

# Assessment of Levels of Complement Factors C3 and C4 in Lupus Nephritis Flare in Resource Limited Setup

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## ABSTRACT

**Background:** Renal biopsy, an invasive and costly procedure, is required to diagnose lupus nephritis. In some studies, complement levels C3 and C4 are being utilized as a diagnostic tool to distinguish whether active urine sediment, rising proteinuria and loss of renal function are attributable to systemic lupus erythematosus (SLE) activity or other non-immunological causes.

**Aim:** To find out the frequency of altered levels of complement factors C3 and C4 in lupus nephritis with nephritic or proteinuric flare.

**Methods:** The present observational study was conducted in the Department of Nephrology, Services Hospital, Lahore. A total of fifty patients of lupus nephritis showing proteinuric, nephritic or mixed flare were enrolled. Demographics and clinical parameters were noted and C3 and C4 levels were measured. Both males and females were included in the study. This study was approved by the Institutional Review Board of the Services Institute of Medical Sciences.

**Results:** Out of a total of 50 patients, 19 were males while 31 were females. The mean age of the patients was  $29.40 \pm 6.66$  years. Low levels of complement factor C3 were discovered in 12(24%) of the patients, whereas low levels of complement factor C4 were detected in 18(36%) of the patients. Proteinuric flare was found in 17 patients, nephritic flare in 25 patients whereas both types of flare were found in a total of 8 patients.

**Conclusion:** One-third of the patients with nephritic type flare of lupus nephritis had low levels of C3 or C4 in our study. Further studies with larger sample size are needed to assess the potential of complement levels C3 and C4 as tools to diagnose lupus nephritis flare.

**Keywords:** Complement, C3, C4, Systemic lupus erythematosus, Lupus nephritis flare

## INTRODUCTION

Lupus nephritis is a deadly chronic autoimmune illness that affects a variety of organ systems, including the nervous, dermal, cardiovascular, and renal systems<sup>1</sup>. It's incredibly diversified, with a wide range of symptoms displayed by disease flares. It is an autoimmune illness because it affects several aspects of the immune system and results in the development of autoantibodies against a variety of targets, including double-stranded DNA (dsDNA), ribonucleic acid-binding proteins (RBPs), and phospholipids<sup>2</sup>. Anti-dsDNA antibodies are the most specific of them. They are present in the blood several years before the disease clinically manifests. Their increased levels usually occur in conjunction with lower complement C3 and C4 levels, indicating a disease flare. Hence these tests are used extensively by nephrologists in deciding whether to increase, decrease or maintain the immunosuppressive medications of the patients with lupus nephritis who are on follow-up treatment<sup>3,4</sup>.

In this study, we endeavored to find the frequency of altered C3 and C4 levels in patients of proteinuric, nephritic and mixed flare of lupus nephritis. In a resource-limited setup of a third world country like ours, it is very difficult to get all the tests indicating lupus nephritis flare done in every patient. Therefore, alternative biochemical variables may be used as a diagnostic tool so that the treatment can be started right after instead of going for other high cost and invasive diagnostic tests.

## METHODS

This descriptive study was carried out at outdoor and indoor facilities of the Services Hospital Lahore, Nephrology Department. A total of fifty patients, diagnosed as cases of lupus nephritis flare, either proteinuric, nephritic or both were included in the study after getting approval from the institutional review board of the Services Institute of Medical Sciences. This study recruited both male and female genders after receiving written informed consent. Blood samples were drawn to measure the altered levels of complement of serum creatinine levels from the baseline and active urine sediment i.e greater than 10 urinary red blood cells per high power field and/ or presence of one or more urinary red blood cells cast high factors C3 and C4. Nephritic

flare was defined as the rise of 30% power field (HPF)<sup>5</sup>. Proteinuric flare was defined by the increase in urinary protein to creatinine ratio (PCR) of at least in patients with non-nephritic syndrome or the doubling of nephrotic proteinuria (i.e baseline PCR > 3) with stable renal function i.e serum creatinine levels less than 1.2mg/dL<sup>5</sup>. Those excluded from the study were the lupus nephritis patients in remission as defined by normal renal function i.e., serum creatinine levels less than 1.2mg/dL, urinary protein to creatinine ratio of less than 0.5 and urinary red blood cells less than 10 per high power field. We excluded the patients having a history or signs and symptoms of urinary tract infection, hypotension, dehydration, urinary tract obstruction and acute coronary syndrome or taking nephrotoxic drugs<sup>6,7</sup>. SPSS version 25 was used to evaluate the data. For each variable, descriptive statistics were used. For all quantitative measures such as age, serum creatinine, urinary protein to urinary creatinine ratio, complement levels and the quantity of urinary red blood cells per high power field, mean and standard deviation was calculated. For qualitative characteristics like gender and flare type, frequencies and percentages were determined.

## RESULTS

A total of 50 patients were included in the study out of which 31 (62%) were female and 19(38%) were male. The mean age of patients was  $29.40 \pm 6.66$  years, it was observed that 20(40%) were 15-27 years old while 30(60%) were 28-50 years old. Mean serum creatinine was  $2.40 \pm 1.32$ mg/dL. Low levels of complement factor C3 were found in 12 (24%) patients whereas low levels of complement factor C4 were found in 18(36%) patients (Fig. 1).

Proteinuric flare was found in total 17(34%) patients, Nephritic flare was found in 25(50%) patients whereas both types of flares were found in 8(16%) patients (Figure-2).

Table-1 demonstrates that low C3 levels were found more in females (75%) than in males (25%). Similarly, low levels of C4 were found more in females (72.2%) as compared to males (27.8%). Also, lower C3 and C4 levels were found more in patients having nephritic type of flare as compared to those having proteinuric and mixed flare (75%, 50% and 25% respectively). We did not find any variation in C3 and C4 levels while comparing different age groups.

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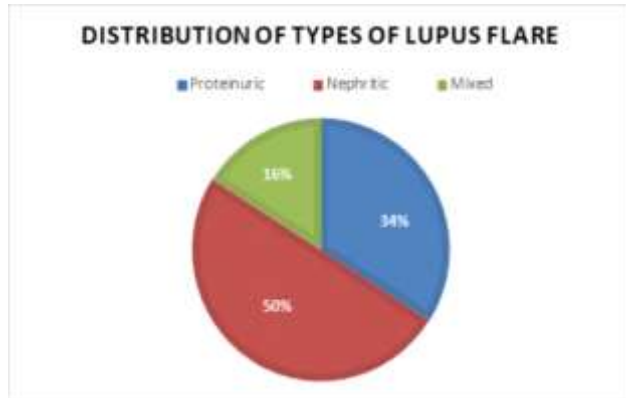


Figure 1: Percentage distribution of low levels of C3 and C4 in total population of the study.

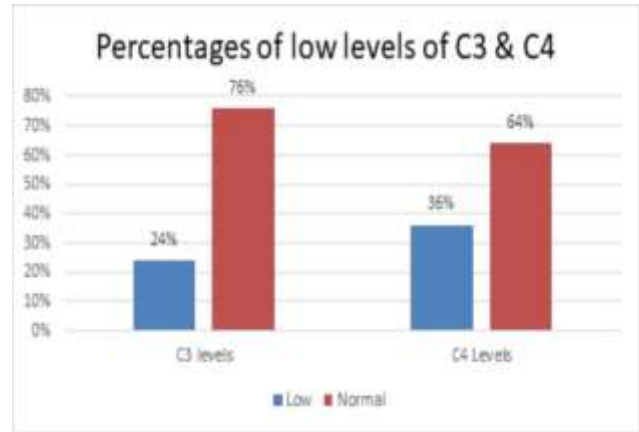


Figure 2: Distribution of different types of flare in total population of study

Table-1: Distribution of C3 and C4 levels in relation to Gender, Type of flare and Age

Variables	Construct	C3 levels				C4 levels			
		low levels		Normal levels		low levels		Normal levels	
		Count	N %	Count	N %	Count	N %	Count	N %
Gender	Male	3	25.0%	16	42.1%	5	27.8%	14	43.8%
	Female	9	75.0%	22	57.9%	13	72.2%	18	56.3%
Type of flare	Proteinuric	0	0.0%	17	44.7%	6	33.3%	11	34.4%
	Nephritic	9	75.0%	16	42.1%	9	50.0%	16	50.0%
	Both	3	25.0%	5	13.2%	3	16.7%	5	15.6%
Age	15-27 years	6	50.0%	14	36.8%	8	44.4%	12	37.5%
	28-50 years	6	50.0%	24	63.2%	10	55.6%	20	62.5%

**DISCUSSION**

In daily clinical practice, the activity of lupus nephritis is assessed by severity of clinical signs and symptoms, laboratory tests like urinary protein to urinary creatinine ratio, urinary active sediment (glomerular red blood cells and casts), low levels of complement factor C3 and C4, anti dsDNA levels, increment of serum creatinine from baseline and renal biopsy findings like glomerular neutrophils, presence and number of glomerular cellular crescents, glomerular necrosis and karyorrhexis, large subendothelial immune deposits and interstitial inflammation<sup>8,9,10</sup>. Out of these, complement factor C3 and C4 levels are comparatively less costly. So, in this study, we tried to find a frequency distribution of altered levels of lupus nephritis flare. We found low levels of C3 and C4 in 24% and 36% of the study population respectively. We need more cohort studies to find the correlation of C3 and C4 levels with lupus nephritic flare. If a strong correlation is found, treatment can be suggested to be started right after obtaining these levels instead of going for other costlier serological tests and renal biopsy as in a resource-limited setup of a third world country like ours it is very difficult to get all the tests indicating lupus nephritis flare done in every patient. In a study done by H. Julkunen et al. in 2012, it was shown that C1q, C3 and C4 levels correlated better with lupus nephritis activity than anti ds DNA levels<sup>3</sup>. In a study done by Moroni G et al in Italy in 2008, C1q, C3, C4, and Anti dsDNA were measured to detect lupus nephritis flare. None of the four immunological tests showed a high enough positive predictive value in this investigation. -C1q was 38%, anti-dsDNA was 31%, C3 was 28% and C4 was 31%<sup>5</sup>. These findings are also in line with what we found in our research. Another study was done by N. Hussain et al in our country. Out of 52 patients with systemic lupus erythematosus, 17 were diagnosed as having lupus nephritis. Normal levels of C3 with low levels of C4 were the most common result (35.39%). 23.5% of patients showed normal levels of C3 and C4 while 29.4% of patients showed low levels of both C3 and C4<sup>11</sup>. These results partially favour the results of our study. Another study by J M Esdaile et al in Canada found no utility of routine testing of C1q, C3, C4 and anti dsDNA levels for predicting lupus nephritis flare. The sensitivity of all four tests in their investigation was about 50%, while the specificity was less than 75%. Positive and negative probability ratios for anti-dsDNA, C1q binding, and C4 were all close to 1.0, suggesting that routine testing has minimal clinical relevance. The probability ratios for a positive test for C3 were close to

2.0, fulfilling the clinical significance threshold. These results are partially consistent with our study.

**CONCLUSION**

One-third of the patients with nephritic type flare of lupus nephritis had low levels of C3 or C4 in our study. Cross-sectional studies with a larger sample size are needed to assess the diagnostic potential of complement levels C3 and C4 in the diagnosis of lupus nephritis flare. However, in female patients with nephritic type of flare, we found low levels of C3 and C4 more frequently.

**Conflict of interest:** Nil

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