Research Article

Hypothyroidism in Children with Steroid-Resistant Nephrotic Syndrome and its Correlation with Clinicopathological Parameters at a Tertiary Care Center in Pakistan

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Abstract

Objective: To find out the frequency of hypothyroidism in steroid-resistant nephrotic syndrome children and to correlate thyroid hormone status with laboratory parameters.

Methods: The study was carried out at Pediatric Nephrology, Sindh Institute of Urology and Transplantation, Karachi, Pakistan from July 2019 to July 2020. In all, 73 children (1-18 years) with steroid-resistant nephrotic syndrome were enrolled. Their demographic, laboratory and histopathological data including thyroid hormone profile at diagnosis were recorded. Clinicopathological features of hypothyroid and euthyroid children were compared. Thyroid profiles of hypothyroid children at diagnosis and after 3 months of immunosuppressive therapy were compared and 3-month profiles were also correlated with degree of proteinuria/response to treatment. Statistical analysis was performed by SPSS version 20.0.

Results: Of 73 children, 20 (27%) were hypothyroid: 18 (90%) subclinical and 2 (10%) with overt hypothyroidism at diagnosis. None of these had anti-thyroid antibodies or abnormal findings on ultrasound. At end-point, 3/20 (15%) were lost to follow up. Of remaining 17 children, 6 (35%) patients who achieved remission of proteinuria had normal thyroid hormones and among the remaining 11 (65%) patients with persistent proteinuria, 6/11 (54.5%) had normal thyroid stimulating hormone values, 4/11 (36.4%) had subclinical hypothyroidism while only one patient (9%) had overt disease. A statistically significant correlation was observed between the thyroid hormones, degree of proteinuria, low serum albumin and high cholesterol.

Conclusion: Hypothyroidism was observed in nearly one third sizeable of children with steroid-resistant nephrotic syndrome. Persistent proteinuria may lead to subclinical or overt hypothyroidism, necessitating monitoring of thyroid function tests.

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Key Words: Children, hypothyroidism, nephrotic syndrome, steroid resistance.

Introduction:

Massive proteinuria in nephrotic syndrome (NS), whether steroid sensitive (SSNS) or steroid resistant (SRNS), occurs due to a disruption in the glomerular filtration barrier (GFB) and subsequent impaired uptake of filtered protein as tubular resorptive capacity is overwhelmed. The plasma proteins including thyroid hormone binding proteins (TBG, albumin) that serve to maintain the thyroid hormones (T3, T4) in normal ranges during physiological states are lost[1,2]. As a consequence, the urinary excretion of

free and bound forms of thyroid hormones is increased in proportion to the degree of proteinuria. In turn, the secretion of thyroid stimulating hormone (TSH) is increased in order to normalise the circulating hormone levels[3].

It has been shown in children with SSNS that there is a state of mild or subclinical hypothyroidism during proteinuria which normalizes on achieving remission[4]. With inadequate compensation in thyroxine production under the influence of increased TSH, overt hypothyroidism may also occur. The uncontrolled proteinuria in children with SRNS may progressively damage the tubular epithelial cells resulting in decreased re-absorption of thyroid hormones and eventually causing overt hypothyroidism that would need treatment[5]. The frequency of subclinical hypothyroidism in SRNS children is reported to be 20-33% in different studies[5,6]. Thyroid hormones play a crucial role in growth and development, normal functioning of central nervous system, sodium and water homeostasis and influence the risk of developing hypertension and dyslipidaemia[7].

SRNS children have a prolonged clinical course and there is limited data about thyroid functional status specifically in this patient group as they may have persistent proteinuria for few months before achieving remission. In addition, very few studies have addressed the outcome of thyroid functional status with the treatment and outcome of the underlying SRNS[5]. Importantly, there is no information on the prevalence of hypothyroidism in children with SRNS in Pakistan. This gap in knowledge compelled us to carry out this study. The objective of the current study was to determine the frequency of hypothyroidism in SRNS children at the time of diagnosis. Secondary objectives were to compare the laboratory and histopathological features of hypothyroid and euthyroid children, to compare the thyroid hormone levels at the time of diagnosis and at 3-month follow-up and to correlate the later with the degree of proteinuria at 3-month follow-up in hypothyroid children.

Methods:

This descriptive, observational study was carried out at Pediatric Nephrology Department of Sindh Institute of Urology and Transplantation (SIUT), Karachi, Pakistan from 1st July 2019 to 31st July 2020 after obtaining approval from the institutional scientific and ethical review committees. Written informed consent was obtained from the guardians before recruitment for children and assent from adolescents. All children and adolescents with persistent proteinuria and steroid resistance at 6 weeks were labeled as SRNS and were included in the study. These were investigated for the presence of clinical or biochemical hypothyroidism. Blood samples were obtained for serum creatinine, total cholesterol, serum albumin and thyroid function tests. Quantification of proteinuria was done by obtaining spot urinary protein to creatinine ratio (UPCR). All patients also underwent renal biopsy to determine the underlying histopathology before starting immunosuppressive therapy of SRNS. The later included one of calcineurin inhibitors (CNIs), such as cyclosporine in the majority of cases. A few cases were treated with Tacrolimus as the first line therapy. Subclinical hypothyroidism was defined as an increase in serum TSH above the upper limit of the normal range with a normal serum FT4 level, while overt hypothyroidism was defined as a low serum FT4 and an increased serum TSH above the upper limit of normal range. Based on the previous study on the frequency of subclinical hypothyroidism in SRNS of 33% with margin of error 8% and 95% confidence interval, a sample size of 73 patients was calculated for this study.

Those children who were initially SSNS but later on developed SRNS during a subsequent relapse (secondary SRNS) were also included in the study. Children with secondary NS (Lupus nephritis, Henoch schonlein purpura nephritis), abnormal renal functions, hypothyroidism of autoimmune origin, congenital nephrotic syndrome (CNS) or children with critical illness requiring ICU management were excluded from the study. Serum TSH and free T4 (FT4) levels were measured using the electro-chemiluminescence immunoassay (ECLIA) method. Beckman Coulter Access Immunoassay Systems kits were used for TSH and FT4. For the reference range, the normal range of each kit was used. In patients with abnormal thyroid hormone profile, thyroid

peroxidase antibody (anti-TPO), thyroglobulin antibody and ultrasound of thyroid were done to rule out autoimmune cause. Those with biochemical hypothyroidism were followed up for 3 months to see whether their thyroid profile improved with the treatment given for SRNS.

Standard definitions according to KDIGO 2012 guidelines were used to define SRNS, Complete remission (CR) and Partial remission (PR)[8]. The reference range of serum TSH was taken as 0.34-5.6 uIU/ml[9] and serum FT4 as 0.58-1.64 ng/dL[10].

The data was entered and analyzed in SPSS version 20. Continuous variables were presented as mean and standard deviation (SD) and categorical variables were reported as frequencies and percentages. Normally distributed variables were compared between hypothyroid and euthyroid SRNS children by unpaired t-test. Thyroid profiles of hypothyroid SRNS patients at diagnosis were compared with those at 3-month follow-up using the paired t-test. Correlation between thyroid hormones and UPCR, serum total cholesterol and albumin was done by Pearson's Correlation Coefficient. A p-value ? 0.05 was den-oted as statistically significant.

Results:

Out of a total of 73 children with SRNS, 20 (27 %) were found to have hypothyroidism. Among them, 18 (90%) children had subclinical hypothyroidism and 2 (10%) had overt hypothyroidism at diagnosis. The demographics, baseline laboratory parameters and histopathologic diagnoses of both euthyroid and hypothyroid children are shown in Table 1. The mean age of hypothyroid children was 5.3 ± 3.8 years. There were 14 males and six females, with male to female ratio of 2.3:1. The mean total serum cholesterol was 390 ± 149.2 mg/dl and mean serum albumin, 1.8 ± 0.8 g/dl. Mean TSH was 9.9 ± 3.7 uIU/ml and mean FT4, 0.9 ± 0.3 ng/dL in this cohort of children. None of these patients, however, had goitre on clinical examination or thyromegaly on ultrasound. Antithyroid antibodies (anti-TPO and anti-thyroglobulin antibody) were absent in all hypothyroid patients. The most common histopathological pattern observed in hypothyroid patients was IgM nephropathy as compared to the most common pattern in the overall studied cohort of FSGS (Table 1). There was statistically significant difference in the levels of serum total cholesterol, serum albumin, and TSH in hypothyroid children as compared to euthyroid ones ($p = 0.017$, 0.014 and < 0.001, respectively), as shown in Table 1.

At 3-month follow-up, of the 20 hypothyroid children, 3/20 (15%) were lost to follow-up. Among the 17 patients who completed follow-up, 11 (65%) did not achieve remission but the thyroid profiles of about half of them (6[54.5]%) normalised. Of the remaining, 4/11 (36.4%) children continued to have subclinical hypothyroidism while only 1 (9%) child had overt hypothyroidism and was treated with hormone replacement. All the 6 (35.3%) children who achieved either PR or CR had normal TSH levels at 3 months ($p = 0.02$, $p = 0.009$, respectively). Table 2 shows the thyroid hormonal status of hypothyroid children both at diagnosis and at 3-month follow-up according to degree of proteinuria/response to treatment of NS, categorized as no remission, CR and PR. FT4 values were within normal limits in all three groups. There was a statistically significant difference in the TSH values of patients with CR or PR and even in those with no remission, as compared to their baseline values. On the other hand, FT4 levels showed significant difference in CR group.

In children who achieved remission, there was about equal proportion of histopathological diagnoses with FSGS in 2 (33.3%) patients and IgM nephropathy in 4 (66.6%).

Table 3 depicts the relationship between serum thyroid hormone levels, serum total cholesterol, serum albumin and UPCR at 3-month follow up. A significant and positive correlation was observed between serum TSH levels and UPCR ($p = 0.002$) and total cholesterol ($p = 0.001$). FT4 showed a significant but negative correlation with total cholesterol ($p = 0.017$) and significant positive correlation with serum albumin ($p = 0.045$). No significant relationship was found between serum TSH and serum albumin levels. Similarly, FT4 levels showed no significant relationship with UPCR.

Table 1: *Demographic, clinical and biochemical parameters of steroid-resistent nephrotic syndrome (SRNS) patients at the time of diagnosis according to thyroid hormone status (n = 73).*

Variables	SRNS patients with hypothyroidism (n= 20)	SRNS patients with euthyroid status (n=53)	p-value
Age	5.3 ± 3.8	6.2 ± 3.6	0.361
Gender (M/F)	14/6	39 /14	0.759
Serum Total Cholesterol	390 ±149.2	305.5± 125	0.017*
Serum Albumin	1.82±0.84	2.4± 0.87	0.014*
Thyroid stimulating hormone	9.9± 3.7	3 ± 1.3	<0.001*
FT4	0.9± ±0.3	1.06± 0.3	0.243
Histopathological diagnoses			
Minimal change disease	2	5	0.999
Focal segmental glomerulosclerosis	8	30	0.205
IgM nephropathy	9	7	0.003
Mesangial proliferative GN	0	1	NA
Membranous GN	1	7	0.432
Membranoproliferative GN	0	3	NA

*P value < 0.05
NA = not applicable. Note: analysis using unpaired T-test

Table 2: *Comparison of thyroid profile of hypothyroid SRNS children at the time of diagnosis and at 3-month follow-up (n = 17).*

Variables	Mean ± S.D	Δ (Difference)	p-value
Complete remission (UPCR<0.2 mg/mg) n = 3			
TSH at diagnosis	6.8±0.9	5.03	0.009*
TSH at 3 months	1.8±0.4		
FT4 at diagnosis	0.94±0.25	0.16	0.03*
FT4 at 3 months	1.1±0.24		
Partial remission (UPCR 0.2-2 mg/mg) n=3			
TSH at diagnosis	14.6±4.3	11.8	0.02*
TSH at 3 months	2.73±1.64		
FT4 at diagnosis	1.04±0.36	0.15	0.45
FT4 at 3 months	0.88±0.11		
No remission (UPCR >2 mg/mg) n=11			
TSH at diagnosis	9.6±3.5	4.63	0.02*
TSH at 3 months	5±4.5		
FT4 at diagnosis	0.93±0.23	0.04	0.6
FT4 at 3 months	0.88±0.23		

*P value < 0.05

Table 3: *Correlation of thyroid hormone levels at 3-month follow up with laboratory parameters.*

Variables	TSH	FT4	T. Cholesterol	S. Albumin	UPCR
TSH	-------	r = - 0.39 p = 0.114	r = 0.608 p = 0.009	r = -0.460 p = 0.063	r = 0.679 p = 0.002
FT4	r = -0.396 p = 0.114	---------	r = -0.567 p = 0.017	r = 0.491 p = 0.045	r = -0.325 p = 0.201

*P value< 0.05
r = correlation coefficient, TSH = Thyroid stimulating

Discussion:

This study reports the frequency of hypothyroidism in SRNS children at the time of diagnosis of the later illness at a single center in Pakistan and to compare thyroid function status in this group of children after 3 months of treatment for SRNS. Non-immune biochemical hypothyroidism was observed in slightly more than one fourth of children with SRNS in this cohort. More or less similar frequencies were also reported by Sharma et al and Kapoor et al[5,6]. A slightly higher frequency (33.3%) was reported in an Indian study by Marimuthu et al.[7] These results indicate that the frequency of hypothyroidism in SRNS children is more or less similar in different studies from different parts of the world. Most other studies are however cross-sectional studies. Only few studies have examined the trend in change of thyroid hormone status with treatment and outcome of the underlying glomerular disease.

Our patients showed subclinical and overt hypothyroidism only in a state of persistent proteinuria. At the 3-month follow up, it was found that those patients, who were initially hypothyroid, became euthyroid, as they went into CR or PR. This finding was consistent with a previous study by Afroz et al[4]. Sharma et al. also noted improvement in thyroid function parameters with remission of proteinuria[5]. However, this is contrary to the observation of Kapoor et al. who reported subclinical hypothyroidism even in states of PR and CR[6]. One potential factor that may lead to this discrepancy is the nature of treatment given for underlying SRNS. It has been reported that glucocorticoids interfere with the regulatory mechanisms of thyroid hormones and may affect thyroid hormone levels. However, the major determinant of thyroid hormonal alterations is the degree and selectivity of proteinuria.

It was also seen that TSH of slightly more than half of patients normalized with normal FT4 despite persistent proteinuria. The exact reason for this finding is not clear. One explanation might be that the quantity of proteinuria decreased in these individuals after treatment and there was selective proteinuria resulting in a decrease in thyroid binding globulin excretion in urine, which in turn improved TSH levels. This study also showed a positive and significant correlation between serum TSH levels, UPCR and serum cholesterol levels as reported by Sharma et al.[5] Likewise, serum albumin concentration showed a significant but negative correlation with UPCR[5]. However, we did not observe a significant relationship between serum FT4, serum TSH and serum albumin levels.

Regarding histopathological correlation with subclinical hypothyroidism in our patients, the dominant pattern was IgM nephropathy followed by FSGS. This is in contrast with the findings of a study in adults, which reported membranous nephropathy as the dominant pattern in subclinical hypothyroidism[11]. This difference could be explained by different histopathologic spectrum of NS in children and adults, where membranous nephropathy is more common in the later group[12].

Although, only one patient in our study warranted referral for treatment of persistent overt hypothyroidism, a brief discussion of treatment of later seems appropriate. There is no documented role of levothyroxine in subclinical hypothyroidism as shown by some studies. Lazar, et al. studied 3510 patients with subclinical hypothyroidism over 5 years and concluded that in children, initial normal or slightly increased serum TSH levels are likely to remain normal or normalize spontaneously in the absence of specific treatment[13]. Similarly, a Korean pediatric study reported 31 patients with NS and found that thyroid hormones' levels normalize after remission, regardless of levothyroxine therapy[14]. IPNA guidelines for SRNS recommend treating children with TSH level >10 mIU /l and low FT4 while they suggest periodical monitoring of thyroid function tests in subclinical hypothyroidism[15] Treatment with levothyroxine supplementation is reserved for children with autoimmune cause and in selected cases of persistent elevation of TSH >10 mlU/ml[16,17].

There are certain strengths and few limitations in the current study. The strengths are a fairly large sample size of a uniform racial population of children and follow-up, albeit of very short duration. In 3 months, the future course of SRNS is generally clarified, which may remit or continue to progress. Most other studies on this subject are cross-sectional studies. The limitations are that it is a single center based study and follow-up is short. There is no control group to compare the results of hypothyroidism.

Despite these limitations, we were able to demonstrate a relationship between thyroid hormonal levels and the degree of proteinuria in SRNS children.

Conclusion:

In conclusion, children with SRNS with persistent nephrotic-range proteinuria, low serum albumin (< 2.5 g/dl) and high serum cholesterol levels (> 200 mg/dl) are more likely to have hypothyroidism and therefore, such children should be screened for thyroid function. The majority of children have subclinical hypothyroidism and hormone replacement therapy should be reserved only for those children who have overt hypothyroidism.

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Conflict of Interest: The authors declare no conflict of interest
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