Original Research Article

Clinicopathological profile of Membranous nephropathy-A single centre experience

Abstract:

The present study discusses about Paradigm shift in Membranous nephropathy. Recently, using laser microdissection of glomeruli and mass spectrometry analysis of the proteins in the biopsies of patients with MN, Sethi et al. discovered another antigen, exostosin, associated with autoimmune etiologies of the disease. Human embryonic kidneys showed the presence of NELL-1 in the extracellular component and possible deposition in the glomerular basement membrane.In patients tested, the glomerular basement membrane and subepithelium stained positively for NELL-1.In biopsies of patients with a diversegroup of glomerular diseases and in those with primary MN, NELL-1 was not present who were PLA2R or THSD7A positive. It can be safely concluded that pts. With NELL1 deposit do not seem to be at a higher risk for malignancies.

Keywords: NELL1, Membranous nephropathy,malignancies,IgG levels

Introduction-

Membranous nephropathy (MN) is a prevalent cause of nephrotic syndrome in adults and can either occur as a primary condition or be associated with various underlying etiologies characterized by unique antigens and antibody complexes localized in the subepithelial region of glomeruli (1). Beck et al.'s identification of the phospholipase A2 receptor (PLA2R) antigen within these deposits, along with immunoglobulin, has spurred significant clinical and research advancements in primary MN. Currently, PLA2R is implicated in 70% to 80% of primary MN cases, while THSD7A (thrombospondin type 1 domain containing 7A) is associated with 1% to 5% (1). THSD7A is a large transmembrane glycoprotein expressed by podocytes, triggering a predominantly IgG4 immune response akin to PLA2R. However, the exact correlation between THSD7A antigen presence and disease activity remains unproven, although it is notably more prevalent among females and associated with certain malignancies.

Recent advances using laser microdissection of glomeruli and mass spectrometry have unveiled additional antigens linked to autoimmune mechanisms in MN, such as exostosin (2). Sethi et al. identified NELL-1 (neural epidermal growth factor-like 1 protein) as a novel antigen in cases negative for PLA2R and THSD7A, categorizing it as a distinct cause of primary MN (3). NELL-1, encoding a 90-kDa protein of 810 amino acids, is highly expressed in osteoblasts and has been

detected in the extracellular matrix of human embryonic kidneys, suggesting potential deposition in the glomerular basement membrane. Immunohistochemical staining has confirmed NELL-1 presence in the glomerular basement membrane and subepithelium of affected patients.

Unlike PLA2R-positive MN, where malignancies are rare and considered coincidental, the association between NELL-1 and malignancies remains unclear (4). Initial studies did not detect tumors in patients with NELL-1-positive MN; however, whether NELL-1 positivity in some cases correlates with tumor presence, similar to THSD7A, is yet undetermined. Thus, the classification of NELL-1 as a primary MN etiology or its association with malignancies requires further investigation

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Objectives:-determination of clinicopathological profile of membranous nephropathy

Methodology: A study was conducted in the department of nephrology, at Sawai Man Singh medical college Jaipur wherein a data of 440 patients with renal pathology was collected. A retrospective observational study was conducted with data from January 2023 to December 2023 in Patients presenting with nephrotic range proteinuria and whose renal biopsy is suggestive of membranous nephropathy. However, the study excluded patients with known lupus nephritis, diagnosed case of malignancy, or any feature suggestive of disease causing secondary membranous nephropathy.

A thorough history was taken of the enrolled patients along with their informed consent. The patients creatinine was measured at presentation and regularly at subsequent visits along with dipstick(tetra bromophenol blue) from proteinuria. Renal biopsy was performed using 20G renal biopsy gun with the help of ultrasound by a nephrologist and samples were subsequently sent for light microscopy and immunofluorescence staining for the type of deposits was done. All patients underwent routine investigations and were tested for viral markers including HIV, HEPATITIS B, HEPATITIS C. Quantitative serum pla2r antibody levels were also done in patients without deposition of PLA2R and THSD7A. All the patients underwent thorough workup for malignancy including chest xray, ultrasonography of abdomen, complete blood count, stool routine and microscopy, pap smear as per indication. For remission criteria used were decrease in urinary protein and decrease in serum creatinine at 6 months(if previously elevated). No cases of relapse and post renal transplant was enrolled in the study. The patients were categorized as per their risk profile as suggested by kdigo(1). All patients underwent serum pla2r antibody test at the initiation of treatment and if elevated after 6 months a repeat test was conducted. The patients were given treatment as per kdigo protocols(1). Urinary protein with dipstick was checked quarterly i.e. at 0,3,6,9,12 months. Excessive data recording was kept using excel data sheets and no breach in data privacy was ensured.

Result: Out of 440 patient biopsied, 52 came out to be having MN with female preponderance of 29 patients. All the patients fell into moderate and high risk profile as per kdigo recommendation.

Table1:Agedistributionofstudiedpatients

<20yrs	20-29yrs	30-39yrs	40-49yrs	50-59yrs	>60yrs	Total
0	3	8	17	18	6	52

Table2:patient category as per risk profile

Moderate risk	High risk	Total
20	32	52

Table3:Agedistributionofpatients with NELL1 deposit

<20yrs	20-29yrs	30-39yrs	30-39yrs	40-49yrs	50-59yrs	>60yrs	Total
0	0	1	0	1	1	4	7

Table4:GFRdistributionofstudiedpatients

GFR(ml/min/m^2)	Number
>60	32
45-59	12
30-44	8
15-29	0
<15	0

Of these 52 patients 7(13.5%) were seen associated with NELL1 deposition in glomeruli with negative PLA2R levels in serum. The THSD7A was found positive in 3 patients while no deposition was found in 2 patients. Conservative treatment alone using ACEi/ARBs was used in 12 patients while 40 patients received immunosuppressive and conservative therapy. Immunosuppression included the use of injection rituximab 500mg weekly for 4 weeks and oral tab methylprednisolone at 1mg/kg (max dose 60mg/kg) to be tapered over the next 3 months.

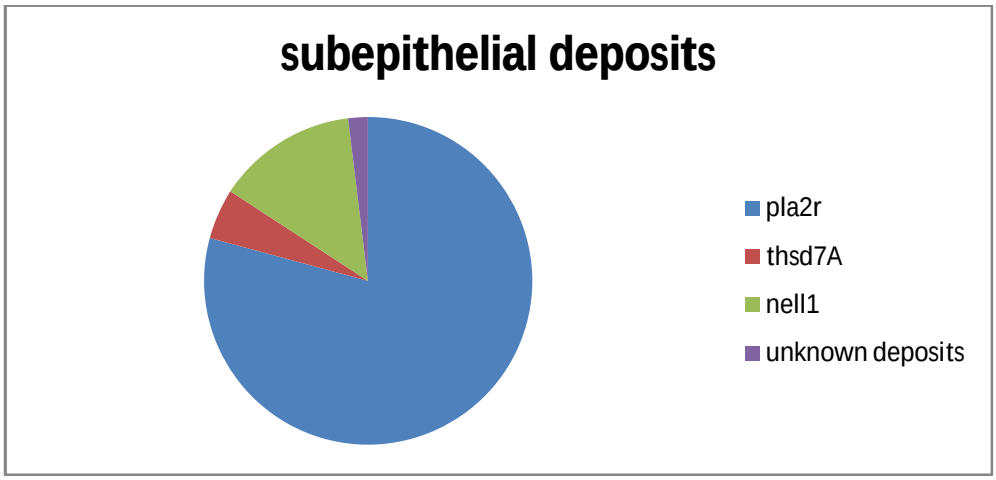

Figure 1: distribution as per type of deposit.

The patients were followed up for the subsequent 6months with achievement of sub-nephrotic range proteinuria in 45 out of 52 patients. GFR also improved from mean 51ml/min/m^2 to 58ml/min/m^2 at 6months of treatment. The patients were closely monitored but no signs suggestive of malignancy was found in 1 year after initiation of therapy.

Amongst the 7 patients with NELL1 deposits 5 were female while the remaining were male. 2 patients with Nell 1 positivity had deranged renal functions with creatinine 2.4 and 2.8 subsequently. All the 7 patients were treated with injection rituximab 500mg/week for 4 weeks along with oral prednisolone at 1mg/kg with subsequent tapering over the next 3 months but remission was achieved in only 4 patients while there was no improvement in serum creatinine levels. All the treatment resistant patients were started on immunosuppressive therapy including injection cyclophosphamide and oral prednisolone. Incidentally, all the 7 patients with NELL1 glomerular deposition also had IgG1 deposits as compared to patients with pla2r deposits were predominantly IgG4 deposits were found. Using various screening methods no signs of malignancy was found in NELL1 patients however, 3 patients with pla2R positivity were found to have malignancy. Two patients had carcinoma of colon while one had lung tumor that turned to be small cell carcinoma.

Discussion:

Membranous nephropathy (MN) is a distinctive glomerular condition and the predominant cause of idiopathic nephrotic syndrome in nondiabetic adults. Approximately 80% of cases are classified as primary MN (PMN), with the remaining 20% associated with secondary MN, linked to systemic diseases or exposures (5). Primary MN is primarily mediated by antibodies targeting the M-type phospholipase A2 receptor (anti-PLA2R) (85%), thrombospondin type 1 domain containing 7A (THSD7A) (3%–5%), or other as yet unidentified mechanisms (10%) (6,7).

Our study observed a prevalence of 76% PLA2R-positive patients and 5% THSD7A-positive patients, while 3% were attributed to unidentified mechanisms. This lower incidence of

unidentified mechanisms might be attributed, in part, to the absence of testing for NELL1 deposition in previous studies. As noted by Sanjeev Sethi (8), recent research has identified novel types of MN associated with EXT1, EXT2, NELL1, Sema3B, and PCDH7, each representing distinct disease entities with unique clinical and pathologic characteristics. Future clinical testing for these novel agents is expected to further reduce the percentage of cases classified under unidentified mechanisms.

In parallel to the study by Nicole Andeenet. al.(2) the patients of NELL1 positivity do present with nephrotic range proteinuria with only few presenting with decreased gfr (29%). As analogus to the study by A.G. Kattah et. al(3)where the primary deposit was found to be IgG4 in primary MN associated with pla2r antibodies, our study also had the similar findings. The predominant deposition of IgG1 in NELL1 positive cases has also been studied previously by Tiffany N. Caza(4). Our study was in correspondence to the same.

Inoppose to the study by Tiffany N. Caza(4) thepatients in our study with NELL1 positivity did not respond adequately to the standard immunosuppressive and conservative therapy. However with the sample size so small it would be demanding to determine the treatment responsiveness of such patients.

In contrast to the study by Xiaoying Hu et. al(5) our study did not find any correlation with malignancy. However, since the sample size is small other studies might differ from the findings of our study. Moreover, the followup of the study was only 1 year and would be rudimentary to deny the association of NELL1 with malignancies. As quoted by SyedaBehjat Ahmad(6) "whether NELL-1 is truly a primary MN etiology or in some populations related to a malignant tumor remains to be determined."

While most instances of NELL1-associated MN are likely to be categorized as idiopathic/primary MN (9), the authors suggest categorizing NELL1 MN as secondary MN, given its association with various secondary diseases documented in prior studies (9). Moreover, standard immunosuppressive therapies have shown reduced efficacy in NELL1 MN

CONCLUSION:

Based on the findings of study it can be concluded that patients with NELL1 deposit belong to an older age group as compared to overall patients of MN. Patients with NELL1 deposit cannot be differentiated from patients of primary MN nor on the basis of clinical features and nor on the GFR deposit or amount of proteinuria. The pts. With NELL 1 deposit also appear to be treatment resistant, however further subsequent research are required for the treatment strategies. Moreover, it can be safely concluded that pts. With NELL1 deposit do not seem to be at a higher risk for malignancies.

Limitations of study: the study was a single centre study with patients evaluated from last 1 year only. With such a small sample size, the extrapolation of results to the general would be deafening. The association of NELL1 MN with malignancy needs long follow up and regular

evaluation. Response and relapse of NELL1 MN associated with traditional treatment of MN needs to be further instigated before any conclusion can be drawn.

Refrences

1. Rovin BH, Adler SG, Barratt J, Bridoux F, Burdge KA, Chan TM, et al. Executive summary of the KDIGO 2021 Guideline for the Management of Glomerular Diseases. Kidney International. 2021 Oct 1;100(4):753–79.

2. Andeen NK, Kung VL, Avasare RS. NELL1 Membranous Nephropathy: Clinical associations provide mechanistic clues. Front Nephrol [Internet]. 2024 Mar 26 [cited 2024 Jun 25];4. Available from: https://www.frontiersin.org/journals/nephrology/articles/10.3389/fneph.2024.1323432/full

3. Kattah AG, Alexander MP, Angioi A, De Vriese AS, Sethi S, Cosio FG, et al. Temporal IgG Subtype Changes in Recurrent Idiopathic Membranous Nephropathy. American Journal of Transplantation. 2016 Oct 1;16(10):2964–72.

4. Caza TN, Hassen SI, Dvanajscak Z, Kuperman M, Edmondson R, Herzog C, et al. NELL1 is a target antigen in malignancy-associated membranous nephropathy. Kidney International. 2021 Apr 1;99(4):967–76.

5. Hu X, Wang G, Cheng H. Specific antigens in malignancy-associated membranous nephropathy. Front Med (Lausanne). 2024;11:1368457.

6. Ahmad SB, Appel GB. Antigens, antibodies, and membranous nephropathy: a decade of progress. Kidney International. 2020 Jan 1;97(1):29–31.

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