IMMUNODEFICIENCY

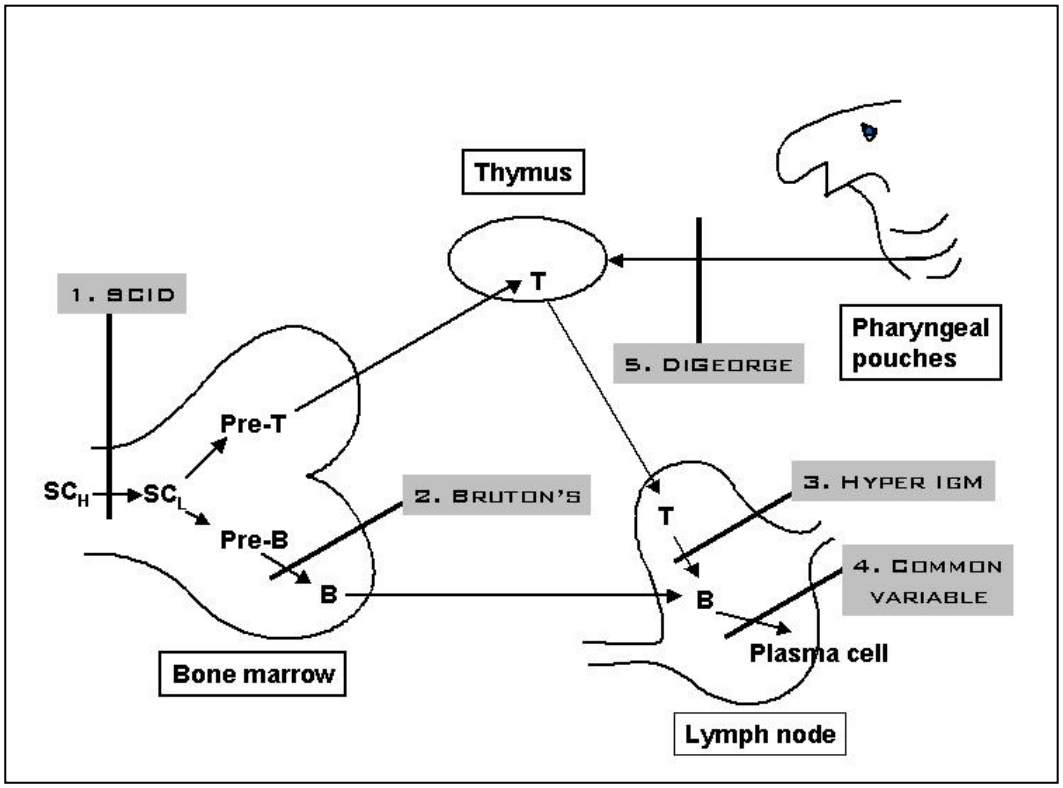
CATEGORIES OF IMMUNODEFICIENCY STATES. ►Immunodeficiency can be primary or secondary. In primary immunodeficiency, there are mutations in genes required for normal development of parts of the immune system. Variable penetrance—the degree to which a mutation is expressed phenotypically—means that children with these mutations may have symptoms along a spectrum of seriousness. If the thymus or bone marrow were congenitally dysfunctional that would result in primary immunodeficiency, while the immunodeficiency that follows treatment with immunosuppressive drugs, or that is seen in patients with advanced cancer, measles, or AIDS, is *secondary* to those conditions.

Another way of putting it is that immunodeficiency can be congenital or acquired, depending upon whether the condition existed at birth or not. Acquired immunodeficiency will usually be secondary to some other condition.

Immunodeficiency can also be temporary and self-limited, as in the transient hypogammaglobulinemia of infancy, or during treatment with cancer chemotherapy drugs; or it can be permanent, or last until we can come up with a cure.

Remember also that deficiency of any component of the immune/inflammatory systems can lead to impaired immunity: in addition to the lymphocyte deficiencies we'll discuss here, there are deficiencies in complement components, in phagocytic cell functions, etc.

PRIMARY IMMUNODEFICIENCY DISEASES (PIDD). These can affect T cells or B cells selectively, or both kinds of cells. It is largely by studying the clinical syndromes associated with immunodeficiency diseases that we know what T and B cells are really important for in humans. The following diagram shows sites of **DEVELOPMENTAL BLOCKS** in conditions that are discussed on the next page.



Block 1: If there are low numbers of T and B cells, it is as if there is a block in the development of the lymphoid stem cell, or its further maturation. These conditions are the worst of the PIDD states and are called **Severe Combined Immunodeficiency Disease,** or **SCID**. Children with the most profound deficiencies rarely survive untreated beyond a year (they are to some extent protected in the neonatal period by maternal IgG). There is lymphopenia of both T and B cells, absent thymic shadow on X-ray, and tonsils are small; mitogen responses and serum immunoglobulins are low. SCID is a ►group of diseases with a similar phenotype. More than half the cases are X-linked recessive. In the commonest of these (SCID-X1), the defect is in ►the gene for the gamma chain that forms part of the receptors for IL-2 and other cytokines necessary for lymphoid development, or their signaling pathways. The rest of SCID cases are autosomal recessive. Most of these patients lack the enzyme adenosine deaminase (ADA); so adenosine accumulates in all cells but impairs lymphocyte development selectively. Among the rarest globally are defects in V(D)J recombination, although that is the most common form of SCID in Navajo and Apache children (incidence about 50/100,000 births.)

Block 2: If there are normal T cells but low to absent B cells, there is a developmental block between the pre-B cell and the B cell. Most patients have pre-B cells in their bone marrow but are deficient in B cells and antibody (serum IgG less than 10% of normal, IgA and IgM virtually undetectable). The disease is called **X-linked (Bruton) Agammaglobulinemia**, in which a protein tyrosine kinase gene, *btk*, normally expressed in pre-B and later B cells, is defective. Boys with Bruton have bacterial infections manifesting as pneumonia and chronic diarrhea. Enteroviruses, which gain entry through mucous membranes unprotected by IgA, may also be a problem; among these is poliovirus. ►These kids are the reason we no longer use oral polio vaccine in America. Incidence about 0.4/100,000 births.

ASK YOURSELF: Why? Wouldn't we particularly *want* to immunize such kids?

Block 3: A rare patient will have high IgM with low IgG and IgA; in such patients there is a defect in the IgM-to-IgG switch mechanism. The Tfh cell has an accessory molecule (CD154 or CD40-ligand) that interacts with CD40 on B cells, signaling them to switch classes (see diagram in T cell unit). If either CD40 or CD40L is defective, the B cell is driven hard but can't receive the instruction to switch past making IgM. This is called **X-linked hyperIgM syndrome**.

Block 4: It can happen that there are normal numbers of pre-B cells and B cells, but the B cells are difficult to trigger to make specific antibody. This condition is called **Common Variable Immunodeficiency (CVID)**, and is actually a group of about 20 conditions, involving abnormal cell-cell communication or immune regulation. This condition is relatively milder than the others, and is diagnosed for the first time in people 20-40 years old. The main phenotype is recurrent bacterial infection. There is increased risk for lymphoma, enteropathy, or autoimmunity. The causes are unknown, but include defects in innate immunity, B and T cells.

Block 5: The thymus is a two-component organ. The lymphoid part comes from precursors in the bone marrow, as we already know; the stroma is derived in the embryo from the endoderm and ectoderm of the 3rd and 4th pharyngeal pouches. If these develop abnormally the stroma will not support thymic lymphoid development, and the patient will have absent T cells with normal B cells. The condition is the ►**DiGeorge Syndrome,** and the cause is a large deletion on chromosome 22. The parathyroids also derive from the pharyngeal pouches, so this diagnosis is sometimes made in infancy when there are unexplained convulsions controllable by calcium. The heart develops abnormally, too. Cell-mediated immunity is depressed; viral and fungal infections are common. Incidence about 30/100,000 births. ►**Nude mice** have quite a different mutation, but also fail to make a thymic stroma (and hair) and so they have no T cells, and are immunologically similar to DiGeorge kids.

INFECTIONS. T cell deficiencies are associated with severe infections with intracellular pathogens, including viruses, certain bacteria, and yeasts and fungi, especially *Candida albicans* and *Pneumocystis jirovecii*. B cell deficiency is characterized by infections with "high-grade" (extracellular, pyogenic = pus-producing) bacterial pathogens such as *Staphylococcus aureus*, *Haemophilus influenzae* and *Streptococcus pneumoniae*. ►All of this makes good sense if you remember antibody and T cell-mediated mechanisms. But keep in mind that every infection involves multiple immune responses, so these generalizations are not hard and fast rules. Gut organisms may be abnormal in either type of disease, so diarrhea and malabsorption are frequent complaints, as is decreased growth rate.

TRANSIENT HYPOGAMMAGLOBULINEMIA OF INFANCY. This is a self-limiting condition, noticed about 3 to 6 months after birth and lasting up to 18 months (in a few cases, much longer). These children are slow to get their production of IgG going. They present mostly with recurrent and persistent Gram-positive bacterial infections, but may get just about anything. ►Perhaps 15% of all chronic diarrhea in infants is due to this condition.

SELECTIVE IgA DEFICIENCY is the most common immunodeficiency disease, with a prevalence of about 200 in 100,000. Although it is usually asymptomatic, the patient may have diarrhea and sinopulmonary infections, or an increased frequency and severity of allergies. There is a familial tendency, and although several mechanisms have been proposed, none is yet clearly established. It is 10-15 times more frequent (or more frequently diagnosed) in people with celiac disease.

OTHER PRIMARY IMMUNODEFICIENCIES[1]. ~150 other conditions have been described.

SECONDARY IMMUNODEFICIENCY. Clinicians and patients need to keep in mind that some treatments are toxic to the immune system and that some diseases themselves are immunosuppressive. Drugs used in the therapy of autoimmune and inflammatory conditions, such as corticosteroids and some monoclonal antibodies, can be profoundly suppressive, and patients treated with these drugs should be warned to keep away from people with infectious diseases (chicken pox, for example, can be devastating in an immunosuppressed person). ►Many viral illnesses, especially measles, mononucleosis, and cytomegalovirus (CMV) infection, are immunosuppressive, and secondary infection is common. **Acquired Immune Deficiency Syndrome** or **AIDS** is the most serious condition involving secondary immunodeficiency.

A puzzling new condition: A cohort of people in SE Asia have developed sudden, rampant mycobacterial infections which their T cells don't control; they have an acquired form of immunodeficiency. It turns out that they are *autoimmune*: they are making autoantibodies to their own IFNγ.

TREATMENT OF IMMUNODEFICIENCY.

1. Isolation = bubbles. (This is impractical for extended periods.)

2. Prophylactic antibiotics. These are used in combinations, which you change monthly.

ASK YOURSELF: Why change them monthly?

[1] Excellent review by CU authors: Yoshikazu Morimoto & John M. Routes (2008). Immunodeficiency Overview. Prim Care Clin Office Pract 35:159–173.

3. Human immunoglobulin, where B cell function is deficient, as in X-linked agammaglobulinemia and common variable immunodeficiency. This must be given approximately monthly. It is pooled from many donors, and is usually about 99% IgG, with a half-life of 3 weeks. The standard of treatment now is a form for IV use (IVIG) from several manufacturers; effective but expensive, and often in short supply. Recently, a preparation for slow subcutaneous infusion (SCIG) that can be done at home has been approved.

Note: Caution must be used when giving immunoglobulins to people with selective IgA deficiency. The IgA in the preparation may be foreign to them, provoking an allergic or immune complex reaction.

4. Transplantation. In DiGeorge, fetal thymus or cultured thymic stromal cells have been used to try to minimize the risk of graft-versus-host disease. Some success is claimed; better diagnosis would aid in the selection of appropriate cases. This child shows some of the features of DiGeorge, including hypertelorism, down-slanting eyes, fishmouth deformity, micrognathia, and low-set ears. He was born with the heart defect Tetralogy of Fallot (which surgeons repaired soon after birth), hypocalcemia, and absent T cells. He received a fetal thymus graft. He developed GvH disease but responded to treatment and had a T cell count of at least 500. The thymic stroma was the donor's but the T cells were his.

For SCID, bone marrow transplantation had about a 50% success rate, but graft-versus-host disease (*see* Type IV Immunopathology) was a severe problem. Current practice uses purified hematopoietic stem cells rather than whole bone marrow. In some cases no pretreatment of a severely immunodeficient recipient is needed; if they have some immune function, rejection of the grafted cells is a risk, and the recipient is "conditioned" by chemotherapy or radiation before the HSC are given. ►Sibling donors are the best, and a good Class II MHC match is imperative.

For adenosine deaminase-deficient patients, transfusions of ►*irradiated* (gamma, or x-ray) red cells can be helpful, because red blood cells contain a lot of ADA.

> **ASK YOURSELF:** Why do we stress the word *irradiated*—is transfused blood normally irradiated? How do you think these transfusions work for the patient?

Purified ADA, stabilized with polyethylene glycol ("PEGylated"), is also available for use as a drug. ►The first-ever successfully gene replacement therapy in humans has been done in ADA-deficient and SCID-X1 children. But problems have arisen: In the first series of about 12 kids, 3 developed leukemia. The vector bearing the normal gene preferentially inserted itself near oncogenes, activating their expression. Better lentiviral vectors have, it seems, obviated that problem, and kids are once again being given replacement gene therapy.

Gene therapy link to cancer cases

Scientists confirm DNA treatment gave 2 boys leukemia

BIRMINGHAM, England, May 5 (AP) — A revolutionary gene therapy treatment that cured 10 French boys of a deadly inherited disorder known as "bubble boy disease" gave two of them leukemia, scientists said.

DR. SALIMA HACIEN-BEY-ABINA said Sunday that genetic tests have confirmed that the treatment, the first time that gene therapy has cured a disease, triggered the cancer in the toddlers. The boys are responding well to anticancer therapies, she added. Experts said it is now clear that the virus used to carry the needed gene into the children's bodies landed in a bad place. Scientists had always feared that cancer might occur if the virus used in the therapy lodged near certain genes that control cell growth and affected those too.

Addressing a conference of the European Society of Human Genetics, Hacien-Bey-Abina of the Necker Hospital for Sick Children in Paris said tests have shown that in the first toddler stricken with leukemia, the correcting gene landed inside a cancer-promoting gene called LMO-2. In the second toddler, the gene landed near the LMO-2 gene.

RETROVIRUS VECTORS PROBLEMATIC

"Apart from the tragedy of those two kids, I think this has put an enormous skid under a great deal of gene therapy, on this whole business of using retrovirus vectors" to get the needed genes into the body, said Andrew Read, a professor of genetics at Manchester University and chair of the scientific committee for the conference.

"That is a perfectly general problem of retroviral vectors and not a problem of the particular virus they used or the particular disease they treated and I cannot see how in principle you can get rid of that risk," said Read, who was not involved in the research.

The boys were given the treatment starting in 1999 when they were babies, ranging in age from one month to 11 months. The first toddler who developed leukemia appeared healthy until August last year — 30 months after treatment — when scientists found leukemia-like overproduction of white blood cells. After that finding, the United States suspended its three gene therapy studies for the disorder. A second child then developed leukemia in December, 34 months after getting the therapy, Hacien-Bey-Abina said.

2 HAD 'BUBBLE BOY' DISEASE

The boys were born with severe combined immunodeficiency, or SCID, a rare inherited disease that occurs in about 1 in 75,000 births. The best known victim was David, Houston's famous "bubble boy" who lived in a germ-proof plastic enclosure until his death at age 12 in 1984.

The French boys had X-linked SCID, a severe form that strikes only boys. It is the most common form of SCID, accounting for about half of cases.

The disease involves a genetic mutation that leaves them without certain proteins crucial to developing disease-fighting immune cells. Without treatment, sufferers die very young. Many babies with the disorder are saved with bone marrow transplants, but they need monthly infusions of immune globulin, antibodies culled from donated blood, for the rest of their lives.

DAVID, THE BOY IN THE BUBBLE. David Vetter was born in Houston in 1971. An older sister was healthy, but a brother had died of infection early in life. Since SCID was suspected, David was delivered by caesarian section and placed in a sterile incubator, which in time grew into the famous bubble. He probably had SCID-X1.

David was kept in the bubble for 12 years because no marrow donor with a good HLA match could be found, and for other reasons. The advances in monoclonal antibody technology of the '80s allowed his doctors to try treating his (mismatched) sister's marrow with anti-CD3 and complement, to rid it of mature T cells which cause GvH, and then transfuse it into David. Infection still is a great problem in bone marrow transplantation, before the marrow has a chance to "take;" major culprits are latent viruses of the herpes family, like CMV (cytomegalovirus) and EBV (Epstein-Barr virus). We'll take up the story from there in class.

WORKUP FOR IMMUNODEFICIENCY. When immunodeficiency is suspected, the family history and patient's history provide valuable clues. The physical examination is also important: How is the patient growing? Are tonsils visible, lymph nodes palpable? Are there, as in DiGeorge, associated abnormalities?

Malaria is not the *only* cause of headache.
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TESTS. You'll find it useful to keep three principles in mind.

First, if you have a good concept of the way in which the immune system fits together, you will be able to choose tests in a logical sequence. I guess this is true of any system. Look at the table and see if you agree that it follows logic that you agree with.

Second, go from procedures which test a large integrated system to tests which are more limited. For example, if a skin test with an antigen that produces good Th1-mediated immunity is positive, it tells you that the patient can: process antigen in APCs, recognize antigen, expand a T cell clone, activate T cells, secrete lymphokines, and respond to lymphokines; which is a lot more information than can be had by measuring $CD4^+$ cells.

Third, go from cheap or easy tests to more expensive or painful ones. We will go into tests in more detail later (*see* Diagnosing the Immune System).

A LOGICAL APPROACH TO WORKING UP DEFECTIVE IMMUNITY.

DEFECTIVE	INITIAL TESTS	MORE ADVANCED TESTS
B cells	• Quantitative IgG, IgA, IgM levels • Specific Abs to prior immunizations • ABO isohemagglutinins	• Ab responses to novel Ags • Sequence suspect genes • Lymph node biopsy
T cells	• Skin test with recall Ag panel • Total lymphocyte count • CD3, CD4, CD8 counts	• Mitogen responses, MLR, cytokine measurements • Sequence suspect genes
Phagocytes	• WBC count, differential, morphology • NBT test, oxidative burst	• Assays for phagocytosis, chemotaxis • Sequence suspect genes
Complement	• CH_{50} • Assay for C1inh (inhibitor)	• Individual complement component levels

Learning objectives for Immunodeficiency

1. Draw an outline diagram of lymphocyte development. On the diagram, indicate if possible locations of abnormalities of development in:
   - DiGeorge syndrome
   - Severe combined immunodeficiency (SCID)
   - X-linked (Bruton) hypogammaglobulinemia
   - Common variable immunodeficiency

2. Characterize the infections you would expect in a pure B cell deficiency and in a pure T cell deficiency.

3. Describe the clinical features which, although not immunological, are associated with DiGeorge syndrome.

4. Discuss the incidence of selective IgA deficiency, and the associated syndromes.

5. Describe the immunological problem of the Nude Mouse, and name the human immunodeficiency condition it resembles.

6. Name the enzyme which is absent in some cases of SCID. Discuss approaches to replacing this enzyme that are currently used.

7. Discuss transplantation therapy in immunodeficiency diseases. Include a consideration of side effects.

8. Given a child with recurrent infections, describe in principle tests which could be done to determine if there is a T, B or combined immunodeficiency, or a PMN, macrophage or complement problem.

9. On a diagram of a lymph node, label T and B cell areas.

10. Describe the contents of commercial gamma globulin and indicate the conditions in which it can be useful replacement therapy. Compare and contrast intramuscular and intravenous therapy.

11. Name two viruses which are immunosuppressive in humans. Discuss a possible mechanism for the immunosuppression caused by one of these viruses.