Content available at: https://www.ipinnovative.com/open-access-journals

Panacea Journal of Medical Sciences

Journal homepage: http://www.pjms.in/

Original Research Article

Varying zinc levels in pediatric nephrotic syndrome patients and its correlation with remission and relapse: An observational study

Rahul Jaiswal[1], Anubha Shrivastava[1], A D Tiwari[1], R K Yadav[1], Manisha Maurya[1], Nandita Mishra[1,*]

[1] *Dept. of Pediatrics, Moti Lal Nehru Medical College, Prayagraj, Uttar Pradesh, India*

ARTICLE INFO

Article history:
Received 20-07-2021
Accepted 06-09-2021
Available online 30-04-2022

Keywords:
zinc levels
Nephrotic syndrome
Remission
Relapse

ABSTRACT

Background: Zinc deficiency, common in children of developing countries, leads to susceptibility to infections. Relapses in nephrotic syndrome are precipitated mostly by infections. Most of morbidity and mortality in nephrotic patients is during nephrotic range proteinuria. Management of factors which reduce duration of relapse, can decrease morbidity in pediatric nephrotic patients. We aimed to study serum zinc levels, in patients of nephrotic syndrome, at onset/relapse and at remission and tried to find out correlation between serum zinc levels and time to achieve remission.

Materials and Methods: Observational study was conducted in tertiary centre of North India over 12 months. Consecutive pediatric patients, with initial episode/relapse of NS were enrolled. Patients were treated as per standard protocol. Time taken for remission was noted. Serum zinc level was estimated first at confirmation of initial episode/relapse and then at remission. Data were summarized as mean ± standard deviation. Groups were compared by paired t test. Pearson correlation analysis was done to assess association between initial zinc level and time to attain remission.

Results: 68 patients were screened for inclusion in the study but only 49 qualified for final analysis. Serum zinc level was statistically different in all patients at enrolment and at remission. Pearson correlation analysis showed an insignificant and inverse correlation between time to remission and serum zinc level ($r=0.14$, $p>0.05$) suggesting that, as serum zinc level at enrolment decreases, the time to remission lengthens.

Conclusion: Zinc level at initial episode/relapse was found to have negative correlation with time to attain remission.

1. Introduction:

The annual incidence of nephrotic syndrome (NS) is 1.2-16.9 per 100,000 children.[1] The pathogenesis of minimal change disease (MCD) is unclear, but there is a strong evidence of immune dysregulation, chiefly involving cell mediated immunity (CMI).[2] Abnormalities in function of T cell subset (especially regulatory T cells) have been variably reported in MCD. Lymphocyte adenosine deaminase activity (as a marker of CMI), demonstrated changes both in active and remission stage of NS.[3] Finding of increased plasma levels of IgE, relatively normal IgG4 with decreased IgG1 and IgG2 and association with atopy suggest type 2 cytokine bias in MCD.[4] NS is also associated with abnormal immunoglobulin levels. There are reports regarding involvement of complement system and B lymphocytes in pathophysiology of NS, although none of the immunological biomarkers evaluated were undeniably linked to changes in glomerular permeability and proteinuria.[3] Some studies also suggest link between

* *Corresponding author*.
E-mail address: drnandita19@gmail.com (N. Mishra).

https://doi.org/10.18231/j.pjms.2022.018

urinary chemokines (such as IL-8/CXCL8 and MCP-1/CCL2) and changes in glomerular permeability and/or the deterioration of glomerulopathies.[5] In recent years, evidence of MCD to be a podocytopathy has emerged and role of a circulating permeability factor is also postulated.[6]

Zinc deficiency is associated with high mortality in developing country. Low zinc level has been described in children with severe malnutrition, malabsorption and nephrotic syndrome due to either lack of intake, decreased absorption or loss of zinc in stool and urine.[7] Zinc has pivotal role in immune regulation. Due to its deficiency, oxidative defence is weakened. Excessive generation of reactive oxygen species is one of the incriminated mechanisms in pathogenesis of progression of renal injury.[8] Basis for infection triggered relapse are still unclear but dysregulation of immune mechanism may have some role. Evidence of perturbed CMI, its association with atopy, elevated levels of IgE and upregulated gene expression for interleukin (IL-4, IL-13) suggest T helper 2 (Th2) cytokine bias.[9] Zinc deficiency probably leads to down-regulation of T-helper 1 (Th1) cytokines, a relative Th2 bias and increased risk of infection. Zinc supplementation leads to decreased episode of infections, presumably due to augmentation of gene expression for IL 2 and interferon, thereby restoring Th1-Th2 imbalance.[10] Decreased infections lead to lower chances of relapses, hence lower morbidity and mortality, due to augmentation of immune response. But zinc supplementation as preventive strategy for NS has not been found strongly in Cochrane.[11]

We aimed to measure levels of zinc during initial episode/relapse and during remission in nephrotic patients and find correlation between initial zinc level and time to remission. This may give a hint to involvement of immune response in pathogenesis of NS and requirement of zinc supplementation to shorten the duration of relapse.

2. Materials and Methods

This observational study was conducted in Department of Pediatrics of a tertiary care centre of North India over 12 months, which included enrolment, analysis and documentation. Ethical clearance was obtained from institutional ethical committee. Consecutive patients, in the age group of 1-14 years, with generalized edema, who were admitted to emergency department or attending out patient department of Department of Pediatrics were screened for the study. The procedure of the study was explained to parents/caretakers in their own language. Written and informed consent was taken from parents/guardian before enrolment. Bedside urinary protein heat coagulation test (BSUP) was done to look for proteinuria. Those with nephrotic range proteinuria i.e., BSUP 3+/4+ underwent biochemical analysis (24-hour urinary protein >1000 mg/m^2/d, hypoalbuminemia $\leq$ 2.5 g/dl and hypercholesterolemia >200 mg/dl) to confirm first episode of NS. Those patients who were already known case of NS, their home BSUP record was revised to confirm relapse. Relapse was defined as BSUP 3+/4+ for three consecutive days, having being in remission previously; it was further divided into two- infrequent relapse and frequent relapse. Patients having frequent relapses were those, who had two or more relapses in initial six months or four or more relapses in any twelve months. All the patients were grouped in three categories; patients with initial episode (IE), infrequent relapsers (IFR) and frequent relapsers (FR). Patients with clinical evidence of systemic disease (e.g., tuberculosis, diabetes, chronic liver disease, malabsorption syndrome), congenital NS, infantile NS, those taking zinc and zinc containing supplement within three months prior to study period, or already on steroid, those with episodes of acute respiratory infections (ARI) and diarrhea three months prior to study and those with history of recurrent attacks of ARI & diarrhea were excluded from the study. Detailed history, anthropometry and physical examination of each case was recorded systematically on a standard proforma.

Patients with first episode of NS were treated with prednisolone at 2mg/kg/day in two divided doses daily for six weeks, followed by 1.5mg/kg/day single dose every alternate day for six weeks. Relapse patients were treated with prednisolone at 2mg/kg/day in two divided doses daily till remission, followed by 1.5 mg/kg/day single dose every alternate day for four weeks. Remission was defined as BSUP nil or trace for three consecutive days. Patients who did not remit on prednisolone at 2mg/kg/day for four weeks were labelled as steroid resistant NS and were excluded from study. Time taken for remission was noted in each patient. Those patients who did not complete treatment, were excluded from final analysis. Serum zinc level was done initially at confirmation of initial episode/relapse and was repeated at remission. Two millilitre sample was collected in plain vial for it. Sample was centrifuged and serum was separated for zinc level analysis by atomic absorption spectrophotometry. Other relevant blood investigations e.g., lipid profile, kidney function test and serum albumin were also done.

Data were entered into Microsoft excel spreadsheet and then analysed by SPSS 24.0. and Graph Pad Prism version 5. Data were summarized as mean ± standard deviation. Groups were compared by paired t test. A two-tailed paired t-test was used at α=5% and considered $p<0.05$ as statistically significant. Pearson correlation analysis was done to assess association between initial zinc level and time to attain remission.

3. Results

All 68 patients were screened for inclusion in the study but only 49 got qualified for final analysis (Figure 1). Baseline characteristics of the enrolled patients is in Table 1. Various

Table 1: Baseline characters of children with nephrotic syndrome

	IE(n=19)		IFR(n=20)		FR(n=10)		Total(n=49)	
Age (in years)	**Number**	**%**	**Number**	**%**	**Number**	**%**	**Number**	**%**
<5	12	63.16%	5	25%	0	0	17	34.7%
5-10	5	26.32%	15	75%	8	80%	28	57.1%
>10	2	10.53%	0	0	2	20%	4	8.2%
				Gender				
Male	12	63.16%	14	70%	7	70%	33	67.3%
female	7	36.84%	6	30%	3	30%	16	32.7%
				Residence				
Rural	10	52.6%	18	90%	5	50%	33	67.3%
urban	9	47.4%	2	10%	5	50%	16	32.7%

IE = initial episode; IFR = infrequent relapse; FR = frequent relapse

Table 2: Biochemical findings at enrolment and at remission (Initial episode) p-value <0.05 = statistically significant; SD = standard deviation

n=19	**Assessed at**	**Mean**	**SD**	**Minimum**	**Maximum**	**Median**	**p-value**
VDRL (mg/dl)	Enrolment	100.52	45.18	7.83	186.0	95.0	0.32
	Remission	85.76	44.73	7.96	179.0	90.0	
HDL (mg/dl)	Enrolment	58.70	17.59	26.80	82.72	65.43	0.63
	Remission	62.02	24.0	25.60	105.58	60.30	
TG (mg/dl)	Enrolment	469.46	170.59	231.79	931.20	443.20	<0.0001
	Remission	158.63	84.89	79.20	447.84	132.30	
Cholesterol (mg/dl)	Enrolment	437.64	144.48	272.00	777.00	453.13	<0.0001
	Remission	155.15	94.99	46.20	409.50	110.00	
Serum urea (mg/dl)	Enrolment	43.83	25.41	17.13	117.90	36.64	0.39
	Remission	37.57	18.94	22.48	96.80	29.29	
Serum creatinine (mg/dl)	Enrolment	0.70	0.27	0.18	1.07	0.71	0.72
	Remission	0.73	0.20	0.34	0.96	0.78	
Serum albumin (g/dl)	Enrolment	2.59	0.58	1.65	3.89	2.40	<0.0001
	Remission	3.36	0.36	2.86	4.48	3.24	
Serum zinc (μg/dl)	Enrolment	49.32	33.25	4.00	165.00	43.00	0.0002
	Remission	104.00	47.61	29.00	213.00	98.00	

VLDL= very low-density lipoprotein; HDL= high density lipoprotein; TG = triglyceride

Table 3: Biochemical findings at enrolment and at remission (Infrequent relapse) p-value <0.05 = statistically significant; SD = standard deviation

n=20	**Assessed at**	**Mean**	**SD**	**Minimum**	**Maximum**	**Median**	**p-value**
VLDL (mg/dl)	Enrolment	73.64	40.57	4.59	132.20	82.50	0.44
	Remission	84.01	42.90	16.00	178.00	80.30	
HDL (mg/dl)	Enrolment	57.52	18.80	9.91	90.00	58.40	0.68
	Remission	55.21	16.65	22.67	80.15	54.30	
TG (mg/dl)	Enrolment	281.22	128.50	104.20	531.92	261.68	0.0002
	Remission	135.40	94.87	36.20	412.00	109.20	
Cholesterol (mg/dl)	Enrolment	286.95	140.85	76.20	612.00	235.45	0.0002
	Remission	139.96	78.13	41.20	380.37	116.20	
Serum urea (mg/dl)	Enrolment	33.96	22.52	14.34	102.40	25.20	0.70
	Remission	31.79	11.04	16.27	53.50	28.99	
Serum creatinine (mg/dl)	Enrolment	0.84	0.31	0.34	1.60	0.86	0.43
	Remission	0.78	0.20	0.43	1.10	0.78	
Serum albumin (g/dl)	Enrolment	2.79	0.70	1.30	4.41	2.75	0.0021
	Remission	3.55	0.76	2.32	5.87	3.39	
Serum zinc (μg/dl)	Enrolment	46.75	27.58	4.00	127.00	43.00	<0.00001
	Remission	144.90	74.95	20.00	369.00	134.00	

VLDL= very low-density lipoprotein; HDL= high density lipoprotein; TG = triglyceride

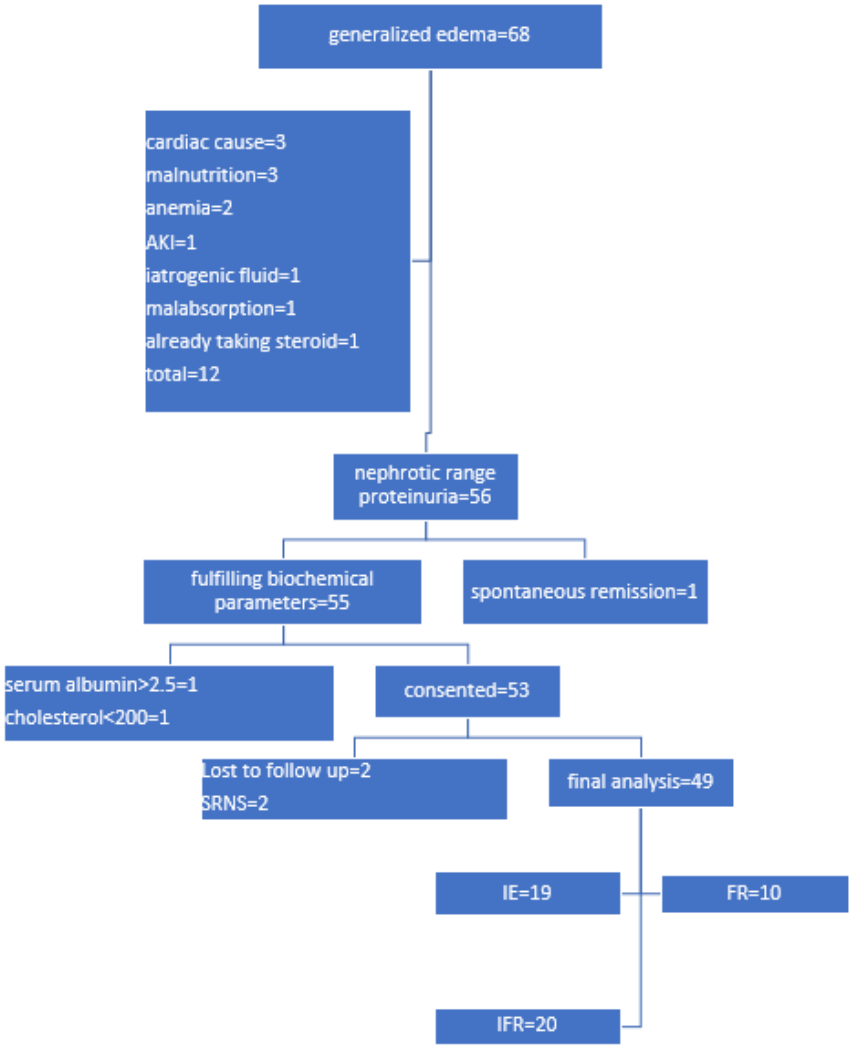

Fig. 1: Patient recruitment. AKI = acute kidney injury
IE = initial episode; IFR = infrequent relapse; FR = frequent relapse

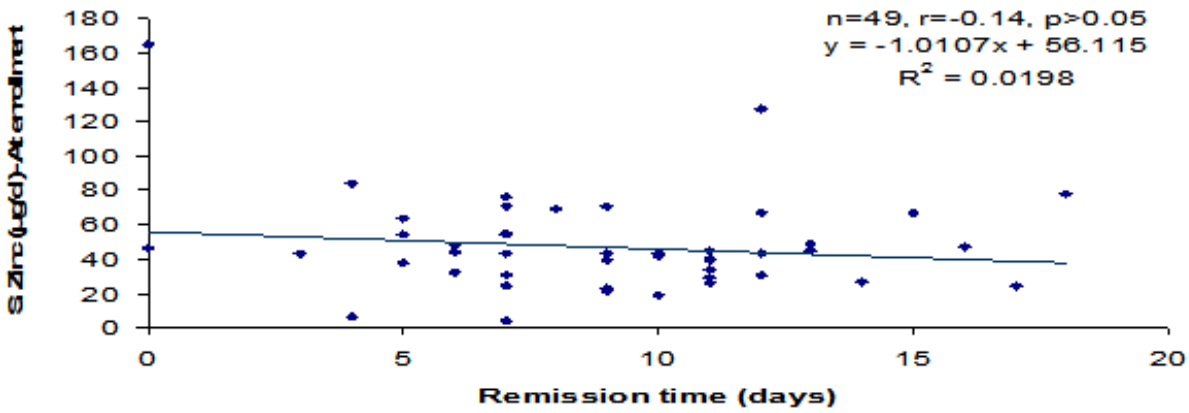

Fig. 2: Pearson correlation graph between zinc level at enrolment and time to attain remission.

Table 4: Biochemical findings at enrolment and at remission (Frequent relapse) p-value <0.05 = statistically significant; SD = standard deviation

n=10	Assessed at	Mean	SD	Min.	Max.	Median	p-value
VDRL (mg/dl)	Enrolment	71.17	44.78	3.39	132.20	68.00	0.97
	Remission	71.87	28.00	42.00	108.70	67.10	
HDL (mg/dl)	Enrolment	50.99	20.93	9.40	90.52	48.00	0.59
	Remission	56.08	20.35	32.20	102.00	55.19	
TG (mg/dl)	Enrolment	276.45	113.20	145.65	427.00	262.60	0.0025
	Remission	133.25	61.49	92.20	291.15	107.60	
Cholesterol (mg/dl)	Enrolment	244.56	86.52	139.56	452.08	222.20	0.0050
	Remission	138.77	59.02	75.00	244.38	115.20	
Serum urea (mg/dl)	Enrolment	38.27	11.22	22.75	54.60	36.25	0.96
	Remission	38.52	9.70	26.42	59.90	36.95	
Serum creatinine (mg/dl)	Enrolment	0.72	0.17	0.44	0.98	0.69	0.29
	Remission	0.82	0.25	0.32	1.10	0.93	
Serum albumin (g/dl)	Enrolment	2.78	0.69	1.80	3.93	2.66	0.08
	Remission	3.22	0.34	2.64	3.70	3.32	
Serum zinc (μg/dl)	Enrolment	44.30	17.83	21.00	78.00	43.50	<0.00001
	Remission	104.30	29.50	52.00	151.00	104.50	

VLDL= very low-density lipoprotein; HDL= high density lipoprotein; TG = triglyceride

biochemical parameters in all the three categories of patients at enrolment and at remission are enumerated in Tables 2, 3 and 4. In patients with initial episode and infrequent relapses, levels of triglyceride, cholesterol and albumin was statistically different at enrolment and at remission. In frequent relapsers, only levels of triglyceride and cholesterol was statistically different at enrolment and at remission. Serum level of zinc was statistically different in all patients at enrolment and at remission.

Correlation between serum zinc level at enrolment and time to remission is summarized graphically in Figure 2. Pearson correlation analysis showed an insignificant and inverse correlation between time to remission and serum zinc level (r=0.14, p>0.05) suggesting that as serum zinc level at enrolment decreases, the time to remission increases. However, the correlation was statistically not significant. On subgroup analysis; correlation between initial serum zinc level and time to remission showed a significant and negative correlation (r=6.082, p<0.05) in patients with initial episode suggesting that as serum zinc level decreases, time to remission increases and vice versa. Correlation in IFR showed, insignificant and positive correlation between serum zinc level and time to remission (r=0.862, p>0.05). Correlation in FR patients also showed an insignificant and positive correlation between initial serum zinc level and time to remission (r=2.225, p>0.05).

4. Discussion

In present study, there was significant difference in mean value of serum zinc during relapse and remission (p value<0.05). It was low at relapse but normal at remission. Correlation study do point out that low serum zinc level at relapse led to greater time required to attain remission, however the values were not statistically significant.

The serum for assessment of zinc level was stored (at -20°C) for an average of one month before analysis as samples needed to be transported to another centre for analysis. Changes, if any, brought about by this storage is not accounted for, in the analysis. Zinc level at remission was considered to be representative of normal baseline level for individual patients, which may not be true in all scenarios.

Almosawi concluded hypozincemia can occur in chronic renal problem like NS. Low serum zinc level was commoner in frequent relapsers.[12] Asim Mumtaz found that there was high prevalence of zinc and copper deficiency in patients suffering from NS. Causes of hypozincemia and hypocuperemia were hypoalbuminemia and raised 24-hour urinary protein losses. Other probable factors were decreased dietary intake and increased loss of trace metals in urine.[13] Lindeman also studied serum zinc concentrations and found it to be decreased in patients with a variety of clinical disorders including nephrotic syndrome, cirrhosis and renal insufficiency. He found that symptoms of acute zinc deficiency (anorexia, dysfunction of smell and taste, mental and cerebellar disturbances) and chronic zinc deficiency (growth retardation, anemia, testicular atrophy and impaired wound healing) are common in these patients.[14] However, to our knowledge there is no study that found out correlation between the low zinc status and persistence of proteinuria.

5. Conclusion

Overall zinc level was found to have negative correlation with remission time. Therefore, it can be concluded that zinc supplementation with standard steroid therapy can

be tried to decrease relapse duration. Characterization of profile of immune response might help in development of specific and individualized therapies, leading to clinical improvement and better prognosis. However, large scale, well designed, prospective studies across different setting are needed to substantiate the recommendation of zinc along with standard steroid therapy in idiopathic nephrotic syndrome.

6. Sources of Funding

No financial support was received for the work within this manuscript.

7. Conflict of interest

The authors declare they have no conflict of interest

References

1. Noone DG, Iijima K, Parekh R. Idiopathic nephrotic syndrome in children. *Lancet*. 2018;392(10141):61–74. doi:10.1016/S0140-6736(18)30536-1.
2. Bagga A, Mantan M. Nephrotic syndrome in children. *Indian J Med Res*. 2005;122(1):13–28.
3. Mishra OP, Garg R, Z A, Usha. Adenosine deaminase activity in nephrotic syndrome. *J Trop Pediatr*. 1997;43(1):33–7. doi:10.1093/tropej/43.1.33.
4. Culver EL, Sadler R, Bateman AC, Makuch M, Cargill T, Ferry B, et al. Increases in IgE, Eosinophils, and Mast Cells Can be Used in Diagnosis and to Predict Relapse of IgG4-Related Disease. *Clin Gastroenterol Hepatol*. 2017;15(9):1444–52.
5. Wde FP, Brito-Melo GE, Guimarães FT, Carvalho TG, Mateo EC. Simões e Silva AC. The role of the immune system in idiopathic nephrotic syndrome: a review of clinical and experimental studies. *Inflamm Res*. 2014;63(1):1–12.
6. Vivarelli M, Massella L, Ruggiero B, Emma F. Minimal Change Disease. *Clin J Am Soc Nephrol*. 2017;12(2):332–45.
7. Javed F, Asghar A, Sheikh S, Butt MA, Hashmat N, Malik BA, et al. Comparison of Serum Zinc Levels Between Healthy and Malnourished Children. *Ann Punjab Med Coll (APMC)*. 2009;3(2):139–43.
8. Dwivedi J, Sarkar PD. Study of oxidative stress, homocysteine, copper & zinc in nephrotic syndrome: therapy with antioxidants, minerals and B-complex vitamins. *J Biochem Tech*. 2009;1(4):104–7.
9. Mathieson PW. Immune dysregulation in minimal change nephropathy. *Nephrol Dial Transplant*. 2003;18(S6):26–9.
10. Prasad AS. Effects of zinc deficiency on Th1 and Th2 cytokine shifts. *J Infect Dis*. 2000;182(1):62–8. doi:10.1086/315916.
11. Wu HM, Tang JL, Sha ZH, Cao L, Li YP. Interventions for preventing infection in nephrotic syndrome. *Cochrane Database Syst Rev*. 2004;(2):3964. doi:10.1002/14651858.CD003964.
12. Almosawi QM. Serum zinc level in children with relapsing nephrotic syndrome. *Int J Curr Res*. 2016;8(11):42328–30.
13. Mumtaz A, Anees M, Fatima S, Ahmed R, Ibrahim M. Serum Zinc and Copper levels in nephrotic syndrome patients. *Pak J Med Sci*. 2011;27(5):1173–6.
14. Lindeman RD, Baxter DJ, Yunice AA, King RW, Kraikit S. Zinc metabolism in renal disease and renal control of zinc excretion. *Prog Clin Biol Res*. 1977;14:193–209.

Author biography

Rahul Jaiswal, Junior Resident

Anubha Shrivastava, Associate Professor

A D Tiwari, Professor

R K Yadav, Assistant Professor

Manisha Maurya, Associate Professor

Nandita Mishra, Assistant Professor

Cite this article: Jaiswal R, Shrivastava A, Tiwari AD, Yadav RK, Maurya M, Mishra N. Varying zinc levels in pediatric nephrotic syndrome patients and its correlation with remission and relapse: An observational study. *Panacea J Med Sci* 2022;12(1):91-96.