Complement Split Product C_3d As A Predictor Of Systemic Lupus Erythematosus Activity In Comparison To C_3, C_4, CH_{50}

Thesis

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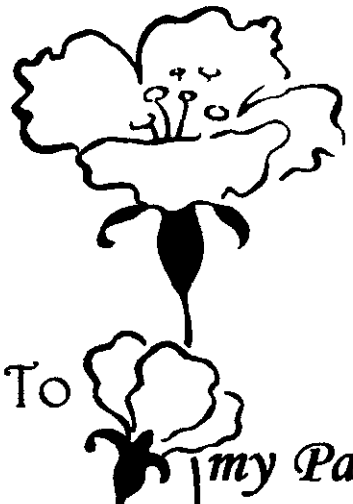
Dalia Fayez



Dedication

To

my Parents

Husband, &

my lovely Daughters

Dalia Fayez
(1997)

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Introduction and Aim of work

Introduction

Systemic lupus erythematosus (SLE) is an autoimmune disease characterized by a wide variety of clinical manifestations and serological abnormalities. So far, the assessment of SLE depends on the patient's condition once first seen by his doctor.

Attempts have been made to correlate the clinical condition of lupus patients with various laboratory measures including major markers of inflammation like the erythrocyte sedimentation rate and C-reactive protein, however, still remains uncertain ***(Senaldi et al., 1988)***.

Autoantibodies, immune-complexes, and complement which are directly involved in the pathogenesis of the disease have been studied extensively. The value of measuring anti-double stranded DNA antibodies to assess disease activity is controversial. The differentiation between reversible organ dysfunction (activity) and irreversible organ damage is a pre-requisite for potential recovery. The laboratory markers of disease activity especially the double stranded DNA and few complement may be normal in the presence of renal disease ***(Hay et al., 1993)***.

Clinical activity and exacerbation of SLE flares and remission are still difficult to predict and standardize. Studies about complement turnover may be of help for this assessment. Consequently, other measures are suggested which indicate complement specially C3d.

C3d is thought to be useful in the renal subset of SLE particularly the C3d / C3 ratio which provides a sensitive marker for disease activity ***(Rother et al., 1993)***.

Aim of the Work :

The aim of this work is to assess the validity of C3d as a sensitive marker of disease activity in comparison to C3, C4, CH50.

Review of Literature

Immune Complexes and Complement

Definition :

Immune complex disease is a group of diseases thought to be mediated by the deposition of immune complex in specific organ or tissue sites including the glomerulus of the kidney and blood vessel wall ***(Michael et al., 1994)***.

These immune deposits are thought to arise from Ag-Ab complexes formed in the circulation and activate the complement cascade. Portions of C9, C4, and C3 complement component modify the structure of the immune complex, inhibiting precipitation in tissues or solubilizing already deposited complexes ***(Moxley and Ruddy, 1993)***.

Once deposited in tissues, they activate a variety of potent soluble mediators of inflammation, such as complement proteins, causing an influx of polymorphonuclear neutrophils and monocytes. Complement fragment bound to the immune direct interaction with complement receptors on various cell surfaces modifying the processing of immune complexes and enhancing their delivery to phagocytic cells ***(Schifferli and Peters, 1986)***.

Complexes activate a variety of cells with surface membrane receptor for immunoglobulin and directly induce the release of toxic products of oxygen with cytokines, various proteases and other enzymes causing irreversible tissue damage ***(Steinberg, 1992)***.

While the specific etiology of these diseases is variable, they share a common pathophysiology. The clinical features of these diseases are quite diverse ranging from mild cutaneous eruption to severe organ involvement with pericarditis, glomerulonephritis, and vasculitis ***(Michael et al., 1994)***.

Pathophysiology :

The introduction of noxious materials into an individual is often followed by an immune response. Specific antibody produced as a response binds to antigen forming immune complexes which are phagocytosed and destroyed by neutrophils and macrophages locally of tissue sites and if released into circulation they are cleared from circulation by phagocytic cells of RES ***(Michael et al., 1994)***.

However, at times, these complexes are deposited in tissues causing inflammation and tissue damage. The biologic activity of the complexes has been studied; not all immune complexes are alike in their capacity to induce tissue damage, as antigens differ in size, charge, and ability to bind to macromolecules. Antibodies differ in isotype, valence, affinity, relative charge, and ability to activate complement ***(Moxley and Ruddy, 1994)***.

It has been shown that the isotype of antibody affect biologic activity. Thus, IgG - IgM containing complexes activate the classic complement pathway and IgA containing complexes often activate the alternative complement pathway. In contrast, cell surface IgE complexes are capable of mediating the degranulation of mast cells by a non-cytotoxic, complement-independent mechanism. The size of immune complexes in the

circulation is an important parameter of toxicity. In general, larger complex causes more tissue damage than the smaller complexes, the size is related to the concentration and molar ratio of antibody and antigen as well as the avidity of Ab for Ag. The ratio of Ag to Ab may range from Ab excess to Ag excess through Ag - Ab equivalence. In marked Ab excess, Ag valences are saturated and in general, the complexes are small, while on antigen excess, antibody combining sites are saturated, chances for lattice formation are limited and again the complexes are small. Immune complexes formed at moderate Ag excess are thought to be most pathogenic, perhaps because they are most efficient at activating the various mediator system such as the complement cascade ***(Michael et al., 1994)***.

The interaction of antigen with antibody in the circulation results in the formation of soluble macromolecule complexes. These complexes localize in tissues for anatomic and physiologic reasons and not because of any immunologic specificity; therefore, their physical properties are of great importance. The size and solubility of the complexes, their concentration, their ability to engage and activate the complement system, and the duration of their presence in the circulation are all significant determinants ***(Wener and Mannick, 1986)***. The strength of the bond between antigen and antibody is responsible for the stability of the complex. Low affinity antibody binds less tightly or to a less number of sites on the antigen when compared with precipitating antibody. Antigens bound by low affinity antibody are eliminated more slowly and theoretically have a greater opportunity to produce kidney damage ***(Virgil and Woods, 1993)***.

Thus, the size, Ag to Ab ratio, quality of the Ab are probably more important determinants of nephrotoxicity than the absolute quantity of circulating immune complexes ***(Wener and Mannik, 1986)***.

Complement is an important mediator of the damage caused by immune complexes and by autoantibodies ***(Shumark and Rock, 1984)***. Although Ag - Ab complexes undoubtedly cause injury in experimental models of nephritis, it remains controversial whether deposition of circulating complexes or their formation in situ is the more important ***(Rees, 1983)***.

The charge of Ag and Ab also appears to be important in determining the pathophysiologic effect of the complexes, the positively charged immune complexes tend to deposit in the renal glomeruli. This is presumably due to the fact that the glomerulus presents a negatively charged surface to the circulation. Also, the basement membrane of skin has negatively charged surface ***(Singh, 1993)***.

Anatomic features are important for immune complex deposition. Glomerular endothelial cells are separated by tiny fenestrations. Intact immune complexes can pass through them and subsequently localize against the glomerular basement membrane where they appear as subendothelial dense deposit. Whether macromolecular complexes can penetrate the basement membrane and deposit in the subepithelial region or if Ag - Ab pass individually and then combine there, also the negatively charged material is thought to regulate the permeability of glomerular capillary wall to circulating macromolecules as discussed before ***(Wener and Mannick, 1986)***.

The venerated concept that immune complexes cause disease began with the observation of ***Von Pirquet (1911)*** on serum sickness, and then many retrospective studies that evidence the role of immune complexes in serum sickness. ***Moxley and Ruddy (1993)*** reported that initial injection of horse serum is followed by urticaria, fever, edema and arthralgia by 8 - 12 days. The reaction to second injection was more rapid and more severe. The injection of Ag is followed by a period of intravascular equilibration and then by intravascular - extravascular equilibration lasting several days ***(Michael et al., 1994)***.

This period is followed by progressive decline in the level of antigen in the circulation representing the degradation of injected serum protein followed by clearance of Ag from the circulation beginning at 7 - 8 days and this is due to the development of an immune response, i.e. formation of Ag - Ab complexes and subsequent clearance from the circulation by cells of RES ***(Walport, 1993)***. During this period, there is fall in complement serum level and pathological changes occur in large arteries as renal glomeruli, joints, and cardiac vessels. There is edema and swelling of endothelial cells, infiltration of monocytes and few granulocytes with deposition of immunoglobulin and C3 along basement membrane in a typical granular pattern ***(Michael et al., 1994)***. As regard arteries, there is marked intimal proliferation of endothelium followed by degradation o internal elastic lamina and adventitia with resulting fibrinoid necrosis of the vessels. By immunofluorescence microscopy, immunoglobulin, Ag and C3 were found roughly in the region of internal elastic lamina which are rapidly phagocytosed by neutrophils present in the lesion. Acute serum sickness is present only as long as these circulating immune complexes persist and resolves rapidly once