ELSEVIER

Induction of Transplant Tolerance by Intrathymic Inoculation of Synthetic MHC Class I Allopeptides

N.C. Chowdhury, B. Murphy, M.H. Sayegh, M.A. Hardy, and S.F. Oluwole

SUCCESS OF organ transplantation between genetically disparate individuals is dependent on long-term use of non-specific immunosuppressive agents that cause significant morbidity and mortality. The ultimate goal in clinical transplantation is the development of strategies to induce specific unresponsiveness to allografts without the impairment of host defense mechanisms. One such strategy that has recently received major attention is the use of intrathymic (IT) injection of alloantigens with peritransplant immunosuppression.[1] We have previously shown that IT injection of purified T-cells[2] or 3M KCl extracts of T-cells[3] induces donor-specific transplant tolerance to cardiac, small bowel, and islet allografts in rodents. This study extends these findings to the use of well-defined polymorphic MHC class I allopeptides derived from the hypervariable domain of RT1.A^u (WF MHC class I).

MATERIALS AND METHODS

We synthesized a total of 6 class I or RT1.A peptides using published sequences for RT1.A^u "Wag"[4] (Biopolymer Laboratory, Brigham and Women's Hospital, Department of Medicine). Whereas 3 of the 6 synthetic RT1.A^u peptides [residues 36–76, 67–85 and 136–159] were immunogenic, 3 others [residues 93–109, 110–125 and 167–182] were non-immunogenic to ACI responders. Naive and the transiently immunosuppressed ACI recipients were given IT injection of 150, 300, and 600 μg RT1.A^u [WF MHC class I] allopeptides of an equal mixture of 3 residues [93–109, 110–125, and 167–182]. The recipients were transiently immunosuppressed with intraperitoneal 0.5 mL rabbit anti-rat lymphocyte serum [ALS, Sera Lab, Accurate, Westbury, NY] at time of injection of IT-allopeptides. Seven days later, the animals were transplanted with heterotopic cardiac allografts obtained from WF (RT1^u) or third party Lewis (RT1^l).

RESULTS

While IT injection of 100 and 300 μg of a mixture of the non-immunogenic class I MHC allopeptides on day −7 relative to cardiac transplant did not significantly prolong graft survival in naive ACI recipients [MST of 9.8, and 12.3 days vs. 10.5 days in controls], 600 μg allopeptides injected IT resulted in significant although modest prolongation of graft survival to an MST of 19.5 days. Furthermore, IT injection of 600 μg allopeptides combined with 0.5 mL ALS on day −7 led to permanent acceptance (>200 days) of cardiac allografts compared to survival of 24.2 days in ALS alone treated ACI controls. In contrast, similar treatment led to acute rejection of third party [Lewis] cardiac allografts, thus confirming donor-specificity. Intravenous injection of 600 μg allopeptides combined with ALS did not prolong graft survival (MST of 26.8 days). The long-term unresponsive ACI recipients (>100 days) challenged with second-set cardiac grafts accepted permanently donor-type (WF) grafts while rejecting the third party (Lewis) grafts, a finding that confirms acquired systemic tolerance.

DISCUSSION

We have now shown that IT inoculation of 600 μg of non-immunogenic MHC class I peptides resulted in permanent cardiac allograft survival in the low responder combination of WF-to-ACI. This observation confirms the effectiveness and practicality of this approach. These data further confirm that transient immunosuppression of the recipient to modulate or delete the mature T-cells in the peripheral circulation is an essential step in the prevention of allograft rejection prior to the emergence of a clone of newly formed Ag-tolerant host T-cells from the foreign Ag-containing thymus. Our results confirm that IT inoculation of synthetic MHC class I peptides, similar to what has been reported for MHC class II peptides in renal allograft model,[5] can induce acquired thymic tolerance. We plan to compare the effectiveness of IT injection of immunogenic versus non-immunogenic synthetic allopeptides in future studies.

REFERENCES

1. Posselt AM, Barker CF, Tomaszewski JE, et al: Science 249:1293, 1990

2. Chowdhury NC, Fawwaz RA, Oluwole SF: J Surg Res 54:368, 1993

3. Oluwole SF, Chowdhury NC, Jin MX, et al: Transplantation 56:1523, 1993

4. Walter L, Tiemann C, Heine L, et al: Immunogenetics 41:332, 1995

5. Sayegh M, Perico N, Imberti O, et al: Transplantation 56:461, 1993

From the Department of Surgery, Columbia University College of Physicians & Surgeons, New York, New York, and the Laboratory of Immunogenetics and Transplantation, Brigham & Women's Hospital, Harvard Medical School, Boston, MA.

Grant Support: NIH Grant HL 14799.

Address reprint requests to Nepal C. Chowdhury, 1227 White Plains Road, Apartment #633, Bronx, NY 10472.

0041-1315/97/$17.00
PII S0041-1345(96)00469-1