Immunology 2017: Lecture 16 handout

Graft versus host disease

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INTRODUCTION

We learned in the previous lecture that self reactive T cells are deleted through several tolerance mechanisms, of which central tolerance is the most important. However, each person's antigens are different from another and my T cells recognize my Ag as self and yours as nonself. This is why transplantation of an organ, or tissue from one person to another causes rejection.

Rejection is a complex phenomenon that involves cellular and antibody mediated reactions leading to destruction of the graft. Here the immune system of the recipient attacks the graft.

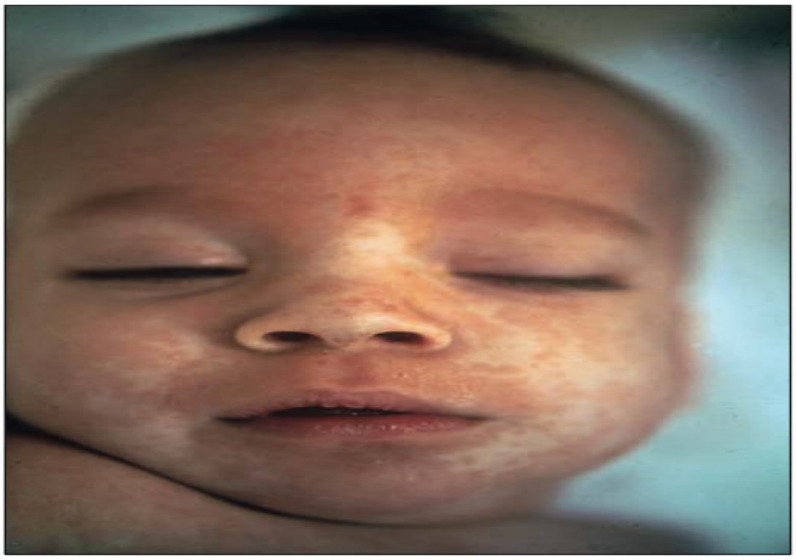
However, if we transplant bone marrow to a person, this bone marrow will contain mature T cells that will attack the recipient's tissues!

Please do not confuse these two situations! In **graft rejection** the recipient's immune system is rejecting the foreign graft. Whereas in bone marrow transplantation the donor (the foreign) is attacking the host. This is called **graft versus host disease.**

BONE MARROW TRANSPLANTATION

Bone marrow transplantation is a useful therapy for some forms of *leukemia, bone marrow failure (aplastic anemia) , and primary immunodeficiency diseases.* More recently, other sources of hematopoietic stem cells, such as peripheral blood stem cells and umbilical cord blood, have also been used for these purposes.

Bone marrow and most other sources of hematopoietic stem cells *contain mature T* lymphocytes, which may recognize the tissues of their new host as foreign and cause a severe inflammatory disease in the recipient. This is known as graft-versus-host disease (GVHD) and is characterized by **a rash, which often starts on the face (see image on next page) , diarrhea, pneumonia, and liver damage**.



To achieve successful engraftment of bone marrow and avoid rejection of the transplant by the host, the immune system of the recipient must be destroyed and the recipient rendered immunocompromised. This is usually accomplished with radiation or the injection of radiomimetic drugs such as busulfan, and the use of immunosuppressive drugs (cyclophosphamide, fludarabine).

GVHD occurs not only when there is a mismatch of classical MHC class I or class II molecules but also in the context of disparities in minor histocompatibility antigens; such minor differences are likely to be present in all donor-recipient pairs other than identical twins, even when HLA-matched.

Mature CD4 T cells in the graft that are activated by allogeneic molecules produce a 'cytokine storm' that recruits other T cells, macro phages, and natural killer (NK) cells to create the inflammation characteristic of GVHD.

*Although B cells may also be present in GVHD inflammation, they do **not** have a significant role in causing or sustaining GVHD*.

GVHD is arbitrarily called '**acute**' if it occurs less than 100 days after the transplant, and '**chronic**' if it develops after 100 days. Chronic GVHD is a more severe and difficult-to-treat problem.

The presence of alloreactive T cells in the donor bone marrow is usually detected in routine laboratory testing by the **mixed lymphocyte reaction (MLR**) in which lymphocytes from the potential donor are mixed with irradiated lymphocytes from the potential recipient. If the donor lymphocytes contain alloreactive T cells, **these will respond by cell division**. Although the MLR is routinely used for the selection of donors it does not accurately quantify alloreactive T cells.

MLR test

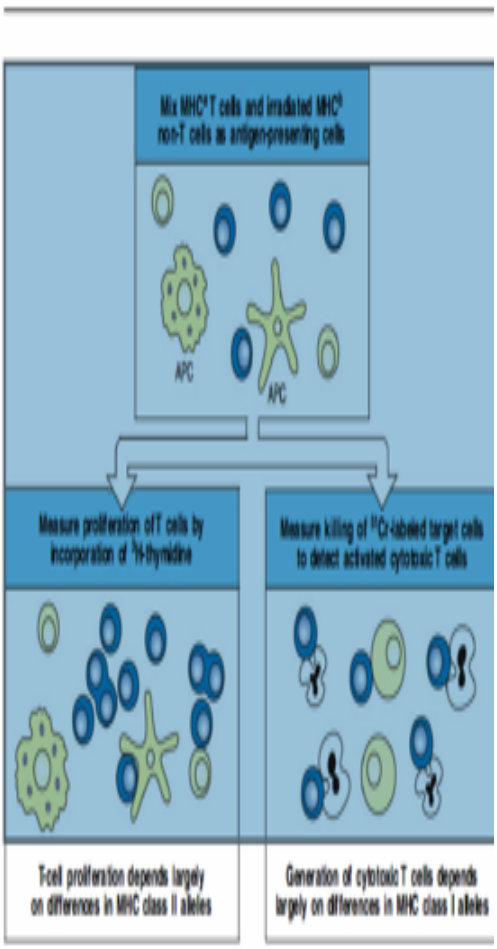


Fig. 6.3 The mixed lymphocyte reaction (MLR) can be used to detect histoincompatibility. Lymphocytes from the two individuals who are to be tested for compatibility are isolated from peripheral blood. The cells from one person (yellow), which also contain antigen-presenting cells (APCs), are either irradiated or treated with mitomycin C; they will act as stimulator cells but cannot now respond by DNA synthesis and cell division to antigenic stimulation by the other person's cells. The cells from the two individuals are then mixed (top panel). If the unirradiated lymphocytes (the responders, blue) contain alloreactive T cells, these will be stimulated to proliferate and differentiate to effector cells. Between 3 and 7 days after mixing, the culture is assessed for T-cell proliferation (bottom left panel), which is mainly the result of CD4 T cells recognizing differences in MHC class II molecules, and for the generation of activated cytotoxic T cells (bottom right panel), which respond to differences in MHC class I molecules. When the MLR is used to select a bone marrow donor, the prospective donor's cells are used as the responder cells and the prospective recipient's cells as the stimulator cells.

The idea of the teast is easy: you mix donor and recepient T. Recepient cells are irradiated so they cannot react to donors' Ag. There wil be some APC within recepient T. If donor T recognize donor's self Ag thy react by proliferation to start an immunologic reaction. So if they proliferate we know they will cause problm when transplanted.

THE CASE

John had aplastic anemia. His bone marrow had very few cells and his red cell, platelet, and white cell precursors were almost completely absent.

Aplastic anemia is ultimately fatal but can be cured by a successful bone marrow transplant.

John had an HLA-identical 1 1 -year-old brother who could be the bone marrow donor. John was admitted to the Children's Hospital and given a course of immunosuppressant drugs to eradicate his own lymphocytes. He was then given bone marrow cells obtained from his brother's iliac crests. He was also started on cyclosporin A to prevent GVHD.

John did well for 3 weeks after the bone marrow transplant and was then sent home to recover. However, in spite of GVHD prophylaxis with Cyclosporin A, on the 24th day after the transplant he was readmitted to hospital with a skin rash and watery diarrhea consistent with acute GVHD.

John was treated with corticosteroids. His skin rash faded, but the intestinal symptoms did not abate and the diarrhea became more profuse. He was given rabbit antithymocyte globulin {ATG) for two consecutive days. This brought about a 90% decrease in the volume of his stool and the intestinal bleeding stopped. Two weeks later, John was sent home, with continuing treatment with low doses of corticosteroid, and his GVHD remained under control.

DRAFT VERSUS HOST DISEASE:

For a recipient to develop GVHD, the **graft must contain immunocompetent cells**, the **recipient must express major or minor histocompatibility molecules that are lacking in the graft donor**, and **the recipient must be incapable of rejecting the graft**.

The first clinical manifestation of GVHD is a bright red *rash* that characteristically involves the palms and soles. The rash usually begins on the face and neck and progresses to involve the trunk and limbs, particularly the palms and soles. The rash may itch a great deal and its onset may be accompanied by fever. After the skin manifestations appear, the *gastrointestinal* tract becomes involved and profuse watery diarrhea is produced.

Liver function tests may become abnormal and reveal destruction of hepatic tissue .Eventually, other tissues such as the *lungs* and *bone marrow* become sites of GVHD inflammation.

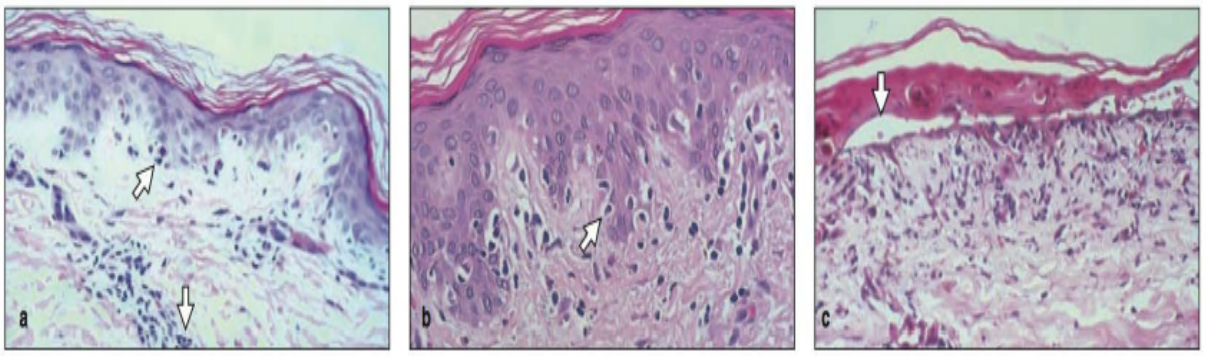


Fig. 11.3 GVHD in the skin. Panel a: early GVHD in the skin. Lymphocytes are emerging from blood vessels (lower arrow) and adhering to the basal layer of the epidermis (upper arrow). Panel b: the basal cells of the epidermis begin to swell and vacuolate. Their nuclei become condensed (dark staining) as these cells die (arrow). Panel c: advanced destruction of the skin by GVHD, with sloughing of the epidermis (arrow). Photographs kindly provided by Robert Sackstein.

The only satisfactory therapy at present for GVHD is elimination of the T cells that initiate the reaction, either by immunosuppressive drugs or, by T-cell-depleting agents.

Types of donors:

In the above case the donor was HLA identical.. and still GVHD occurred (due to minor differences in MHC)

It's not always possible to find HLA identical donor. So we may use:

1. **haploidentical (that is, halfmatched) donor** (typically, one of the two parents) .here there is a very high risk of GVHD. For this reason, the bone marrow from haploidentical donors is manipulated to deplete the mature T lymphocytes (or to purify the stem cells only) before attempting the transplant.

2. **Matched Unrelated Donors (MUDs)** has also become current practice at many centers and there is also a significant risk of GVHD with this type of transplant. Some bone marrow transplant centers use T-cell depletion also for transplantation from MUDs, but this approach is not followed by the majority of centers. **Transplantation with unmanipulated bone marrow from MUDs is associated with a higher rate of engraftment and may provide a 'graft-versus-leukemia' reaction,** in which ***donor-derived mature T cells may kill the residual leukemic cells of the recipient,*** making relapse less likely in the case of bone marrow transplants for the treatment of leukemia.

Questions:

1. Bone marrow is often depleted of T cells before transplantation, to try and avoid GVHD. However**, <u>in the treatment of leukemia by bone marrow transplantation, T cells in the graft can have a beneficial effect.</u>** How do you explain this?

Answer: The engrafted T cells recognize allogeneic antigens on the recipient's hematopoietic cells and thus will **attack the leukemic cells**. *One such antigen, HB- 1, which is a B-cell lineage marker, is expressed by acute lymphoblastic leukemia cells, which are B-lineage cells, and by B lymphocytes transformed by Epstein-Barr virus (EBV).* Graft (donor) T cells can attack these leukemic or lymphoma cells and helps in treating the disease and decreasing recurrence.

2. CD4 T cells in the graft that recognize foreign histocompatibility molecules become activated and produce the cytokine interferon (IFN)-y. This helps sustain and increase GVHD. Why?

Answer: **IFN -y induces the expression of MHC molecules on cells**; this makes GVHD worse, because it provides more targets (more MHC) for the donor T cells.

3. John was given ATG (in addition to steroids) to control acute GVHD. Do you know any other ways to achieve in vivo depletion of T lymphocytes?

Answer: In vivo T-cell depletion can also be achieved with **the intravenous injection of monoclonal antibodies, such as anti-CD3.** This approach is also used to treat initial graft rejection in recipients of solid organ transplantation, but it is associated with a higher risk of lymphoproliferative disease induced by the Epstein-Barr virus. **Monoclonal antibodies targeting activated, but not resting, T lymphocytes have been also used in the treatment of GVHD, and include anti-CD25 and anti-CD40L monoclonal antibodies**

4. Why are the skin and intestinal tract the major sites of GVHD?

Answer: One reason could be that the **skin and intestine express a higher level of MHC** molecules than other tissues. **The intestinal tract is also likely to be damaged by the preparative cytotoxic treatments** given to destroy the recipient's bone marrow (this is because it has a high proliferative rate andchemotherapy targets highly proliferative tissue). *This damage in the intestine induces the production of cytokines; as well as inducing MHC molecules, these can also drive GVHD and make the tissue susceptible to immunological attack*.