SIGNIFICANCE OF COMPLEMENT PROFILE AND COMPLEMENT RECEPTOR 1 EXPRESSION IN RBC AND KIDNEY TISSUE IN IMMUNE COMPLEX MEDIATED DISEASE

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ABSTRACT

A study was conducted to evaluate role of complement proteins and complement receptor 1 (CR1) in pathogenesis of Systemic lupus erythematosus (SLE) and Immune complex (IC) mediated glomerulonephritis. C3, C4, C3d and CH100 in serum, CR1 in renal biopsies and RBC showed these parameters to be of great diagnostic and prognostic values in Immune complex mediated diseases. Our study revealed an overall decrease in levels of CR1, C3, C4 in IC mediated as compared to non - IC mediated disease. Whereas C3d in case of SLE 247 ± 39 AU/L including IC mediated Glomerulonephritis (ICGN) 208 ± 51 AU/L was found to be significantly increased (P < 0.05) than normal control 46 ± 6 AU/L. There was no appreciable increase in case of non - 1C mediated GN (61 ± 12 AU/L) CRI among SLE patients (261 ± 141/E) and IC mediated group (270 ± 107/E) was found to be significantly lower (P < 0.05) than normal control (627 ± 132/E) and non - IC GN (550 ± 86/E). C4 values among SLE, patients were found to be 191 ± 104 mg/L as compared to control (286 ±110 mg/L). The kidney biopsy of type III and type IV lupus nephritis revealed a complete absence of CR1 in contrast to minimal change diseases. Thus this study revealed that above parameters could be a valuable tool for distinguishing IC versus non-IC mediated kidney diseases.

Key Words: Complement receptor 1 (CR1) Glomerulonephritis, SLE.

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INTRODUCTION

Complement system comprises of multimolecular self assembly of 30 plasma proteins implicated as principal mediators of inflammation, promoting opsonisation and may damage host tissue when there is persistent activation of complement by antigen-antibody complex and deposited in tissues. The receptor for cleavage fragment of third component of human complement C3b receptor (CR1) is expressed abundantly in erythrocytes, polymorph, monocyte, T and B cells, glomerular podocytes and soluble form in plasma. It plays major role in the disposal of immune complexes from circulation. It also serves as regulatory protein by preventing classical and alternative pathway of complement.[1,2]

It serves as co-factor for factor I mediated cleavage of C3 to C3bi and C3dg, C4 into C4c and C4d. The CRI function is important for clearance and neutralization of immune complex (IC) in liver and spleen, thus prevents deposition in small blood vessels and kidney tissue. The deficiency of complement proteins impair the RBC immune complex processing and clearing mechanism and predisposes individuals to immune complex/ auto immune diseases. A deficiency of C2, C4, Clq or C3 deficiency is associated with pyogenic infections, glomerulonephritis and systemic lupus erythematosus (SLE).[3] Deficiency of E-CR1 may impair the function of erythrocyte IC clearing and processing mechanism. Deficiency may predispose to the development of IC mediated diseases like SLE, rheumatoid arthritis, acute glomerulonephritis, Sjogren's syndrome. In IC mediated glomerular injury is caused by trapping of circulating Ag-Ab complexes in glomeruli.[4]

Studies pertaining to the localization of the receptor in case of SLE and IC mediated glomerulonephritis (ICGN) where tissue injury is very pronounced could help in providing conclusive evidence of the role of CR1 as a regulator of complement activation.[2] Hence the aim of present study was to:

1. Quantitate CR1 expression on RBC in SLE, ICGN and non ICGN.

2. Perform immunohistochemical localization of CR1 in kidney tissue in order to evaluate the relationship between CR1 expression in RBC and kidney tissue in diseased states.

3. Estimate the serum complement levels which includes C3, C4, C3d, CH100 and its correlation with CR1 and disease activity.

MATERIALS AND METHODS

Fifty-one subjects included in the study consisted of normal controls (n=14) and patients suffering from diseases such as SLE (n=7) IC mediated (n=10) and non-IC mediated glomerulonephritis (n=20). Controls were staff and students of AIIMS, New Delhi and relatives of patients. Patients were selected out from out patient department of Rheumatology clinic, AIIMS, which met the American Rheumatism Association criteria (1982) for selection of SLE. Biochemical investigations like serum creatinine, 24 hr urinary protein, microscopic examination of casts in urine and histopathological diagnosis were undertaken to divide patients into two groups : Immune complex mediated glomerulonephritis (ICGN) and non-IC mediated glomerulonephritis (non-ICGN).

Quantitation of CR1 receptors on erythrocytes were measured with patients using ELISA (Anthos Instrument, Austria), coating the wells with patients RBC and mouse monoclonal antibodies to human CR1 receptor.[5] Total hemolytic

complement activity (CH100), C3, C4 was estimated by single radial immunodifiusion techniques using standard calibration curve. C3d levels were quantitated by an ELISA using rabbit anti human C3d. The glomerular CR1 expression in normal kidney tissue or of renal diseases with immune complex deposits were examined based on immunofluorescent techniques.[6]
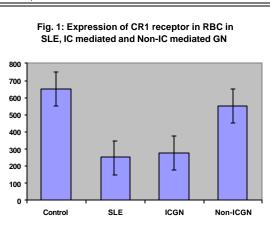
RESULT

Table I shows serum levels of C3, C4, CH100 and C3d in healthy control, IC mediated and non-IC mediated gloinerulonephritis. As evident from the table there is five fold increase of C3d in SLE and ICG'N as compared to the non-IC mediated GN and healthy control. Values of C3, C4, CH100 in SLE, ICGN are lower as compared to the healthy control. There was no significant differences between the levels of C3, C4, CH100 and C3d between non-ICGN and healthy control. CR1 expression in RBC (CR1/RBC) among SLE, ICGN, Non-ICGN has been presented in fig 1. Significant lowering of CR1 is evident in SLE (261 ± 141) and ICGN (270 ± 107) as against healthy control (627 ± 132). No significant change in CR1 expression had been found between non ICGN (550 ± 86) and control.

Table I : Serum complement level (C3, C4, CH100 and C3d) in SLE, IC mediated and non-IC mediated GN

*   *P < 0.01 in comparison to healthy control*
*[a]   *P < 0.05 in comparison to Non-IC GN*

Fig. 1: Expression of CR1 receptor in RBC in SLE, IC mediated and Non-IC mediated GN

DISCUSSION

Recent studies have highlighted significant role of complement proteins and complement receptors in pathogenesis of immune complex mediated diseases. Enumeration of CR1 expression on erythrocyte has long been recognized as of great diagnostic and prognostic values in the pathogenesis of several immune complex mediated diseases such as SLE, Rheumatoid arthritis and essential mixed cryoglobulinemia (EMC).[7] Result from the present study show that a statistically significant inverse relationship exist between the RBC CR1 receptor number and the disease severity. Whereas 50% decrease among SLE and ICGN was observed, no appreciable difference was seen in non-ICGN as compared to healthy control. The decrease of CR1 expression in SLE and ICGN could be because of certain acquired mechanisms during progression of the disease severity, as rapid degradation of CR1 receptors occurs due to proteolysis by macrophages proteases activated by the reaction of their complement receptor.[8]

The decrease expression of CR1 on erythrocyte correlated significantly with kidney involvement as evident from the immunofluorescence studies of the biopsy of lupus nephritis.[9] A recent study show that an early decrease in complement receptor

expression that was progressive was demonstrated before any major clinical manifestation of nephritis could be detected.[10] Our study supports these findings.

The study of complement profile viz C3, C4, CH100 and C3d was undertaken to observe the extent of complement activation during the disease states. Marginal decrease of C3, CH100 and significant decrease of C4 levels found in SLE and ICGN as compared to the non -ICGN and healthy control consistence with the earlier findings.[11] The C3d level were significantly higher in SLE and ICGN groups. This shows that C3d could be used as a reliable marker of complement activation in the above disease conditions.[12]

Hence, the chosen parameters (CR1/E, C3, C4, CH100, C3d and Immunohistochemical localization) could be of value as diagnostic and prognostic tool for SLE, IC mediated, versus non-IC mediated kidney diseases.

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