenopathy, she had already ceased taking an immunosuppressant without any sign of GVHD and she had sufficient number of CD4 cells (504 / μ l). Furthermore, reactivation of *T. gondii* does not usually involve lymph nodes. All these findings suggested that the patient had re-infection of toxoplasma from the environment.

Increased FDG uptake in lymph nodes of patients who underwent HSCT should be interpreted with caution in differentiating toxoplasmosis from recurrence of an initial hematological malignancy or post-transplant lymphoproliferative disorder. Diagnosis should be confirmed by histopathological evaluation together with serological transition of anti-toxoplasma antibodies.

References

1. Montoya JG and Liesenfeld O. Toxoplasmosis. *Lancet* 2004; 363(9425): 1965-1976.
2. Martino R, Maertens J, Bretagne S et al. Toxoplasmosis after hematopoietic stem cell transplantation. *Clin Infect Dis* 2000; 31(5): 1188-1195.
3. Chandrasekar PH and Momin F. Disseminated toxoplasmosis in marrow recipients: a report of three cases and a review of the literature. *Bone Marrow Transplant Team. Bone Marrow Transplant* 1997; 19(7): 685-689.
4. Tenter AM, Heckeroth AR, and Weiss LM. *Toxoplasma gondii*: from animals to humans. *Int J Parasitol* 2000; 30(12-13): 1217-1258.
5. Matsuo Y, Takeishi S, Miyamoto T et al. Toxoplasmosis encephalitis following severe graft-vs.-host disease after allogeneic hematopoietic stem cell transplantation: 17 yr experience in Fukuoka BMT group. *Eur J Haematol* 2007; 79(4): 317-321.
6. Gajurel K, Dhakal R, and Montoya JG. Toxoplasma prophylaxis in haematopoietic cell transplant recipients: a review of the literature and recommendations. *Curr Opin Infect Dis* 2015; 28(4): 283-292.
7. McCabe RE, Brooks RG, Dorfman RF, and Remington JS. Clinical spectrum in 107 cases of toxoplasmic lymphadenopathy. *Rev Infect Dis* 1987; 9(4): 754-774.
8. de Medeiros BC, de Medeiros CR, Werner B, Loddo G, Pasquini R, and Bleggi-Torres LF. Disseminated toxoplasmosis after bone marrow transplantation: report of 9 cases. *Transpl Infect Dis* 2001; 3(1): 24-28.
9. Fricker-Hidalgo H, Bulabois CE, Brenier-Pinchart MP et al. Diagnosis of toxoplasmosis after allogeneic stem cell transplantation: results of DNA detection and serological techniques. *Clin Infect Dis* 2009; 48(2): e9-e15.
10. Roemer E, Blau IW, Basara N et al. Toxoplasmosis, a severe complication in allogeneic hematopoietic stem cell transplantation: successful treatment strategies during a 5-year single-center experience. *Clin Infect Dis* 2001; 32(1): E1-8.
11. Slavin MA, Meyers JD, Remington JS, and Hackman RC. Toxoplasma gondii infection in marrow transplant recipients: a 20 year experience. *Bone Marrow Transplant* 1994; 13(5): 549-557.
12. Goebel WS, Conway JH, Faught P, Vakili ST, and Haut PR. Disseminated toxoplasmosis resulting in graft failure in a cord blood stem cell transplant recipient. *Pediatr Blood Cancer* 2007; 48(2): 222-226.
13. Derouin F, Pelloux H, and Parasitology ESGoC. Prevention of toxoplasmosis in transplant patients. *Clin Microbiol Infect* 2008; 14(12): 1089-1101.
14. Mulanovich VE, Ahmed SI, Ozturk T, Khokhar FA, Kontoyiannis DP, and de Lima M. Toxoplasmosis in allo-SCT patients: risk factors and outcomes at a transplantation center with a low incidence. *Bone Marrow Transplant* 2011; 46(2): 273-277.
15. Small TN, Leung L, Stiles J et al. Disseminated toxoplasmosis following T cell-depleted related and unrelated bone marrow transplantation. *Bone Marrow Transplant* 2000; 25(9): 969-973.
16. Meignan M, Itti E, Gallamini A, and Younes A. FDG PET/CT imaging as a biomarker in lymphoma. *Eur J Nucl Med Mol Imaging* 2015; 42(4): 623-633.
17. Tateno T, Onozawa M, Hashiguchi J et al. Disseminated toxoplasmosis after hematopoietic stem cell transplantation showing unusual magnetic resonance images. *Transpl Infect Dis* 2017; 19(4).
18. Gajurel K, Dhakal R, and Montoya JG. Toxoplasmosis in hematopoietic cell transplant recipients. *Transpl Infect Dis* 2017; 19(5).

Figure Legends

Fig. 1. Cervical lymphadenopathy detected by PET-CT.
¹⁸F-FDG PET-CT showed focal FDG uptake at the right cervical lymphadenopathy with SUVmax of 15.4 (arrows).

Fig. 2. Histopathologic findings of lymph node biopsy.
Histopathologic findings consisted of lymphofollicular enlargement with hyperplastic germinal centers (center), paracortical aggregation of monocytoïd B cells (left part), and epithelioid cell microgranulomas (right part). These findings were suggestive of toxoplasmic lymphadenitis (Piringer-Kuchinka lymphadenitis).
Haematoxylin and eosin staining; original magnification, x40.

Fig. 3. PCR detection of *Toxoplasma gondii* B1 gene.
The *T. gondii* B1 gene was amplified by nested PCR using the primer set TOXO B1 F0: 5'-GGAAGTGCATCCGTTTCATGAG-3', and TOXO B1 R0: 5'-GCAGCGACTTCTATCTCTGTG-3' for the 1st PCR and TOXO B1 F1: 5'-TGCATAGGTTGCAGTCACTG-3' and TOXO B1 R1: 5'-TCTTTAAAGCGTTCGTGGTC-3' for the 2nd PCR. β globin was amplified as an internal control using the primer set β globin F: 5'-ACACAACTGTGTTCCTAGC-3' and β globin R: 5'-GGAAAATAGACCAATAGGCAG-3'. Cervical lymph node (LN) before HSCT, which was diagnosed as LN infiltration of mycosis fungoides, was negative for *T. gondii*. However, the swollen LN after HSCT was positive for *T. gondii*. The amplified *T. gondii* B1 gene product was verified by direct sequencing.

Fig. 4. Anti-toxoplasma antibodies.
Anti-toxoplasma IgG, but not IgM, was positive before HSCT. When cervical lymph node swelling was detected 14 months after HSCT, both anti-toxoplasma IgG and IgM became positive. The IgG titer was increased and the IgM titer was decreased at 16 months after HSCT.

Figure 1 Cervical lymphadenopathy detected by PET-CT

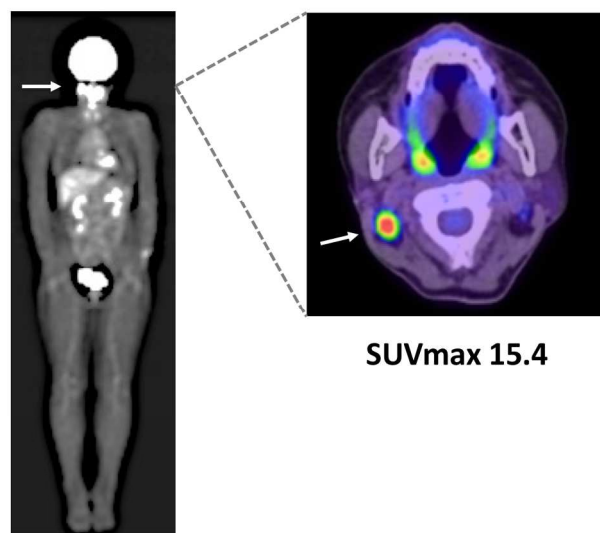


Figure1

254x190mm (200 x 200 DPI)

Figure 2 Pathological findings of lymph node biopsy

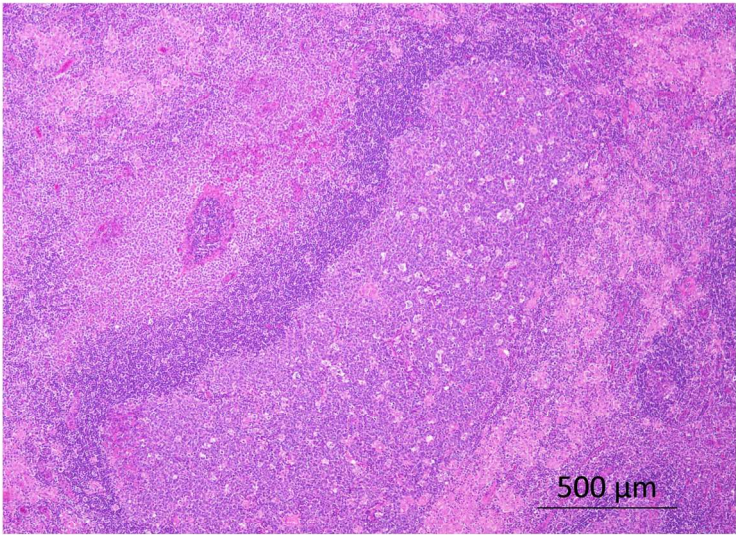


Figure2
254x190mm (200 x 200 DPI)

Figure 3 PCR detection of *Toxoplasma gondii* B1 gene

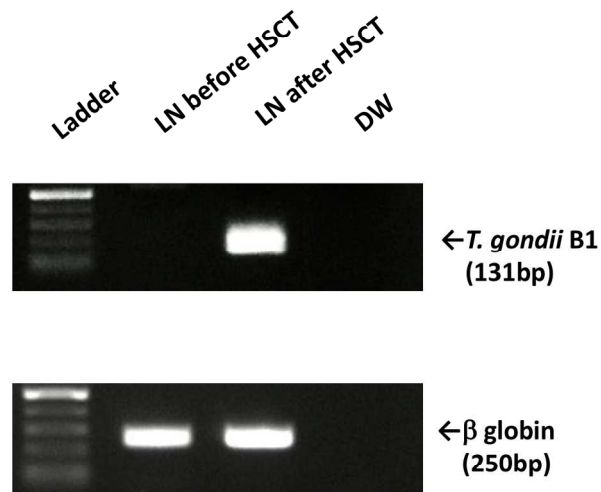


Figure3

254x190mm (200 x 200 DPI)

Figure 4 Anti-toxoplasma antibodies

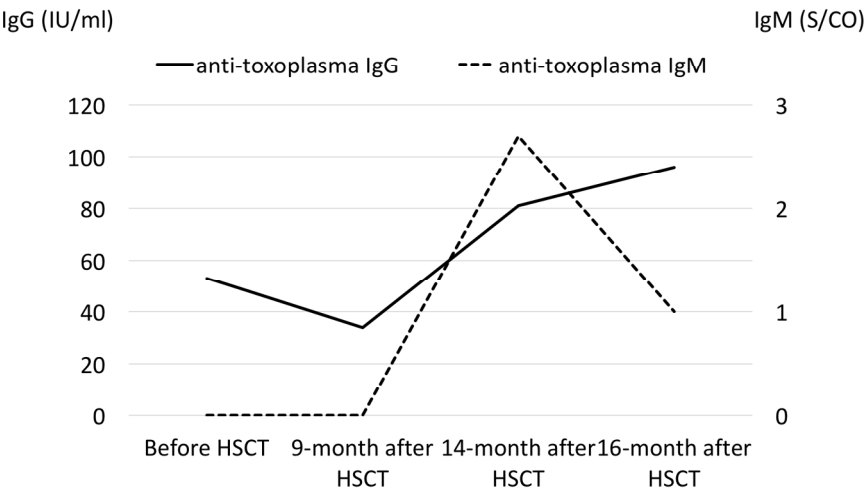


Figure4

254x190mm (200 x 200 DPI)