MEASUREMENT OF IMMUNE COMPLEXES IN RHEUMATOID ARTHRITIS BY PLATELET AGGREGATION TEST

THESIS

Submitted For The Partial Fulfilment of The

Master Degree " M.Sc. "

' GENERAL MEDICINE '

BY

Mohammad Mohammad EL.Husseiny Khalil

M.B. , B.CH.

Supervised by

Prof. Mohammad Diaa El.Din Soliman
Prof. of Medicine
Ain Shams Faculty of Medicine

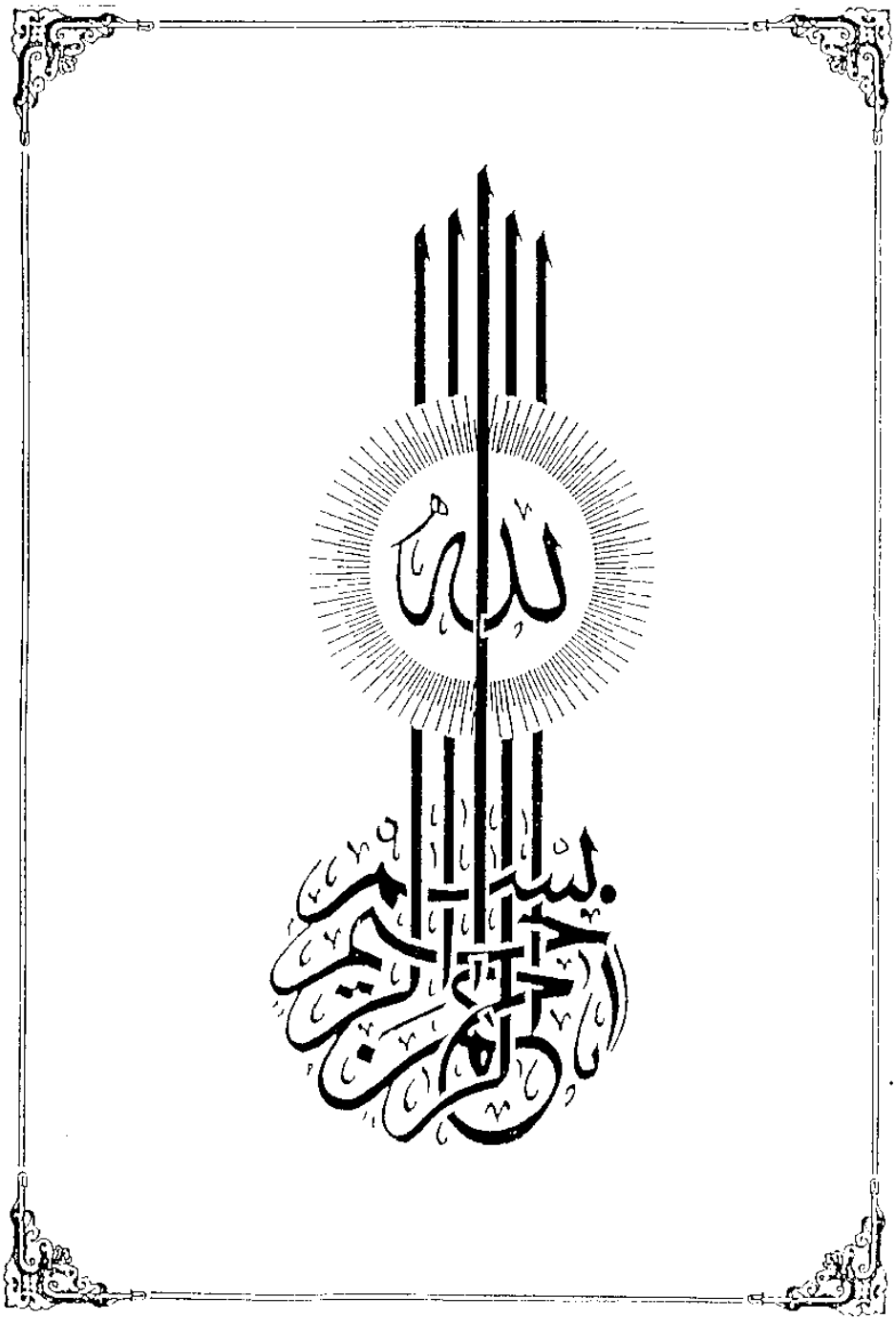
Prof. Wagih Naguib Ibrahim
Prof.of Clinical Pathology
Ain Shams Faculty of Medicine

Dr. Kamal Abdel Monaem Ali Amer
Assist. Prof. of Clinical Pathology
Military Medical Academy

Faculty of Medicine
Ain Shams University
CAIRO
1983

ACKNOWLEDGEMENT

I wish to express my thanks and appreciations to Prof. Dr. Mohammad Diaa El-Din Soliman, Prof. of Medicine, Ain Shams University, for suggesting the idea and plan of this study, his patience, kind and continuous support , through the whole work. His exceptionally cordial and able assistance can not be denied. To Prof. Dr. Wagih Naguib Ibrahim, Prof. of Clinical Pathology, Ain Shams University, I acknowledge my indebtedness and record my gratitude, for clarifying certain aspects of the practical part and for his many helpful suggestions and constructive creticism . I am also indebted to Colonel Dr. Kamal Ali Amer, Assist. Prof. of Clinical Pathology, Military Medical Academy , for his invaluable assistance, during the protracted preparation of this work and for revising the whole manuscript. My thanks are due and in full to the staff of Maadi Armed Forces Hospital, particularly Colonel Dr. Sami Hassan and Colonel Dr. Mahmoud Sami, for allowing me to make use of the rheumatology out-patient clinic, to study the cases of this work . I must also place on record the help given by the Librarians , to locate the references necessary in completing this work. A good share of my thanks goes to my



family, without whose cooperation and understanding, this work could not have been finished. And above all, thank God for completing this work .

.....

INDEX OF CONTENTS

INTRODUCTION AND LITERATURE REVIEW

RHEUMATOID ARTHRITIS

HISTORIC :

In ancient times, many forms of arthritis were considered together, under the general category of rheumatism, but it is clear from clinical descriptions, available to the medical historian, that rheumatoid arthritis 'RA' has afflicted mankind for a considerable period of time. Hippocrates himself may have recognized the disease in early times (Copeman, 1964) . Thomas Sydenham described a disorder, which he called rheumatic polyarthritis (Sydenham, 1909), but it was not until the latter part of the nineteenth century that Garrod used the term RA and a clinical impression of the disease began to be formed. From 1650 to 1850, all rheumatic diseases were treated categorically under the general term gout ; but in 1859, Garrod noted that elevation of blood uric acid was not present in RA . Nowadays RA is defined as a systemic disease of connective tissue , in which the striking manifestation is its tendency to produce lesions in joints and periarticular structures. Extraarticular features such as subcutaneous nodules , episcleritis , pleuritis and neuropathy , as well as

constitutional symptoms of fever, fatigue and weight loss emphasise the systemic nature of the disease (Tannenbaum and Hawkins, 1976).

DIAGNOSIS

The clinical presentation and course of RA is very variable. This has necessitated a committee of the American Rheumatism Association (ARA), to propose criteria for classic, definite and probable RA. These criteria are outlined as follows ;

1- Morning stiffness lasting 1 hour or more .

2- Pain on motion or tenderness in at least one joint.

3- Swelling (soft tissue thickening or fluid - not bony overgrowth) in at least one joint continuously for not less than 6 weeks.

4- Swelling of at least one other joint (any interval of joint free symptoms between the two joint involvements may not be more than three months).

5- Symmetrical joint swelling with simultaneous involvement of the same joint on both sides of the body. Distal interphalangeal involvement will not satisfy this criterion .

6- Subcutaneous nodules over bony prominences, on extensor surfaces or in juxta-articular regions.

7- X-ray changes typical of RA (which must include at least bony decalcification localised to, or greatest around, the involved joints and not just degenerative changes. Degenerative changes do not exclude the diagnosis.

8- Positive sheep red cell agglutination test. (Any modification will suffice that does not give more than 5% positive results in control subjects).

9- Poor mucin precipitate from synovial fluid (with shreds and cloudy solution).

10- Characteristic histologic changes in the synovium, with three or more of the following : marked villous hypertrophy ; proliferation of superficial synovial cells often with palisading ; marked infiltration of chronic inflammatory cells (lymphocytes or plasma cells predominating) with tendency to form "lymphoid nodules " ; deposition of compact fibrin either on the surface or interstitially ; foci of cell necrosis.

11- Characteristic histologic changes in nodules showing

granulomatous foci with central zones of cell necrosis, surrounded by proliferated fixed cells, and peripheral fibrosis and chronic inflammatory cell infiltration predominantly perivascular.

Classical RA requires at least seven criteria and the joint symptoms and signs must be continuous for at least six weeks.

Definite RA requires at least five criteria. The joint symptoms and signs must be continuous for at least six weeks.

Probable RA requires at least three criteria with continuous joint symptoms of at least four weeks.

PREVALENCE AND AETIOLOGY :

PREVALENCE

RA afflicts an estimated 2-3 % of population . It occurs in all races and at all ages, with peak incidence in the 4 th and 5 th decades. Approximately two to three times more females than males acquire this illness (Panush, 1979) . Female preponderance is explained, if one thinks of the disease as one in which immunologic

events exercise a fundamental role. The capacity for females to surpass males in a broad variety of measurements of specific immune responsiveness is now well established. Thus, elevations of either specific antibody production or quantitative levels of various immunoglobulins (Igs) have often been recorded when females are compared to males, and an enhanced primary as well as secondary immune response is also noted when females are compared to males (Ainbender et al., 1968). In addition cell mediated immunity is more vigorous in females than in males (Brent and Medawar, 1966; Kongshaun and Bliss, 1970) and all things considered, it may be reasonable to suggest that these interesting sex differences in the immune response may influence male to female expression of RA itself.

AETIOLOGY:

The aetiology of rheumatoid disease ' RD ' remains unknown. However, it has been suggested that RD is a "two phase " disease. Phase one is characterised by an antigenic alteration in the synovium, produced by an infectious agent or perhaps by an intrinsic metabolic abnormality

in connective tissue. Phase two is a self perpetuating immunologic reaction to the new antigens of other exogenous or endogenous origin (Glynn, 1972). The numerous studies aimed at isolating infectious agents, such as bacteria, mycoplasma and viruses from either synovial fluid or synovial membrane have yielded inconclusive results. However, in patients with juvenile RA, elevated IgM and IgG rubella antibody levels have been demonstrated. Rubella virus specific antigen has been detected in the synovial tissues of about one - third of these patients (Ogra et al., 1975). Also, there is raised antibody level to measles virus in RA sera. Rheumatoid factor 'RF' appears in some bacterial infections such as subacute bacterial endocarditis and titre falls after treatment with antibiotic. RA like models can be induced in animals with mycoplasma (Golding, 1978). Thus, infection as a cause of RA has a fluctuating popularity and if an infectious agent is an aetiologic factor in RA, it may exist in a defective or noncytopathogenic state, as the case with slow or transforming viruses (Barland, 1973).

Many investigators have proposed that an interaction between an infectious agent and the immune system in a

genetically susceptible host is important in the pathogenesis of RA. Although there is a slight tendency for diffuse connective tissue disorders and immunologic abnormalities to occur in families of RA patients, no consistent genetic pattern has been established. There is no evidence of an increased frequency of a given HLA haplotype in patients with RA (Lies et al., 1972). The immunologic role in RA may be evidenced by the infiltration of synovial membrane by lymphocytes and plasma cells and detection of high levels of circulating Igs, RFs and occasionally antinuclear factors (ANFs). RF is demonstrated, as well, in lymph nodes and synovial membranes . Immune complexes ' ICs ' formed by IgG, RF and complement (C) are demonstrated in synovial fluid leucocytes . C level is low in synovial fluid, being used up to form IC. Vasculitis in RA resembles serum sickness. However, injection of RF into normal persons does not produce RA, though high titres may persist, for several weeks (Golging, 1978) . There is a rare form of seronegative polyarthritis, occuring in adult onset hypogamma-globulinaemia, primarily affecting large joints and accompanied by tenosynovitis of the hands and feet. The

condition resembles RA, but the joints show no erosions, and the joint effusions contain no polymorphs and have normal C levels. They have recurrent chest infections and they show dramatic response to gamma-globulin treatment (Webster et al, 1976).Familial tendency or at least genetic influence has been observed in RA by many investigators (Bunin et al,1964) .The most releaving is twins study particularly monozygotic twins. Concordance appears in 30 to 40 per cent of monozygotic pairs who were followed up for long times. Meyerowitz et al studied eight cases of discordant monozygotic twins, in 1968. These cases had RD onset at childhood and remained discordant for 23 years. Baum and Fink (1968) reported two concordant and two discordant twin-pairs, in a study on four pairs of monozygotic twins. Tannenbaum and Hawkins (1976) have shown two of two concordant monozygotic twin pairs. In both , disease onset was at childhood. Thus, not all pairs reported showed rheumatoid concordance. This may indicate that other host or perhaps environmental factors have an important role in determining actual expression of the disease. Recent work by Stastny (1974; 1976& 1979) has provided a link between HLA or histocompatibility types

to RA, in particular the HLA. D4 phenotype. An association with HLA.DRWR has also been reported and appears to be associated with the severer forms of the disease. Apart from infection, immunology and genetics as main factors influencing the aetiology of RD, other less important factors co-exist. The disease occurs following trauma to a joint.It is more prevalent in temperate than in tropical climates or Eskimos. The disease is more frequent, following mental trauma, explaining a psychologic basis for the disease. A hormonal role may influence the disease as the disease is more common in females, with remissions during pregnancy and exacerbations at the menopause. Acromegaly is associated with rheumatoid-like arthritis. Adrenocortico-trophic hormones ' ACTH ' produce marked decrease in disease activity. However, adrenocortical activity shows no constant abnormality. RA is uncommon in association with hyper-thyroidism and the disease may show exacerbation, after thyroidectomy (Golding, 1978).

IMMUNOLOGIC ASPECTS AND PATHOGENESIS OF RA

IMMUNOLOGIC ASPECTS

Although there is no firm evidence that the primary abnormality in RA is an immunologic one, there is little doubt now that immunologic processes play a central role in the pathogenesis of the synovitis.For many years, it has been recognised that patients with RA often show the presence in their sera of RF or antibodies to gamma globulins (Ralph and Williams, 1980). They may therefore be directed against IgG , IgA , IgM, IgD or IgE. Usually, only those antibodies directed against IgG are reffered to as RFs, in patients with RA (Panush, 1979). These antibodies were first recognised by Cecil et al. (1931) during agglutination reaction studies, using streptococci and later by others, using various types of sensitised particles (Waaler, 1940; Rose et al., 1948). Early studies identified RF as 19 S IgM antibodies.Subsequent work revealed that RFs are not restricted to 19 S IgM antibodies, but also occur as IgG, IgA, IgD and 7 S IgM antibodies (Stage and Mannik, 1972 - 1973) . To detect