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Research paper

Association of environmental exposure to heavy metals with chronic kidney disease (CKD) in patients from White Nile province, Sudan

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Abstract

This research was focused on assessment of chronic kidney disease (CKD), due to the contamination of water, in White Nile Province, Sudan with heavy metals, agrochemicals, hard water, ionicity, and climate changes. Nevertheless, the levels of any of the pollutants or conditions reported in this water are inconsistent not correlated with the prevalence of the disease and are too low to cause (CKD). The most common cause of chronic kidney disease (CKD) among all groups was diabetes mellitus (46.50%); followed by hypertension (46.50%); chronic glomerulonephritis (7.64), while other causes account for only (9.55%). The highest prevalence of CKD was recorded in Eld – Duiem, followed by Allahamed localities, with a percentage of (22.9% and 17.8%), respectively. The highest prevalence of the cases was recorded in patients of the range age of 31-50 years, followed by patients from the group 51-70 years old, and least in the group of 71-90 years old. Famers recorded a frequency of (20.38%) and sponsors recorded a frequency of (17.20%), while, among groups of other occupation a high frequency of (42%) was recorded. The level of education reflected a high frequency of CKD among the individuals of primary education, followed by individuals of secondary education, while the lowest frequency was recorded among illiterates. People living in red brick building, recorded the highest number of CKD patients (142 patients) while only (5 patients) who live in wool building were recorded. On the other hand, high prevalence of CKD (113 patients) was recorded in groups who do not take herbs, while the more patients (44) were recorded among individuals who take herbs. All results showed significance of ($P \leq 0.05$).

Keywords: *Algae Toxins; Anthropogenic Activities; Drinking-Water Supplies; Functional Geographical Distribution; Glomerular Tissue.*

1. Introduction

Chronic kidney disease is typically defined by a progressive loss of functional glomerular tissue followed by proteinuria and loss of glomerular filter function, but, in contrast, there is no signature feature that characterizes CKDu. The pollution of water and food through human waste and anthropogenic activities, including industrial waste and agricultural runoff, is a mounting problem worldwide. Water pollution from microbes causes identifiable diarrheal illnesses. The consumption of water contaminated with heavy metals, fluoride, and other toxins causes insidious illnesses that lead to protracted, non-communicable diseases and death. Chronic kidney disease of unusual/uncertain/unknown etiology is one such example, began to manifest in the mid-1960s in several dry-zonal agricultural societies in developing economies that are located around the equator. Several potential causes have been postulated, including heavy metals, fluoride, cyanobacterial and algae toxins, agrochemicals, and high salinity and ionicity in water, but no specific source or causative factor has been identified for CKD of multifactorial origin (CKDmfo). Three large studies conducted in the recent past failed to find any of the postulated components (heavy metals, cyanobacterial toxins, fluoride, salinity, or agrochemicals) at levels higher than those deemed safe by the World Health Organization and the US Environmental Protection Agency. At the reported low levels in water and with the heterogeneous geographical distribution, it is unrealistic to expect any of these components individually could cause this disease. However, the additive or synergistic effects of a combination of factors and components, even at lower exposure levels, together with malnutrition and harmful behaviors, and/or a yet unidentified (or not investigated) toxin, can cause this epidemic high on biomass energy sources, and congestions are evident for many of the social services when they exist. Health problems comprise child infection with cough, fever, diarrhea and vomiting, and malaria in adolescents while bilharzias infection is reported. State's support to all social services is imperative for food-security improvement, with priority given to Algabalain, Alsalam and Tandalti in health and education services, Rabak, Algabalain, Alsalam, Kost and Alduim localities in clean water provision, and the State at large for provision of non-biomass energy sources and sanitation services [14]. Both the incidence and prevalence of chronic renal failure have risen constantly over the last 3 decades, which is now a growing public health problem. Identifying risk factors associated with the disease is essential in order to prevent it from affecting even more patients. Studying the toxic effects of heavy metals on the human body has become especially important in the last

50 years, given that large amounts of these products were disposed of as industrial waste and they are not biodegradable, remaining in the environment for long periods of time. For this reason, despite the fact that strict regulations enforced mainly in Europe and North America limit the disposal of heavy metals, high levels of these elements are still present in soil and sediment, resulting in chronic exposure in the general population. Heavy metals are a poorly defined group of elements. Some are necessary for the human body, such as iron (Fe), cobalt (Co), copper (Cu), manganese (Mn), molybdenum (Mb) and zinc (Zn). It is unknown whether the other metals – lead (Pb), cadmium (Cd) and arsenic (As) – serve any purpose in the body, but they do have a direct effect on the kidneys and they are particularly nephrotoxic, even at "normal" levels. There is no clear evidence of nephrotoxicity due to other metals such as uranium and mercury [8]. Intestinal absorption of divalent metals such as Cd and Pb is facilitated by divalent metal transporter 1 (DMT-1). DMT-1 is located in the duodenum, erythrocytes, liver and cells in the proximal convoluted tubule (PCT). This protein transports Fe and has a high affinity for other divalent metals such as Cd, Ni (nickel), Pb, Co, Mn, Zn and Cu [9]. Decreased intake of Fe and Zn results in increased expression of DMT-1, which increases intestinal absorption of Cd and Pb and therefore toxicity by these metals [1]. Experiments in cell lines in which DMT-1 expression has been blocked suggest that there is a different Pb transporter [5]. Heavy metals are metabolised in the liver, where they bind to low molecular weight proteins (<10kDa) called metallothioneins (MT). These proteins are widely distributed throughout the body and contain a large quantity of the amino acid cysteine, which gives them a high affinity for reacting with and storing metals such as Zn, Cd, Hg (mercury), Cu, Pb, Ni, Co and Fe. The main function of MT is to store essential metals such as Zn and Cu in the intracellular medium and transfer them to metalloproteins, transcription factors and enzymes. MT also play a role in the elimination of free radicals and in cellular repair and regeneration processes [7]. Increased intracellular levels of Cd and Pb increases MT expression, and MT knockout mice are more susceptible to toxicity from these metals [18]. A previous study showed that Lead (Pb), mercury (Hg), and cadmium (Cd) are common heavy metal toxins and cause toxicological renal effects at high levels, but the relevance of low-level environmental exposures in the general population is controversial. A total of 1,797 adults who participated in the KNHANES (a cross-sectional nationally representative survey in Korea) were examined, and 128 of them (7.1%) had chronic kidney disease (CKD). Our study assessed the association between Pb, Hg, Cd exposure, and CKD. Blood Pb and Cd levels were correlated with CKD in univariate logistic regression model. However, these environmental heavy metals were not associated with CKD after adjustment for age, sex, BMI, smoking, hyperlipidemia, hypertension, diabetes, and these metals in multivariate logistic regression models. We stratified the analysis according to hypertension or diabetes. In the adults with hypertension or diabetes, CKD had a significant association with elevated blood Cd after adjustment, but no association was present with blood Pb and Hg. The corresponding odds ratio [OR] of Cd for CKD were 1.52 (95% confidence interval [CI], 1.05-2.19, P=0.026) in adults with hypertension and 1.92 (95% CI, 1.14-3.25, P=0.014) in adults with diabetes. Environmental low level of Pb, Hg, Cd exposure in the general population was not associated with CKD. However, Cd exposure was associated with CKD, especially in adults with hypertension or diabetes. This finding suggests that environmental low Cd exposure may be a contributor to the risk of CKD in adults with hypertension or diabetes [12]. Although environmental and consumer regulations have reduced the overall levels of heavy metal contamination over the last several decades, low levels of exposure remain widespread. In addition to lead paint that remains common in older homes, the aging infrastructure of water systems throughout the United States confers daily exposure to lead for many individuals [3]. Similarly, cadmium is in many products, including batteries, pigments, metal coatings, plastics, and cigarette smoke, and exposure occurs through ingestion of contaminated foodstuffs or inhalation [2]. Although low levels of exposure may be of less clinical significance for healthy adults, children are particularly susceptible, in part due to higher proportions absorbed across the gastrointestinal tract and rapid cell metabolism. Whether CKD may confer a similar heightened susceptibility is largely unknown. Studies demonstrating an association between lead [13] and cadmium [11] and kidney disease have primarily been interpreted as causative due to the toxic effect on proximal tubular function. However, the possibility of reverse causality, whereby impaired renal function results in higher circulating blood levels and thus greater long-term toxicity to other organs, has not been fully clarified. Furthermore, because heavy metal toxicity disproportionately affects minoritized racial and ethnic populations, who simultaneously have higher rates of CKD, understanding the hazard from environmental toxins is of public health urgency.

2. Materials and methods

2.1. Study area

This study was carried out during the period (April 2016 - October 2020) within Eld – Duiem locality in the White Nile State, Sudan. The State is home to significant numbers of refugees of about (153,000 in 2006) forming nearly 10% of the population; and also, a transit point for Internally displaced people s(IDPs) returning to the south and south of Kordofan. Most of these Internally displaced peoples (IDPs) have been settled in many camps.

2.2. Study population

The population of this study area was of different multi - ethical groups. A total of 157 patients (77 males and 80 females aged, 10 years to 80 years), with early stage (preserved renal function), were randomly selected from the outpatient clinic in the hospital. Another 157 healthy persons (77 males and 80 females, ages 19 to 80 years) were selected as control. Weight and height were recorded for each individual and body mass index (BMI) was calculated (kg/m^2). Inclusion criteria for case selection included confirmed renal failure and signing the inform consent. Exclusion criteria for case selection included patients with: Macro-albuminuria, congestive cardiac failure, diabetic nephropathy ketonuria, urinary tract infection and pregnant women.

2.3. Demography data

Data on demographic information, lifestyle risk factors, and personal medical history of patients were gathered by designed standard questionnaires and recorded by trained staff. Information included the following:

2.3.1. Patient's age, sex and time of onset and duration of renal failure

2.3.2. Total duration of renal failure, the drugs the patient was taking, the dosages and the regularity of the treatment

2.3.3. All details regarding the presenting complaints

2.4. Research ethics

The proposal of this study was approved by the Research Committee in the Faculty of Medicine and Health Science, University of Al-butana. Verbal informed consent was obtained from all study subjects (3.7.20016).

2.5. Collection and analysis of water samples

2.5.1. Water sampling

Water was pumped from bore holes for 2 minutes before obtaining 500 ml sample for analysis. Water from the dams was sampled 10-15 cm below the water surface using labeled acid washed plastic containers to avoid unpredictable changes in characteristic as per standard procedures of [16]. Samples were labeled according to the source and the region from they were obtained. The water was acidified with 2 ml of analytical grade nitric acid in order to preserve metals and avoid precipitation [6]. The samples were stored at a 5ºC temperature awaiting the transportation to the laboratory for analysis.

2.5.2. Water digestion and analysis

A volume of 100 ml of the sample was measured using a 100 ml volumetric flask and put in a conical flask with 5 ml of concentrated nitric acid. The mixture was heated slowly on a hot plate and evaporated to about 20 ml ensuring that the water did not boil. A further 5 ml of concentrated nitric acid was added and the beaker was covered with a watch glass while heating continued. Nitric acid continued to be added until the solution appeared light colored and clear. Another 2 ml of concentrated hydrochloric acid was added and heated slightly to dissolve any remaining residue. Few drops of hydrogen peroxide were also added to ensure complete digestion. The solution was filtered and the filtrate was transferred to a 100 ml volumetric flask and made up to the mark with distilled water [10].

2.6. Methods

Water regimented or treated to make it potable the house and main source of cooking fuel and different cooking techniques: (1) soaking, (2) pressure cooking, and (3) normal cooking. Mineral content was analyzed using flame photometry and nitro-vanado-molybdate colorimetry.

2.6.1. Collection and analysis of soil samples

2.6.1.1. Collection of soil samples

Sampling sites were chosen in line with anthropogenic sources of heavy metals. Samples were taken from Abo Qroon, Alhduab, Al-Dawim, Al-Quitam, Al-Quitaina, allahamd, Kosti, Rabak, shabasha, the mandarib, and Arshul Mountain. Twenty-four soil samples (three replicates) were collected from the surface from various locations to cover the industrial area in the White Nile state, and 10 meters to1km far from the bank the White Nile for areas that are covered by the flood. At each sampling point, approximately 0.5 kg of soil was collected from 0-10 cm depth using a stainless-steel sampler. Soil samples along the roads were collected at a distance of one meter away from the road and within an area of one square meter. Three samples were collected from each point, thoroughly mixed in a clean plastic container. To obtain a representative each sample was dried, crushed and sieved with 2 mm mesh before being stored in a labeled polythene bag prior to analysis. The soil samples were labeled according to the regions from which they were obtained.

2.6.1.1.1. Soil digestion and analysis

One gram of each well mixed sample was put into 250 ml glass beaker and digested with 24 ml of nitric and hydrochloric acids and then evaporated to near dryness. The soil samples were then dissolved in 10 ml of 2% nitric acid, filtered and then diluted to 100 ml with distilled water according to [15]. One gram of randomly selected soil powder was spiked with three different concentrations of heavy metals one at a time (1.0, 1.5, 2.0 ppm) each run in with the ICP9000 machine. Stock solution for each of the heavy metals Lead Zinc Manganese Cadmium and Chromium was prepared and used to generate a calibration curve for each metal, before the measurement of the concentration in the soil and water samples. The digestion method and atomic absorption spectroscopy analysis were validated by recovery method. One gram of randomly selected soil powder was spiked with three different concentrations of heavy metals one at a time (1.0, 1.5, 2.0 p.p.m.) each run in with the ICP9000 machine. This was followed by the digestion of the spiked samples and determination of metal concentration using ICP9000. Blank or unspiked samples were digested through the same process and analyzed by same AAS. The amount that was recovered after digestion of the spiked samples was used to calculate % recovery [17]. Buck scientific (210 VGF) ICP-MS machine was used for analysis. Its parameters were set according to the specifications given in the manufactures manual including lamp current and fuel system of air/acetylene flame. The AAS machine had a picking meter that indicated when the optimum conditions had been realized. Its optimization was automatic.

3. Statistical analysis

Values were presented at mean ± Standard deviation (SD) in mg of all measure variables. Students t-test and one-way ANOVA with Dunnett' spots test were performed using Graph Pad Prism version 6 for windows, Graph pad software, San Diego California USA, www. Graphpad.com. p values ≤ 0.05 were considered significant.

4. Results

Table 1: The Frequency of Renal Failure (CKD) Recorded Among Patients from Different Localities

Locality	Frequency	Percent (%)	Valid Percent	Cumulative Percent
Arshul Mountain	10	6.4	6.4	72.0
Shabasha	10	6.4	6.4	92.4
Ahamonta	11	7.0	7.0	14.6
Kosti	11	7.0	7.0	79.0
Rabak	11	7.0	7.0	86.0
Abo Qroon	12	7.6	7.6	7.6
Al-Quitana	16	10.2	10.2	47.8
Allahamd	28	17.8	17.8	65.6
Al-Dawim	36	22.9	22.9	37.6
Total	157	100.0	100.0	603

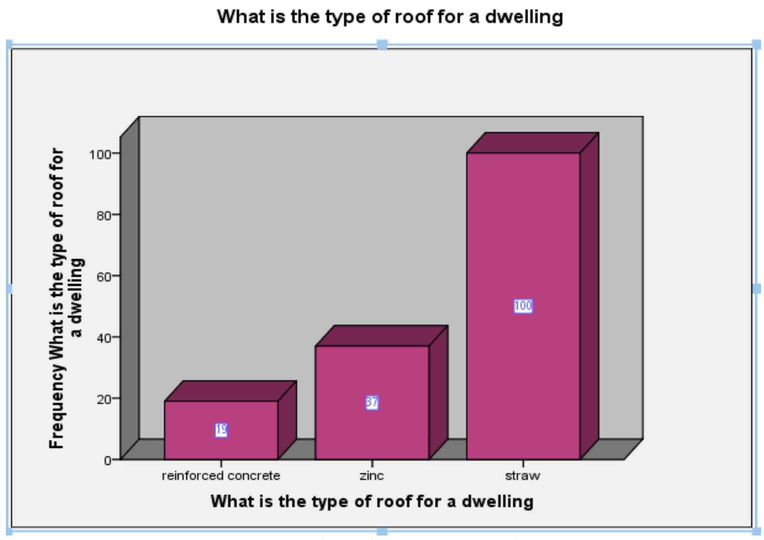

Fig. 1: Diagrammatic Representation Showing the Frequency (%) of Roof Material Used for A Dwelling Recorded Among Patients Most Using Straw Roof (100%).

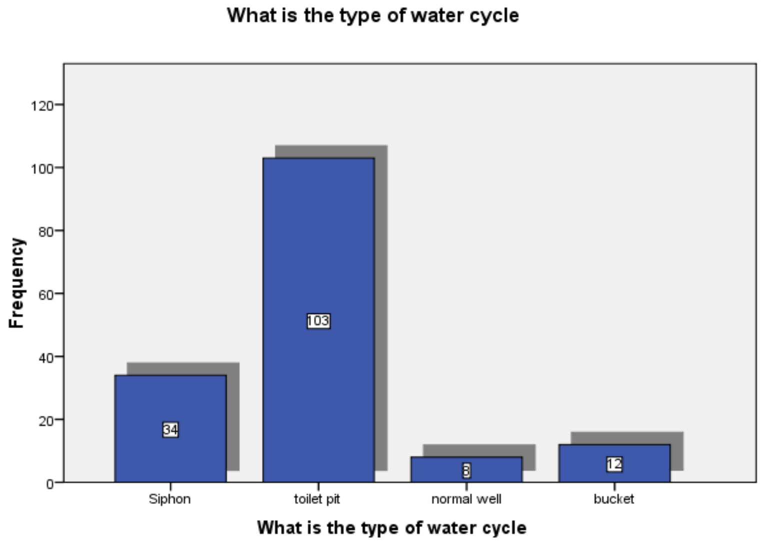

Fig. 2: Diagrammatic Representation Showing the Frequency (%) of Renal Failure (CKD) Recorded Among Patients Using Different Type of Water Cycle.

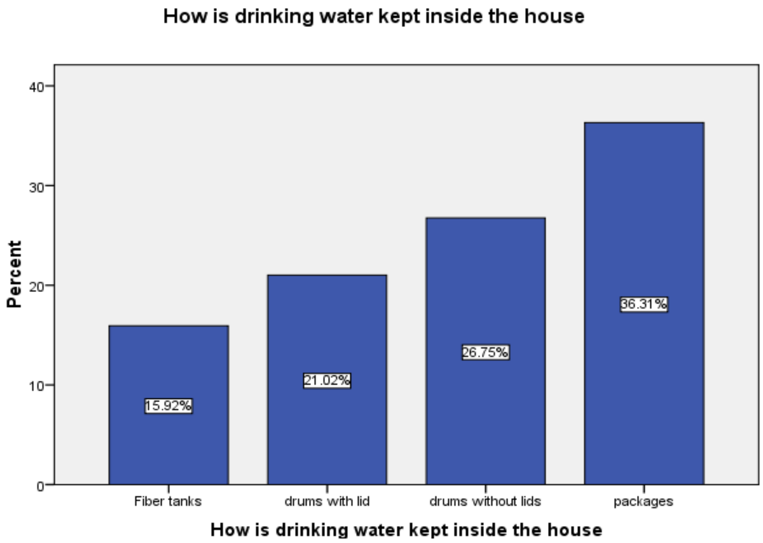

Fig. 3: Diagrammatic Representation Showing the Frequency (%) of Renal Failure (CKD) Recorded Among Patients in Accordance on How Drinking Water Is Kept Inside the House.

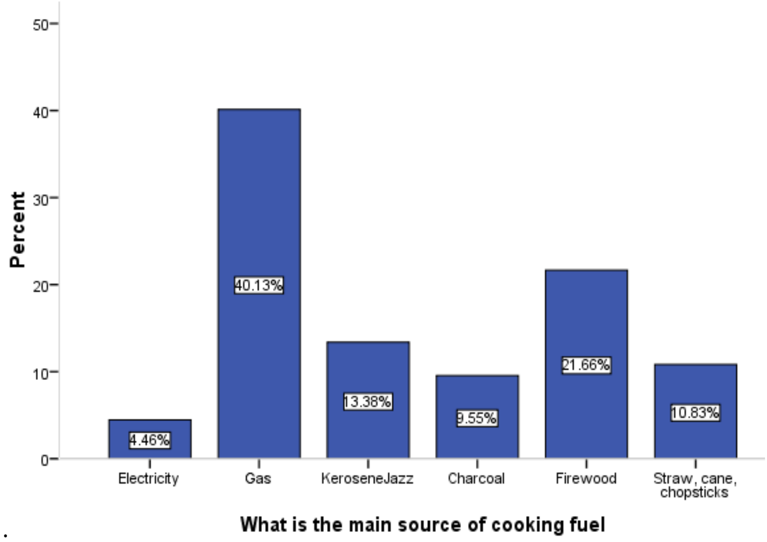

Fig. 4: Diagrammatic Representation Showing the Frequency (%) of Renal Failure (CKD) Recorded Among Patients in Accordance to the Main Source of Cooking Fuel.

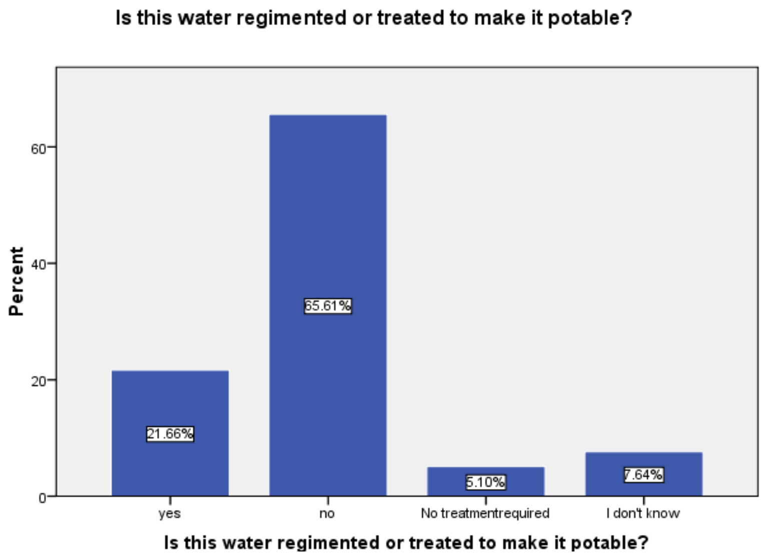

Fig. 5: Diagrammatic Representation Showing the Frequency (%) of Renal Failure (CKD) Recorded Among Patients in Accordance to the Procedure for Making Water Potable; Regimented Or Treated.

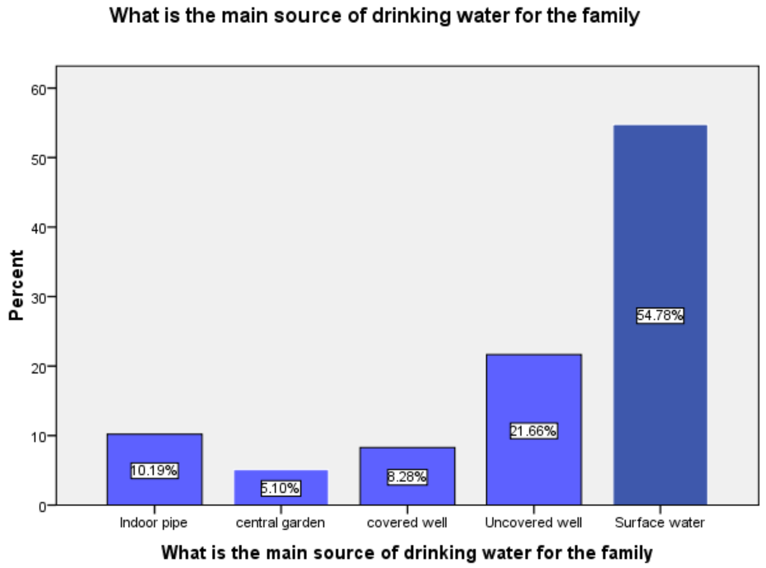

Fig. 6: Diagrammatic Representation Showing the Frequency (%) of Renal Failure (CKD) Recorded Among Patients in Accordance to the Main Source of Family Drinking Water.

Fig. 7: Diagrammatic Representation Showing the Frequency (%) of Renal Failure (CKD) Recorded Among Patients in Accordance to the Place Where Food Cooked for Family Members.

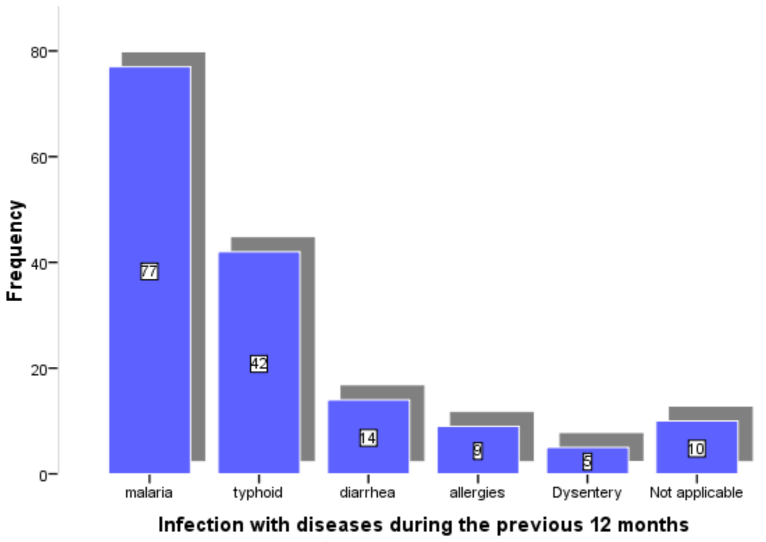

Fig. 8: Diagrammatic Representation Showing the Frequency (%) of Renal Failure (CKD) Recorded Among Patient Infected with Disease During the Previous 12 Month.

5. Discussions

This study provides new estimates of chronic kidney disease (CKD) prevalence – including CKD of unknown etiology (CKDu) – across nine districts most affected by CKD in White Nile state, including an examination of rural households and historical reliance on groundwater consumption. A carefully designed household survey provides information on whether these households self-reported having a member in the decade prior to 2016, who had been clinically diagnosed with CKD. Households were classified according to whether or not they had used groundwater (from household wells, agro-wells or springs) as their primary source for drinking or cooking for at least five years between 1999 and 2018. More than 98% of households reported having consumed groundwater as their primary source of drinking or cooking water for at least five of those years and >15% of households reported having at least one CKD-affected member in the ten-year period up to 2018, but these numbers varied across and within various districts. Table 1, showed that the frequency of renal failure (CKD) recorded among patients from different localities was increased successively as follows; Al-Dawim, Allahamd, Al-Quitana, The mandarib, Abo Qroon, Rabak, Kosti, Ahamonta, Shabasha, then finally Arshul Mountain. The reported characteristics of symptomatic individuals revealed that the incidence of CKD was significantly higher among females (62%) than males (38%), roof for a dwelling recorded among patients most using straw roof (100%) Fig.1. About (103%) of pateints using toilet pit the different of type water cycle Fig. 2. About (36.31%) packages drinking water kept inside the house Fig. 3. About (41.13%) using Gas the main source of cooking fuel Fig. 4. (65.61%) select the option no of regimented water or treated to make it potable Fig. 5. About (54.78%) surface water the main source of drinking for family show in Fig. 6. These findings in water pollution goes together with the contamination of the food chain; thus, the two cannot be separated. Once the chemicals and toxins get into the human food chain, long-term disastrous health consequences are inevitable, and a negative vicious cycle is established. One of the goals should be to break this negative vicious cycle and re-establish the healthy environment, food-chain and health [4], about (82%) traditional kitchen (tackle) where is food cooked for family members show in Fig. 7. about (77%) of patients infected with malaria during the previous 12 month. About the (46%) of patients are fair during the previous 12 month show in Fig. 8. The current study strongly supports the hypothesis that CKDu in Sri Lanka is a drinking-water-related disease in farmers who have a history of spraying glyphosate. These data illustrate the scale of CKD in the most-affected districts of White Nile State on an aggregate basis as well as revealing differences across districts and at the sub-district level.

6. Conclusions

Research has focused on the contamination of water with heavy metals, agrochemicals, hard water, ionicity, climate change, and so forth. Nevertheless, the levels of any of the pollutants or conditions reported in water in White Nile State are inconsistent not correlated with the prevalence of the disease, and are too low to be the sole cause of CKD-mfo. Meanwhile, several nephrotoxins prevalent in the region, including medications, leptospirosis, toxic herbs, locally grown tobacco, and petrochemicals, as well as the effects of changed habits occurred over the past four decades have not been studied to date. Taken together, the geographical distribution and overall findings indicate that combinations of factors and/or their interactions are likely to precipitate CKD-mfo, which kills more than 1,000 people annually in Sudan; most victims are middle-aged male farmers. The current epidemic of CKD-mfo is causing a large-scale human suffering and economic hardship to victims and their families, similar to the harm done by other common diseases induced by environmental factors, such as gas; the main source of cooking fuel , infection with malaria during the previous 12 month, using straw roof, and exposure to asbestos. The solution does not lie in the expansion of dialysis centers but in promptly intervening with effective programs to prevent this disease. When one person in a family is chronically ill, the entire family is burdened; the effects are particularly devastating for economically deprived families, which include the vast majority of families in Sudan and other affected regions in the country. Water pollution goes together with the contamination of the food chain; sedimented-groundwater (from household wells, agro-wells or springs) as the primary source for drinking or cooking for at least five districts. Thus, the two cannot be separated. Once the chemicals and toxins get into the human food chain, long-term disastrous health consequences are inevitable of causes renal disease, and a negative vicious cycle is established. One of the goals should be to break this negative vicious cycle and re-establish the healthy environment.

7. Recommendations

Provision of clean potable water, broader education, prevention of pollution, and long-term environmental preservation efforts are the ways ahead to decrease the incidence of CKD. Creating multiple Task Forces would not have any meaningful impact on preventing and eliminating this deadly disease from White Nile State. Other recommendations include; increasing the requirements of prevention of the escalating incidence of CKD in White Nile State and other affected countries. Activating governmental dedication and commitment efforts. The establishment and funding of a new entity, "CKD-Alleviation Authority," which needs to be fully independent of ministries and government departments. The introduction and enforcement of legislation on pollution prevention and control. Development and incorporation of sustainable agricultural methods. The provision of safe and clean drinking water and safe sanitary facilities to all communities.

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