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ORIGINAL ARTICLE

A comparison of 1995 WHO classification with 2003 ISN/RPS classification of lupus nephritis: a single centre observation

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Abstract

Background: In the past, lupus nephritis was histologically classified according to the 1995 WHO Classification. With the introduction of the 2003 ISN/RPS Classification, many nephropathology services converted to this new classification. This study was undertaken to compare both classification systems in a single centre practice. *Methods:* 103 consecutive adequate renal biopsies initially reported as lupus nephritis in the Department of Pathology, Faculty of Medicine, University of Malaya were reassessed using the criteria of both the 1995 WHO Classification and the 2003 ISN/RPS Classification. *Results:* The relative prevalence for each class using the WHO Classification were: Class I (1%), Class II (8.7%), Class III (6.8%), Class IV (60.2%), Class V (20.4%), Class VI (2.9%) while the prevalence using the 2003 ISN/RPS Classification were: Class I (1%), Class II (8.7%), Class III (6.8%), Class IV (61.2%), Class V (21.3%), Class VI (1%). Both classifications were essentially comparable with regards to Classes I, II and III. The differences in Classes IV, V and VI were significant in potential to alter patient management. The identification of segmental lesions (Class IV-S) over and above a global nephritis (Class IV-G) deserves more focused clinicopathological studies to gauge whether these groups have different clinical manifestations and outcomes. With regards Class V, the ISN/RPS system, by requiring that all mixed classes be stipulated in the diagnostic line, minimizes the chances of patients missing out on additional treatment. The ISN/RPS system has stricter criteria for Class VI, which again minimizes patients missing out on therapy. On the whole, the ISN/RPS system is more user-friendly as criteria are more clearly defined which translates to more benefits to patient care.

Keywords: systemic lupus erythematosis, renal pathology, glomerulonephritis, renal classification

INTRODUCTION

Lupus nephritis (LN) is an immune-mediated nephritis occurring in 40%-80% of patients with systemic lupus erythematosus (SLE). The diagnosis, treatment, monitoring and prognosis of LN is based on the combination of clinical manifestations, biochemical alterations and renal biopsy findings of the affected individuals. As the morphology of lupus nephritis is diverse, the World Health Organisation (WHO) introduced a classification system in 1974, revising it in 1982 and again in 1995 with the aim of standardising the histological diagnosis across different reporting centres.[1] However, ambiguities in classification categories and diagnostic terminologies led to problems in inter-observer agreement. The new classification system proposed by the International Society of Nephrology/Renal Pathology Society (ISN/RPS) in 2003 aimed at reducing the perceived ambiguities as well as providing a better pathophysiological basis for treatment.[2]

Since its introduction, the ISN/RPS classification system has gained popularity in both research and medical fields due to its clearer classification categories.[3,4] However, it has yet to be adopted by WHO as the international / sole classification system for LN and some centres around the world are still using the 1995 WHO Classification. The Department of Pathology, University of Malaya which provides the histopathology diagnostic service for its teaching hospital, the University of Malaya Medical Centre, has been making a transition from the 1995 WHO classification to the ISN/RPS system. This present study was carried out to examine

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whether the WHO 1995 revised classification and ISN/RPS 2003 classification systems are comparable, and where the change may impact on patient management.

MATERIALS AND METHODS

All renal biopsies reported as lupus nephritis by the Department of Pathology, Faculty of Medicine, University of Malaya between 1st January 2007 and 30th June 2009 were included in the study. The histological slides of the aforementioned cases were retrieved from department archives and re-classified according to both the 1995 WHO Classification System and the ISN/RPS 2004 Classification System. The criteria of these classification systems are summarized in Tables 1 and 2 respectively.

Multiple histological levels of the biopsies, stained using Haematoxylin & Eosin, Periodic Acid Schiff, Periodic Acid Methanamine Silver and Masson Trichrome were assessed under light microscopy. Slides which were faded and rendered unreadable were re-stained accordingly. Data for immunofluorescence were also reviewed based on the archived records. No data for electron microscopy were available for review, as electron microscopy was not routinely performed for lupus nephritis biopsies in this centre.

Cases that showed, upon review, a discrepancy in classification from the original reports, or discordance in the two classifications were then reviewed a second time with two investigators and a consensus reached.

Kidney biopsies were originally done on patients with informed consent for the purpose of providing a diagnosis. Further consent was not obtained specifically for this study as they were analysed in a retrospective manner.

TABLE 1: WHO Histological Classification of Lupus Nephritis (1995)[1]

Class I	Normal glomeruli a. Nil (by all techniques) b. Normal by light microscopy, but deposits by electron or immunofluorescence microscopy
Class II	Pure mesangial alterations a. Mesangial widening and/or mild hypercellularity (+) b. Moderate hypercellularity (++)
Class III	Focal segmental glomerulonephritis (associated with mild or moderate mesangial alterations) a. With “active” necrotizing lesions b. With “active” and sclerosing lesions c. With sclerosing lesions
Class IV	Diffuse glomerulonephritis (severe mesangial, endocapillary or mesangiocapillary proliferation and/or extensive subendothelial deposits) a. Without segmental lesions b. With “active” necrotizing lesions c. With “active” and sclerosing lesions d. With sclerosing lesions
Class V	Diffuse membranous glomerulonephritis a. Pure membranous glomerulonephritis b. Associated with lesions of class II c. Associated with lesions of class III d. Associated with lesions of class IV
Class VI	Advanced sclerosing glomerulonephritis

TABLE 2 : ISN/RPS 2003 Classification of Lupus Nephritis[a,2]

Class I	**Minimal mesangial lupus nephritis** Normal glomeruli by light microscopy but mesangial immune deposits by immunofluorescence
Class II	**Mesangial proliferative lupus nephritis** Purely mesangialhypercellularity of any degree or mesangial matrix expansion by light microscopy, with mesangial immune deposits. No subepithelial or subendothelial deposits by light microscopy
Class III	**Focal lupus nephritis** Active or inactive focal, segmental or global endo- or extracapillary glomerulonephritis involving <50% of all glomeruli, typically with focal subendothelial immune deposits irrespective of mesangial alterations Class III (A): Active lesions Class III (A/C): Active and chronic lesions Class III (C): Chronic lesions
Class IV	**Diffuse lupus nephritis** Active or inactive diffuse, segmental or global endo- or extracapillary glomerulonephritis involving >50% of all glomeruli, typically with focal subendothelial immune deposits. Divided into diffuse segmental (IV-S) lupus nephritis when >50% of all glomeruli show segmental lesions and diffuse global (IV-G) lupus nephritis when >50% of all glomeruli show global lesions. Segmental is defined as a glomerular lesion involving less than half of the glomerular tuft and global when it involves more than half of the glomerular tuft. IV-S (A): Diffuse segmental lupus nephritis with active lesions[b] IV-S (A/C): Diffuse segmental lupus nephritis with active and chronic lesions[b,c] IV-S (C): Diffuse segmental lupus nephritis with chronic lesions[c] IV-G(A): Diffuse global lupus nephritis with active lesions[b] IV-G(A/C): Diffuse global lupus nephritis with active and chronic lesions[b,c] IV-G(C): Diffuse global lupus nephritis with chronic lesions[c]
Class V	**Membranous lupus nephritis** Global or segmental subepithelial immune deposits or their morphologic sequelae by light microscopy and by immunofluorescence or electron microscopy irrespective of mesangial alterations. May occur in combination with class III or IV, in which case, both are diagnosed
Class VI	**Advanced sclerosing lupus nephritis** >90% of all glomeruli globally sclerosed without residual activity

[a]Minimum of 10 glomeruli required
[b]Active lesions: Endocapillaryhypercellularity, karyorrhexis, fibrinoid necrosis, rupture of glomerular basement membrane, cellular or fibrocellular crescents, wire loops, hyaline thrombi.
[c]Chronic lesions: Glomerular sclerosis, fibrous adhesions, fibrous crescents.

RESULTS

Demographic profile

Of the 109 cases retrieved, six were excluded from the study because the biopsies contained less than ten glomeruli, the minimum number of glomeruli needed for the ISN/RPS Classification. Of the remaining 103 renal biopsies, 89 biopsies were from female patients and the remaining 14 biopsies were from male patients. The mean age at which the renal biopsies were preformed was 29 years (range 10 years to 59 years). The biopsies were obtained mainly from Chinese (56 cases/54.37%), Malay (33 cases/32.04%)

TABLE 3: Categorisation of study biopsies using the WHO 1995 and ISN/RPS 2003 Classification Systems[a]

WHO	Number (%)	ISN/RPS	Number (%)
I	1 (0.97)	I	1 (0.97)
IIa	4 (3.88)	II	9 (8.7%)
IIb	5 (4.85)		
IIIa	3 (2.91)	III (A)	3 (2.91)
IIIb	4 (3.88)	III (A/C)	4 (3.88)
IIIc	0	III (C)	0
IVa	2 (1.94)	IV-S (A)	2 (1.94)
IVb	34 (33.00)	IV-S (A/C)	0
IVc	22 (21.36)	IV-S (C)	1 (0.97)
IVd	4 (3.88)	IV-G (A)	34 (33.01)
		IV-G (A/C)	25 (24.27)
		IV G (C)	1 (0.97)
Va	5 (4.85)	V	5 (4.85)
Vb	0		
Vc	3 (2.91)	V + III	3 (2.91)
Vd	13 (12.62)	V + IV	14 (13.59)
VI	3 (2.91)	VI	1 (0.97)
All classes	103 (100)	All classes	103 (100)

[a]Categorisation is not a direct comparison especially in Class IV

and Indian (11 cases/10.68%) patients. The remaining 3 cases were from other ethnic groups.

Histological categories

A distribution of the biopsies classified by both systems is summarized in Table 3.

All the biopsies that were reported as Class I, II (Fig. 1A) or III (Fig. 1B) according to the 1995 WHO Classification remained in their respective classes when reclassified using the 2003 ISN/RPS Classification. Cases which were reported as either Class IVa or Class IVb under the WHO System were reclassified into either Class IV-G (A) (Fig. 1C) or Class IV-S (A) (Fig. 1D) in the ISN/RPS classification system. Similarly cases which were reported as Class IVd under the WHO System converted to either Class IV-G(C) or Class IV-S(C) under the ISN/RPS System. It was however interesting to note that all the WHO Class IVc when reclassified under the ISN/RPS Classification became Class IV-G (A/C). Only the WHO Class Va biopsies remained as pure Class V under the ISN/RPS Classification System (Figs. 1E &1F). All other Class V biopsies in the WHO Classification required an additional class notification under the ISN/RPS system.

Of the three cases which were Class VI under the WHO Classification, only one remained as a Class VI Lupus Nephritis under the ISN/RPS Classification. The remaining two were reclassified to a pure Class IV G (A/C) and a mixed Class V + Class IV-G (A/C).

Of the 103 biopsy samples, only 51 cases were recorded as having fresh tissue sent for immunofluorescence. Of these, immunofluorescent deposits in either the mesangium or capillaries were found in 42 cases for C3 and IgG, 34 cases for IgA and 32 cases for IgM.

DISCUSSION

The preponderance of the female gender and ethnic Chinese in the study population is not surprising as systemic lupus erythematosus has been well documented to be more common in Chinese females in this country.[5,6] The overall Class distribution of lupus nephritis in this study mirrors that of others in the same region[7] and of previous studies from this centre[8,9] with the exception of a lower prevalence of Class II and higher prevalence of lupus Class V. Being a referral centre for both treatment and assessment of lupus nephritis, this changing pattern may be due to changes in biopsy practice and not

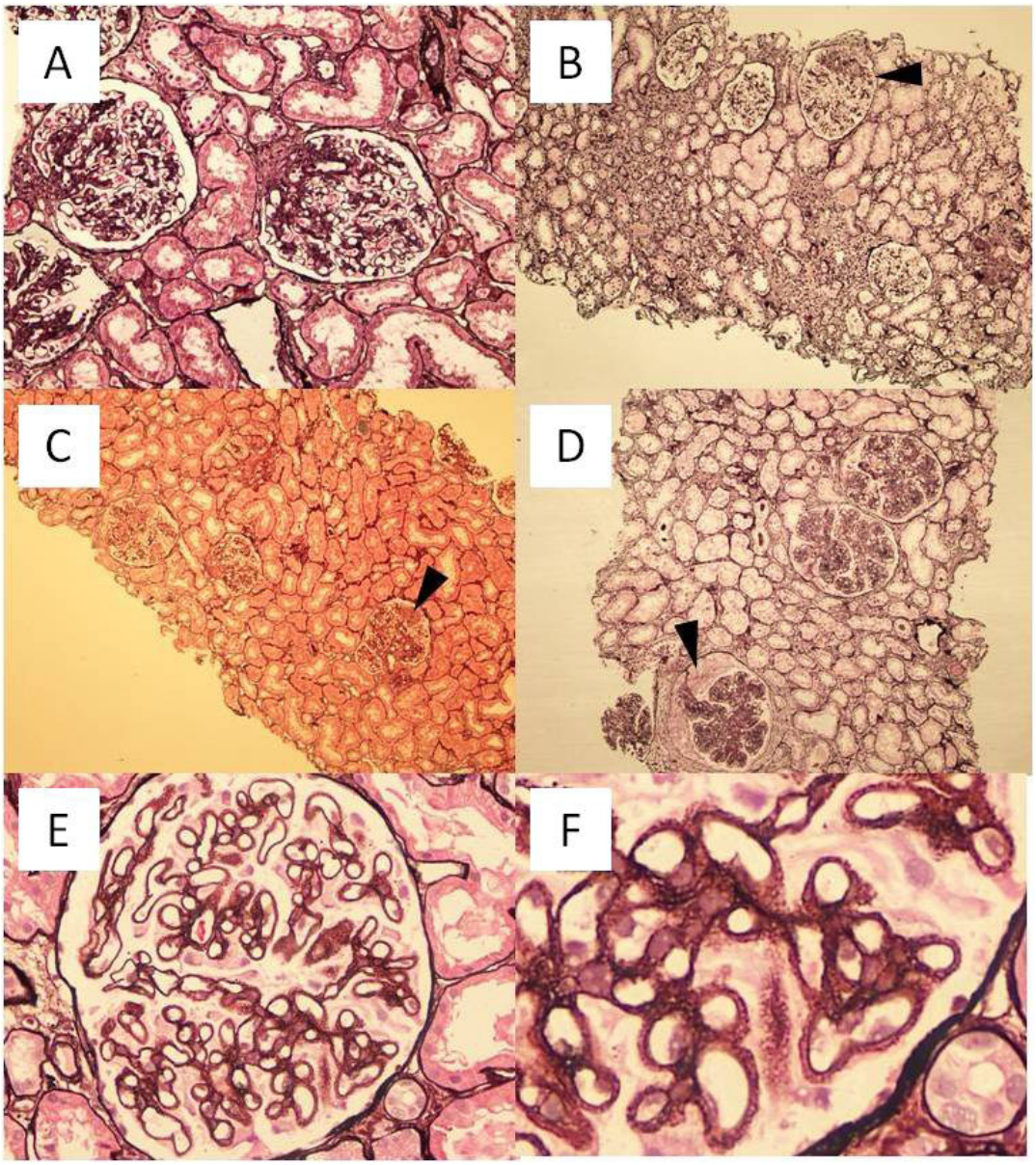

FIG. 1: Photomicrographs of lupus nephritis. (A) Lupus nephritis ISN / RPS Class II.Glomeruli showing only mesangial proliferation. The capillaries remaining widely patent and unremarkable. No spikes or tram-tracks present (PAAg x200). (B) Lupus nephritis ISN / RPS Class III (A). A single glomeruli with active segmental changes (arrow) (PAAg x100). (C) Lupus nephritis ISN / RPS Class IV-S (A). Diffuse proliferative changes with segmental glomerular lesion (arrow) (PAAg x100). (D) Lupus nephritis ISN / RPS Class IV-G (A). There is diffuse glomerulonephritis with global activity. A cellular crescent is also noted in one of the glomeruli (arrow) (PAAg x100). (E & F) Lupus nephritis ISN / RPS Class V: Glomerulus showing numerous spikes and chain-links (PAAg x400 and x600).

a true evolution in disease patterns. However, it is also important to note that lupus nephritis has a heterogenous morphology and is subject to sampling bias. It is therefore important to use more than one modality in the assessment of renal biopsies for lupus nephritis. In this study, we are fortunate to have multiple levels of tissue for assessment using different stains although immunofluorescence was not available for all cases and electron microscopical examination was not routinely performed.

Class I lupus nephritis

This study was not able to demonstrate any issues with regards to classification by either the WHO or ISN/RPS system, due to the small number (only one) that fell into this category. Conceivably, with the adoption of the ISN/RPS system, there will be an even lower prevalence of Class I LN as biopsies that are totally normal in both appearance and with negative immunofluorescence will no longer be accepted as LN.

Class II lupus nephritis

Superficially there was no discordance in the classification of the 9 cases in this category. However, 4 were categorized as IIa and 5 as IIb in the WHO system. The ISN/RPS system simply categorized all 9 as Class II. It is thus simpler to use as it does away with assessing for the severity of mesangial cellularity, as long as there is no endothelial proliferation, capillary deposits or capillary occlusion. It remains to be seen whether recording the severity of mesangial cellularity in Class II lupus nephritis in the WHO system has any clinical relevance or advantage over the ISN/RPS system. A further refinement of Class II may be useful, in view of the recent report by that Class II cases who progressed to Class III or IV had a significantly heavier immune-complex deposition in the glomeruli.[10]

Class III lupus nephritis

We observed a good concordance in Class III LN by both WHO and ISN/RPS systems. The 3 cases classified as WHO IIIa were classified ISN/RPS III (A) while the 4 classified as WHO IIIb were classified ISN/RPS III (A/C). The 1995 WHO classification stipulated Class III for focal segmental glomerulonephritis with necrotizing lesions creating an ambiguity between Class III and Class IV. In our study, "focal" was interpreted as <50% of glomeruli showing morphological changes (based on the 1974 classification). Hence, in this comparison there was less room for differences between the two classifications. Notably, both systems identified 3 additional cases of Class III LN which occurred in association with Class V LN.

Questions have been raised as to whether there is clinical relevance in recognizing global and segmental subtypes in Class III. A recent study has not found a significant difference in serum creatinine, proteinuria, activity, chronicity and proportion of endocapillary proliferation between these two subtypes.[11]

Class IV lupus nephritis

Class IV LN constituted the largest category (60%) of LN in this study. Superficially, it appeared that there was concordance between the WHO and ISN/RPS systems for Class IV LN in that all 62 cases classified as WHO IV were also classified as ISN/RPS Class IV. Notably, because the ISN/RPS system allows for mixed classes to be specifically stipulated in the diagnostic line, there were 14 additional cases with Class IV changes that had coexisting Class V changes.

More importantly, the ISN/RPS system requires a separation of biopsies showing segmental acute and chronic lesions against a background of diffuse nephritis. The WHO system has no provision for that. Only one of the four biopsies originally classified as WHO IVd was categorized as ISN/RPS Class IV-S (C). This may be an important distinction as cases with segmental lesions over and above a diffuse nephritis may connote a different pathophysiology, such as more vasculitic features, and a different clinical outcome.[11] Questions have been raised as whether the ISN/RPS Class IV-S (A; A/C and C) cases are more akin to Class III than Class IV. Vandepapelière *et al*[12] found that the activity indices of IV-G and IV-S were more similar to Class III while Gao *et al*[13] found that IV-S had a higher prevalence of fibrinoid necrosis than IV-G. Several other studies support a significant difference in renal function between the segmental and global subclasses in Class IV, with IV-S being more favourable.[11,14] A Korean study indicated that pretreatment proteinuria was higher in IV-G than IV-S, and also that IV-G patients responded less well to induction therapy with IV cyclophosphamide pulse than IV-S.[15]

Class V lupus nephritis

Although good concordance was noted between the two systems for Class V (membranous) nephritis, there are some differences of clinical relevance. While the WHO classification did allow for Class V to be present with other classes of lupus nephritis, it did not emphasize the "dual pathology" that was present in the biopsy, as such "mixed" classes are categorised as "subclasses" of Class V. In the ISN/RPS classification, both classes of lupus nephritis are reported in the diagnostic line highlighting the dual nature of the disease. The separation of "mixed" classes in the diagnostic line may be of clinical importance as treatment for "mixed" LN may differ from pure

membranous LN. Hence the ISN/RPS system minimizes the chances of patients missing out on additional treatment strategies.[16,17]

Class VI lupus nephritis

From the results, the main discordance between the two classification systems arose in Class VI. This is due to differences in defining Class VI between the two systems. With the WHO Classification, it was possible to diagnose advanced sclerosing lupus nephritis in biopsies with just over half the glomeruli showing global sclerosis. However, using the ISN/RPS Classification, Class VI now requires at least 90% of the glomeruli having sclerosis in the absence of any activity. The addition of the exclusion criteria is central in identifying patients who have a less advanced and more treatable form of lupus nephritis. Hence, it is the recommendation of this study that persons with WHO Class VI lupus nephritis be reassessed using the 2003 ISN/RPS classification.

CONFOUNDING ISSUES

The ISN/RPS classification is not without its difficulties. While it addresses the issue of specimen adequacy by stipulating the minimum number of glomeruli, it however remains silent as to whether subcapsular glomeruli, which have a higher proportion of sclerotic glomeruli particularly in the elderly, are to be used in the assessment. Similarly, the classification is also silent whether glomerular changes such as global sclerosis secondary to hypertension, diabetes mellitus and other pathologies should be counted. While including it into the count may be reflective of the overall renal function, it is not reflective of the disease attributed to lupus nephritis. In the absence of florid secondary changes (e.g. Kimmelstiel-Wilson nodules, onion skin vasculopathy, etc) or a positive clinical history, separating other aetiologies from lupus-affected glomeruli may be difficult.

Sampling bias in a renal biopsy may result in the absence of subendothelial deposits in the biopsy viewed under light microscopy with presence of the same in the immunofluoresence sample. In such cases, the immunofluorescence sample will be depended upon to make the diagnosis of Class III or IV lupus nephritis.

The ISN/RPS classification is more focused on glomerular changes. Although vascular and interstitial changes are graded as mild, moderate and severe, there is less clarity regarding the cut-off criteria for their assessment, leading to a broad degree of subjectivity.

A recent evaluation of the ISN/RPS classification indicated that interobserver variability for Class III and IV remains substantial, and proposed several areas for improvement along the lines of refinement of criteria for assessments along the lines of the Oxford IgAN Classification.[18]

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