Random proteinuria screening in elementary school children in Jos metropolis, Nigeria

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Abstract

Background: Proteinuria in children may represent a benign condition or a serious underlying renal disease or systemic disorder.
Methods: A total of 249 pupils were selected for screening from three different primary schools in Jos metropolis, Plateau State, Nigeria. Proteinuria was determined qualitatively using bromophenol blue method impregnated in combi 10 dip stick. Those positive for proteinuria were quantified using 3% sulphur salicylic acid technique.
Results: Out of the 249 pupils who participated in this study, 12 (4.8%) had proteinuria [5 (2.0%) females and 7 (2.8%) males respectively]. Urinary levels of above 150mg/dl or 5mg/ml gave a positive result with the dipstick and were considered to increase the risk for kidney disease.
Conclusion: Prevalence of proteinuria in elementary school children in Jos metropolis is on the increase. More work should be done on evaluation of urinary protein creatinine ratio in children to assist in diagnosis of renal disease in children.

Keywords: Proteinuria, kidney disease, elementary school children.

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Introduction:
Early detection of proteinuria has more often than not been underrated as abnormal rise in proteinuria is a significant risk factor for both renal disease and for cardiovascular morbidity and mortality. The kidneys play a vital role in the maintainence of good health and hence taking good care of it would prevent chronic kidney failure[1]. Early detection of various diseases in life cannot be over emphasized as it usually prevents adverse undesired effects especially in low-income families and the economy of developing countries like Nigeria. Early detection of diseases has the advantage of preventing disease progression, helps the less privilege in a society and cuts down the cost of medical bills while enhancing the health of families.

Proteinuria is described as the urinary protein excretion of greater than 150mg/day or 5mg/dL[2]. Proteinuria of more than 0.15g/day may indicate an underlying kidney disease[3].

Normal barriers to protein filtration begin in the glomerulus, which consists of unique capillaries that are semi permeable but are effective barriers to plasma proteins. The adjacent basement membrane and visceral epithelial cells of the glomerulus are covered with negatively charged proteoglycans[3].

Proteins cross to the tubular fluid in inverse proportion to their size and charge. Proteins with a molecular weight of less than 20kilodaltons pass easily across the glomerular capillary wall. Conversely, albumin, with a molecular weight of 65kilodaltons and a negative charge is restricted from crossing. The smaller proteins are reabsorbed at the proximal tubule after they cross the glomerulus leaving an undetected quantity in the urine[4].

Proteinuria is mostly asymptomatic so the majority of individuals with proteinuria may not know about it until it is detected during a routine checkup. Some students come from homes where visits to hospitals are not encouraged due to religious beliefs or due to poverty. Some adults end up with renal diseases that could have been detected earlier in life and treated if they had the priviledge to be screened at an early age.

"Despite the well known limitations, currently the most widely used biomarkers for the detection of chronic kidney disease and acute kidney injury are proteinuria, serum creatinine and blood urea nitrogen"[5] according to Rosner et al.In a study done in Jos by Akor et al[6], proteinuria was the commonest urinary abnormality found in school children.

Proteinuria is also known as proteins in urine or albuminuria and it gives a clue to the state of the kidneys and can be a prognostic tool for detection and management of kidney disease. Therefore, this research

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was aimed at determining the prevalence of proteinuria among school children in Jos, Nigeria.

Materials and Methods

This was a screening study carried out in three primary schools located within the Jos metropolis of Plateau State,Nigeria. Ethical Clearance was obtained from the ethical committee of the Jos University Teaching Hospital. The 2006 Nigeria provisional census put the population of Plateau state at 3,128,712 with 1,585,679 females.

Plateau state lies between latitude 7° and 11° North and longitude 70° and 25° East. The capital city is a pear shaped upland known as Jos Plateau. It stretches for approximately 104km from north to south and 80 km from east to west covering an area of about 8,600km. This region has a height of 1,200m above sea level

The study was carried out on both male and female pupils between the ages of six and twelve years. Consent was obtained from the Head Teacher and parents of the respective schools involved. A questionnaire was also administered which was completed by the parents of the pupils and returned before the collection of urine samples.

The study was carried out on 450 pupils; (150 pupils from each of the three schools that were visited making 450 pupils in all).

The pupils were randomly selected from the three schools.A structured questionnaire was administered and filled by consenting respondents. The sample size was obtained using the Thursfield formula of 1997;

$n = (1.96)^2 P_{exp} (1- P_{exp}) / d^2$.
Where n = sample size
p=expected prevalence (proteinuria = 3.5% Akor et al., 2009)[6]
d = desired absolute precision of 5%
$n = (1.96)^2 X\ 0.035\ X\ (1-0.035) / (0.05)^2$
$n = 3.842\ X\ 0.035\ X\ 0.965 / 0.0025$
$n = 52$

A chemically clean universal container was given to the pupils into which a random urine sample about 20mls was collected and submitted within few minutes of voiding. The urine was examined visually and chemically , (this entailed observation of colour, pH, odour and specific gravity). Proteinuria was detected using dipstick (tetrabromophenol blue strip) and positive samples were subjected to protein quantification technique using 3% sulfur salicylic acid test[7]. Pupils with a high proteinuria were rescreened in the next 3days to confirm the proteinuria. All pupils whose parents gave consent were enrolled for this study while pupils below the ages of 6 years and above the ages of 12 years are excluded. The method used was the tetrabromophenol blue dipstick method and 3% sulfosalicylic acid method was used to quantify the proteins found in the urine of children that were positive for the dipstick.The method used is based on the principle that the peptide bond of proteins react with tetrabromophenol in an acidic medium to produce a colour which is directly proportional to the concerntration of protein present in urine[7].

The procedure entailed dipping the test end of the strip into freshly voided urine for about 1 second. The excess urine was drained off along the edge of the container and the colour of the strip was compared with the colour chart on the container within 60 seconds. Presence of proteins was indicated by a change in colour from yellow to bluish green depending on the concentration of protein present.

The Sulfosalicylic acid (SSA) method assay for proteinuria detection was based on the principle that 3% sulphosalicylic acid causes precipitation of proteins with resultant production of turbidity and can be measured using a spectrophotometer[8]. In this method, 3 test tubes were setup, the blank which contained 0.2ml of normal saline, standard tube which contained 0.2ml of protein standard solution and the test which contained 0.2ml of urine. 0.8mls of 3%SSA was added to all the tubes, the solution was well mixed and allowed to stand for 10minutes at room temperature. The standard and test were read against the blank at 450nm with glass cuvette of 1cm light path.

Concerntration of protein in urine was calculated using the formular below:

$$\text{Conc. of test} = \frac{\text{abs of test}}{\text{abs of std}} \text{ X conc. of std}$$

Where:
conc = concentration
abs = absorbance
std = Standard

Data was analysed using simple descriptive statistical methods.

Results

Two hundred and forty nine out of the 450 pupils who were administered questionnaires returned completed questionnaires giving a response rate of 55.3%. Out of the 249 pupils enrolled for this study; 127 were males (51%) while 122 were females (49%) with a male/female ratio of approximately 1:1 as shown in Table 1. The age group 8-9years had the largest number of participants [112 (45%)] while 6-7 year olds were the fewest 19.3%]. Parents of the children were mostly gradutes (45%) with only few (5.6%) having no form of education. The source of drinking water for 81.5% of the participants was tap water (water board treatment center) while 14.9% had

water from well, 2.4% had source of drinking water from borehole and 1.2% got water mainly from sachet.

Out of the 122 (49.0%) females screened, 5 (2.0%) were positive for proteinuria while 117 (47%) were negative. On the other hand, 7 (2.8%) males out of the 127 (51.0%) were positive for proteinuria while 120 (48.2%) were negative for proteinuria. From this, 237 (95.2%) were negative and 12 (4.8%) were positive for protenuria, with a chi square of 0.32 and p value of 0.6.

The ages of pupils enrolled in this study ranged between 6 and 11 years with a mean age of 8.8±1.6years. The 48 (19.3%) pupils that were between 6 and 7years old were all negative for proteinuria while 8 to 9 year olds had 6 (2.4%) pupils positive for proteinuria and 106 (42.6%) pupils negative. Pupils between the ages of 10 and 11 year olds had 6 (2.4%) positive for proteinuria and 83 (33.3%) pupils negative for proteinuria (p= 0.3).

Table 1. Characteristics of 249 primary school pupils screened for proteinuria in Jos, Nigeria

Variable	Total n(%)	Proteinuria+ve n(%)	Proteinuria -ve n(%)	p
Male	127 (51)	7 (3.8)	120 (48.2)	0.60
Age group (years)				
6-7	48 (19.3)	0 (0.0)	48 (19.3)	0.50
8-9	112 (45)	6 (2.4)	106 (42.6)	
10-11	89 (35.7)	6 (2.4)	83 (33.3)	
Educational status of parents				
None	14 (5.6)	2 (0.8)	12 (4.8)	0.08
Primary	15 (6.0)	2 (0.8)	13 (5.2)	
Secondary	36 (14.5)	1 (0.4)	35 (14.1)	
Tertiary	112 (45.0)	5 (2.00	107 (43.0)	
Postgraduate	72 (28.9)	2 (0.80	70 (28.10	
Source of drinking water				
Borehole	6 (3.4)	1 (0.4)	5 (2.00	0.97
Sachet water	3 (1.2)	0 (0.0)	3 (1.2)	
Tap water	203 (81.5)	10 (4.0)	193 (77.5)	
Well	37 (14.9)	1 (0.4)	36 (14.5)	
Body mass index				
Underweight	7 (2.8)	0 (0.0)	7 (2.8)	0.01
Normal	180 (72.2)	5 (2.0)	175 (70.3)	
Overweight	57 (22.8)	6 (2.4)	51 (20.5)	
Obese	5 (2.0)	1 (0.40	4 (1.6)	

The data in Table 1 also shows the relationship between body mass index and proteinuria. The body mass index was grouped after it was calculated from the height and weight of the pupils into underweight [7 (2.8%) pupils], normal weight [180 (72.3%) pupils], overweight (57 (22.8%) pupils] and obese [5 (2%) pupils]. All 7 (2.8%) underweight pupils were negative for proteinuria, 5 (2%) Pupils with normal weight were positive for proteinuria while 175 (70.3%) pupils were negative. 6 (2.4%) pupils of the overweight group were positive while 51 (20.5%) pupils were negative for proteinuria. The obese category had 1 (0.4%) positive pupil and 4 (1.6%) negative pupils for proteinuria with Chi square of 8.6 and the p value 0.03.5 (2.0%) pupils from parents with tertiary education tested positive for proteinuria while 2 (0.8%) pupil from a parent with primary school level of education was positive for proteinuria. The non educated, primary and post tertiary educated parents had 2 (0.8%), 2 (0.8%) and 2 (0.8%) positive pupils respectively. 70 (28.1%) of pupils who tested negative for proteinuria were from post tertiary educated parents, 107 (43.0%) proteinuria negative pupils were from tertiary educated parents while non-educated parents had 12 (4.8%) pupils testing negative and primary and secondary educated parents had 13 (5.2%) and 35 (14.1%) negative pupils respectively.

Discussion

The participation rate by both parents that consented and pupils that responded after distribution of questionnaires and consent forms was 55.3%. This was lower than the 89% participation rate reported by Onifade and Grange in a similar study in southern Nigeria who worked amongst rural primary school children in Lagos[9]. Our low participation rate reported might have been due to the less consent parents gave the pupils and a decline in the knowledge of the importance of proteinuria screening in the development of renal diseases and lack of personal benefits by either parents or pupils [9].

In our study, the prevalence of proteinuria amongst primary school children was 4.8%. This value was higher than the 0.2% Hajar et al got in Lebanon[10] and the 3.5% obtained in Jos by Akor et al [6]. The high value in our study may have been due to; orthostatic proteinuria as random urine sample was used in our study instead of the usual early morning midstream urine which was used by other researchers. It may have also been due to the high incidence of infectious agents in the study area such as Schistosoma haematobium, Plasmodium species, filarial worms and viral hepatitis each of which may lead to the development of nephritis with high proteinuria levels as reported by Adesola et al in a similar study in Ile-ife, Nigeria[12]. On the other hand, our study recorded a lower prevalence when compared to the 7.5%, 7.7% and 16% by Onifade and Grange[13] who worked in western Nigeria together with Adesola et al[9] and Patil et al[12] who worked in India respectively. Unlike some other studies, in our study more males (2.8%) presented with proteinuria than females (2.0%) though it was not statistically significant p = 0.6. The male female ratio was approximately 1:1. Our findings were consistent with an earlier report from Onifade and Grange in west Nigeria where more males presented with proteinuria than

females although their participants were adults[13]. They attributed the increase incidence in males to the contributions of Schistosomiasis to proteinuria and that schistosome was mostly prevalent in males than in females [13]. Findings from Akor et al [6] were not in line with our study as they reported more females with proteinuria than males[11]. They attributed the higher incidence in females to contamination in the urine from the female genital tract[11]. A recent study in India by Patil et al showed that sex had no relationship with the proteinuria status[15].

Our study suggests that proteinuria was predominant in pupils between the ages of 8 and 11 years (4.8%). This may have been because more pupils between the ages of 8 and 11 years were screened. However, in a similar study in India by Patil et al, proteinuria was predominant in children between the ages of 10 and 13 years which is not far from the 8 to 11 years in our study ans it was also not significant in their study. Patil et al also proposed that age had no relationship on the status of proteinuria in a mass screening programme conducted in India[15].

Our finding that pupils with overweight were more prone to developing proteinuria (2.4%) agrees with previous reports by Toto et al in 2010. He reported that there is an association between body mass index and proteinuria saying there is a graded increase in the magnitude of urine total proteins with respect to increasing body mass index[16]. This he suggested may be due to an alteration in podocyte structure and function, glomerular capillary hypertension and adipodiocytes derived cytokines, which increase glomerular permeability to proteins leading to proteinuria [16],this finding was significant statistically $p = 0.03$.

Our study showed that most pupils with proteinuria (2.0%) came from parents with tertiary education. This may have occurred since most of the enrolled pupils were from parents with tertiary education. This increase in number of tertiary educated parents may be due to the level of awareness concerning education in our environment.

From our findings, only 4.0% of pupils who positive for proteinuria drank tap water. This finding of ours was different from that of Chen et al who reported more proteinuria positive cases from people that drink well water due to arsenic poisoning in well water that increases the incidence of proteinuria in China where his work was done. The variance might be because of higher industrial activities and wastes resulting in higher levels of arsenic compound in China where their study was carried out and the low levels of arsenic compounds in Nigeria where our study was carried out[17].

In conclusion, we found out that the incidence of proteinuria in children of school age in Jos metropolis is on the increase with the males dominating. Further studies are therefore necessary to establish aetiology, risk factors and other factors necessary for the proper diagnosis, treatment and management of proteinuria in children as these children are the future leaders of our great nation. Efforts should also be made by Government to ensure provision of automated reporting results for proteinuria and a law should be passed to make proteinuria testing routine and compulsory to school children. This would enable more parents to consent to screening their wards and children at an early age.

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