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The group of homogeneous galactan binding immunoglobulins from which several galactan binding heterologous recombinants have been prepared, constitutes and instructive case. Using the known sequences of these proteins, hypothetical, three-dimensional structures for some of them were constructed [*Feldman,* private commun.]. Examination of these structures revealed that several of the more important substitutions among these proteins are at the V_H-V_L contact areas. More specifically, some of the residues that determine the contacts between the domains are subject to a large number of variations as they belong to the J and D segments of the immunoglobulins. Thus, the domains contact forming residues are selected at the gene level by the recombination process of the J_H and D segments to V_H and of J_L to V_L. This early selection of preferential H-L association would affect not only the affinity between the chains but also their mode of antigen binding and the structural rearrangements that are induced by it. On the other hand, the formation of a hybrid which maintains the hapten binding capacity suggest that the substitutions are such that they allow for the structural and functional requirements of the newly formed site.

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Interaction of Complement and Gammaglobulins (Antibodies)

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The interaction of antibodies in immune complexes with the C1 complement (C1) molecule of the classical pathway or components of the alternate pathway is essential for the activation of the complement system. The activated complement system mediates biologically important reactions as cell lysis, anaphylatoxis, immune adherence, phagocytosis, chemotaxis, liberation of enzymes or pharmacologically active mediators from cells and to some extent the degree of immune response via complement receptors on immune cells. The interaction of the C1 molecule with antigen-antibody complexes consists in recognition of the complex, binding and activation of the C1. The C1q subnit of the C1 molecule recognizes and binds to the Fc part (CH2 domain) of IgG and IgM molecules. The trigger for recognition and binding is the transition from 'star' to 'staple' form of IgM molecules, and the close proximity of several IgG molecules bound to antigen. Activation of the attached C1 molecules is only obtained when several of the six arms of the C1q molecule are bound to the Fc portion of the antibody molecule. Activation of the C1 molecule means that the zymogen subunits C1r and C1s in the Ca^{2+} dependent complex with C1q are proteolytically activated to serine proteases. Activated C1r cleaves its substrate C1s. C1s esterase then further activates neighboring C4 and C2 molecules, thus initiating the classical complement pathway. It is unclear, however, how C1r is activated since no apparent proteolytic activity is associated with C1q. Isolated C1r has a tendency to spontaneously activate. It has been speculated that this might reflect an internal property of the C1r molecule where one chain of the C1r molecule might activate the other by cryptic protease sites. The detailed analysis of the spontaneous activation, however, showed that trace contaminations with proteases, even in the purest C1r preparations (> 96% C1r), cause this activation. Inhibition of the contaminating proteases with diisopropyl fluorophosphate leads to stable C1r preparations which are fully active, to reconstitute macromolecular C1 and to activate the classical pathway. The spontaneous activation of isolated C1r is, therefore, no model system for the activation of C1r within the C1 mole-

cule. It can be shown by iodination, Ca^{2+} removal, or monoclonal antibodies, that conformational changes are induced in the C1q and C1r molecule on binding of the C1 molecule to antigen-antibody complexes. It seems, therefore, possible that the events which induce the activation of the C1r molecule need the joint occurrence of simultaneous events in both the C1q and C1r molecule.

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Roles of Antibody and Complement in Phagocytosis and Killing of Extracellular and Intracellular Bacteria by Human Phagocytes

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I have studied the influence of antibody and complement on the interaction of human phagocytes with extracellular and intracellular bacterial pathogens. The extracellular bacteria that I studied were *Escherichia coli,* an encapsulated strain and an unencapsulated mutant. The intracellular bacteria that I studied were *Legionella pneumophila,* the agent of Legionnaires' disease. I have previously reported that *L. pneumophila* multiply intracellularly in human monocytes. These *E. coli* and *L. pneumophila* bacteria were completely resistent to the bactericidal effects of human serum even in the presence of high-titer specific antibacterial antibody.

When unencapsulated and encapsulated *E. coli* were attached to human polymorphonuclear leukocytes (PMN) with concanavalin A, PMN readily ingested unencapsulated but not encapsulated *E. coli.* PMN ingested encapsulated *E. coli* only if serum ligands such as antibacterial or anti-concanavalin A antibodies were added. Thus, attachment by itself results in ingestion of unencapsulated but not encapsulated *E. coli.* Phagocyte Fc or C3 receptors are required for ingestion of encapsulated *E. coli* even when these bacteria are attached to the phagocyte surface.

The *E. coli* strains were investigated in the absence of concanavalin A. Unencapsulated *E. coli* fixed the third component of complement (C3) to their surfaces in the absence of antibody. PMN and monocytes required complement but not antibody to ingest and kill these bacteria efficiently. In contrast, encapsulated *E. coli* fixed C3 only if antibody was present. PMN and monocytes required both antibody and complement to ingest and kill encapsulated *E. coli* efficiently. Thus, under physiologic conditions, PMN and monocytes efficiently ingest and kill *E. coli* only if C3 is fixed to the bacterial surface, and antibacterial antibody is required for C3 fixation to the surface of encapsulated *E. coli.*

To study the role of phagocyte complement receptors, I used antibacterial IgM to fix C3 to the surface of encapsulated *E. coli.* To study the role of phagocyte Fc receptors, I used antibacterial IgG in the absence of complement. PMN and monocytes ingested and killed encapsulated *E. coli* in the presence of IgM and complement, but not in the presence of either serum opsonin alone. In the presence of IgG alone, PMN and monocytes did not ingest or kill encapsulated *E. coli* efficiently; both IgG and complement were required for efficient ingestion and killing. Thus, the complement receptor mediates phagocytosis of complement-coated encapsulated *E. coli* and is the primary mediator of phagocytosis and killing of these bacteria.

L. pneumophila fixed C3 only if specific anti-*L. pneumophila* antibody was present. As with encapsulated *E. coli,* PMN and monocytes required both antibody and comple-