


Maintenance of long remission in adult T-cell leukemia by Tax-targeted vaccine: A hope for disease-preventive therapy

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Adult T-cell leukemia/lymphoma (ATL) is an aggressive lymphoproliferative disease caused by human T-cell leukemia virus type 1 (HTLV-1). Multi-agent chemotherapy can reduce ATL cells but frequently allows relapses within a short period of time. Allogeneic hematopoietic stem cell transplantation (allo-HSCT) following chemotherapy is now a standard therapy for ATL in Japan as it can achieve long-term remission in approximately one-third of recipient ATL patients; however, it also has a risk of treatment-related mortality. Allo-HSCT often induces HTLV-1 Tax-specific cytotoxic T cells (CTL) as well as graft-versus-host (GVH) response in ATL patients. This observation led to development of a new therapeutic vaccine to activate Tax-specific CTL, anticipating anti-ATL effects without GVH response. The newly developed Tax-DC vaccine consists of autologous dendritic cells pulsed with Tax peptides corresponding to CTL epitopes that have been identified in post-allo-HSCT ATL patients. In a pilot study of Tax-DC therapy in three ATL patients after various initial therapies, two patients survived for more than 4 years after vaccination without severe adverse effects (UMIN000011423). The Tax-DC vaccine is currently under phase I trial, showing a promising clinical outcome so far. These findings indicate the importance of patients' own HTLV-1-specific T-cell responses in maintaining remission and provide a new approach to anti-ATL immunotherapy targeting Tax. Although Tax-targeted vaccination is ineffective against Tax-negative ATL cells, it can be a safe alternative maintenance therapy for Tax-positive ATL and may be further applicable for treatment of indolent ATL or even prophylaxis of ATL development among HTLV-1-carriers.

1 | INTRODUCTION

Adult T-cell leukemia/lymphoma is an aggressive lymphoproliferative disease, occurring in a small percentage of HTLV-1-infected individuals.¹ There are four types of ATL: acute, lymphoma,

chronic and smoldering. Among them, the former two are known to have a poor prognosis because of rapid progression, frequent relapse and severe immunosuppression.² The prognosis of indolent ATL (smoldering and chronic ATL) varies widely among individuals. Katsuya et al³ categorized indolent ATL by the levels of

Abbreviations: allo-HSCT, allogeneic hematopoietic stem cell transplantation; ATL, adult T-cell leukemia/lymphoma; CCR4, C-C chemokine receptor 4; CR, complete remission; CTL, cytotoxic T cells; DC, dendritic cells; GVH, graft-versus-host; GVHD, graft-versus-host disease; GVL, graft-versus-leukemia; HAM/TSP, HTLV-1-associated myelopathy/tropical spastic paraparesis; HBZ, HTLV-1 basic leucine zipper; HLA, human leukocyte antigen; HTLV-1, human T-cells leukemia virus type 1; IFN- α /AZT, interferon- α and azidothymidine; IKZF1/3, IKAROS family zinc finger 1 and 3; IL, interleukin; IRF4, interferon regulatory factor 4; NK, natural killer; OS, overall survival; PBMC, peripheral blood mononuclear cell; PD-1, programmed cell death 1; PD-L1, PD-1 ligand 1; PKR, dsRNA-dependent protein kinase; PR, partial remission; PVL, proviral load; sIL-2R, soluble interleukin-2 receptor; Treg, regulatory T-cells.

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sIL-2R α in the serum and indicated the OS at 4 years to be 26.2%, 55.6% and 77.6% for low, intermediate and high-risk groups, respectively. Despite the presence of obvious hematological abnormalities, "watchful waiting" is usually recommended for indolent ATL, unless unfavorable prognostic factors appear, including elevated lactate dehydrogenase or blood urea nitrogen, or decreased albumin levels.²

For acute- and lymphoma-type ATL, multi-agent chemotherapy and subsequent allo-HSCT are commonly used in Japan, achieving long-term remission in one-third of ATL cases.^{4,5} Recently, mogamulizumab⁶ and lenalidomide⁷ have also become available for acute- and lymphoma-type ATL. However, neither of these drugs are approved for indolent ATL yet. Combined IFN- α /AZT therapy is widely used for ATL in other countries and is reported to be effective, especially for indolent ATL.^{8,9}

We recently developed a new therapeutic vaccine, Tax-DC, to activate HTLV-1 Tax-specific cytotoxic T cells (CTL), consisting of Tax peptide-pulsed autologous DC.¹⁰ This was based on the experimental findings that Tax-specific CTL showed anti-tumor effects in animal models of HTLV-1-infected tumors and the clinical observation that Tax-specific CTL were activated in ATL patients after allo-HSCT.¹¹ A clinical study of the Tax-DC vaccine in a small number of ATL patients after various chemotherapy regimens suggests its potential role in achieving long-term remission.¹⁰ These findings indicate the importance of patients' own immunity in maintenance of remission.

In this review, we focus on the Tax-targeted vaccine therapy, which provides a new approach to ATL therapy, which could be extended for treatment of indolent ATL or even ATL prophylaxis. We also discuss the mechanisms of immunosuppression, a key issue underlying ATL development, which is another important target for induction of anti-tumor immunity in therapeutic and prophylactic strategies against ATL.

2 | CURRENTLY AVAILABLE ATL THERAPIES

For acute- and lymphoma-type ATL, multi-agent chemotherapy, mogamulizumab, lenalidomide and HSCT are currently available in Japan. The mechanisms of anti-ATL effects and influences on the host immunity of these therapies are summarized in Table 1.

For the last few decades since the discovery of ATL, various regimens of chemotherapy have been tried for ATL with limited success. The median survival time of ATL patients treated with the latest regimen of multi-agent chemotherapy (VCAP-AMP-VECP) was 12.7 months.¹² Chemotherapy is effective in reducing ATL cells but allows frequent relapses.

Mogamulizumab is a humanized anti-CCR4 antibody that reduces ATL cells mainly through antibody-dependent cellular cytotoxicity by NK cells.¹³ Because Treg partly express CCR4, mogamulizumab also reduces Treg, which is of potential benefit for reducing immunosuppression.¹⁴ However, this is presumably a cause of the trend of severe adverse effects of HSCT in some patients who were previously treated with mogamulizumab.¹⁵ Median survival time of mogamulizumab for relapsed ATL patients was 13.7 months.⁶

Lenalidomide has been used for multiple myeloma and was recently approved for ATL in Japan. Lenalidomide exerts its anti-tumor and immunomodulatory effects through binding with cereblon.¹⁶ Anti-tumor effects involve downregulation of transcription factors including IKZF1/3 and IRF4 in multiple myeloma,¹⁷ while immunomodulatory effects include activation of T- and NK cells that potentially support the anti-tumor immune response.¹⁸ Median survival time for lenalidomide in relapsed or recurrent ATL patients was 20.3 months.⁷

Anti-ATL effects of IFN- α /AZT combination therapy were first reported in 1995,^{8,9} and this regimen has been used for ATL patients in several countries, although it is not yet approved in

TABLE 1 Mechanisms of currently available ATL therapies and Tax-DC vaccine

	Mechanism of anti-ATL effect	Effects on host immune system	Adverse effects
Chemotherapy	Induction of cell death in dividing cells	Immune suppression	Cytopenia
Mogamulizumab	Killing of CCR4 ⁺ cells through ADCC by NK cells ¹³	Reduction of Treg	Infusion reactions, skin rash ⁶
Lenalidomide	Downregulation of IKZF1/3, IRF4 and so forth by binding cereblon (multiple myeloma) ^{a,16,17}	Enhancement of T-cell and NK cell activity ¹⁸	Cytopenia ⁷
IFN- α /AZT	Activation of p53 pathway and suppression of Tax expression ²⁰	Unknown	Cytopenia ^{8,21}
Allo-HSCT	Elimination of recipient hematopoietic cells	Induction of GVH and Tax-specific CTL responses ²⁵	GVHD
Tax-DC vaccine	Killing of HTLV-1-infected cells	Activation of Tax-specific CTL response ¹⁰	Fever, skin rash ¹⁰

ADCC, antibody-dependent cell-mediated cytotoxicity; allo-HSCT, allogeneic hematopoietic stem cell transplantation; ATL, adult T-cell leukemia/lymphoma; AZT, azidothymidine; CCR4, C-C chemokine receptor 4; CTL, cytotoxic T cells; DC, dendritic cells; GVH, graft-versus-host; GVHD, graft-versus-host disease; IKZF1/3, IKAROS family zinc finger 1 and 3; IRF4, interferon regulatory factor 4; NK, natural killer; Treg, regulatory T cells.

^aReported in multiple myeloma.

Japan. The reported 5-year OS rates for IFN- α /AZT were 28%, 0% and 100% for acute, lymphoma and indolent types of ATL, respectively.¹⁹ Mechanisms of anti-ATL effects of IFN- α /AZT therapy include suppression of Tax expression and activation of the p53 pathway.²⁰ However, IFN- α /AZT therapy frequently allows relapse of ATL after cessation of treatment.²¹

To overcome the frequent relapses in aggressive types of ATL, allo-HSCT has been used for ATL patients combined with multi-agent chemotherapy in Japan,⁴ although early attempts at autologous HSCT in ATL patients were unsuccessful.²² In a retrospective analysis of 386 ATL cases treated with allo-HSCT, the 3-year OS was 33%, while allo-HSCT also caused treatment-related mortality in a similar number of patients.⁵ The reduced intensity conditioning regimens of allo-HSCT slightly improved the treatment-related mortality,²³ while the survival curve still rapidly declined in the first 1-2 years, before reaching a plateau.²⁴

3 | ACTIVATION OF TAX-SPECIFIC CTL AFTER ALLO-HSCT

In allo-HSCT, the hematopoietic system of a recipient ATL patient is replaced with donor-derived hematopoietic cells. GVH response is the main anti-tumor mechanism of allo-HSCT, where the GVH responder T cells eliminate residual ATL cells together with other recipient cells. However, GVH response also causes GVH disease that can be fatal. Therefore, immunosuppressants are required for some time after allo-HSCT. After hematopoietic chimerism is established, the newly constituted system of T-cell tolerance begins to control the GVH response. In addition, activation of CD8⁺ HTLV-1 Tax-specific CTL is often observed in ATL patients following allo-HSCT, especially after removal of immunosuppressants.²⁵ This is likely to be a result of de novo T-cell responses by the donor-derived immune system against Tax-expressing cells in the recipient tissues. Although Tax protein is usually undetectable in PBMC

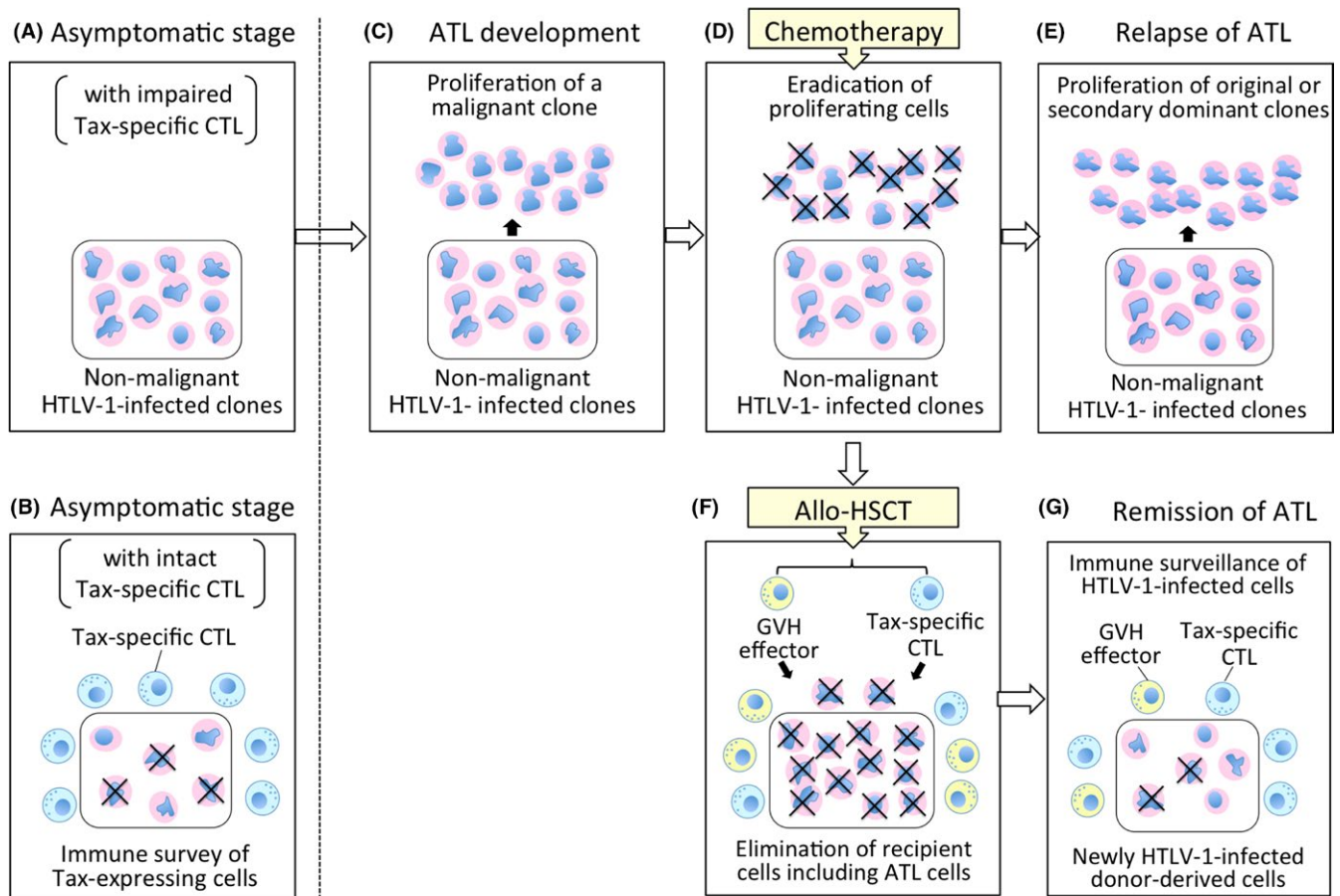


FIGURE 1 Possible dynamics of human T-cell leukemia virus type 1 (HTLV-1)-infected cell clones during adult T-cell leukemia/lymphoma (ATL) development and allogeneic hematopoietic stem cell transplantation (allo-HSCT). Asymptomatic HTLV-1 carriers possess multiple HTLV-1-infected cell clones. Impairment of Tax-specific cytotoxic T cells (CTL) allows expansion of the pool of HTLV-1-infected cell clones (A), while it is controlled in the presence of intact Tax-specific CTL that survey Tax-expressing infected cells (B). At the onset of ATL, one or several HTLV-1-infected clones expand (C). Upon intensive chemotherapy, proliferating malignant ATL cells die (D), but the residual original ATL clones or secondary dominant clones proliferate again at relapse (E). When allo-HSCT is performed, graft-versus-host (GVH) and HTLV-1-specific T-cell responses occur, and recipient hematopoietic cells are eliminated together with residual ATL cells (F). After hematopoietic chimerism is established, Tax-specific CTL survey newly HTLV-1-infected donor-derived cells, contributing to the maintenance of remission (G)

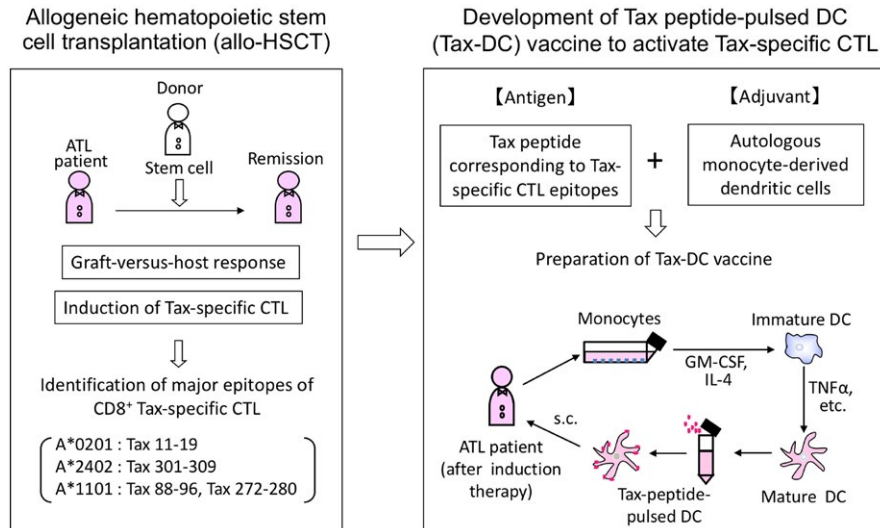


FIGURE 2 Development of Tax-DC vaccine based on findings in adult T-cell leukemia/lymphoma (ATL) patients after allogeneic hematopoietic stem cell transplantation (allo-HSCT). Tax-specific cytotoxic T cells (CTL) are often activated in ATL patients following allo-HSCT. Some of the major epitopes recognized by Tax-specific CTL have been identified (left panel).^{25,26} The Tax-DC vaccine was developed by pulsing synthetic Tax oligopeptides corresponding to the CTL epitopes onto autologous monocyte-derived dendritic cells, and administered s.c. to ATL patients three times with 2-wk intervals (right panel)¹⁰

by serological means in HTLV-1-infected individuals, activation of Tax-specific CTL in ATL patients after allo-HSCT demonstrates the presence of Tax expression in vivo at levels detectable by CTL.

Analysis of Tax-specific CTL in ATL patients after allo-HSCT identified dominant CTL epitopes to be restricted to HLA-A2, -A24 or -A11, respectively.^{25,26} These CTL epitopes are also found in HAM/TSP patients and asymptomatic HTLV-1-carriers, indicating that these epitopes are commonly recognized by CTL as an anti-viral immune response irrespective of disease.^{27,28}

4 | ATL RELAPSE AND ITS PREVENTION BY ALLO-HSCT

It is unclear why ATL relapses so frequently in a short period of time after chemotherapy. This may be related to the fact that HTLV-1-infected individuals possess numerous HTLV-1-infected clones, although most of them are not yet malignant (Figure 1). In asymptomatic HTLV-1-carriers with intact HTLV-1-specific T-cell responses, the immune surveillance system eliminates infected clones expressing viral antigens to some extent. However, under impaired HTLV-1-specific T-cell responses, the infected clones have greater chances of survival and evolution. Among such HTLV-1-infected clones, the dominant malignant clones proliferate during ATL development. After initial chemotherapy, only proliferating cells are eliminated. However, there remain numerous HTLV-1-infected clones. At relapse, the residual original ATL clone or secondary malignant clones may proliferate. The presence of a pool of HTLV-1-infected clones with various proliferation capacities and frequent acquisition of drug resistance²⁹ is assumed to be a reason for such frequent ATL relapse. This scenario is consistent with previous reports that the dominant

ATL clones sometimes change during the disease course and also between tissues.^{30,31}

In contrast, allo-HSCT eradicates recipient-derived cells irrespective of HTLV-1 infection or their capacity to proliferate. Allogeneic HSCT induces GVH response that is supposed to eliminate the residual ATL cells. Tax-specific CTL that are often activated after allo-HSCT may also partly contribute to GVL effect. Tax-specific CTL seem to remain in the long term as memory T-cells and potentially act as an immune surveillance system on newly HTLV-1-infected donor-derived clones after hematopoietic chimerism is established (Figure 1).

5 | TAX-TARGETED THERAPEUTIC VACCINE

The findings in ATL patients after allo-HSCT led to the hypothesis that a vaccine to activate Tax-specific CTL may induce long-term remission in ATL patients without GVHD. Our previous study using a rat model indicated that a vaccine using Tax peptide corresponding to the major epitope of CD8⁺ Tax-specific CTL induced tumor eradication, when appropriate adjuvant was used.³² To overcome the severe immunosuppression in ATL patients, we chose autologous monocyte-derived DC matured in vitro as an adjuvant for anti-ATL vaccine.³³ These DC were pulsed with Tax peptides corresponding to the major Tax-specific CTL epitopes previously identified in post-HSCT ATL patients (Figure 2).

In a small pilot study, the Tax-DC vaccine was administered s.c. to three ATL patients in stable condition after initial therapies, but who could not receive allo-HSCT for various reasons.¹⁰ After three inoculations with Tax-DC vaccine, Tax-specific CTL were clearly activated in these patients without severe adverse effects. A long-term clinical follow up found that two of three patients survived for more than

TABLE 2 Clinical outcomes of the pilot study of Tax-DC vaccine therapy in ATL patients^a

Disease status at enrollment	Patient 1	Patient 2	Patient 3
	SD	PR	PR
Clinical response after vaccination			
8 wk	PR	SD	PR
6 mo	PR	PD	CR
Long-term clinical outcomes ^b			
TTNT	53 mo	15 mo	>5 y
Survival	60 mo	23 mo	>5 y

ATL, adult T-cell leukemia/lymphoma; CR, complete remission; PD, progressive disease; PR, partial remission; SD, stable disease; TTNT, time to next therapy.

^aThree patients with acute ATL were enrolled in the pilot study of Tax-DC vaccine at least 1 mo after previous therapies.¹⁰

^bLong-term clinical outcome represents time after enrollment in the study.

4 years after vaccination (Table 2). Patient 1 maintained PR without additional therapy for 4 years before exacerbation with lymph node swelling that was easily controlled by chemotherapy. However, this patient subsequently suffered repeated infections and died 5 years after vaccination. Patient 2 initially showed stable disease with improved performance status but relapsed at 6 months with lymphoma lacking the ability of viral expression and died 23 months after vaccination, suggesting an ATL clone without Tax expression escaped the host immunity. Patient 3 survived for longer than 5 years after vaccination and remains alive in CR.

The Tax-DC vaccine is currently in a phase I clinical trial, in which an additional three ATL patients treated with Tax-DC vaccine with the same regimen as the pilot study have remained in CR for at least 2 years. Although the effectiveness of Tax-DC vaccination remains to be confirmed by a phase II study, the clinical outcomes of these studies indicate that Tax-specific CTL may contribute considerably to maintenance of remission in ATL patients unless Tax-negative ATL clones emerge at relapse.

6 | TAX EXPRESSION IN ATL CELLS IN VIVO

Expression levels of Tax in vivo have been controversial. Despite the presence of HTLV-1 antibodies and Tax-specific CTL responses in many HTLV-1-infected individuals, viral proteins are undetectable in freshly isolated PBMC by serological means, raising skepticism around the significance of a Tax-targeted immunotherapy.

There are at least two types of ATL that can be separated by the ability of primary ATL cells to express HTLV-1 antigens. In nearly half of ATL cases, HTLV-1 antigens can be spontaneously induced after short-term culture.³⁴ In the remaining ATL cases, ATL cells do not express HTLV-1 antigens even after culture in vitro.

In the former, ATL cells retain the ability to express Tax. A similar reversible type of suppression can be reproduced in vitro by co-culturing IL-2-dependent HTLV-1-infected cells with stromal cells, in which type I IFN responses occur.³⁵ Soluble type I IFN also suppresses HTLV-1 expression. IFN-stimulated genes such as PKR contribute to IFN-mediated suppression of Tax expression primarily at the post-transcriptional level.²⁰ This is consistent with the fact that pX mRNA is barely detectable in primary ATL cells despite undetectable Tax protein.³⁶ Thus, HTLV-1 expression in ATL cells in these cases is not absolutely silent. It remains unclear how Tax-specific CTL control ATL cells with such low levels of viral antigen in vivo. One possibility is the sensitivity of Tax-specific CTL, which can recognize much smaller amounts of antigen than those detectable by flow cytometry.³⁷ It is also conceivable that Tax-specific CTL may eliminate small numbers of Tax-expressing cells that are critical for the growth of the majority of ATL cells. Alternatively, ATL cells may express Tax for only a short period.³⁸

In contrast, ATL cells failing to express Tax cannot be a direct target of Tax-specific CTL. Genetic alteration and epigenetic silencing of HTLV-1 proviruses in ATL cells have been reported in such cases of ATL.³⁹ As these alterations are presumed to increase as the disease stage advances, Tax-targeting vaccines could be more effective in the earlier stages.

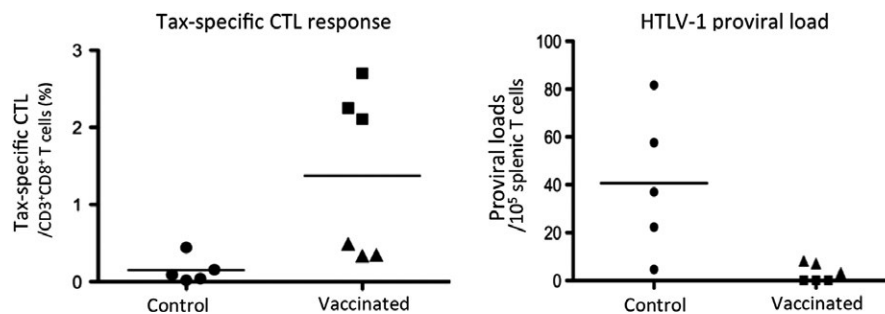


FIGURE 3 Reduction of proviral load by Tax-DC vaccine in a rat model of human T-cell leukemia virus type 1 (HTLV-1) carriers with impaired Tax-specific cytotoxic T-cell (CTL) response. In a rat model of immune tolerance to HTLV-1, oral HTLV-1 infection induces persistent HTLV-1 infection with impaired Tax-specific CTL responses and elevated proviral load (PVL). Tax-DC vaccine consisting of syngeneic bone marrow-derived dendritic cells pulsed with synthetic Tax peptide corresponding to the rat Tax-specific CTL epitope induced Tax-specific CTL responses (left), and reduced PVL (right) in the 3 wk following vaccination, supporting the notion of a potential prophylactic effect of the Tax-targeted vaccine on reducing the risk of adult T-cell leukemia/lymphoma (ATL) (modified from the original article published in the *Journal of Immunology*. Copyright 2017. The American Association of Immunologists, Inc.)⁵⁵

TABLE 3 Vaccine studies to induce HTLV-1-specific T-cell responses in animal models

Antigen	Adjuvant	Animal model	Induced immune response	Anti-HTLV-1/ATL effect in vivo	Reference
Expression vector containing wild type or mutant Tax cDNA	None	Uninfected rats	Tax-specific CTL	Adoptively transferred CTL eradicated HTLV-1-infected lymphoma in nude rats	Ohashi et al ⁵⁶
Epitope peptide of Tax-specific CTL from rats	CpG-ODN	Uninfected rats	Tax-specific CTL	Adoptively transferred CTL eradicated HTLV-1-infected lymphoma in nude rats	Hanabuchi et al ³²
Multivalent peptide consisting of three Tax peptides (tri-Tax) of HLA-A*0201-restricted CTL epitopes with TT3	Nor-MDP in squalene: aralcel A	Uninfected HLA-A*0201-Tg mice	CTL responses to each intended epitope	Reduction of viral replication after challenge with Tax-expressing recombinant vaccinia virus infection	Sundaram et al ⁵⁷
Envelope epitope peptide linked to MVF and multivalent tri-Tax epitope peptide	None	Uninfected squirrel monkeys	Antibody and T-cell responses	Protection of HTLV-1 infection in one of two monkeys after challenge with HTLV-1 infection	Kazanji et al ⁵⁸
HBc chimeric particle incorporating the HLA-A*0201-restricted HTLV-1 Tax-epitope	None	Uninfected HLA-A*0201-Tg mice	Tax 11-19- specific CTL	N.S.	Kozako et al ⁵⁹
Oligomannose-coated liposomes encapsulating Tax 11-19 peptide	None	Uninfected HLA-A*0201-Tg mice	Tax 11-19-specific CTL	N.S.	Kozako et al ⁶⁰
Tax 11-19 peptide with tetanus helper peptide	Incomplete Freund's adjuvant	Uninfected HLA-A2.1/CD11c-DTR-Tg mice	Tax-specific CTL	N.S.	Sagar et al ⁶¹
Recombinant vaccinia viruses expressing HBZ or Tax	None	Uninfected mice, HTLV-1-infected monkeys	HBZ or Tax-specific CTL	Adoptive transfer of HBZ-specific CTL increased survival of mice inoculated with HBZ-Tg mouse-derived lymphoma cells	Sugata et al ⁶²
Lentiviral vector encoding Tax, HBZ, p12 ^I , p30 ^{II}	None	Uninfected mice and rats	Cellular immune response	N.S.	Revaud et al ⁶³
Epitope peptide of Tax-specific CTL from rats	Dendritic cells	Orally HTLV-1- infected rats	Tax-specific CTL	Reduction of HTLV-1 proviral load	Ando et al ⁵⁵

CTL, cytotoxic T cells; DTR, diphtheria toxin receptor; HBc, hepatitis B virus core; HBZ, HTLV-1 basic leucine zipper; HLA, human leukocyte antigen; HTLV-1, human T-cell leukemia virus type 1; MVF, promiscuous T-helper cell epitope from measles virus fusion protein; N.S., not shown; Nor-MDP, N-acetyl-glucosamine-3-acetyl-L-alanyl-D-isoglutamine (muramyl dipeptide derivative); ODN, oligodeoxynucleotides; Tg, transgenic; TT3, promiscuous T-helper epitope from tetanus toxoid.

Additional candidate target antigens for anti-ATL vaccine to induce CTL may be cancer/testis antigens, as NY-ESO-1 mRNA is expressed in 61.4% of ATL cases.⁴⁰ It has been reported that HBZ could also be a target for CTL.⁴¹ However, experimentally induced HBZ-specific CTL hardly react with HTLV-1-infected cells or PBMC from HTLV-1-infected individuals,^{42,43} suggesting the scarcity of HBZ antigen presentation in HTLV-1-infected cells despite abundant HBZ mRNA expression.⁴⁴

7 | CONTROL OF IMMUNOSUPPRESSION AS A THERAPEUTIC OPTION

Tax-specific CTL are mostly undetectable or dysfunctional in ATL patients. In advanced stages of the disease, this may be partly

attributed to the generalized immunosuppression. However, impairment in the Tax-specific CTL response is also observed in early stages of ATL and even in a minor population of asymptomatic HTLV-1 carriers. In these individuals, the immunosuppression is selective against HTLV-1-specific responses, as CTL responses against other pathogens such as cytomegalovirus are comparable with HTLV-1-negative healthy individuals.²⁸

There are several possible mechanisms of immunosuppression in ATL. First, immune tolerance resulting from newborn infection or oral infection through the mother's milk may occur. Induction of HTLV-1-specific T-cell tolerance is well established in a rat model of oral HTLV-1 infection,⁴⁵ which partly explains the epidemiological finding that ATL preferably develops from individuals infected through the vertical route.⁴⁶ Second, T-cell exhaustion due to persistent viral infection is possible, as improvement of Tax-specific

CTL function by blockade of PD-1/PD-L1 interaction has been reported.⁴⁷ PD-L1 is expressed on ATL cells in 7.4% of ATL cases,⁴⁸ and alteration of this gene is frequently found in ATL cells.⁴⁹ Third, IL-10, an immunosuppressive/anti-inflammatory cytokine, is elevated in the serum of ATL patients.^{50,51} IL-10 also promotes proliferation of HTLV-1-infected cells, suggesting its active role in leukemogenesis.⁵²

Because restoration of Tax-specific CTL functions by Tax-DC vaccine seems essential to maintain remission in ATL patients, mitigation of immunosuppression is an important factor in anti-ATL therapy. In this regard, mogamulizumab and lenalidomide may be helpful because they reduce Treg and enhance T-cell immunity, respectively.^{14,18} Although these effects are not antigen-specific, mogamulizumab and lenalidomide may further enhance Tax-specific CTL responses when combined with Tax-targeted vaccine. Immune checkpoint inhibitors may also restore Tax-specific T-cell responses, although care should be taken as they may potentially restore other unwanted autoimmune responses. A recent report indicated that nivolumab unexpectedly promoted ATL progression in indolent ATL,⁵³ suggesting that the PD-1/PD-L1 system may still mediate a negative signal to ATL cells at indolent stages. Because ATL cells are a source of IL-10, decreasing ATL cells by various therapies may reduce IL-10 levels, thus alleviating some immunosuppression.

8 | TAX-TARGETED VACCINE AS A POTENTIAL EARLY AND/OR PROPHYLACTIC THERAPY

There is no approved therapy for indolent ATL in Japan. Intensive chemotherapy may not be suitable, as it induces immunosuppression. Chemotherapy is one of the factors for worse prognosis in

indolent ATL.⁵⁴ IFN- α /AZT is effective for indolent ATL, but requires continuous medication, as cessation of IFN- α /AZT allows relapse.²¹ Because Tax-silencing is presumed to be less frequent in earlier stages, a Tax-targeting vaccine may be a good candidate to treat indolent ATL.

It is also worth considering the use of Tax-targeted vaccination for disease prevention in HTLV-1 carriers at high risk of ATL. In a rat model of oral HTLV-1 infection, HTLV-1-specific T-cell tolerance was established with higher PVL than in rats infected through other routes, resembling the situation in HTLV-1 carriers with impaired CTL responses. This was resolved by Tax-DC vaccination, resulting in activation of Tax-specific CTL responses and reduced HTLV-1 PVL (Figure 3).⁵⁵ This observation supports the idea of prophylactic use of Tax-targeted vaccines in HTLV-1 carriers with impaired Tax-specific CTL responses.

Although it has shown promise, the prototype Tax-DC vaccines are available only for patients possessing HLA-A2, -A24 and -A11 alleles because of the HLA restriction of the CTL, and so further development is encouraged. Various types of vaccines have been reported to induce HTLV-1-specific T-cell response using animal models (Table 3). It should be taken into consideration, however, that HTLV-1 causes not only ATL but also inflammatory diseases such as HAM/TSP. It is unknown whether the responses caused by CTL and/or adjuvants may produce pathogenic inflammation. To develop a disease-preventive vaccine, it is important to define the HTLV-1-infected population at high risk of ATL.

9 | CONCLUSION

In conclusion, the Tax-DC vaccine study provides a new concept in ATL therapy that invokes the patient's own immunity, particularly Tax-specific CTL, following only a short course of vaccination. These CTL

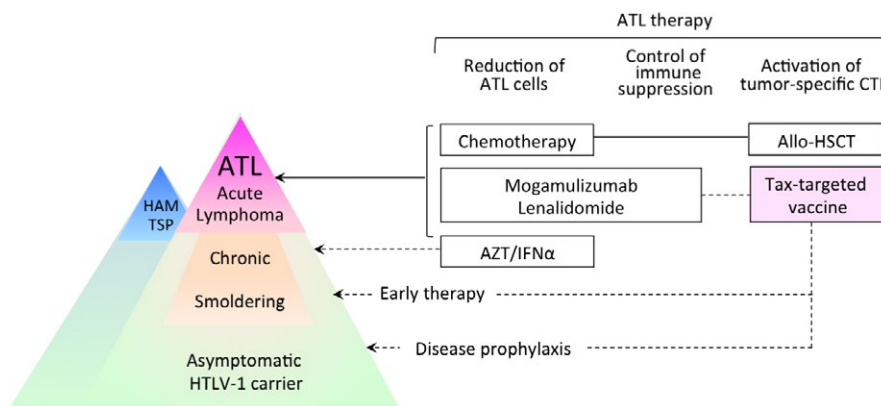


FIGURE 4 Potential position of the Tax-DC vaccine in currently available adult T-cell leukemia/lymphoma (ATL) therapies. Among currently available anti-ATL therapies, allogeneic hematopoietic stem cell transplantation (allo-HSCT) invokes host immunity against ATL, while other therapies reduce ATL cells. Mogamulizumab and lenalidomide also have immunomodulatory effects to support T-cell immunity without specificity. Tax-DC vaccine activates Tax-specific cytotoxic T cells (CTL) to maintain remission when used after other initial therapies. It may also be an appropriate therapy for earlier stages of ATL and potentially contribute to prophylactic strategies against ATL in the future, because of its limited toxicity. Approved (solid line) and not-approved (dotted line) usages in Japan are shown. HAM-TSP, HTLV-1-associated myelopathy/tropical spastic paraparesis; IFN- α /AZT, interferon- α and azidothymidine; HTLV-1, human T-cell leukemia virus type 1

contribute to long-lasting remission by continuously surveying HTLV-1-infected cells, although the CTL become ineffective when Tax-negative ATL cells proliferate in advanced stages. In addition, because of the limited toxicity, the timing of the Tax-targeted vaccine therapy may potentially be shifted to earlier stages of ATL (Figure 4), which may contribute to prophylactic strategies against ATL in the future.

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CONFLICT OF INTEREST

The authors declare no conflicts of interest for this article.

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