THE ROLE OF ANTINEUTROPHIL CYTOPLASMIC ANTIBODIES IN THE DIAGNOSIS OF SYSTEMIC VASCULITIS IN CHILDREN

THESIS

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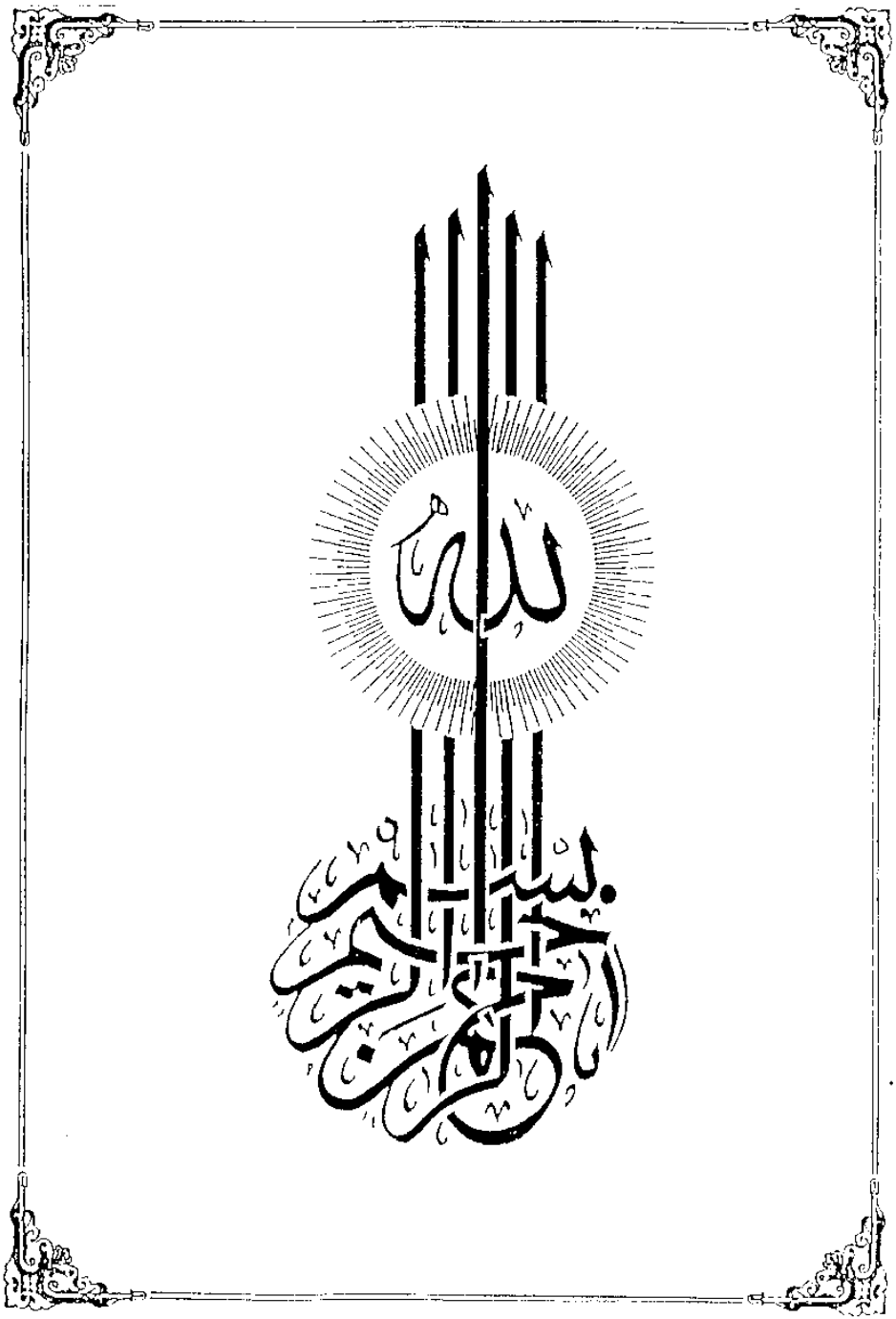

LIST OF ABBREVIATIONS

AECA	:	Antiendothelial cytoplasmic antibodies
ANA	:	Antinuclear antibodies
ANCA	:	Antineutrophil cytoplasmic antibodies
ANTI-MPO	:	Antimyeloperoxidase antibodies
ASO	:	Antistreptolysin O.
BCGF	:	B cell growth factor
C-ANCA	:	Cytoplasmic antineutrophil cytoplasmic antibodies
CRP	:	C-reactive protein
DNP	:	Deoxy ribo-nucleoprotein
ELISA	:	Enzyme linked immunosorbent assay
ESR	:	Erythrocyte sedimentation rate
GBM	:	Glomerular basement membrane
GN	:	Glomerulonephritis
3 HDIP	:	Tritiated diisopropyl Fluorophosphate
HLA	:	Human leucocyte antigen
HSP	:	Henoch Schonlein purpura
HUVEC	:	Human umbilical vein endothelial cell
IGPT	:	Indirect granulocyte phagocytosis test
IL-4	:	Interleukin -4
JRA	:	Juvenile rheumatoid arthritis
MPA	:	Microscopic polyarteritis
n-DNA	:	Native deoxyribonucleic acid

PAN	:	Polyarteritis nodosa
P-ANCA	:	Perinuclear antineutrophil cytoplasmic antibodies
RIA	:	Radioimmuno assay
RF	:	Rheumatoid factor
SLE	:	Systemic lupus erythematosus
SNGN	:	Segmental necrotizing glomerulonephritis
VWF	:	Von willebrand factor

INTRODUCTION

Antineutrophil cytoplasmic antibodies (ANCA), were first reported in 1982 in 8 patients with pauciimmune glomerulonephritis (Davis et al ., 1982). In 1984, Hall and co-workers reported finding ANCA in 4 patients with systemic vasculitis.

In 1984, Woude & collaegues described antibodies in sera of patients with Wegener's granulomatosis which react with cytoplasmic constituents of human peripheral blood neutrophils. The antigen appears to be associated with a 29 KDa serine protease in the primary granules in the cytosol.

Until 1988, only one ANCA subtype had been reported which had a cytoplasmic pattern by indirect immunofluorescence. this was given the designation c - ANCA. Another ANCA subtype with a perinuclear pattern (p-ANCA) which usually was specific for myeloperoxidase (MPO), was reported to be present in cases of SLE by Nassberger et al., in 1990.

Circulating antibodies directed against neutrophil cytoplasmic antigens (ANCA) are specific markers for systemic vasculitis such as Wegener's granulomatosis & microscopic polyarteritis. (Van der Woude et al., 1985; Falk et al., 1989).

Wegener's granulomatosis is a systemic disease characterized by necrotizing granulomas in the upper and lower respiratory tract, in association with necrotizing granulomas in the kidney (necrotizing crescenting glomerulonephritis) and systemic vasculitis. (Facui et al., 1983). (The disease usually run a rapidly fatal course however, with cyclophosphamide and corticosteroids treatment, long term remission may be achieved, provided that the diagnosis is established early).

In Wegener's granulomatosis there is no specific diagnostic test, and the diagnosis depends upon a combination of typical clinical features and characteristic histological appearance which is often inconclusive and this sometimes leads to irreversible organ damage (Pinching et al., 1983).

Recently, ANCA were demonstrated to be specific for Wegener's granulomatosis and a significant correlation between these antibodies and disease activity was observed. (Gross et al., 1986).

Aim of the study:

The aim of the present study is to detect and explore the specificity of antineutrophil cytoplasmic antibodies (ANCA) in patients with systemic vasculitis.

REVIEW

OF THE

LITERATURE

VASCULITIS

Vasculitis is defined as a clinicopathologic process characterized by inflammation and necrosis of blood vessels. Arteries, and sometimes veins, of various sizes and of different locations throughout the body may be involved, resulting in a great diversity of symptoms and findings (Facui, et al., 1978; Hunder et al., 1990).

The clinical spectrum ranges from a primary disease involving exclusively blood vessels to an involvement of vessels as a relatively insignificant component of another systemic disease, (D'Cruz and Hughes, 1992).

The various patterns of the disease depend on the size and location of the affected vessels. When small non muscular vessels are involved, the disease makes the form of Henoch Schonlein vasculitis (anaphylactoid purpura), with involvement of larger muscular arteries the disease is called polyarteritis nodosa, variants include infantile polyarteritis, Wegener's granulomatosis, and probably Kawasaki disease. Some overlap of these syndromes occurs and vessels of various sizes may sometimes be involved in the same patient (Hunder et al.,1990).

In Takayasu arteritis the aorta and other great vessels are the sites of inflammation. Inflammation of the blood vessels also occurs in other rheumatic diseases in children, notably rheumatoid arthritis, lupus erythematosus, dermatomyositis and scleroderma, in hypertension and in vessels exposed to local infection, trauma or thromboemboli (Sigal, 1987; Fauci and Leavitt, 1989).

Classification of Vasculitis

Because vasculitis can potentially involve any blood vessel, a complex and often confusing array of clinical syndromes results. vasculitis is classified as follows:

1. Systemic necrotizing vasculitis:
   - A. Polyarteritis nodosa
   - B. Allergic angitis and granulomatosis
   - C. "Polyangitis overlap syndrome"
   - D. Associated diseases (connective-tissue diseases, hepatitis B, cytomegalovirus infection, hairy cell leukaemia).

2. Small vessel (hypersensitivity) vasculitis:
   - A. Henoch-Schonlein purpura.
   - B. Serum sickness
   - C. Other drug-related vasculitis

D. Vasculitis associated with food, foreign protein, or other exogenous antigens.
E. Vasculitis associated with a systemic disease
F. Hypocomplementemic urticarial vasculitis
G. Congenital deficiencies of the complement system
H. Erythema elevatum.

3. Arteritis of larger arteries
   A. Temporal (cranial) arteries.
   B. Takayasu's arteritis
   C. Large-artery arteritis- complicating diseases such as ankylosing spondylitis, reiter's syndrome, relapsing polychondritis, and Cogan's syndrome.
   D. Aortitis-associated syphilis.

4. Behcet's disease.

5. Wegener's granulomatosis.

6. Thromboangitis obliterans (Buerger's disease).

7. Isolated angitis of the central nervous system.

8. Mucocutaneous lymph node syndrome (Kwasaki's disease)

9. Miscellaneous vasculitis syndromes.
   (Cupps and Fauci, 1981).

Pathophysiology:

The vasculitis are a heterogenous group of clinical syndromes, and therefore no single cause explains the pathophysiology of all of the inflammatory vessel diseases. The best characterized mechanism is immune complex mediated vasculitis. The elements necessary for the expression of this process include:

1. Soluble immune complexes larger than 19S formed in slight antigen excess.
2. Increased vascular permeability with passive deposition of complexes in the vessel wall.
3. Activation of complement with subsequent attraction of polymorphnuclear neutrophils (PMN) to the site of immune-complex deposition.
4. Release of inflammatory mediators and disruption of vascular integrity.

Aberrant regulation of the T-B cell, monocyte-macrophage and endothelial cell function may be important in some of the vasculitides. Other factors that determine disease activity include the class of blood vessel affected, the pattern of organ system involvement, and the relative sensitivity of the involved tissue to the effects of ischaemia, (Fauci et al., 1978, Fan et al., 1980 ; Falk and Jennette , 1991).

Both Henoch Schonlein vasculitis and polyarteritis may follow exposure to drugs or allergens, polyarteritis nodosa has been associated with hepatitis B, vascular damage presumably being caused by immune complexes of the viral antigens and antibody. In serum sickness, vasculitis is caused by deposition of immune complexes. The clinical pattern of some diseases such as Wegener's granulomatosis and Churg-Strauss syndrome with prominent involvement of the respiratory tract, suggest that an inhaled allergen may be responsible. Furthermore, studies of sputum and bronchoalveolar lavage fluid suggest that antineutrophil cytoplasmic antibodies may actually be produced in the lungs of these patients, (Baltro et al., 1991).

Polyarteritis nodosa has been associated with hepatitis B, vascular damage presumably being caused by immune complexes of the viral antigen and its antibody. Genetic factors may also be important as the histocompatibility antigen DR-2,was found more commonly in patients with Wegener's granulomatosis (Elkon et al., 1983).

Patients with systemic vasculitis typically present with malaise, fever, and weight loss but may have had non specific symptoms long before this. For example, patients with Wegener's granulomatosis and Churg-Strauss syndrome may have experienced sinusitis, nose bleeds,

painful red eyes, joint pains or sensory loss for months or years before the condition is diagnosed. Patients with non specific symptoms but without renal impairement present particular difficulties and diagnosis may be considerably delayed (D'Cruz et al., 1989 ; D'Cruz and Hughes , 1992).

In childhood, Henoch Schonlein vasculitis is the most commonly encountered type, polyarteritis and its variants are much rarer in children. In contrast to most other rheumatic diseases, Henoch-Schonlein vasculitis and polyarteritis nodosa predominantly affect males (Brasile et al.,1989).

The systemic vasculitic disorders Wegener's granulomatosis (WG) and microscopic polyarteritis (MPA) have a poor prognosis in the absence of immunosuppressive therapy. The morbidity and mortality are mainly related to the occurrence of severe lung hemorrhage and rapidly progressive glomerulonephritis (Savage et al., 1985 ; Couser , 1988 ; Falk et al ., 1990).

The diagnosis of systemic vasculitis in general, and WG and MPA in particular, has been based on clinical findings and tissue biopsy, since no specific serological test has been available (Haworth et al., 1985). This approach has meant that in patients without classical

clinical features, diagnosis and treatment may be delayed, and unfortunately the management of these potentially treatable vasculitic syndromes is impeded by the lack of specific tests to provide laboratory diagnosis and to allow monitoring of disease activity. Hitherto, only general tests to detect the presence of an active inflammatory response have been of any value (Hind et al.,1984 ; Falk and Jennette , 1991) .

Most rheumatic diseases lack pathognomonic features. Because of this, the physician is forced to rely on the presence of combination of clinical and laboratory manifestations in order to identify specific rheumatic diseases (Falk and Jennette , 1991).

As vasculitis comprise a diverse group of conditions and even there is marked variability of the different cases of the same type of vasculitis, this has prevented the development of a universally accepted classification of vasculitis. Yet, it is apparent that within the group of vasculitis patients, there are several clinical syndromes which appear to be distinct processes(Jennette and Falk , 1990).

In attempts to define such interrelationships, several investigators have described classification of vasculitis (Fauci et al.,1978 ; Alarcon, 1980; Hunder