Establishment of Animal Models Using Experimental Rats for Allogeneic Tissue Transplantation and Quantitative Flow Cytometric Detection of Immunochimera

L Sun,*_M Med (Melb), MD (S'pore)_, B H Lim,** _MBBS, M Med (Surg), FRCS (Edin)_, V Yeow,***_MBBS, FRCS (Edin), FRCS (Glas)_, L X Xin,****_BSc_, P Tan,†_FAMS, M Med_

Abstract

Establishment of animal models used for allogeneic tissue transplantation, MHC phenotyping as well as quantitative detection of immunochimera were carried out in this study. Both Buffalo (BUF) and Lewis (LEW) rats were chosen as the target animals. The rats were treated with single dose cyclophosphamide (Cy) of 25 to 100 mg/kg to mimic the standard conditioning therapy. Total white blood cells (WBC) were monitored daily for up to 8 days: WBC reached the nadir by day 4 and started to recover by day 5 with an obvious rebounce at day 7 of Cy treatment. Flow cytometric techniques were used to determine the haplotypes of the major histocompatibility complex (MHC) as well as quantitative detection of immunochimerism in unfractionated rat WBC. Monoclonal antibodies against the rat class-I MHC antigens RT1A^{ab} and RT1A^{u} were used to label the class-I MHC antigens on total rat WBC. The results showed that the BUF rats were positive for both RT1A^{ab} and RT1A^{u} antigens, whilst the LEW rats were negative for both. Immunochimera was mimicked* in vitro *by serial dilution, ranging from 1/1 to 1/10^{5} of (BUF/LEW) WBC. A sensitivity of 1/10^{4} (BUF/LEW WBC) was achieved. The results showed that there were at least 2 major MHC mismatched loci between BUF and LEW rats and flow cytometry provided a sensitive method for the detection of immunochimera in unfractionated rat WBC. We concluded from this study that both strains of rats could be used as models for allogeneic tissue transplantation across at least two major MHC-mismatches. The sensitivity of flow cytometric method was satisfactory for the detection of immunochimera.

***Ann Acad Med Singapore* 1999; 28:90-4**

Key words: Cyclophosphamide, Detection of immunochimera, Experimental rats, Major histocompatibility complex (MHC)