Personalized Medicine Universe xxx (2018) 1–6

Contents lists available at ScienceDirect

Personalized Medicine Universe

journal homepage: www.elsevier.com/locate/pmu

Review

Cloning and expansion of antigen-specific T cells using iPS cell technology: Possible use of regenerated T cells in personalized medicine

Hiroshi Kawamoto [a, *], Kyoko Masuda [a], Seiji Nagano [a, b]

[a] Laboratory of Immunology, Institute for Frontier Life and Medical Science, Kyoto University, Kyoto, Japan
[b] Department of Hematology and Oncology, Graduate School of Medicine, Kyoto University, Kyoto, Japan

Keywords:
Adoptive immunotherapy
iPS cells
CD8αβ T cells
WT1 antigen
Neoantigen

A B S T R A C T

Recent advances in adoptive immunotherapy using cytotoxic T lymphocytes (CTLs) have demonstrated that CTLs are effective in killing tumor cells *in vivo* for some tumor types. However, a critical issue that CTLs collected from patients are easily exhausted during expansion culture is yet to be addressed. Therefore, we have been developing a strategy that utilizes induced pluripotent stem cell (iPSC) technology, based on the idea that iPSCs produced from antigen-specific CTLs would regenerate CTLs with the same antigen specificity as the original CTLs. We previously succeeded in regenerating melanoma antigen MART1-specific CTLs, and more recently in producing potent CTLs expressing the CD8αβ heterodimer. We are now developing a novel method by which non-T derived iPSCs are transduced with exogenous T cell receptor (TCR) genes. If this method is applied to the allogeneic transfusion setting wherein HLA haplotype-homozygous iPSC stocks are used as the cell source, it will be possible to prepare "off-the-shelf" T cells. We are also considering incorporation of a personalized medicine approach to this allogeneic setting. In such a scheme, genes encoding TCRs specific for neoantigens will be collected from patients and HLA-homo iPSCs will be transduced with these TCR genes. Using such iPSCs, it will be possible to produce allogeneic CTLs expressing autologous TCRs originating from patients.

1. Current status and recent issues in cancer immunotherapy

Cancer immunotherapy has evolved remarkably during the past several years. Immune checkpoint blockade drugs such as anti-CTLA-4 monoclonal antibody (mAb) and anti-PD-1 mAb have shown therapeutic effects [1–3]. However, their therapeutic efficacy reaches only 20–30% and autoimmune-related adverse events are frequently observed as side effects [4].

Delivery of autologous CTLs directly into patients has also shown good results. For example, Rosenberg and colleagues have demonstrated that transfusion of *ex-vivo* expanded tumor-infiltrating lymphocytes (TILs) was effective in melanoma patients [5]. Transfer of T cell receptor (TCR) genes such as NY-ESO1 antigen-specific one, into patients' peripheral T cells also achieved good clinical outcomes [6–8], indicating that targeting a single antigen can be effective for some types of cancer. In addition, T cells enforced to express the chimeric antigen receptor (CAR) have shown enormous efficacy in the treatment of B cell leukemia [9].

In both TCR and CAR engineering, peripheral T cells are transduced by a retrovirus, introducing the risk of tumorigenicity due to the random genomic integration of the transfected gene. Moreover, in the autologous setting, it would be costly to produce T cells, and it is not always easy to obtain potent CTLs from patients.

2. Application of reprogramming technologies for the cloning and expansion of T cells

To address the aforementioned problems, we have thought of a method by which T cells can be expanded. When embryonic stem cells (ESCs) or iPSCs are generated from T cells, the genomic structure of rearranged T cell receptor (TCR) genes should be passed on to the resulting ESCs/iPSCs, and T cells regenerated from these ESCs/iPSCs should express the same TCR as the original T cells

* Corresponding author. 53 Kawahara-cho, Shogoin, Sakyo-ku, Kyoto 606-8507, Japan. Fax: +81 75 751 3839.
E-mail address: kawamoto@infront.kyoto-u.ac.jp (H. Kawamoto).

https://doi.org/10.1016/j.pmu.2018.05.002

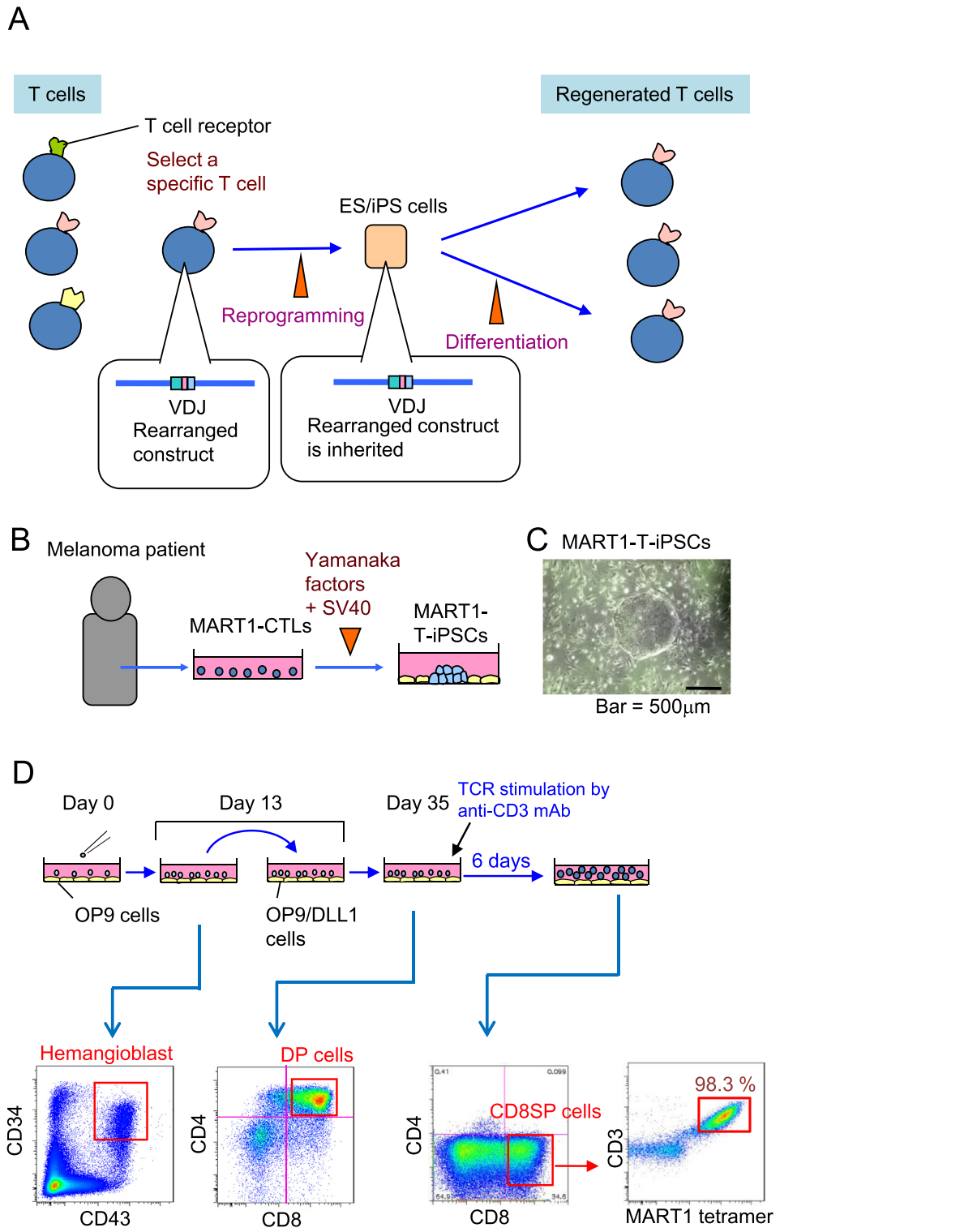

Fig. 1. Regeneration of T cells using iPSC technology. A. The concept of T-iPSC therapy. When T-iPSCs are established from tumor antigen-specific T cells, the rearranged configuration of TCR genes is passed on to iPSCs. T cells regenerated from these T-iPSCs express the same tumor antigen-specific TCRs. B. We established MART1-specific T-iPSCs by transducing MART1-specific T cells derived from a melanoma patient with Yamanaka factors and SV40 using the Sendai virus vector system. C. A colony of MART1-T-iPSCs. D. MART1-iPSCs were sequentially cultured with two types of feeder cells, OP9 and OP9/DLL1. On Day 13, $CD34^{+}CD43^{+}$ hemangioblasts were generated, and on Day 35, CD4/CD8 double-positive (DP) cells were generated, when anti-CD3 mAb was added to induce the generation of mature T cells. CD8 single-positive cells were generated 6 days after anti-CD3 mAb stimulation. Virtually all CD8 single-positive cells expressed a TCR specific for MART1 antigen.

Please cite this article in press as: Kawamoto H, et al., Cloning and expansion of antigen-specific T cells using iPS cell technology: Possible use of regenerated T cells in personalized medicine, Personalized Medicine Universe (2018), https://doi.org/10.1016/j.pmu.2018.05.002