The Influence of Removing Passenger Cells on the Fate of Skin and Parathyroid Allografts

Evidence for Major Histocompatibility Complex Restriction in Transplantation Immunity

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SUMMARY

Studies with mice on the survival of skin grafts incompatible with respect to the histocompatibility-Y transplantation antigen or to skin-specific antigens, as well as studies with rats on the fate of cultured parathyroid allografts, provide evidence for major histocompatibility restriction in transplantation immunity. If confirmed, these studies indicate that islet allografts devoid of passenger leukocytes might survive better in major histocompatibility complex incompatible recipients. DIABETES *31* (Suppl. 4):60–62, 1982.

Although major histocompatibility complex (MHC) restriction with respect to transplantation antigens has been amply and repeatedly demonstrated in vitro,[1–7] there has been no *direct* evidence that it plays a role in sensitizing hosts to skin and other organ allografts in vivo. We believe we have evidence for such a role. Our evidence stems from mouse studies on histocompatibility-Y (H-Y)[8] and skin specific (Skn)[9] antigens as well as from experiments concerned with the ability of cultured parathyroid grafts to be accepted by MHC incompatible and compatible rats.[10]

Whereas C57BL/6 (B6) female mice uniformly reject H-Y incompatible adult skin grafts [their median survival time (MST) is about 20 days], about 75% of such females accept neonatal B6 male skin grafts (comprising about half the integument of a newborn), and about half of these permanently accept subsequently transplanted adult male B6 grafts as well.[8] Similarly, not only are Skn-incompatible neonatal A strain grafts [unlike A strain adult skin grafts (Table 1, experiment 1)] frequently accepted by B6 mice made tolerant at birth of B6/A lymphoid cells, but these grafts too often render their hosts unresponsive of adult strain A skin.[9] However, much to our surprise, we found that whereas these strain A (neonatal and adult) skin grafts (about 1 cm^2) are usually *accepted* when they are retransplanted to secondary tolerant (of B6/A lymphoid cells) B6 animals, after they have resided on their primary tolerant B6 hosts for 100–150 days (Table 1, experiments 2 and 3), this is not the case with similarly sized H-Y incompatible grafts. Twenty-seven of thirty originally neonatal male skin isografts were rejected (MST 32 days) within 80 days when transplanted from one B6 female to another, even after they had survived on their primary hosts for >150 days.

To account for this difference, we propose that allografts can only provoke a strong immune response if they include donor macrophages (or, in the case of skin, Langerhans cells that may serve the same function of antigen processing),[11–13] or if MHC compatible macrophages are available to react with cells bearing the foreign antigens. We believe that the strain A Langerhans cell population of a strain A skin graft maintained on a B6 mouse (tolerant of B6/A lymphoid cells) for 100–150 days is replaced with a B6 Langerhans cell population (the lifespan of the Langerhans cell has been estimated at about 3 mo).[14] Accordingly, when this graft is retransplanted to a secondary tolerant B6 host, its Skn antigens (because they are processed by B6 Langerhans cells) are recognized in association with self (B6) MHC ($H\text{-}2^b$) and not in association with the MHC ($H\text{-}2^a$) of the strain A graft. On the other hand, a neonatal B6 male skin graft maintained on a B6 female for >100 days still possesses a B6 (albeit female) Langerhans cell population and hence is rejected when regrafted to a secondary B6 female host.

Evidence in support of this interpretation of our results stems not only from the fact that strain A skin grafts raised on B6/A F_1 hybrids, i.e., on hosts that are MHC compatible with the grafts, for >100 days are usually either rejected or become highly contracted when retransplanted to B6 mice tolerant of B6/A lymphoid cells (Table 1, experiments 4 and 5), but also from the fact that neonatal strain A grafts retained on tolerant B6 hosts for only 27 days, i.e., a period insufficient for their Langerhans cell population to be completely replaced were, with one exception, rejected when retransplanted to secondary B6 recipients (Table 1, experiment 6).

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TABLE 1
Survival of neonatal or adult strain A skin grafts on B6 hosts (tolerant of A/B6 lymphoid cells) after being maintained for various periods on either tolerant B6 or A/B6 F_1 hybrid mice

Exp.	Source of graft	First host	Maintained (days)	N	Survival (days) on B6 second host*
1	Adult	—	—	11	13, 18, 2 × 19, 20, 22, 27, 31, 32, 2 × >100
2	Neonatal	B6*	100–150	8	8 × >100
3	Adult	B6*	100–150	7	38, 6 × >100†
4	Neonatal	A/B6	100	6	41, 77, 4 × >100‡
5	Adult	A/B6	150	8	2 × 28, 50, 62, 2 × 64, 65, >100
6	Neonatal	B6*	27	5	24, 25, 36, 71, >100§

* Tolerant of A/B6 lymphoid cells.
† One of these grafts became progressively smaller and was scored as rejected at 105 days; another persisted at ~50% of its original size.
‡ One of these grafts became progressively smaller and at 100 days was <5% of its original size; another was ~50% of its original size.
§ At 100 days, this graft was ~60% of its original size.

We also believe it is significant that when five tolerant B6 mice which had retained strain A neonatal skin grafts in excellent condition for >100 days (after they resided for >100 days on primary tolerant B6 hosts) were challenged on the other side of their thorax with a fresh strain A graft, one of these mice rejected both grafts (the fresh adult graft in 16 days and the well-established neonatal graft in 53 days), and the neonatal grafts on two other secondary hosts (one of which rejected its adult graft in 19 days while the graft on the other became progressively smaller) became severely contracted. We suggest that the complete or partial rejections by these mice was a consequence of the fact that because the fresh strain A grafts they were challenged with included normal populations of strain A Langerhans cells, these cells were able to process the foreign A strain Skn antigens (to which they must constantly be exposed) and serve them up to the host in association with a H-2^a haplotype.

We also contend that it may be significant that two B6 mice which were regrafted with fresh strain A adult skin grafts after their neonatal A strain grafts (with B6 Langerhans cells) had been in place for more than 100 days, accepted these grafts without any signs of rejection. This suggests that not only are grafts devoid of MHC compatible macrophages (Langerhans cells) frequently accepted by MHC incompatible hosts, but by one means or another, they may induce unresponsiveness.[15,16]

Further evidence for our hypothesis stems from the fate of cultured parathyroid grafts in rats. Thus, while there is ample evidence that culturing endocrine allografts may dramatically improve their survival,[17–23] we have evidence that this may only apply to MHC *incompatible* grafts. We found that whereas only 1 of 31 Fischer rats accepted freshly transplanted MHC-incompatible ACI parathyroid glands for more than 100 days, after they have been maintained in culture in an O_2-enriched environment for 26 days, they survived for this period in 8 of 31 rats. On the other hand, although 21 of 38 Fischer rats accepted fresh MHC-compatible Lewis parathyroids for more than 100 days, only 10 of 31 hosts challenged with cultured glands accepted them indefinitely. Assuming that the major influence of maintaining parathyroids (and other tissues) in vitro is to eliminate passenger leukocytes, including macrophages, it follows that only when they are transplanted to MHC compatible hosts would a population of MHC compatible (of host origin) macrophages be available to present their foreign antigens to the hosts.

While these experiments do not rule out the possibility that in the absence of MHC compatible macrophages alternate forms of antigen presentation may also sensitize the host, they suggest that if this is the case, these pathways are not as effective.

If the interpretation of our findings is correct, its clinical implications are obvious. Currently and for sound reasons, all efforts are directed towards matching donors and hosts with respect to their MHC. However, if our results are verified, they indicate that if methods become available to remove passenger cells from allografts, they might be more likely to survive in MHC *incompatible* recipients. Thus, with respect to the clinical transplantation of passenger cell-free islet allografts, attempts to match donor and recipient with respect to their MHC antigens may actually be counterproductive.

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