CASE 5 Factor I Deficiency

The alternative pathway of complement activation is important in innate immunity.

The complement system plays a crucial part in the destruction and removal of microorganisms from the body. Pathogens coated with complement proteins are more efficiently phagocytosed by macrophages, and bacteria coated with complement can also be directly destroyed by complement-mediated lysis. The system of plasma proteins known collectively as complement can be activated in various ways (see Fig. 4.1), of which the so-called alternative pathway is important in innate or nonadaptive immunity. This pathway can be activated in the absence of antibody, although even low titers of IgM antibodies against an infecting microorganism will greatly amplify complement activation.

The complement protein C3 is the starting point of the alternative pathway. It is one of the more abundant globulins in blood and is continuously being cleaved at a fairly low rate into a smaller C3a and a larger C3b fragment by a variety of host or microbial proteinases—this is called the 'tickover' of C3. Cleavage exposes a highly reactive thioester bond in the C3b fragment, which enables C3b to bond covalently with the hydroxyl group of serine or threonine in a protein or the hydroxyl group of a sugar on a microbial surface. If C3b fails to attach to a microbial surface, the thioester bond is spontaneously hydrolyzed and the C3b is inactivated (Fig. 5.1).

Topics bearing on this case:
Alternative pathway of complement activation
Factor I cleavage of C3b
Opsonizing activity of C3b
C3a activation of mast cells

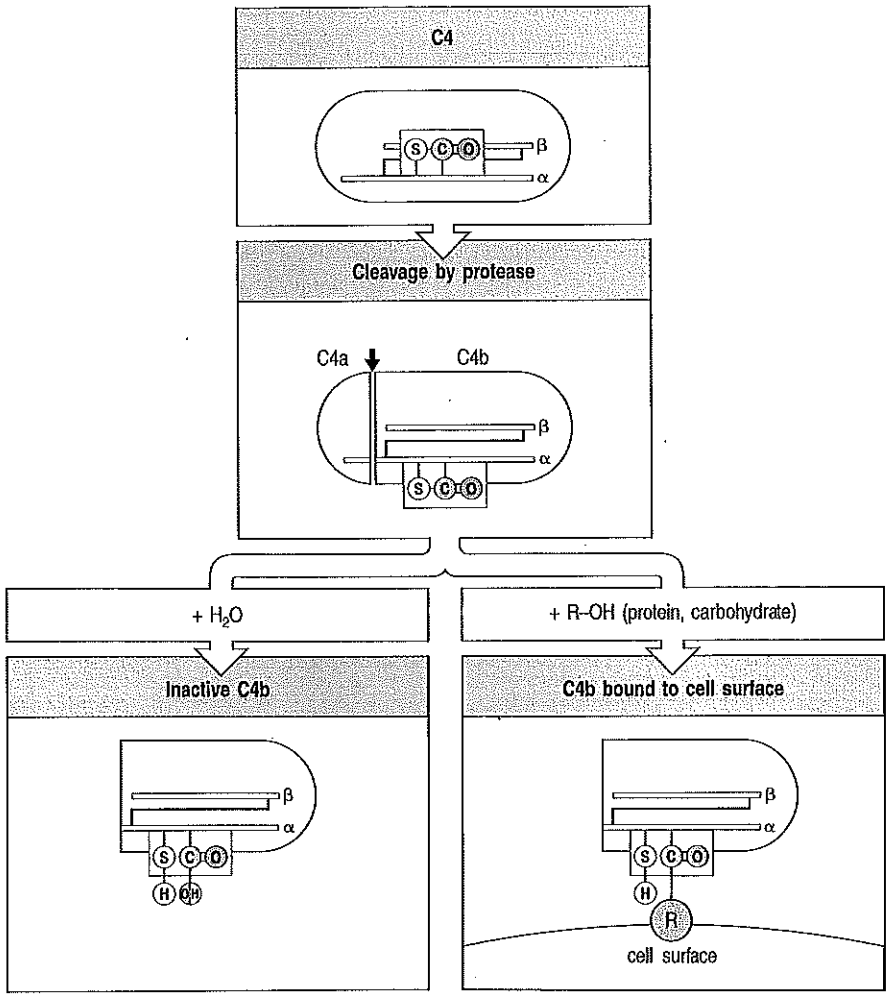


Fig. 5.1 Cleavage of C3 exposes a reactive thioester bond that enables the larger cleavage fragment C3b to bind covalently to the bacterial cell surface. Intact C3 has a shielded thioester bond that is exposed when C3 is cleaved by a proteinase to produce C3b. The highly reactive thioester bond in C3b can react with hydroxyl or amino groups to form a covalent linkage with molecules on the microbial surface. In the absence of such a reaction the thioester bond is rapidly hydrolyzed, inactivating C3b.

The binding of C3b to a microbial surface stimulates the cleavage of more C3 molecules. Another alternative pathway component, factor B, binds to C3b, and in this bound state is cleaved by a pre-existing blood proteinase, factor D, leaving the larger Bb fragment still bound to the C3b. The resulting C3b,Bb complex is an active serine protease, known as the alternative pathway C3 convertase, which specifically cleaves native C3 to make more C3b and C3a (Fig. 5.2).

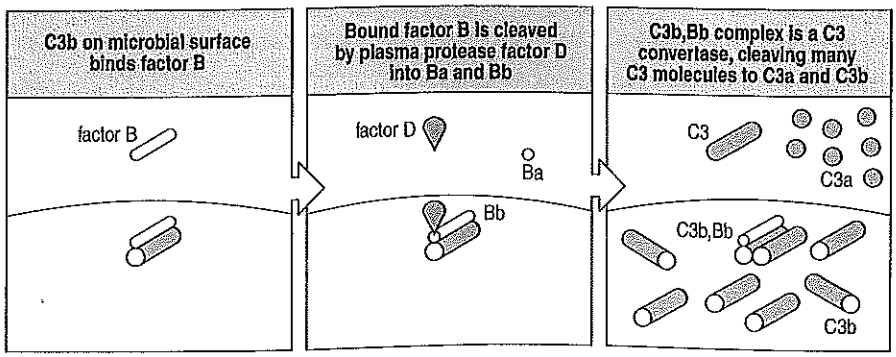


Fig. 5.2 The alternative pathway of complement activation leads to amplification of C3 cleavage. C3b deposited on a microbial surface can bind factor B, making it susceptible to cleavage by factor D. The C3b,Bb complex is the C3 convertase of the alternative pathway of complement activation and its action results in the deposition of many more molecules of C3b on the pathogen surface.

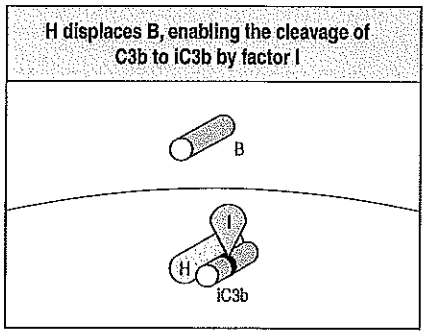


Fig. 5.3 The conversion of C3b to iC3b by factor I. On microbial surfaces, factor H displaces B from the C3b,Bb complex and cleaves C3b to produce iC3b. On host cell surfaces, the complement receptor CR1, which binds C3b, can substitute for factor H in this reaction.

C3b bound to a microbial surface acts as an opsonin by binding to a specific receptor, the complement receptor 3 (CR3), on phagocytes, facilitating the ingestion of C3b-coated particles. But before C3b can act as a ligand for CR3, and thus as an effective opsonin, it has to undergo a further cleavage to a fragment called iC3b, which is effected by a blood serine protease called factor I, acting in conjunction with the blood protein factor H, components of the alternative pathway (Fig. 5.3). iC3b acting at the receptor CR3 can activate neutrophils and macrophages in the absence of antibody (Fig. 5.4). Cleavage of C3b by factor I also has another critical effect. It inhibits the C3 convertase activity of the C3b complex, thus ensuring that supplies of C3 do not become depleted. On host cell surfaces, complement receptor 1 (CR1) may bind to C3b and serve as the cofactor instead of factor H.

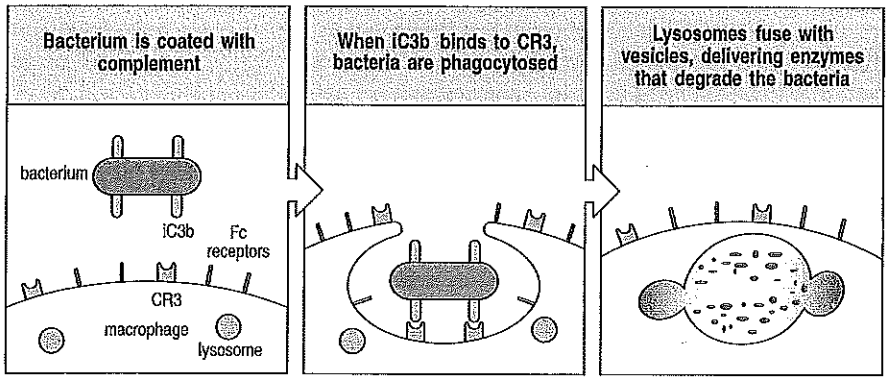
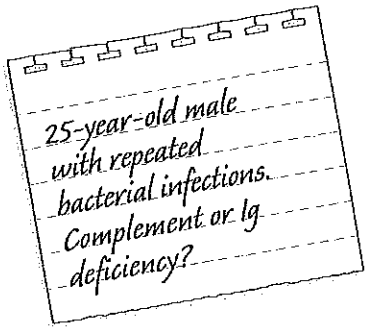
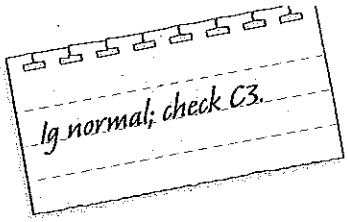


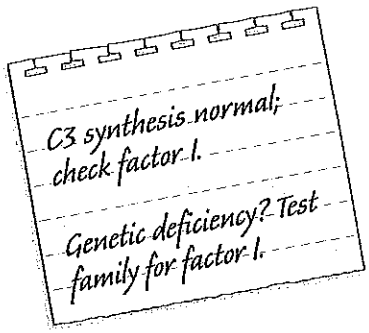
Fig. 5.4 iC3b binds to complement CR3 receptors on phagocytes and stimulates phagocytosis. iC3b on the surface of the pathogen binds to CR3 receptors on the phagocyte. Binding signals the phagocyte to engulf the particle and activates the internal destruction mechanisms.

The case of Morris Townsend: uncontrolled complement activation leads to susceptibility to infection and to hives.

Morris Townsend was admitted to the Brighton City Hospital at age 25 with pneumonia. This was his 28th admission to the hospital in his lifetime. From his first year onwards he had been repeatedly admitted for middle ear infections and mastoiditis. During these episodes, which were successfully treated with antibiotics, a variety of pyogenic (pus-forming) bacteria were cultured from his ears or mastoids, including *Staphylococcus aureus*, *Proteus vulgaris*, and *Pseudomonas aeruginosa*. At age 3 he had a tonsillectomy and adenoidectomy because of enlargement and chronic infection of his nasopharyngeal lymphoid tissue; at age 6 he had scarlet fever. He had also been admitted at other times with left lower lobe pneumonia (when *Haemophilus influenzae* had been cultured from his sputum), an abscess in the groin, acute sinusitis, a posterior ear abscess due to *Corynebacterium* species, skin abscesses with accompanying bloodstream infection (septicemia) due to β-hemolytic streptococci and, on one occasion, septicemia due to *Neisseria meningitidis* (meningococcemia).







On physical examination at his latest admission, Morris was found to be slightly obese but otherwise normally developed. His hearing was poor in both ears and this was attributed to his recurrent ear infections and mastoiditis. He also told doctors that he developed hives all over his body after drinking alcohol or after taking a bath or shower.

A urine analysis yielded normal results. His hematocrit was 43% (normal) and his white cell count was 6000 μl^{-1}. His platelet count was 240,000 ml^{-1} (normal) and his blood clotted normally. His red blood cells gave a strong positive agglutination reaction with an antibody to C3 but no agglutination with an antibody to IgG or IgM. His serum IgG level was 915 mg dl^{-1}, IgA 475 mg dl^{-1} and IgM 135 mg dl^{-1} (all normal). Morris responded normally to an injection of tetanus toxoid; his antibody titer rose from 0.25 to 8.0 hemagglutinating units ml^{-1}. He gave a positive delayed-type skin reaction to mumps and monilia antigens.

Serum levels of C3 were 27 mg dl^{-1} (normal values 97–204 mg dl^{-1}); of this, 8 mg dl^{-1} was C3 and 19 mg dl^{-1} C3b. The serum levels of all other complement components were normal except for factor B, which was undetectable. His serum failed to kill a smooth strain of *Salmonella newport*, even after addition of C3 to the serum to render the C3 concentration normal. To investigate the turnover of C3, Morris was injected with a dose of C3 labeled with the radioactive tracer ^{125}I. The results of this investigation showed that the rate of synthesis of C3 was normal but that C3 was being broken down at four times the normal rate (Fig. 5.5). A test of his serum with an antibody to factor I showed that his serum lacked factor I.

Morris's family had no history of recurrent bacterial infections, but investigations showed reduced levels of factor I in both his parents and in several of his siblings (Fig. 5.6).

Factor I deficiency.

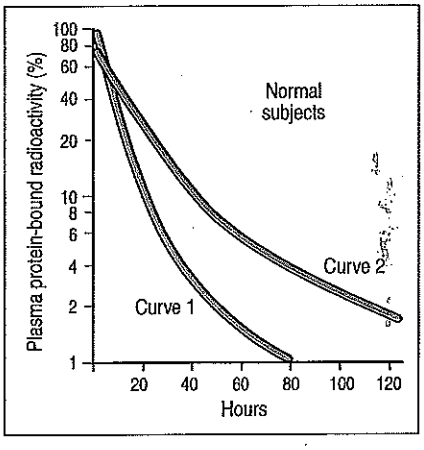


Fig. 5.5 The rate of disappearance of ^{125}I-labeled C3 from the plasma. The rate of disappearance of radioactively labeled C3 from the patient's plasma (curve 1) is much faster than that in normal subjects (shaded area; 11 normal subjects). Curve 2 shows the rate of disappearance of C3 in the patient after the infusion of 500 ml of normal plasma.

Patients such as Morris Townsend with a genetic deficiency in factor I were instrumental in deciphering the mechanism of activation of the alternative pathway of complement. Innate immunity is a first and highly effective means of defense against the common extracellular bacteria that cause pyogenic infections. The lack of factor I means that the alternative pathway C3 convertase is uninhibited and consumption of C3 is greatly accelerated, leading to C3 depletion. The lack of C3, and the nonproduction of iC3b, results in defective opsonization, which is the main means of removing and destroying these bacteria. Thus, factor I deficiency, like the genetic deficiency of C3 itself, results in a greatly increased susceptibility to infections with such bacteria. The clinical findings in factor I deficiency are not unlike those observed in X-linked agammaglobulinemia—a failure of opsonization results in frequent pyogenic infections.

The gene encoding factor I is on chromosome 4. The family of Morris Townsend provides a classic case of the inheritance of a recessive mutation in an autosomal gene (see Fig. 5.6). His parents, some of his siblings and two nephews are heterozygous for the defect; they produce roughly half the normal amounts of factor I, which is sufficient to prevent any clinical symptoms. Morris appears to be the only family member who is homozygous for the defect, and who thus exhibits symptoms.

Two interesting facts emerge from his clinical history. He sustained recurrent hives and had one bout of meningococcemia. It is easy to understand why he had hives. He was constantly cleaving C3 to C3a and C3b. C3a binds to mast cells and, among other things, causes the release of histamine and hence hives. The interesting question is why he did not have hives all the time and why they became problematic only after the ingestion of alcohol or exposure to hot and cold water. We must suppose that he had tachyphylaxis, or end-organ unresponsiveness, to histamine. Only when histamine release was increased, as by alcohol consumption or sudden changes in ambient body temperature, did the symptoms appear.

Morris Townsend's meningococcemia in particular is symptomatic of a deficiency in components of the alternative pathway of complement action. There are two common human pathogens in the bacterial genus *Neisseria: Neisseria gonorrhoeae* and *Neisseria meningitidis*. The former causes the sexually transmitted disease gonorrhea; the latter causes septicemia and meningitis and can be rapidly fatal. Patients have died from septic shock within 20 minutes of the onset of the symptoms of meningococcemia. Patients with genetic defects in the alternative pathway of complement activation or in the terminal components of complement sustain overwhelming and repeated infection with *Neisseria*. Deficiencies in the alternative pathway components factor D and properdin (factor B deficiency has never been observed in humans) were discovered because these patients developed recurrent meningococcemia. Similar observations have been made in patients with deficiencies of the later-acting C5, C6, C7, C8, and C9 components. These clinical observations highlight the importance of the bactericidal action of complement in controlling septicemia due to *Neisseria*.

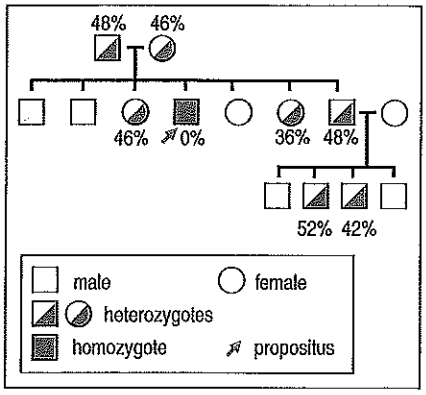


Fig. 5.6 Autosomal recessive inheritance of the factor I defect. The numbers indicate percentages of normal level of factor I in serum. No number indicates normal levels. The blue arrow indicates the patient, Morris Townsend.

Questions.

1 *Morris Townsend's clinical course has improved with age and he now has far fewer infections than he had as a child and adolescent. How do you explain this?*

2 *From the radiolabeled C3 experiment, we found that Morris Townsend catabolized the C3 very quickly but that his rate of synthesis of C3 was normal. What do you anticipate would happen if we repeated the experiment with radiolabeled factor B?*

3 *Morris Townsend was given a large dose of pure factor I intravenously. What changes would you predict to occur in his serum proteins?*

4 *What other genetic defect in the alternative pathway might lead to the same clinical and laboratory results as factor I deficiency?*

5 *Why did Morris' red blood cells agglutinate with antibody to C3?*