

Transplantation of Human Peripheral Stem and Progenitor Cells to Humanized Mouse Model for Systemic Lupus Erythematosus

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Introduction: Systemic Lupus Erythematosus (SLE) is an autoimmune disease characterized by damaged and dysregulated immune system due to breakdown in the selection process during clonal growth of immune cells. Studies have shown that patients with systemic lupus erythematosus (SLE) display altered gene expression patterns and increased double-stranded DNA breaks within their hematopoietic stem and progenitor cells (HSPC). However, the current animal models for SLE found in the existing literature predominantly emphasize the use of peripheral blood mononuclear cells (PBMC) over HSPC for the creation of humanized mouse models. Nevertheless, these prior models were constrained by the limited efficiency of human cell engraftment and limited PBMC ability to replicate the capacity of HSPC to generate human SLE cells that can engraft host mice, thus making the transplant protocol inadequate.

Patients and Methods: Transplantation was initiated by extracting HSPC from stable SLE patients by leukapheresis. The collected cells were assessed for purity before storage at -80°C . Humanized mice were conditioned with cyclophosphamide and total-body irradiation before receiving the HSPC transplant. After transplantation, the mice were administered human granulocyte-colony stimulating factor and sacrificed to evaluate autoantibodies and HSPC in their bone marrow and blood samples. Statistical analysis was performed using Student's *t*-test and one-way ANOVA.

Results: Upon human stem cells engraftment into mice, we found a noteworthy presence of HSPC, as evidenced by the positive expression of hCD45, hCD34, and/or hCD44, and the production of human antinuclear antibodies. The results indicated that the transplanted mice generated reactive autoantibodies against human cells, similar to that observed in the human donor patient. These findings shed light on the survival and behavior of transplanted human stem cells in a mouse model.

Conclusion: In this study, we successfully induced immunodeficiency in mice for xenotransplantation with human peripheral stem cells.

Keywords: autoimmune, experimental, humanized mouse, stem cells

Introduction

Systemic Lupus Erythematosus (SLE) is an autoimmune disease characterized by the destruction of the patient's healthy cells and tissues caused by their immune response. During clonal growth of immune cells, a breakdown in the selection process can give rise to abnormal immune cells that have the potential to target healthy cells. It leads to a damaged and dysregulated immune system and the subsequent development of SLE, an autoimmune disorder. Ongoing research is still striving to gain comprehensive insights into the specific role of hematopoietic stem cells in SLE.¹⁻³ There is evidence suggesting that these cells may significantly impact the development of SLE in humans, particularly concerning their aging process.⁴ The loss of self-tolerance leads to an autoimmune response against the cells and tissues. Another study suggested that hematopoietic stem and progenitor cells (HSPC) in SLE patients exhibit significant changes in gene expression and increased dsDNA breaks.⁵ Developing effective treatments for systemic lupus erythematosus is challenging owing to the limited specificity in targeting aberrant immune cells, as the precise pathogenesis of the disease

remains unclear. Mouse model is needed to gain a better understanding of the disease and to test treatment options without directly involving human subjects. The development of a human disease model typically involves the use of experimental animals with appropriate characteristics and conditions. Noteworthy advancements have been achieved in comprehending the pathogenesis of Systemic Lupus Erythematosus (SLE) by conducting research on spontaneous or artificially induced mouse models that are prone to developing lupus.^{6–8}

Despite distinct difference in genetic background, mouse models have been invaluable in exploring the genetic and environmental factors underlying autoimmunity and in testing novel therapies that can then be translated to clinical trials in human patients. At present, two primary methods are employed in the construction of humanized mouse models of SLE. The methods currently being explored are either the transfer of peripheral blood mononuclear cells (PBMCs) from individuals with systemic lupus erythematosus (SLE) to immunodeficient mice or the utilization of human hematopoietic stem cells (HSCs) in immunodeficient mice, followed by an intraperitoneal (i.p.) injection with pristane to trigger lupus.⁶ In vivo investigations of human immunology have been made easier by the use of humanized mice, which successfully reassemble human immune systems in immunodeficient animals. This is particularly useful when studying human-specific infectious illnesses and cancer. Various studies have been conducted to investigate the pathogenesis of human SLE in animal model; however, the usage of HSPC humanized mouse model was less often mentioned in the literature compared to PBMC humanized mouse models.^{9–11}

However, these prior models were limited by the insufficient efficiency of human cell engraftment, the requirement for substantial quantities of PBMCs from SLE patients who frequently experience lymphopenia, or the lack of human anti-nuclear autoantibody production. Consequently, the utilization of HSPCs as an alternative to PBMCs may prove advantageous due to the self-renewal and differentiation capabilities of HSPCs, potentially necessitating fewer cells from patients compared to PBMCs. Furthermore, HSPCs possess the capacity to generate a more diverse range of immune cell types, potentially offering a more comprehensive representation of human SLE pathogenesis.^{5,7}

Materials and Methods

HSPC Preparation

SLE patient with active disease was invited to voluntarily contribute their peripheral blood stem cells through a secure leukapheresis procedure. Upon obtaining their consent, selected patient underwent comprehensive assessments of physical status, complete blood counts, and coagulation assays both before and after the leukapheresis procedures, ensuring the continuous safety and well-being of the participating patient. Selected patient is 22-year-old female who presented with symptoms consistent with SLE such as seizure, oral ulcer, pleural effusion, and alopecia. This resulted in a EULAR/ACR total score of 18, classifying her as having systemic lupus erythematosus according to EULAR/ACR criteria with SLEDAI score 19. She had recently diagnosed with SLE and her condition was stable during leukapheresis procedure. Patient's lab profile is provided in the [Supplementary 1](#). The volunteer subsequently underwent apheresis at the Haemonetics MCS-9000 Apheresis Unit, utilizing a specialized stem cell collection bag unit configured according to the protocol for collecting HSPC. The collected bag was further processed to eliminate platelet and other non-mononuclear cell contamination through gradient separation, following the manufacturer's protocol (Lymphoprep). Subsequently, the mononuclear layers were isolated and stored at -80°C in Cell Banker freezing medium.

Humanized Mice Conditioning and Transplantation Process

Mice were procured from the veterinary pharmacy center, in Surabaya, Indonesia, and acclimated in our local cages for two weeks. The 6- to 8-week-old female ddY mice were specifically used for this model, accommodated in a meticulously controlled negative-pressure environment within the animal facilities at the Faculty of Medicine, Brawijaya University. The procedures strictly adhered to the protocols established by the Research and Ethics Committee of Saiful Anwar Hospital. Specifically, 6- to 8-week-old female ddY mice underwent intraperitoneal (i.p.) injections of cyclophosphamide at a dosage of 80 mg per kilogram of body weight, administered twice with a one-week interval. Following this, Total Body Irradiation (TBI) was conducted using Cobalt-60 isotope at a dose of 5 Gy, administered 24 hours after the last cyclophosphamide injection. After a post-irradiation period of 60 days, the transplanted mice underwent a tail vein

injection of 5×10^6 peripheral blood stem cells obtained from an SLE patient, while the wild-type (WT) mice received i.p. injections of PBS. Following the aforementioned procedures, the treated mice were additionally administered human granulocyte-colony stimulating factor (G-CSF/Leukogen) at a dosage of 100 $\mu\text{g/kg}$ body weight per day. This supplementation was commenced on the same day as the peripheral blood stem cell injections and was consistently administered at the same dosage for a total duration of 5 days. At 48-hour and 7-day intervals post-peripheral blood stem cell injection, the mice were humanely euthanized. Subsequently, blood samples were collected, and bone marrow was extracted for analysis utilizing the Sysmex XN-3000 hematology analyzer (Sysmex, Japan) and the Beckman-Coulter Navios flow cytometer (Beckman Coulter).

Stem Cells Evaluation in Humanized Mice

Blood was acquired from the mice through cardiac puncture as part of the terminal procedure, which was conducted under anesthesia using isoflurane. Peripheral complete blood counts were determined using the Sysmex XN-3000 hematology analyzer (Sysmex, Japan). To obtain serum, blood samples were placed in SST tubes (SST 3 Vacutainer, BD Biosciences, USA) and allowed to clot for 30 minutes at room temperature. Subsequently, the samples were centrifugation at 4400 rpm for 15 minutes at room temperature. The resulting serum was then subjected to analysis for the human anti-nuclear antibody (Maglumi X3, Snibe, Shenzhen) and anti-dsDNA antibody (Alegria ORG204S, Orgentec, German). Additionally, the serum was subjected to mouse anti-nuclear antibody and anti-dsDNA antibody analysis using assays provided by Eiyue, China (cat# FY-EM6379, cat# FY-EM6458).

Bone marrow was harvested by flushing the marrow of a single femur from each mouse, followed by resuspension, which was achieved through repeated passage through an 18-gauge needle. Subsequently, both blood and bone marrow cells were subjected to staining for 30 minutes at 4°C in the absence of light. Anti-human antibodies, including CD45-APC-Cy7, CD34-FITC, and CD44-PE (Biolegend, USA), were added to the cell suspension and were stained according to the manufacturer's instructions. The stained cells were analyzed on a Cytotflex flow cytometer (Beckman Coulter) using FlowJo version 10 (FlowJo, Beckton-Dickinson, OR, USA). All procedures have been approved by Dr. Saiful Anwar General Hospital Medical Research Ethical Committee(400/281 /K.3/302/2023). This study was conducted in accordance with the principles outlined in the Declaration of Helsinki, and participant provided written informed consent prior to their inclusion in the research.

Statistical Analysis

The Student's *t*-test and one-way ANOVA were employed for comparisons using Prism version 9 from GraphPad Software, San Diego, CA. The data are presented as mean $\pm$ SEM. In the figures, statistical significance is denoted as $*p < 0.05$.

Results

Mice Subjected to Pre-Treatment Involving a Combination of Cyclophosphamide and Total Body Irradiation Exhibited the Development of Human Stem Cells Following Xenotransplantation

To induce immunodeficiency and facilitate xenotransplantation of human cells, ddY mice underwent two administrations of cyclophosphamide combined with 5 Gy of total body irradiation (TBI). We introduced 5×10^6 cells of HSPC through the tail vein of pre-treated mice, accompanied by an additional intraperitoneal injection of G-CSF. One week was allocated to evaluate the homing capability of stem cells to the bone marrow and the autoantibody production of the transplanted cells (Figure 1). At 48 hours post HSPC infusion, a subset of mice was euthanized to assess the presence of human stem cells in both peripheral blood circulation and the bone marrow compartment.

In comparison to the wild-type (WT), the mice subjected to pre-treatment and HSPC transplantation demonstrated a noteworthy presence of human stem cells and progenitor cells, as evidenced by the positive expression of hCD45, hCD34, and hCD44 (Figure 2A and B).

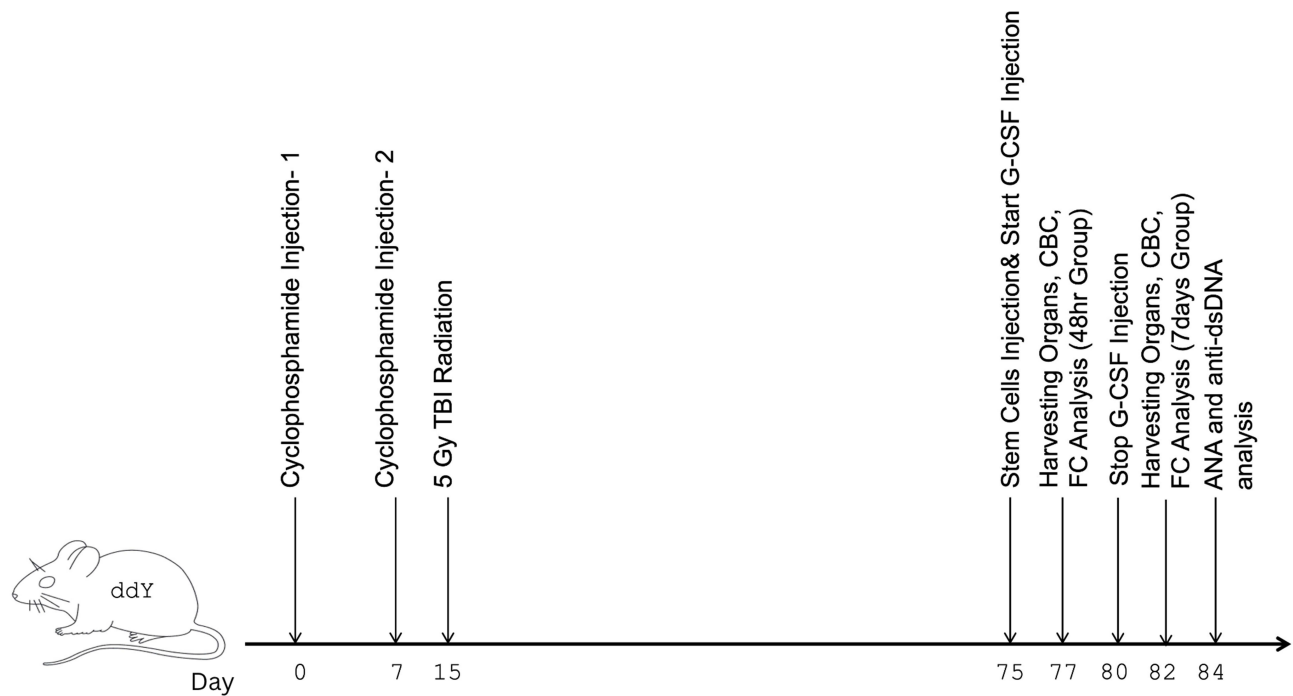


Figure 1 Research Timeline. Mice were conditioned with 2 doses of cyclophosphamides, TBI following SC injection intravenous in mice's tails and G-CSF Injection. 48 hours and 7 days later mice were sacrificed and evaluated for human cells and autoantibody production.

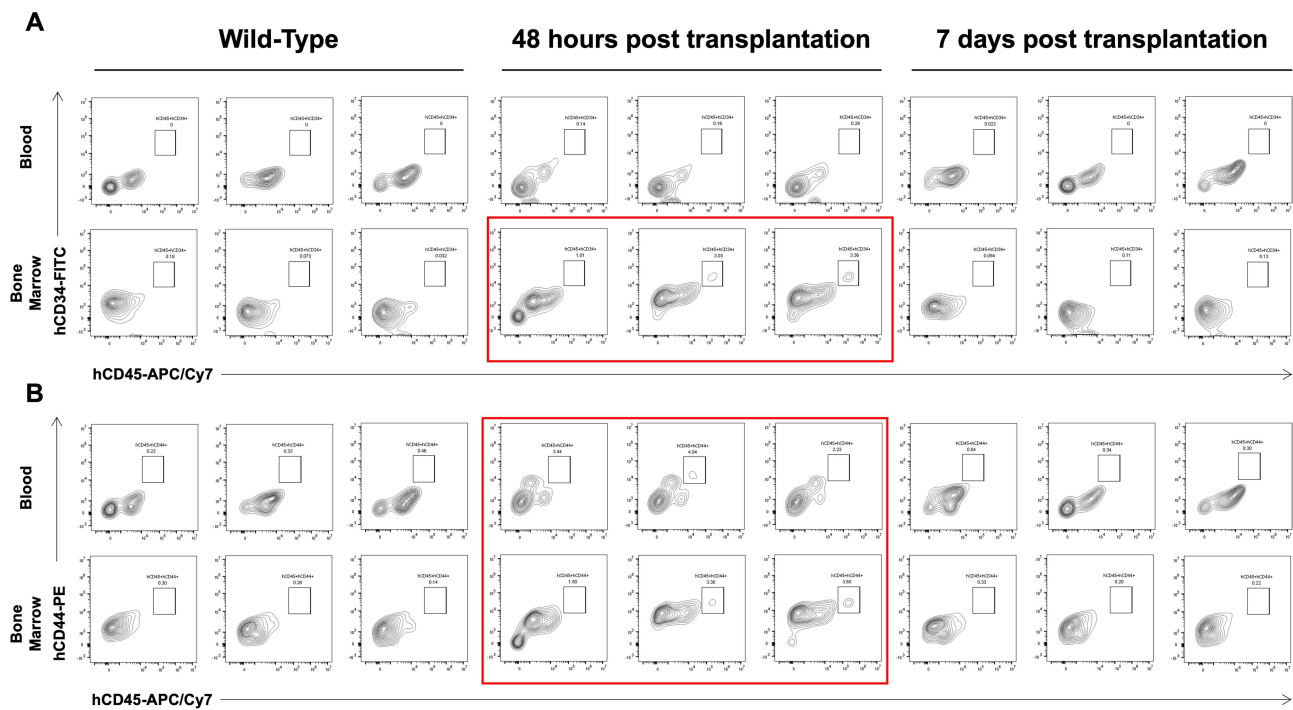


Figure 2 Scattergram for Flow-cytometry for (A) CD34⁺CD45⁺ expression and (B) CD44⁺CD45⁺ expression for 3 different mice groups namely wild-type (without any treatment), humanized mice (transplanted with HSPC) at 48 hours and at 7 days post-transplantation. For each group there are 2 types of samples (peripheral blood and bone marrow) from 3 mice. Interestingly the human HSPC population was detected strongly in the 48 hours after transplantation indicate by red boxes.

Interestingly, upon withdrawal of G-CSF after 5 days, the presence of human stem cells in both compartments reverted to baseline levels, akin to the WT mice. Further examination of peripheral blood smears revealed distinctive characteristics of blood cells. The native lymphocytes from wild-type mice (Figure 3A) exhibited significantly different

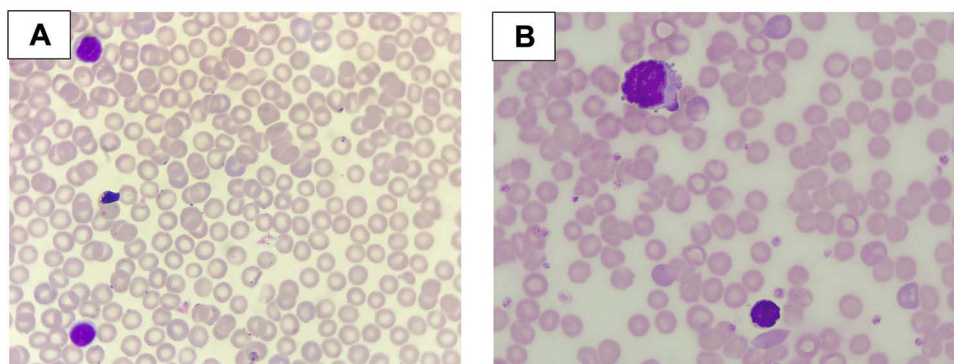


Figure 3 Blood smear of mice showing the morphology of mice lymphocytes in (A) from wild-type mice that starkly contrasted with the morphology of transplanted human stem cells (B) from humanized mice model.

morphological characteristics compared to the transplanted human stem cells in recipient mice, which were distinguished by their large size, basophilic cytoplasm, and occasional membrane blebbing (Figure 3B).

Transplanted Mice Develop Severe Cytopenia

Following treatment and transplantation with HSPC, the mice experienced significant reductions in white blood cell and platelet counts, as depicted in Figure 4A–C. Although there was a slight decrease in hemoglobin levels, it did not reach statistical significance (Figure 4A). During the same period, the white blood cell count decreased by approximately 50% from its baseline level (Figure 4B), and the platelet count experienced a substantial decrease, becoming completely undetectable by day 7 (Figure 4C).

Autoantibodies Targeting Human Nuclear Antigens and dsDNA Were Identified in Transplanted Mice

To assess whether the graft cells generate reactive autoantibodies against human cells, as observed in donor patients, we subjected the mice serum to test for human anti-nuclear antibody (ANA) and human anti-double stranded Deoxyribonucleic acid (anti-dsDNA) antibody. The donor samples had previously tested positive for the ANA test but negative for the anti-dsDNA antibody. The human ANA level was found to be higher in transplanted mice than in wild-type (WT) (Figure 5A), while the human anti-dsDNA antibody levels remained similar (Figure 5B).

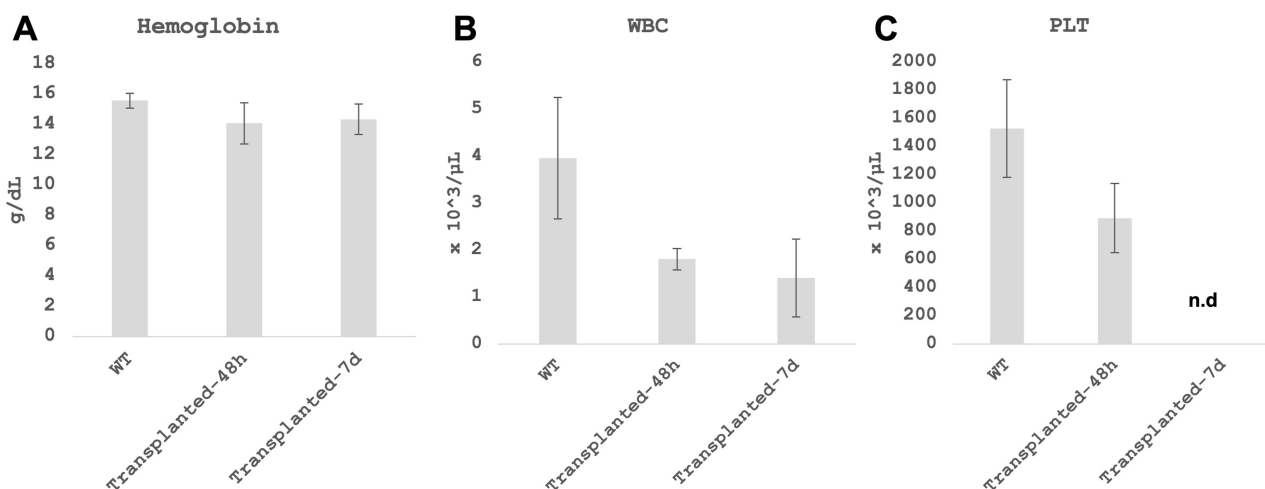


Figure 4 CBC Profile of humanized mice compare to wild-type (control) group (A) Hemoglobin, (B) WBC, (C) Platelet. n.d. denotes not detected.

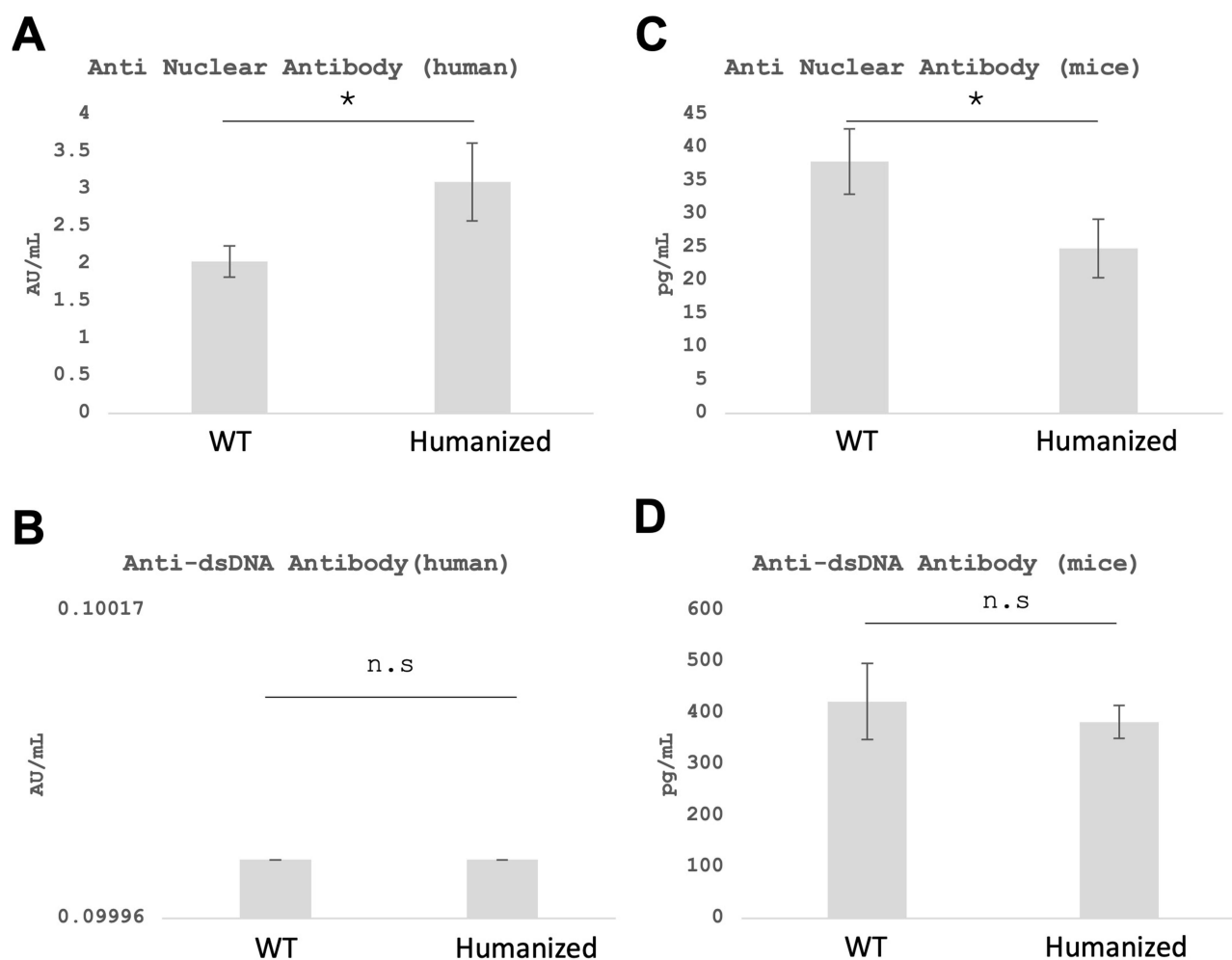


Figure 5 Serological Profile of Humanized Mouse Model. (A and B) indicating anti human antibody, (C and D) indicating anti mice antibody. Statistical significance is denoted as * $p < 0.05$.

To confirm the specificity of reactivity to human cells, we additionally conducted enzyme-linked immunosorbent assay (ELISA) tests to detect mouse anti-nuclear antibodies and mouse anti-dsDNA. In contrast to the human antibody findings, the concentration of mouse ANA was markedly lower in transplanted mice than in WT (Figure 5C). Meanwhile, there was no significant difference in mouse anti-dsDNA antibody levels (Figure 5D). Together with the presence of viable B-cells from the donor, these results suggest that graft stem cells have survived within the host mice, resulting in the production of a substantial level of autoantibodies reactive to human nuclear antigens, in line with the observed patient profile.

The Presence of Granulocyte-Colony Stimulating Factor (G-CSF) Was Supporting the Survival of Graft Cells Within the Host Mice

Administering a daily subcutaneous injection of 100 μ g per kilogram of body weight to host mice appeared to enhance the survival of transplanted human stem cells. At 48 hours post-transplantation, human stem cells were readily identified in both peripheral blood and bone marrow. After discontinuing G-CSF administration on day 5, the presence of human stem cells in both peripheral blood and bone marrow was significantly reduced, indicating that G-CSF was supporting the expansion and mobilization of human stem cells in host mice (Figure 1).

Discussion

The excessive production of antinuclear autoantibodies (ANA) leads to the formation of immune complexes, triggering inflammation that lasts chronically, which is a defining characteristic of SLE.¹² However, it is essential to continue exploring the correlation between various clinical symptoms and laboratory test findings.¹³ The significance of using animal models in studying SLE is underscored by the ongoing uncertainty regarding its exact cause.¹⁴ Our study has effectively created immunodeficiency in ddY mice for xenotransplantation with human peripheral stem cells. This was confirmed by a 50% decrease in the white blood cell count compared to the wild-type mice. Following xenotransplantation, these mice exhibited a significant presence of human stem cells, demonstrated by positive expressions of hCD45, hCD34, and/or hCD44. This finding is in agreement with that of Andrade et al, who also showed that humanized mouse models of systemic lupus erythematosus (SLE) have shown the presence of human CD45⁺ cells in their mouse model.¹¹ Human CD34⁺ hematopoietic stem cells were found after transplanting mice into another study that created an HSC-pristane-humanized SLE mouse model, indicating successful reconstitution of the human immune system.⁷

Moreover, the presence of cytopenia in humanized mouse models helps replicate the haematological abnormalities seen in SLE patients, enhancing the model's relevance for studying the disease pathogenesis and testing potential treatments.⁶ Seven days after transplantation and cessation of G-CSF administration, there was a notable decrease in the quantity of transplanted human stem cells (hCD45⁺, hCD34⁺, and hCD44⁺) in both peripheral blood and bone marrow. Therefore, it can be inferred that G-CSF plays a vital role in the proliferation and mobilization of human stem cells in host mice. This finding is in agreement with findings from Patterson & Pelus in which 90% of the hematopoietic stem cells were depleted in the bone marrow, but they still suspected localization to the spleen and thus can escape collection.¹⁴

One sign of resemblance to SLE in the transplanted mice is the presence of human antinuclear antibodies (ANA). In contrast to the WT, these humanized mice developed autoantibodies against human nuclear antigens, which mirrored the autoantibody profile of SLE patients in our investigation. Both groups exhibited comparable levels of mouse anti-dsDNA antibodies; however, the mouse ANA demonstrated lower antibody levels in the transplanted group, potentially due to the impaired ability of the mice's immune cells to produce sufficient antibodies. This phenomenon may be attributed to the transplanted group undergoing multiple treatments prior to transplantation, which weakened their immune system. Additionally, the human anti-dsDNA exhibited consistent serological patterns with the SLE patient who provided the stem cells. These results, along with the fact that humanized peripheral blood smears contain viable stem cells, imply that the transplanted stem cells survived in the host mice and produced a significant amount of autoantibodies reactive to human nuclear antigens that are in line with the patient profile that has been observed. A study carried out by Gunawan et al on mice also demonstrated that humanized mouse models of systemic lupus erythematosus (SLE) yielded positive serum antinuclear antibody (ANA) test results. This result highlights the ability of these models to replicate important immunological aspects of SLE, which is indicative of a breakdown in immune tolerance and a surplus of autoantibodies.⁷

The current study is subject to a few limitations. Firstly, the number of eligible donors is limited due to the current scarcity of SLE patients meeting the criteria. Additionally, this research did not explore the tissue phenotype of the humanized model, leaving uncertain whether inflammation in the tissue or leukocyte homing in the secondary lymphoid organs took place. The suspected homing leukocyte is similar to Kokkinopoulos et al finding who noted a significant alteration in the homing patterns of CD34⁺ cells derived from the peripheral blood of patients with systemic lupus erythematosus (SLE), suggesting the potential migration of these cells in secondary lymphoid organs.¹ These constraints mean that additional studies on a larger number of SLE donor samples with different immunological serological profiles (ex: anti-dsDNA) are required to compare whether the mice's serological profile consistently resembles that of the donor. To check for any indications of inflammation or leukocyte homing in the kidney, spleen, and lymph nodes, we are also thinking of evaluating these organs.

Conclusion

Systemic lupus erythematosus is an autoimmune disease that destroys healthy cells and tissues. Ongoing research aims to elucidate the role of hematopoietic stem cells in systemic lupus erythematosus (SLE) and to understand the role of hematopoietic stem cells in SLE, which may impact its development in humans. Recent studies on animal models have

shown a preference for using peripheral blood mononuclear cells (PBMCs) over hematopoietic stem and progenitor cells (HSPCs) in creating humanized mouse models. Consequently, there is limited information available on the development of animal models for experimental purposes using HSPCs. This study successfully induces immunodeficiency in ddY mice for xenotransplantation with human peripheral stem cells, revealing a 50% decrease in white blood cell count compared to wild-type mice. Upon transplantation of human stem cells into mice, we found a noteworthy presence of HSPC and the production of human ANA antibody this is similar to the serological profile of human donors. Cessation of HSPC after termination of G-CSF injection showing the importance of G-CSF for the proliferation and mobilization of human stem cells in host mice. In conclusion, this humanized mouse model shows a potential for a disease model that may represent clinical variation among SLE patients. This model may be used for a thorough investigation of the underlying mechanisms of disease pathogenesis in SLE. Nevertheless, a great deal more work is required to create a humanized mouse model through the procedure of transplanting human stem cells into host mice.

Abbreviations

PBMC, peripheral blood mononuclear cells; HPSC, hematopoietic stem and progenitor cells; SLE, systemic lupus erythematosus; ANA, anti-nuclear antibody; ANOVA, Analysis of Variance; G-CSF, Granulocyte-colony stimulating factors; HSC, hematopoietic stem cells; i.p, intraperitoneal; PBS, phosphate buffered saline; mononuclear cells; dsDNA, double-stranded Deoxyribonucleic acid; TBI, total body irradiation.

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Disclosure

The authors report no conflicts of interest in this work.

References

1. Kokkinopoulou I, Banos A, Grigoriou M, et al. Patrolling human SLE haematopoietic progenitors demonstrate enhanced extramedullary colonisation; implications for peripheral tissue injury. *Sci Rep.* **2021**;11(1):1–14. doi:10.1038/s41598-021-95224-y
2. Hormaechea-Agulla D, Le DT, King KY. Common sources of inflammation and their impact on hematopoietic stem cell biology. *Curr Stem Cell Rep.* **2020**;6(3):96–107. doi:10.1007/s40778-020-00177-z
3. Tang X, Wang Z, Wang J, Cui S, Xu R, Wang Y. Functions and regulatory mechanisms of resting hematopoietic stem cells: a promising targeted therapeutic strategy. *Stem Cell Res Ther.* **2023**;14(1):1–18. doi:10.1186/s13287-023-03316-5
4. Gao L, Slack M, McDavid A, Anolik J, Looney RJ. Cell senescence in lupus. *Curr Rheumatol Rep.* **2019**;21(1):1–8.
5. Chen J, Liao S, Zhou H, et al. Humanized mouse models of systemic lupus erythematosus: opportunities and challenges. *Front Immunol.* **2022**;12:816956. doi:10.3389/fimmu.2021.816956
6. Gunawan M, Her Z, Liu M, et al. A novel human systemic lupus erythematosus model in humanised mice. *Sci Rep.* **2017**;7(1):1–11. doi:10.1038/s41598-017-16999-7
7. Alves da Costa T, Lang J, Torres RM, Pelanda R. The development of human immune system mice and their use to study tolerance and autoimmunity. *J Transl Autoimmun.* **2019**;2:100021. doi:10.1016/j.jtauto.2019.100021
8. Shultz LD, Lyons BL, Burzenski LM, et al. Human lymphoid and myeloid cell development in NOD/LtSz-scid IL2R γ null mice engrafted with mobilized human hemopoietic stem cells. *J Immunol.* **2005**;174(10):6477–6489. doi:10.4049/jimmunol.174.10.6477
9. Mauermann N, Sthoeger Z, Zinger H, Mozes E. Amelioration of lupus manifestations by a peptide based on the complementarity determining region 1 of an autoantibody in severe combined immunodeficient (SCID) mice engrafted with peripheral blood lymphocytes of systemic lupus erythematosus (SLE) patients. *Clin Exp Immunol.* **2004**;137(3):513–520. doi:10.1111/j.1365-2249.2004.02559.x
10. Andrade D, Redecha PB, Vukelic M, et al. Engraftment of peripheral blood mononuclear cells from systemic lupus erythematosus and antiphospholipid syndrome patient donors into BALB-RAG-2 γ -IL-2R γ mice: a promising model for studying human disease. *Arthritis Rheum.* **2011**;63(9):2764–2773. doi:10.1002/art.30424
11. Ameer MA, Chaudhry H, Mushtaq J, et al. An overview of systemic lupus erythematosus (SLE) pathogenesis, classification, and management. *Cureus.* **2022**;14(10):1.
12. Lam NCV, Brown JA, Sharma R. Systemic lupus erythematosus: diagnosis and Treatment. *Am Fam Physician.* **2023**;107(4):383–395.
13. Huang X, Zhang Q, Zhang H, Lu Q. A contemporary update on the diagnosis of systemic lupus erythematosus. *Clin Rev Allergy Immunol.* **2022**;63(3):311–329. doi:10.1007/s12016-021-08917-7
14. Patterson AM, Pelus LM. G-CSF in stem cell mobilization: new insights, new questions. *Ann Blood.* **2017**;2(5):10. doi:10.21037/aob.2017.06.02

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