



## Impact of Hardness, Fluoride and Nephrotoxic Trace Metal Contaminants in Drinking Water Sources on CKDu Progression in Uva Province, Sri Lanka

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### Abstract

Chronic Kidney Disease with unknown etiology (CKDu) has impacted adversely the health sector as well as the socioeconomic sector in Sri Lanka. During the past two decades, CKDu escalated to dry and intermediate zone in Uva Province at an alarming rate. Therefore, this study aimed to investigate the impact of physicochemical parameters including nephrotoxic trace elements on CKDu progression in Uva province. A questionnaire-based survey was conducted to study the demographic and other lifestyle-related factors of CKDu patients in the identified prevalent areas. Along with that, water samples were collected from the drinking water sources of the relevant patients. Water samples were analyzed for physicochemical parameters i.e.; pH, Electrical Conductivity (EC), Total Hardness (TH), Nitrate, Fluoride (F) and heavy metals following standard protocols. Statistical analysis was conducted by IBM SPSS software version 20. The survey data confirmed that CKDu is prominent within the male population who engage in agricultural activities as an occupation and whose drinking water source is shallow-dug wells. Out of all the DS areas, the highest mean TH and Mg<sup>+2</sup> ion content were recorded in TDS and WDS areas. The highest F content was recorded in the TDS area. Correlation data of F content revealed a significant positive relationship ( $p < 0.05$ ,  $r^2 = 0.69$ ) with Mg<sup>+2</sup> ion content whereas nephrotoxic heavy metals showed a negative correlation with F content in water. The results suggest that the increased hardness along with high F content in drinking water may positively affect the progression of CKDu in Uva province.

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### Introduction

Chronic Kidney Disease of unknown etiology/origin (CKDu) is a kind of nephropathy that emerged in the mid-'90s in several tropical to sub-tropical regional countries like Nicaragua, Andhra Pradesh in India, El Salvador, Egypt, Costa Rica and North Central Province (NCP) in Sri Lanka [1,2]. It has escalated progressively over the past two decades within low socioeconomic paddy farming communities in Sri Lanka. It is demarcated into specific geological areas of the country, especially dry and intermediate zone. This disease is identified in individuals in later stages when experiencing a kidney impairment in proximal tubules and the interstitium which slowly progresses to renal failure and most importantly the disease is asymptomatic in its early stages [2]. The CKDu was first reported by NCP and over the past two decades, it has been escalating and prominent its prevalence over Uva Province and some other nearby districts at an alarming rate [3].

Due to the characteristics of geological areas where the CKDu exists, most of the hypotheses for the causative agent turned toward environmental factors. Although nephrotoxic contaminants in drinking water, are often considered a major factor that causes CKDu, the root cause is still unrevealed. The study describes in this paper investigates the impact of physicochemical parameters including nephrotoxic heavy metals on CKDu in Uva Province, Sri Lanka.

### Methodology

#### Site selection

Three Divisional Secretariats (DS) administrative areas were selected for the study that has been identified with the highest number of CKDu occurrence from both the districts in Uva Province, namely; Mahiyangana DS (MDS), Rideemaliyadda DS (RDS), Kandaketiya DS (KDS) from Badulla district and Wellawaya DS (WDS), Thanamalwila DS (TDS), Siyambalanda DS (SDS) from Monaragala district.

**Table 1:** Physicochemical properties of water samples in CKDU endemic areas in Uva Province.

| Parameter               | Mahiyangana<br>Mean ± SD     | Rideemaliyadda<br>Mean ± SD  | Kandaketiya<br>Mean ± SD     | Wellawaya<br>Mean ± SD       | Thanamalwila<br>Mean ± SD    | Siyamblanduwa<br>Mean ± SD   |
|-------------------------|------------------------------|------------------------------|------------------------------|------------------------------|------------------------------|------------------------------|
| pH                      | 7.43 ± 0.28 <sup>a</sup>     | 7.87 ± 0.51 <sup>a</sup>     | 6.94 ± 0.37 <sup>a</sup>     | 7.18 ± 0.63 <sup>a</sup>     | 7.35 ± 0.43 <sup>a</sup>     | 7.38 ± 0.26 <sup>a</sup>     |
| EC (µS/cm)              | 252.18 ± 93.14 <sup>b</sup>  | 378.22 ± 177.97 <sup>a</sup> | 390.77 ± 153.80 <sup>a</sup> | 341.12 ± 184.24 <sup>a</sup> | 386.72 ± 168.68 <sup>a</sup> | 236.92 ± 90.19 <sup>b</sup>  |
| TH (mg/L)               | 188.04 ± 143.86 <sup>b</sup> | 145.22 ± 79.04 <sup>b</sup>  | 156.07 ± 66.16 <sup>b</sup>  | 277.32 ± 121.57 <sup>a</sup> | 197.76 ± 107.73 <sup>b</sup> | 79.89 ± 70.49 <sup>c</sup>   |
| Mg <sup>2+</sup> (mg/L) | 11.69 ± 10.25 <sup>b</sup>   | 8.74 ± 10.42 <sup>c</sup>    | 12.74 ± 4.29 <sup>b</sup>    | 27.21 ± 18.97 <sup>a</sup>   | 30.47 ± 18.54 <sup>a</sup>   | 8.52 ± 7.43 <sup>c</sup>     |
| F (mg/L)                | 1.34 ± 0.44 <sup>b</sup>     | 0.32 ± 0.23 <sup>c</sup>     | 0.35 ± 0.34 <sup>c</sup>     | 1.14 ± 1.44 <sup>b</sup>     | 2.09 ± 2.35 <sup>a</sup>     | 0.69 ± 0.73 <sup>c</sup>     |
| Cr (µg/L)               | 0.36 ± 0.88 <sup>b</sup>     | 0.45 ± 0.51 <sup>b</sup>     | 0.48 ± 0.45 <sup>b</sup>     | 0.48 ± 0.29 <sup>b</sup>     | 0.80 ± 0.28 <sup>a</sup>     | 0.49 ± 0.42 <sup>b</sup>     |
| Mn (µg/L)               | 125.09 ± 332.46 <sup>a</sup> | 115.33 ± 383.53 <sup>a</sup> | 11.33 ± 21.68 <sup>c</sup>   | 7.55 ± 12.15 <sup>c</sup>    | 1.81 ± 2.64 <sup>d</sup>     | 62.69 ± 89.91 <sup>b</sup>   |
| Fe (µg/L)               | 39.94 ± 61.28 <sup>b</sup>   | 29.63 ± 22.93 <sup>b</sup>   | 34.05 ± 27.37 <sup>b</sup>   | 40.07 ± 30.62 <sup>b</sup>   | 41.92 ± 34.31 <sup>b</sup>   | 103.93 ± 256.51 <sup>a</sup> |
| Co (µg/L)               | 0.54 ± 1.10 <sup>a</sup>     | 0.27 ± 0.55 <sup>a</sup>     | 0.35 ± 0.42 <sup>a</sup>     | 0.20 ± 0.10 <sup>a</sup>     | 0.36 ± 0.23 <sup>a</sup>     | 0.29 ± 0.49 <sup>a</sup>     |
| Ni (µg/L)               | 1.66 ± 3.63 <sup>a</sup>     | 1.30 ± 1.25 <sup>a</sup>     | 1.27 ± 0.74 <sup>a</sup>     | 0.82 ± 0.66 <sup>a</sup>     | 1.52 ± 1.16 <sup>a</sup>     | 0.76 ± 0.80 <sup>a</sup>     |
| Cu (µg/L)               | 1.63 ± 1.89 <sup>b</sup>     | 0.83 ± 0.89 <sup>c</sup>     | 1.98 ± 2.87 <sup>a</sup>     | 1.40 ± 2.08 <sup>b</sup>     | 2.83 ± 6.06 <sup>a</sup>     | 0.91 ± 0.42 <sup>c</sup>     |
| Zn (µg/L)               | 42.11 ± 72.38 <sup>a</sup>   | 12.47 ± 11.93 <sup>b</sup>   | 39.67 ± 67.45 <sup>a</sup>   | 12.48 ± 8.62 <sup>b</sup>    | 7.81 ± 9.45 <sup>b</sup>     | 10.56 ± 16.55 <sup>b</sup>   |
| As (µg/L)               | 0.37 ± 0.30 <sup>c</sup>     | 0.23 ± 0.15 <sup>c</sup>     | 1.12 ± 1.06 <sup>a</sup>     | 0.55 ± 0.70 <sup>b</sup>     | 0.61 ± 1.02 <sup>b</sup>     | 0.27 ± 0.19 <sup>c</sup>     |
| Cd (µg/L)               | 0.61 ± 0.40 <sup>a</sup>     | 0.13 ± 0.13 <sup>b</sup>     | 0.65 ± 0.56 <sup>a</sup>     | 0.25 ± 0.21 <sup>b</sup>     | 0.07 ± 0.04 <sup>c</sup>     | 0.06 ± 0.07 <sup>c</sup>     |
| Ba (µg/L)               | 79.60 ± 56.65 <sup>b</sup>   | 40.19 ± 25.37 <sup>c</sup>   | 66.42 ± 33.56 <sup>b</sup>   | 116.05 ± 69.47 <sup>a</sup>  | 78.10 ± 36.64 <sup>b</sup>   | 60.32 ± 56.38 <sup>b</sup>   |
| Pb (µg/L)               | 1.55 ± 3.04 <sup>a</sup>     | 0.29 ± 0.24 <sup>c</sup>     | 0.84 ± 0.68 <sup>b</sup>     | 0.32 ± 0.18 <sup>c</sup>     | 0.26 ± 0.14 <sup>c</sup>     | 0.36 ± 0.12 <sup>c</sup>     |

All values are means of triplicate determination ± SD

Different letters in the same column showed statistical difference according to Tukey's test at  $p < 0.05$

A total of 114 CKDU patients were selected for the survey, who resides in the selected DS areas from both districts in Uva province. The selected patients were above 18 years old and residents in the same area for more than 10 years. Demographic data of households were collected through a questionnaire. Data related to the drinking water sources, previous records of having hypertension, disease conditions like diabetes mellitus, snake bites, use of agrochemicals in day-to-day life, and daily water consumption details were also recorded.

### Water sampling and analytical methods

Water samples were collected from 145 water resources that are being used for consumption (either drinking or cooking) and irrigation purpose by respondents including dug wells (n=138), and tube wells (n=7). Water samples were directly filtered into sterile polyethylene vials using microfiltration (pore size - 0.22 µm) and followed by acidification with high purity 10% HNO<sub>3</sub>. Then samples were stored and transferred under cold conditions (<7°C) for analysis of trace elements including As, Cd, and Pb which are known as nephrotoxic heavy metals.

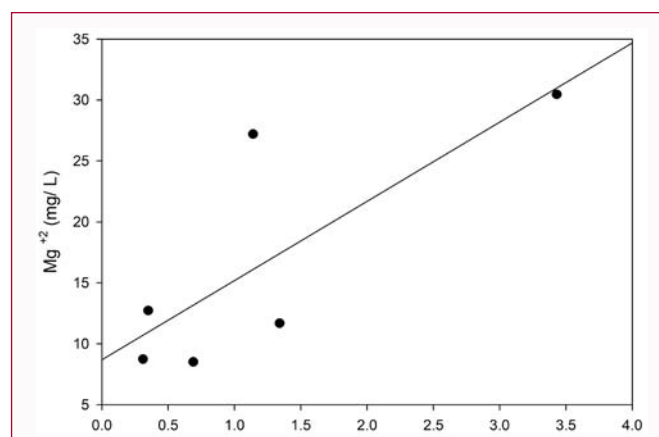
Parameters i.e.; pH, Electrical Conductivity (EC), and Dissolved Oxygen (DO) were measured on-site using a multi-parameter (HACH, Loveland, Colorado, USA). The Total Hardness (TH) and Fluoride (F) content of water samples were measured by EDTA titrimetric method (APHA -234°C) and SPANDS method using a UV visible Spectrophotometer (HACH DR 2700). Trace elements including major heavy metals (As, Cd, and Pb) were analyzed using Inductively Coupled Plasma-Mass Spectrometry (ICP- MS) (Agilent 7900, USA) (APHA, 1998). All the analyses were carried out in triplicated samples and statistical analysis was conducted by IBM SPSS software version 20.

## Results and Discussion

The study population (n=114) represents 79.67% male and 20.33% female respondents and the average age of them was 42.7 (range: 23-78) of years. Out of the male respondents, 92.4% of the

study population in endemic areas were engaged in farming for more than 10 years and they have engaged in applying agrochemicals for their cultivations without any safety precautions.

Drinking water source of the majority of people in all study areas (MDS-87.09%, RDS-75%, KDS-67%, WDS-82.3%, TDS-78.4%, SDS-81.4%) were groundwater (shallow well water). They had been using the same water source for more than 10 years for drinking purposes and the water had not been subjected to purification. The average daily water consumption of the respondents was 5.4 ± 1.7 L. Interestingly, 62% of the endemic area population had used an average amount of 1.2 L of water, that was directly consumed by a small pit dug in the paddy field or the small stream flows nearby the farming field when they get thirsty during the fieldwork. The current study revealed that the CKDU prevalence among males was predominant in Uva Province and 91.7% of them are farmers by occupation. Shallow-dug wells are used to fetch water for consumption by the majority of the people, due to pipe-borne water or purified water facilities being lacking in the rural agricultural areas of the region.



**Figure 1:** Correlation plot of Mg<sup>2+</sup> ion content with a fluoride content of water samples.

Hardness in water is one of the important factors for CKDu, which is attributed to the  $\text{Ca}^{+2}$  and  $\text{Mg}^{+2}$  ions [2]. Higher mean TH,  $\text{Mg}^{+2}$  ion content and F values were recorded in MDS, WDS and TDS areas in Uva province (Table 1). Out of all the DS areas, the highest mean TH and  $\text{Mg}^{+2}$  ion content were recorded in TDS (TH-  $197.76 \pm 107.73$  mg/L,  $\text{Mg}^{+2}$  -  $30.47 \pm 18.54$  mg/L) and WDS (TH-  $277.32 \pm 121.57$  mg/L,  $\text{Mg}^{+2}$  -  $27.21 \pm 18.97$  mg/L) areas whereas the highest F content ( $2.09 \pm 2.35$  mg/L) was recorded in TDS area. Interestingly, water samples collected from all DS areas are classified as hard water according to the SLSI standards. Further, recorded F content in TDS, WDS and MDS areas was higher than the SLSI permissible level (1 mg/L). Correlation data of F content revealed a significant positive relationship ( $p < 0.05$ ,  $r^2 = 0.69$ ) with  $\text{Mg}^{+2}$  ion content (Figure 1) in the study samples. A previous study reported the formation of nephrotoxic  $\text{MgF}^+$  complex in water, due to the high levels of hardness along with the high levels of F ions in water [4].

However, the measured nephrotoxic heavy metal levels in the water samples were far below the permissible levels for drinking water according to SLSI and WHO guidelines. Further, the correlation data of F content with As, Cd and Pb exhibited a negative correlation. The results of the current study aligned with a previous study [5], which express the low level of heavy metals in CKDu endemic areas in NCP. Another study reported a low level of heavy metal content in drinking water in Monaragala district [6].

## Conclusion

The prevalence of CKDu in Uva Province is becoming dominant in the area at a rapid rate. High levels of  $\text{Mg}^{+2}$  ions increased the hardness of drinking well water and the high level of fluoride is

shown to be a major threat to the progression of CKDu in residents. Prolonged consumption of high fluoride and  $\text{Mg}^{+2}$ - containing water along with even minute amounts of heavy metals may adversely affect the scenario. Moreover, further studies need to be carried out with more samples and clinical data to ascertain the cause of the etiology.

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