LUPUS NEPHRITIS: CORRELATION OF CLINICOBIOLOGICAL MANIFESTATION WITH HISTOLOGICAL TYPE

Andreea Munteanu, A. Caraba, Alina Pacurari, I. Romosan

DEPARTMENT OF INTERNAL MEDICINE, UNIVERSITY OF MEDICINE AND PHARMACY „VICTOR BABEŞ” TIMIȘOARA

Summary
Lupus nephritis (LN) is one of the most severe manifestations of SLE, being a major cause of morbidity and mortality. Renal involvement plays a decisive role in the development of the disease, renal failure being one of the main death causes in SLE. The incidence of LN varies, depending on the diagnostic methods used. When diagnosis is established on clinicobiological criteria, the incidence of LN ranges between 60 – 80%; a diagnosis based on the histological exam of the renal needle-biopsy tissue shows an incidence ranging between 95 – 100%. The aim of the study was to prove the existence of a correlation between the histological types of renal impairment and the clinicobiological parameters. The study was conducted on 30 patients (all women) with LN hospitalised in the Medical Department of the Clinical Railways Hospital, Timisoara, between January 2007 – December 2011. The lab tests determined: proteinuria, haematuria, serum creatinine, creatinine clearance, anti-dsDNA antibodies, circulating immune complexes, C3, C4, IgG. Kidney needle-biopsy was performed using the Tru Cut device and the tissue specimens were stained with hematoxylin eosin and PAS and analysed under an optical microscope. The mean age of the study group was 37 years. No correlation was found between the histological type of renal impairment and haematuria (p=0.07), proteinuria (p=0.067), creatinine (p=0.11), creatinine clearance (p=0.11). Anti-dsDNA antibodies were positive in 100% of the patients, their titers being correlated with the disease activity and with the histological type involved. There was a tight correlation between the values of the C3c complement (p<0.001), the C4 (p<0.001) and the histological type. The histological exam confirms the positive diagnosis of lupus renal disease, determines the type and activity stage, thus providing prognostic data and determining appropriate therapy.

Key words: systemic lupus erythematosus, lupus nephritis, renal histopathological exam.

Introduction
Systemic lupus erythematosus (SLE) is a chronic, inflammatory, multisystem disorder associated with the production of antinuclear, anticytoplasmic and antimembrane antibodies as a consequence of an impaired immune tolerance and the development of autoimmune phenomena. SLE represents the prototype of the autoimmune disease (Mason et al., 2008).

The clinical picture is extremely varied, reflecting the chronic inflammation of the various organs and systems. The most common targets of LES are the skin, joints and kidneys, but practically any organ or system may present LES-induced morphofunctional anomalies. Immune anomalies include the presence of autoantibodies (the most specific being antinuclear antibodies – ANA - and anti-double stranded DNA (dsDNA) antibodies), anti-gammaglobulinemia, hypocomplementemia. The histological exam of the affected organs shows the presence of autoimmune deposits and complement fractions (Kotzin., 1996).

Lupus nephritis (LN) is one of the most severe manifestations of SLE, being a major cause of morbidity and mortality. Renal involvement plays a decisive role in the development of the disease, end-stage renal failure being one of the main death
causes in SLE (Contreras et al., 2005; Appel et al., 2007).

Lupus nephritis may involve all renal structures: glomeruli (the most severe involvement), tubes, the interstitium, vessels. Although glomerular involvement has a significant diagnostic importance, some patients presented severe tubulo-interstitial or vascular damage despite only slight glomerular impairment (Caraba et al., 2009).

The clinicobiological picture of LN may vary from isolated urinary disorders to rapidly progressive renal failure. The histopathological exam is mandatory for a correct diagnosis, prognosis and therapeutic conduct in LN (Cagnoli, 2003).

The aim of the study is to point out existing correlations between the histopathological findings and the clinicobiological parameters.

Material and methods

The study was conducted on 30 patients (all women) with SLE-induced renal involvement. The patients were hospitalised in the Medical Department of the Clinical Railways Hospital, Timisoara, between January 2007 – December 2011.

All the patients met the ARA lupus criteria. The exclusion criteria were: pregnancy, SLE-associated bleeding diathesis, uncontrolled high blood pressure, the patient’s refusal.

The clinicobiological exam of the patients was performed upon diagnosis (inclusion in the study group) and assessed the following:

a) Index of renal involvement:
- proteinuria (Turbidimetry method, normal values NV<200mg/24h)
- haematuria (Addis-Hamburger method, NV H<1000 red blood cells/minute)
- serum creatinine (spectrophotometry –enzyme colouring, NV 0.6-1.0 mg/dl )
- creatinine clearance (ml/min/1.73/mp) = urine creatinine (mg/dl) x urine volume(ml)/ serum creatinine (mg/dl) x collecting time (minutes =1440 minutes/24h)

b) Immunological tests:
- immune circulating complexes (ICC) (Elisa Enzyme Immunoassay, NV <16microg/ml)
- anti- dsDNA antibodies (FEIA immunofluorescence, NV <10UI/ml)
- C3c and C4( immunoturbimetry C3 - NV 90-180mg/dl, C4 NV 10-40 mg/dl), IgG (immunoturbimetry NV 700-1600mg/dl)

c) Renal biopsies:
- renal needle biopsy was performed with a thick needle using a Tru Cut device. The biopsy specimens were stained with hematoxylin and eosin and PAS, and were analysed under an optic microscope. The specimens were classified according to the ISN/SPN classification of lupus nephritis (2003).

d) Statistical analysis:
- all the values were expressed as mean values ± standard deviation (SD). The values were compared using the ANOVA test, p < 0.05 being the statistically significant value.

Results

The study group consisted of 30 female patients, aged between 25 – 51 years, mean age 37.83 ± 7.03.

The clinicobiological parameters used in assessing the renal function are shown in Table nr.1.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Value (mean ± SD)</th>
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<tbody>
<tr>
<td>Age (years)</td>
<td>37.83±7.03</td>
</tr>
<tr>
<td>Serum creatinine (mg/dl)</td>
<td>1.50±1.37</td>
</tr>
<tr>
<td>Proteinuria (mg/24h)</td>
<td>2651±1244.70</td>
</tr>
<tr>
<td>Haematuria (red blood cells/minute)</td>
<td>2536±1822.59</td>
</tr>
<tr>
<td>Creatinine clearance (ml/min)</td>
<td>87.7±26.40</td>
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</tbody>
</table>
Immunoassay showed low values of the C3c complement in 60% of the patients and of the C4 in 75 per cent of the patients. Anti-dsDNA antibodies were positive in all the patients (100%). Immunoglobulin levels were high in 90% of the patients.

Following the renal needle biopsy, LN was diagnosed as follows: type II – 20% of the patients; type III – 13% of the patients; type IV – 44% of the patients; type V – 23% of the patients (Table nr. 2).

<table>
<thead>
<tr>
<th>Histological class</th>
<th>% affected patients</th>
</tr>
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<tbody>
<tr>
<td>II (mesangial lupus nephritis)</td>
<td>20%</td>
</tr>
<tr>
<td>III (focal lupus nephritis)</td>
<td>13%</td>
</tr>
<tr>
<td>IV (diffuse lupus nephritis)</td>
<td>44%</td>
</tr>
<tr>
<td>V (membranous lupus nephritis)</td>
<td>23%</td>
</tr>
</tbody>
</table>

The comparison between the degree of renal involvement and the histological type of lupus nephritis is shown in Figures 4 – 7.

### Table nr.2

Fig.1 Mesangial lupus nephritis (optical microscopy)

Fig.2 Focal lupus nephritis (optical microscopy)

Fig.3 Diffuse lupus nephritis (optical microscopy)

Fig.4 Mean values of proteinuria correlated with the histological type (p=0.067)

Fig.5 Mean values of haematuria correlated with the histological type (p=0.07)
Discussion

Systemic lupus erythematosus represents the prototype of the autoimmune disease associated with a production of antibodies targeting nuclear, cytoplasmic and membrane antigens (Wallace et al., 2002).

Immune anomalies include the presence of autoantibodies (the most specific ones being the antinuclear, anti-dsDNA antibodies). The histological exam of the impaired organs reveals the presence of immune complexes and complement fractions. The kidneys represent the main target for ICC (Edworthy, 2005).

Lupus nephritis (LN) is one of the most severe and most common manifestations of SLE, being a major cause of morbidity and mortality (Romosan et al, 2002). Renal impairment plays a crucial role in the development of the disease, renal failure being one of the main death causes in SLE (Moroni et al., 2004).

The incidence of LN varies according to the diagnostic methods used. If diagnosis is established solely on clinicobiological criteria (proteinuria, haematuria, cylindruria, sodium retention, high blood pressure, oedema syndrome) the incidence of LN varies between 60 – 80%. On the other hand, if diagnosis is based on the histological exam of the kidney biopsy tissue obtained by kidney needle-biopsy, the incidence of LN will vary between 95 – 100% (Zabaleta, 2003; Cameron, 1999).

From a clinicobiological point of view, LN may manifest itself in various ways. Proteinuria is present in almost every patient, with variable intensity, reaching the nephrotic stage. Electrophoresis reveals the presence of non-selective or mixed glomerular proteinuria, seldom only tubular, depending on the histological structures predominantly involved. Another urinary sign is microscopic haematuria which may, rarely, be macroscopic. High blood pressure is associated with severe LN, and oedemas indicate the presence of nephrotic syndrome. Nicturia is characteristic for tubulointerstitial involvement, while azotemia indicates renal failure. In some cases, LN may start as an acute renal failure, commonly associated with other severe manifestations (myocarditis, disorders of the central nervous system) (Edworthy, 2005).

Following biopsy, once the diagnosis was established it showed that 44% of the patients belonged to histological type IV of LN.

The lowest values of proteinuria were found in patients belonging to histological type II LN, while the highest value were found in patients belonging to type V (p = 0.067), although they were not statistically significant. (Fig 1).

Haematuria was found in all histological types (p = 0.07). (Fig. 2).

Histological type IV (diffuse LN) had the highest values of serum creatinine and the lowest values of creatinine clearance.
clearance, while these values were regulated in histological type V (membranous LN). P was statistically insignificant both for creatinine (p = 0.11) and for creatinine clearance. (p = 0.11).

Anti-dsDNA antibodies, highly specific markers for SLE, were positive in all the patients (100%). Antibody titer is closely connected to the disease activity stage, playing an important role in monitoring treatment (Alba et al., 2003). Although anti-native DNA antibodies are correlated with the histological type and with the activity stage, this correlation is not strong enough to impose an intensified therapeutic conduct when higher levels of these antibodies are found.

There was a tight correlation between C3c complement values (p<0.001) and C4 (p<0.001) and the histological type involved.

No strict correlation was found between the clinicobiological manifestations and the histological type of LN. Several studies have shown that a diagnosis established solely on the basis of clinicobiological data can be inconclusive (Mittal et al., 2005; McLaughlin et al., 1994; Caraba, 2006)

Conclusions
There is no strict correlation between the clinicobiological manifestations and the histological type of LN. Even the patients without clinically manifest renal disease often have immune mesangial deposits, therefore the clinicobiological picture cannot be considered predictive for the severity of the histological damage.

The histopathological exam is mandatory for the assessment of any patient with LN as it confirms the positive diagnosis of lupus renal disease, determines the type and activity stage, thus providing prognostic data and determining appropriate therapy.

References
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