Prophylaxis Against a Melanesian Variant of Human T-Lymphotropic Virus Type I (HTLV-I) in Rabbits Using HTLV-I Immune Globulin From Asymptomatically Infected Japanese Carriers

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Molecular variants of human T-lymphotropic virus type I (HTLV-I), which diverge significantly from the so-called cosmopolitan prototypes, have been discovered in Melanesia. In this study, HTLV-I IgG (I-IgG) prepared from seropositive healthy Japanese carriers was evaluated for its protective effect against a Melanesian isolate, HTLV-I\textsubscript{Mel}, in rabbits. Normal IgG (N-IgG) prepared from seronegative healthy Japanese was used as control. Both preparations contained 50 mg/mL of IgG and I-IgG had a high neutralizing antibody titer, as determined by vesicular stomatitis virus–HTLV-I pseudotype assay. Of four experimental groups (A, B, C, and D), each with three rabbits, groups A and B were infused with 10 mL of N-IgG and I-IgG, respectively, and animals were challenged immediately by transfusion of 5 mL of blood from a rabbit infected with HTLV-I\textsubscript{Mel}. Animals in groups C and D were immunized with 10 mL of I-IgG 24 and 48 hours, respectively, after being transfused with 5 mL of blood from the virus-infected rabbit. HTLV-I infection, as determined by seroconversion and verified by polymerase chain reaction, occurred in all rabbits in groups A and D after 2 to 6 weeks, but in none of the animals in groups B and C. These data indicate that I-IgG is protective against HTLV-I\textsubscript{Mel} infection when administered before or within 24 hours of transfusion with virus-contaminated blood. Moreover, our study shows that the neutralizing domains of the so-called cosmopolitan and Melanesian strains of HTLV-I are functionally indistinguishable.

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Results

Four groups (designated A, B, C, and D), each composed of three female rabbits, were studied. Rabbits in groups A and B were initially inoculated intravenously with 10 mL of N-IgG and I-IgG, respectively, and were challenged immediately by transfusion of 5 mL of blood from the HTLV-I-infected rabbit donor. Rabbits in groups C and D were first transfused with 5 mL of blood from the HTLV-I-infected rabbit and then immunized with 10 mL of I-IgG 24 and 48 hours later, respectively.

All rabbits in groups A and D seroconverted for HTLV-I after 2 to 6 weeks, whereas all animals in groups B and C remained seronegative during the observation period of 7 to 9 months. Serial determinations of human and rabbit IgG antibodies to HTLV-I by ELISA indicated that the group A rabbits, inoculated with N-IgG and lacking human anti-HTLV-I antibodies, began to produce rabbit anti-HTLV-I antibodies after 2 to 3 weeks that were maintained thereafter. By contrast, rabbits in groups B, C and D, which were inoculated with I-IgG, rapidly cleared the infused human anti-HTLV-I antibodies from their circulation in 3 weeks. This was followed by the production and persistence of rabbit anti-HTLV-I antibodies in group D rabbits but not in group B and C rabbits. The antibody response curves in four rabbits representing each of the four groups are shown in Fig 1.

Western blot analysis of sera confirmed the presence of rabbit IgG antibodies to HTLV-I p28, p24, and p19 in group A and D rabbits and their absence in group B and C rabbits (Fig 2, upper panel). HTLV-I proviral sequences
were detected by PCR at 10 weeks in all of the six seroconverted rabbits but in none of the six seronegative rabbits (Fig 2, lower panel).

Rabbits in group C that had been protected from HTLV-I<sub>MELS</sub> infection by immunization with I-IgG were challenged 8 months later by transfusion of 5 mL of blood from the same virus-infected rabbit donor. Seroconversion for HTLV-I occurred after 3 to 4 weeks and HTLV-I pX sequences were detected by PCR at 10 weeks in all of these rabbits, verifying the short-term protection afforded by passive immunization.

**DISCUSSION**

In the present study, we showed that IgG from Japanese asymptomatically infected with cosmopolitan strains of HTLV-I was effective in preventing cell-to-cell infection with a Melanesian variant of HTLV-I. In short, our data indicate that cosmopolitan and Melanesian strains of HTLV-I share common neutralizing epitopes that are functionally indistinguishable. This is not unexpected because comparative analysis of env gene sequences of Melanesian and cosmopolitan strains of HTLV-I<sup>18</sup> indicates total conservation of the amino acid sequences of the putative neutralizing epitopes that have been mapped to residues 88-98<sup>20</sup> and residues 191-196.<sup>21</sup> Collectively, these findings will weigh heavily in strategies aimed at developing vaccines and immunoprophylactic measures against HTLV-I infection worldwide.

It should be noted that rabbits could be protected from infection with either cosmopolitan or Melanesian strains of HTLV-I by passive immunization administered before or 24 hours after transfusion of virus-contaminated blood. Immunization administered 48 hours after transfusion was not effective. These observations suggest that transmission of virus from viral genome-carrying lymphocytes to host lymphocytes takes place sometime between 24 and 48 hours after transfusion. Due to the heterologous system, the infused human anti-HTLV-I antibodies declined with a half-life of approximately 7 days, becoming undetectable in 3 weeks, and the transfused allogeneic lymphocytes, including those carrying the viral genome, were postulated to have been rejected within 1 to 2 weeks by the host. As expected, group C rabbits that had been once protected from HTLV-I<sub>MELS</sub> infection by I-IgG could be readily infected by challenge with virus-infected blood after the disappearance of the passively acquired antibodies.

Finally, we have previously shown that milk-borne transmission of HTLV-I can also be prevented by passive immunization of suckling rabbits.<sup>20</sup> In some HTLV-I-endemic regions of Japan, seropositive pregnant women are being advised to feed their newborn infants with formula rather than breast milk. This recommendation may be impractical in countries such as Papua New Guinea and the Solomon Islands. Thus, prophylactic immunization of babies against milk-borne transmission of cosmopolitan and Melanesian strains of HTLV-I may be an interim measure until an effective HTLV-I vaccine is developed.

**REFERENCES**

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