Leucodepletion and immune response mechanisms

J. W. Semple

Department of Laboratory Medicine & Pathobiology, St. Michael’s Hospital, Departments of Pharmacology, Medicine & Laboratory Medicine & Pathobiology, The University of Toronto & The Toronto Platelet Immunobiology Group, Toronto, Canada

Introduction

The recipient and donor mechanisms responsible for mediating alloimmunity and immunomodulation during blood transfusions are complex and most probably superimposed thus creating multilevel fields of potential interaction and regulation. For example, at the recipient level, both the innate and adaptive immune systems can be significantly affected by various types of transfusions [1,2]. In addition, the health status of the recipient plays a critical role in determining how the host will immunologically respond to transfusion. On the other hand, donor transfusion products have many characteristics that can potentially influence recipient immunity and its regulation (e.g. age, leucocyte content, etc.). Understanding these varied parameters culminate in an immune response or not is fundamental to developing safer blood products and better care for recipients.

Many prospective randomized studies have shown that leucoreduction of blood components can reduce the incidence of alloimmunization and platelet refractoriness [3]. Mainly based on these data, leucoreduced components are now recommended for any patient requiring long-term transfusion support such as patients with leukaemia, lymphoma, myelodysplasia or those undergoing stem cell transplantation. Although leucoreduction has shown a clear benefit by reducing the incidence of alloimmunization and platelet refractoriness [3]. Mainly based on these data, leucoreduced components are now recommended for any patient requiring long-term transfusion support such as patients with leukaemia, lymphoma, myelodysplasia or those undergoing stem cell transplantation. Although leucoreduction has shown a clear benefit by reducing the incidence of alloimmunization and platelet refractoriness [3]. Mainly based on these data, leucoreduced components are now recommended for any patient requiring long-term transfusion support such as patients with leukaemia, lymphoma, myelodysplasia or those undergoing stem cell transplantation. Although leucoreduction has shown a clear benefit by reducing the incidence of alloimmunization and platelet refractoriness [3]. Mainly based on these data, leucoreduced components are now recommended for any patient requiring long-term transfusion support such as patients with leukaemia, lymphoma, myelodysplasia or those undergoing stem cell transplantation. Although leucoreduction has shown a clear benefit by reducing the incidence of alloimmunization and platelet refractoriness [3]. Mainly based on these data, leucoreduced components are now recommended for any patient requiring long-term transfusion support such as patients with leukaemia, lymphoma, myelodysplasia or those undergoing stem cell transplantation. Although leucoreduction has shown a clear benefit by reducing the incidence of alloimmunization and platelet refractoriness [3].

Correspondence: John W. Semple, Department of Laboratory Medicine and Pathobiology, St. Michael’s Hospital, 30 Bond St., Toronto, Ontario, M5B 1W8, Canada
E-mail: semplej@smh.toronto.on.ca
in mice translates to an approximate dose in humans of $2.5 \times 10^6$ leucocytes per transfusion. Clinical studies in leukaemic patients subsequently suggested that the minimal threshold of leucocyte contamination in blood products to prevent alloimmunization should be less than $1.5 \times 10^6$ leucocytes [3]. However, several reports subsequently reported that in healthy mice, rats and humans transfused with leucoreduced allogeneic platelets, leucocyte levels as low as 1/µl (approximately $3 \times 10^5$ per transfusion) not only activated recipient T cells [9] but also stimulated IgG antidonor alloantibody responses [10–12].

In 1998, Kao et al. [13] demonstrated that when MHC class II positive APC were depleted from murine peripheral blood mononuclear cells and 10$^5$ depleted cells were transfused weekly into fully allogeneic recipients, the anti-MHC class I antibody response was significantly reduced. This suggests that when relatively small doses of MHC class I molecules are transfused, direct allorecognition of MHC class II + leucocytes may be necessary to enhance the recipient alloimmune response. However, this may not be the case for transfused allogeneic platelets since they themselves deliver a significantly higher dose of donor MHC class I molecules compared with leucocyte transfusions. This exposes the recipient to high doses of MHC class I molecules that can significantly stimulate alloimmunity via indirect allorecognition. We addressed this by using mice with severe combined immunodeficiency (SCID) as platelet donors. SCID mice do not have circulating T or B cells and thus platelets prepared from their plasma can be consistently rendered extremely leucoreduced (< 0.05 WBC/µl) [14]. The SCID mouse platelets expressed high levels of MHC class I molecules and despite undetectable MHC class II positive APC, they were significantly more immunogenic than control platelets containing 1 leucocyte/µl when transfused into allogeneic CBA recipients. Thus, as the class II positive APC numbers are lowered within the platelet product, a biphasic anti-MHC class I antibody response is observed (Fig. 1). This response pattern suggests that leucoreduction may be an active process in that it produces a dose of MHC class II positive APC that can suppress the antibody response against platelets. When these cells are totally depleted, the immunosuppression is apparently relieved and indirect allorecognition of donor platelet antigens proceeds unchecked. The recipient immune mechanism(s) responsible for this WBC-induced reduction of platelet immunity are unknown but may include the long-term engraftment of donor-derived haematopoietic cells (microchimerism), a limited sublethal graft-vs.-host reaction or the transfer of potentially tolerogenic costimulatory molecule-deficient APC resulting in operational tolerance.

To further test how direct allorecognition of donor leucocytes affects platelet MHC class I immunity, we first pulsed recipient BALB/c APC with donor C57BL/6 platelets in vitro to allow for uptake and processing (the indirect pathway) and then transfused them into BALB/c recipients together with varying numbers of intact donor MHC class II positive APC. Similar to the experiments with platelets derived from SCID mice, the donor leucocytes significantly reduced the pulsed APC immunogenicity. APC were titrated into stock platelets from SCID mice and transfused into CBA recipients weekly and IgG antidonor antibodies were measured by flow cytometry. The data is expressed as the mean ± SD of week 5 titres from 10 recipient CBA mice. The arrow at the top represents the shift in IgG antidonor isotype production dependent on the APC content.

In summary, murine models of leucoreduced platelet immunity support the notion that leucoreduction significantly reduces alloimmunization against platelets. What the animal models additionally suggest is that in recipients with a healthy immune system, leucoreduction may not be effective in reducing immunity against platelet-derived alloantigens because normal immunity (indirect allorecognition) is intact. Understanding the nature of this leucocyte-dependent regulation of platelet humoral immunity may be fundamental to designing more effective antigen-specific immunotherapies for those patients who can respond to platelet antigens.

**Acknowledgements**

Supported by grants from the Canadian Blood Services (CBS) R & D Fund (XT0008) and the CBS/Canadian Institutes for Health Research Partnership Fund (#84755).
References


