*Environmental Engineering and Management Journal* August 2020, Vol. 19, No. 8, 1289-1297
http://www.eemj.icpm.tuiasi.ro/; http://www.eemj.eu

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# ASSOCIATION OF NEPHROLITHIASIS WITH DRINKING WATER QUALITY AND DIET IN PAKISTAN

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## Abstract

Bad water quality has serious implications for human health, which may include gastrointestinal, liver, respiratory diseases and even cancer. In view of drinking water hardness in the study area, it was hypothesized that bad water quality has some association with the renal disorders. The possible association between nephrolithiasis, drinking water quality and diet in human subjects in district Abbottabad, Pakistan was investigated. Drinking water samples were collected from different areas (total number, n=100) and analyzed for various physico-chemical properties like turbidity, electrical conductivity, and pH by using standard methods. The concentration of total hardness ranged from 250 $mgL^{-1}$ to 800 $mgL^{-1}$ which exceeded the standard value of 60 $mgL^{-1}$ and the water was found to contain high content of calcium ions. Kidney stones analysis reports of affected subjects (n=100) were examined for the type of stone. The calculi collected showed various shapes and were composed of calcium oxalate (88%), struvite (4%), uric acid (7%) and mix of calcium oxalate and uric acid (1%). Oxalate rich diet was another important risk factor for nephrolithiasis. The patients were consuming 100-300 mg $day^{-1}$ of dietary oxalate. In conclusion, the quality of drinking water and diet habits contributed to nephrolithiasis.

*Key words:* human diet, nephrolithiasis, water hardness, water quality

*Received: November, 2019; Revised final: March, 2020; Accepted: April, 2020; Published in final edited form: August, 2020*

## 1. Introduction

Goal six of sustainable development goals emphasizes the need for clean and accessible water for nations of the world. It is a well-established fact now that poor water and sanitation, its shortage and low quality have serious impacts on economics and livelihood of various human societies around the globe. According to UNO (2018), almost one fourth of the world population have not full access to freshwater resources and "by 2050 at least one in four people is likely to live in a country affected by chronic or recurring shortages of fresh water". The poor drinking water quality poses significant effects on human health not only in Pakistan, but also in many other developing nations (Abbasi et al., 2019). According to WHO (2018), about 2 billion people are drinking water containing fecal contamination worldwide. Poor drinking water quality with imbalanced ionic strength has resulted in many diseases. Various studies pointed out the presence of contaminants which may result in infectious diseases

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(Cheng et al., 2017; Levallois and Villanueva, 2019; Wasana et al., 2017).

Access to clean drinking water is increasingly becoming difficult in many parts of the globe since last twenty five years. In developing countries, around sixty percent population is drinking contaminated drinking water (Aslam et al., 2016; Pandit and Kumar, 2015). At present, economic challenges in many developing countries are compelling to adopt economically viable and sustainable solutions for drinking water and wastewater treatment. As major bottle necks in adopting sophisticated technologies are high energy input, operational cost, and affordability (Mahmood et al., 2013). Due to use of contaminated water, especially hardness values beyond permissible limits, many health issues are arising in Pakistan. Nephrolithiasis is also one of them and it refers to the state of having stones in kidney or in the urinary tract (Sorokin et al., 2017).

Many elements impact renal calculi; among them drinking water quality and especially water hardness and dietary oxalate intake are important factors because these are the most likely means of nephrolithiasis (Guasch-Ferré et al., 2013; Fakhoury et al., 2019). Types of in kidneys stones vary depending upon many factors; calcium oxalate is the most common renal calculus. Kidney stone formation and its reformation are common problems of present time. According to National Institute for Health and Care Excellence (NICE) guidance on urolithiasis, adults are recommended to drink 2.5-3.0 L of water per day (NICE, 2019). However, there is lack of recommendations regarding the quality of water in NICE guidance as hardness of potable water may influence stone recurrence in urolithiasis patients. Other possible factors responsible for nephrolithiasis may include atmosphere, different diseases, unhealthy food, poor sustenance, hereditary susceptibility, obesity and both amount and timing of dietary calcium (Ca) and magnesium (Mg) consumption (Kozisek, 2003; Kale et al., 2014). Type of kidney stone depends on the nutritional habits as well as on other factors, such as the pH of the urine and its chemical composition including the concentration of different dissolved substances in it.

Metabolically calcium ions are eliminated from the body through the urinary tract and under certain circumstances; these can precipitate leading to the formation of urinary stones. Each stone contains 1-5% of carbon-based substances; an inorganic part formed by different types of crystals (Brener et al., 2011). Khan et al. (2019) demonstrated that dissolved salts made hard water at risk for health, especially due to the presence of Ca and Mg. A study by Vinarova et al., (2016) suggested that a huge increment in calciuria in females happens due to hormonal changes with age. In males, it is mostly incited by high Ca, protein and salt consumption.

Previous studies also demonstrated that chances of kidney stone formation in some particular cases may be reduced due to moderately high Ca concentration in the daily eating schedules of subjects (Rodgers et al., 2019; Tavasoli et al., 2019). A lower dietary Ca accompanied by low oxalate content (for instance, green leafy vegetables, different drinks, grain) is prescribed for patients experiencing renal lithiasis because (Curhan et al., 1993; Brener et al., 2011). Blood begins retaining more Ca from water and nourishment than required because of certain inborn issues in individuals with kidney stone. Subsequently, abundant Ca is stored in the walls of the kidney (Khan et al., 2019).

Drinking water quality, diet habits, and kidney stone composition are important aspects that need to be explored in detail so that the prevalence of nephrolithiasis and its association with drinking water quality and diet can be assessed (Tae et al., 2018). The current investigation on possible association of kidney stones and drinking water quality in Hazara region was planned due to increased hardness of drinking water in the region. The current report will certainly help to analyze the possible reasons of nephrolithiasis especially drinking water in other regions of the world with high water hardness. Although nephrolithiasis has been documented in a few cities in Pakistan (Memon et al., 2012); it is serious health risk affecting a large number of populations in many regions of the world. The current study focuses on the possible correlation of nephrolithiasis between drinking water quality, Ca, and oxalate rich diet.

## 2. Material and methods

### *2.1. Subjects*

One hundred patients with kidney stone were selected for this study. The drinking water samples (in use of patients) were also gathered from all enlisted patients. Locations were grouped into ten main stations and each main station was further divided into ten sub-stations (smaller areas) and ten samples were collected from each main station. Station means a locality from where many samples were collected. These samples were transported, stored at 4ºC and analyzed for different physicochemical parameters like water temperature, turbidity and pH within 24h of their collection. Different parameters such as hardness, pH, turbidity and electrical conductivity (EC) were measured in the laboratory according to standard methods (Soleimani et al., 2018). All hundred human subjects ranged from 15-60 years of age. Percentage ratio was used to show frequency of renal stone. Mean age of subjects with renal stones was also recorded (Mean age 37.5 years). Screening reports of blood and urine analysis of subjects were also considered for the analysis of nephrolithiasis.

A special questionnaire was designed under supervision of expert nephrologists and was used to collect all basic information required to assess the possible association of nephrolithiasis with drinking water quality and diet in affected subjects in district Abbottabad. The main focus was to get exact information about routine eating habits of affected subjects. Their earlier background of kidney inflammation or kidney stone formation and its chemical composition, sources of consumption of drinking water in all affected subjects were also recorded (Yousefi et al., 2018). The human subjects were well briefed about the information being collected from them and it was ensured that their identity would not be disclosed. Moreover, the study was approved by the bioethics committee at COMSATS University, Islamabad.

### *2.2. Collection of drinking water samples*

Drinking water samples were collected from enlisted patients from overall hundred sites and different ten sites of the same region were categorized as one location. According to these distributions ten main locations were defined and drinking water samples were evaluated for different parameters and results are shown in map of the sampling area (Fig. 1).

### *2.3. Analysis of drinking water quality*

Drinking water samples were analyzed regarding various physicochemical parameters such as total hardness, pH, EC and turbidity (APHA, 2005). For total hardness, drinking water sample (50 mL) was taken into a conical flask and buffer solution (2 mL) was added along with a few drops of Eriochrome Black-T.

Titration of this solution was done immediately (because it is unstable under alkaline condition) against EDTA solution until the pink color changed to purple). Reaction between EDTA and $Ca^{2+}$ is 1:1 and the formula weight of $CaCO_3$ is 100.0 g $M^{-1}$. Hardness was calculated by the formula according to (Eq. 1):

$$Hardness\ (mg\ CaCO_3 L^{-1}) = \frac{Volume\ of\ EDTA\ used}{Volume\ of\ sample\ taken} \times 1000 \quad (1)$$

Moreover, EDTA titrant (0.01 M) is equivalent to 400.8 µg Ca/mL. So, calcium (Ca) was determined by using the (Eq. 2):

$$Ca\ (mg\ L^{-1}) = \frac{Volume\ of\ EDTA\ used}{Volume\ of\ sample\ taken} \times 400.8 \quad (2)$$

A drinking water sample was taken in a glass beaker and probe of pH meter was dipped in and it was allowed to stand until temperature steadied. It took an hour to get stabilized and readings of all drinking water samples were recorded after stabilization.

Turbidity meter measured the turbidity of drinking water sample. Electrical conductivity (EC) meter measured the EC of drinking water samples. Each drinking water sample was taken in a glass beaker and probe of EC meter was dipped into the beaker and it was allowed to stand until reading steadied. It took a few minutes to get stabilized and readings of all drinking water samples were recorded after stabilization.

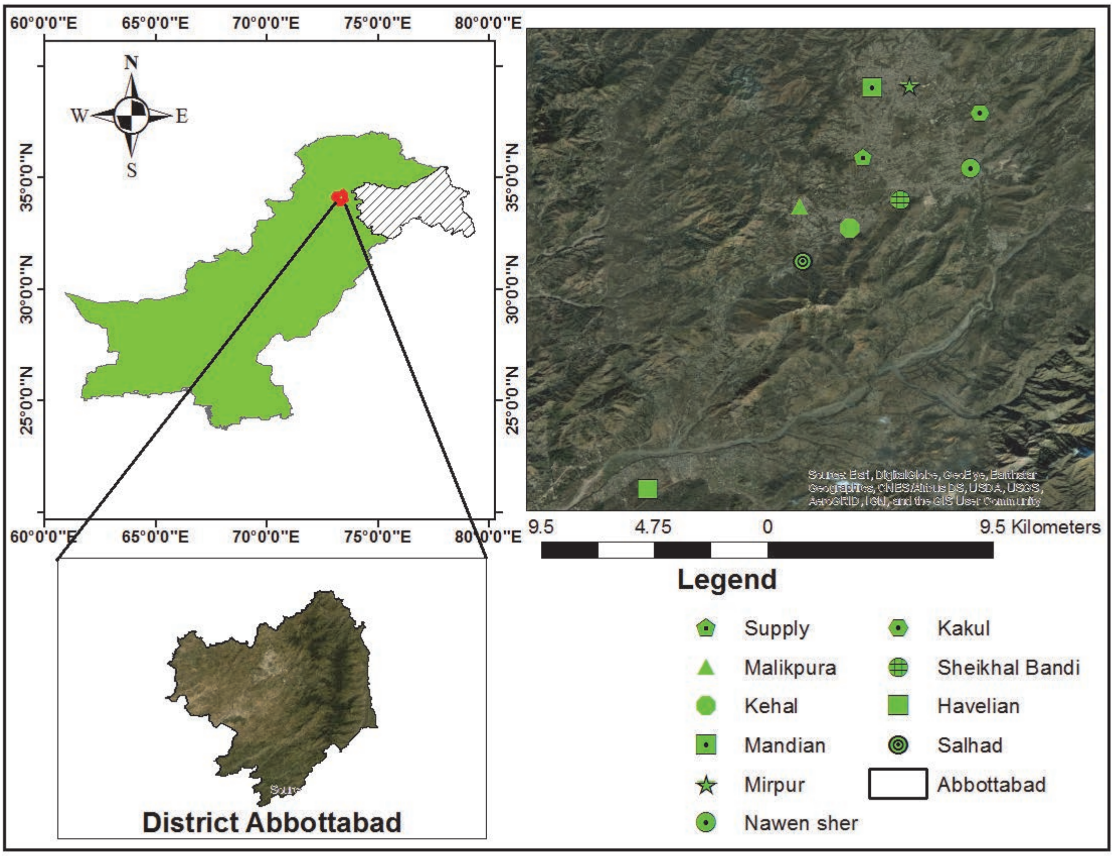


**Fig. 1.** Sampling locations in district Abbottabad

### *2.4. Laboratory analysis for kidney stone composition*

For kidney stone analysis, the presence of oxalate was considered first. Oxalate was determined by the addition of 1 mL of 5% $H_2SO_4$ solution and 2 drops of 1% $KMnO_4$ were added to a small amount of the stone powder. After heating the violet solution, it reduced $KMnO_4$ causing discoloration, hence indicated the presence of oxalate. Furthermore, the presence of phosphate was determined by adding few drops of concentrated $HNO_3$ and 1-2 mL of 5% ammonium molybdate to a small amount of solution. A yellowish color of ammonium phospho-molybdate as a result of heating demonstrated the presence of phosphate in the renal stone (Wilkinson, 2001). A small amount of stone powder was disintegrated in 2N HCl solution and $CO_2$ gas was discharged. Urate (murexid response) was performed in a porcelain capsule by color which demonstrates the formation of purpuric acid through the oxidation of uric acid by $HNO_3$. A drop of 25% $NH_3$ was added and a drop of 10% NaOH and violet color appeared due to development of ammonium purpurate indicating the presence of carbonate (Wilkinson, 2001).

### *2.5. Data treatment and analysis*

A correlation analysis examined the relationship between kidney stone patients and the level of calcium hardness. Basic statistical tests including percentage, ratio, mean and standard deviation and trend line were applied on the data.

## 3. Results and discussion

### *3.1. Types of kidney stone*

The most common kidney stone in the subjects was calcium oxalate. On the basis of chemical composition of kidney stones, it was observed that calcium oxalate stones were prevalent in patients as compared to other types of kidney stones such as struvite, uric acid, and mixture of oxalate and uric acid. Prevalence of four different types of kidney stones in study areas is shown in Fig. 2. The maximum percentage of kidney stones in the tested subjects was calcium oxalate i.e. 88% of kidney patients followed by 7% of patients with uric acid stone, 4% with struvite stone and only 1% of patients with both calcium oxalate and uric acid stones.

Hardness of drinking water indicates the presence of Ca or Mg salts which may cause problem in kidney patients. Fig. 3 shows an abrupt decline in the number of kidney stone patients (calcium oxalate stone) with a decreasing level of drinking water hardness. It showed a significant value of coefficient of correlation ($R^2$ = 0.932), indicating the strong positive relation between two variables (x-y) where variable x indicates level of drinking water hardness and the variable y indicates number of patients.

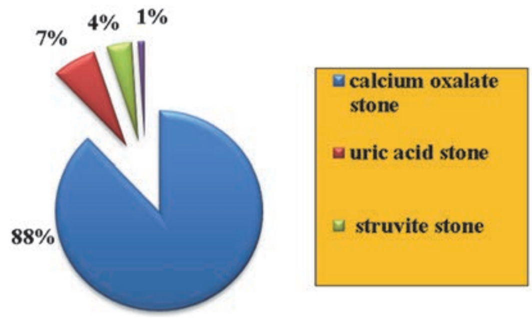


**Fig. 2.** Prevalence of four different types of kidney stones in the tested subjects of study area

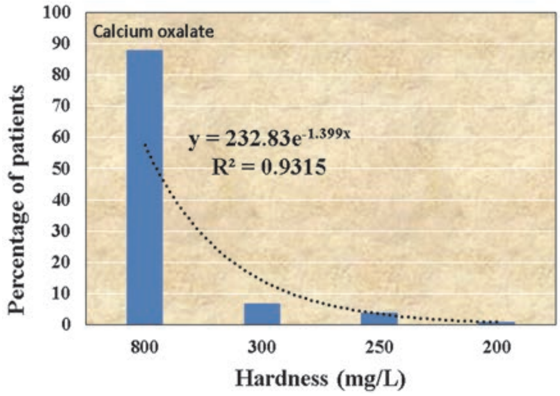


**Fig. 3.** Exponential trend line showing prevalence of kidney stone

### *3.2. Gender wise prevalence of nephrolithiasis*

The maximum prevalence of nephrolithiasis was found in males (54%), followed by females (45%) as indicated by the data presented in Fig. 4. It indicated that consumption of hard water along with oxalate rich diet might be a major source of calcium oxalate kidney stone comparatively more in males compared to the females in study areas.

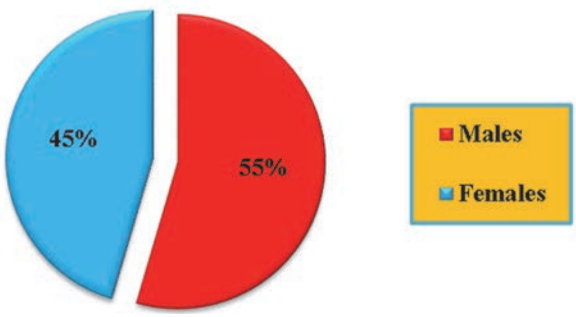


**Fig. 4.** Gender wise prevalence of kidney stones in district Abbottabad

### *3.3. Hardness of drinking water samples*

Field data and analysis of drinking water samples indicated that 15% of subjects were consuming hard water (200-300 mg $L^{-1}$) and 19% were taking very hard water (301-500 mg $L^{-1}$) and 34%

subjects were consuming water hardness with high risk (501-600 mg $L^{-1}$) and 32% were consuming completely deteriorated quality of drinking water (601-800 mg $L^{-1}$) as shown in Fig. 5. Water containing calcium carbonate ($CaCO_3$) below 60 mg $L^{-1}$ is mostly considered as safe. According to Kahlown et al. (2003), the standards regarding various physicochemical properties are as: turbidity < 5 NTU, hardness as $CaCO_3$ < 60 mg $L^{-1}$, total dissolved solids (TDS) < 1000 and pH 6.5 - 8.5.

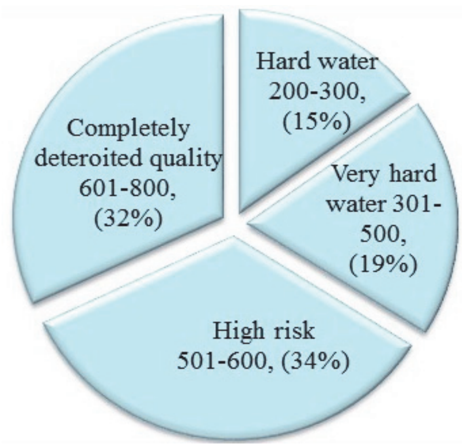


**Fig. 5.** Categorization of drinking water consumption in study areas

The maximum hardness value (599 mg $L^{-1}$) was found in drinking water sample collected from Kehal while the minimum value (483 mg $L^{-1}$) was found in drinking water samples collected from Salhad. The majority of the water samples had calcium hardness value in the range of 500-600 mg $L^{-1}$.

### *3.5. pH values of drinking water*

The mean pH values of drinking water samples collected from the study areas were presented in Fig. 6. The maximum pH value (7.4) was found in drinking water sample collected from Malikpura while that of the minimum pH value (7.2) was observed in drinking water sample collected from Supply. The majority of the water samples had pH value near 7.4 which implied that it was within acceptable range for drinking water. The suitable pH of drinking water ranges from 6.5 to 8.5 as proposed by national quality standards for drinking water quality. Water samples were analyzed for further parameters which revealed high calcium salts in drinking water samples leading to high pH values in the areas under study.

### *3.6. Electrical conductivity of drinking water samples*

Electrical conductivity (EC) indirectly measures the presence of Ca, $HCO_3^{-1}$, $NO_3^{-1}$, $PO_4^{3-}$, $Fe^{2+}$, $Fe^{3+}$, $SO_4^{2-}$ and other ions dissolved in water. Fig. 7 shows the EC of various water samples collected from the study areas. The maximum EC value (574.6 µS $cm^{-1}$) was found in drinking water sample collected from Malikpura while that of the minimum (488 µS $cm^{-1}$) was found in drinking water sample collected from Mandian. The majority of the water samples had EC value around 570 µS $cm^{-1}$. National standard for EC of drinking water is 300 µS $cm^{-1}$. Higher EC various above 300 µS $cm^{-1}$ indicate the presence of pollutants in drinking water which render it unfit for human consumption. Water samples were analyzed for other parameters; high values of calcium hardness were associated with high EC of drinking water samples in the areas under study.

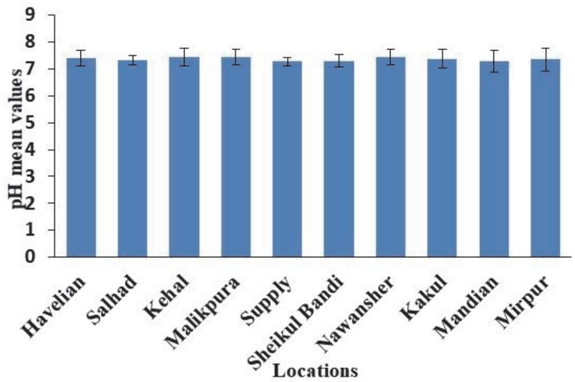


**Fig. 6.** Variation in pH of drinking water samples

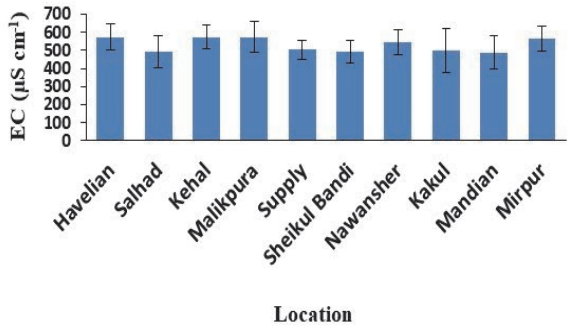


**Fig. 7.** Variations of EC in drinking water samples

### *3.7. Turbidity of drinking water samples*

The presence of silt, sand and mud, bacteria and different germs etc. results in turbid water. Fig. 8 shows that the maximum turbidity (0.4 NTU) was found in drinking water sample collected from Kehal while that of the minimum (0.20 NTU) was observed in drinking water samples collected from Supply. The mean turbidity values of drinking water samples ranged from 0.2 to 0.4 NTU. It might be due to the presence of suspended particulate matter, anthropogenic activities and decrease in water table. The maximum limit for turbidity in drinking water is 5 NTU as proposed by National standards for drinking water.

Dietary oxalate is not an essential nutrient and its higher intake in the body can lead to kidney stone. It is commonly found in plants and humans. Fig. 9 shows that the maximum mean consumption of oxalate (249 mg $day^{-1}$) was found among patients from Kehal, while that of the minimum value (166 mg $day^{-1}$) observed among patients from Mirpur.

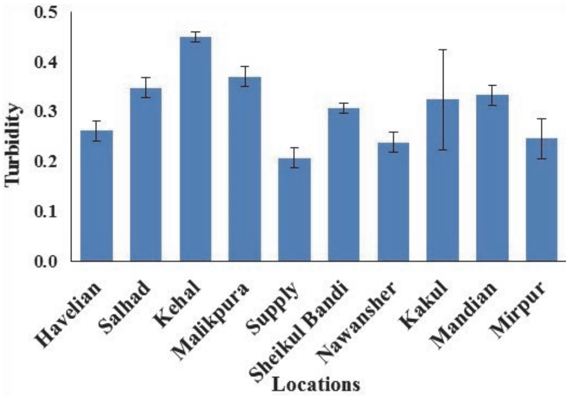


**Fig. 8.** Variation in turbidity of drinking water samples

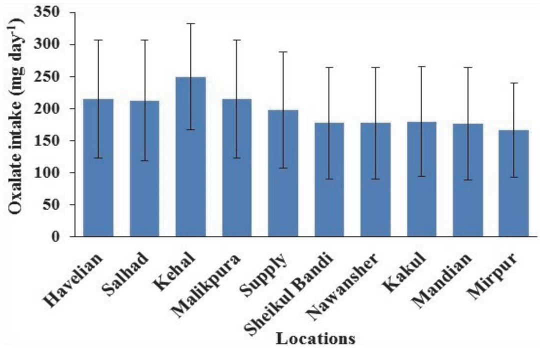


**Fig**. 9. Daily consumption of oxalate rich diet in affected subjects (mg day$^{-1}$)

### *3.8. Oxalate rich food & formation of calcium oxalate stone*

According to National standard, daily oxalate consumption should not exceed 50 mg day$^{-1}$. The relation of water quality with prevalence of kidney stones in study area has been presented in Table 1. These high levels of oxalate intake indicated that high oxalate content of food in the presence of high levels of calcium hardness from drinking water and diet are major possible cause of nephrolithiasis in the study areas. Table 2 presented the data on drinking water intake, urine analysis of affected human subjects indicating the presence of $Ca^{+2}$ and oxalate in urine samples.

On the basis of this diet schedule, 40% of patients were consuming 300 mg oxalates along with 1400 mg Ca on a daily basis. This intake of oxalate is too high, according to National Food Standard and 25% of patients were consuming about 150 mg oxalates day$^{-1}$ along with Ca rich diet, 18% were consuming 120 mg oxalates daily and 17% were taking 100 mg oxalates daily indicating that oxalate rich diet may be a major cause of nephrolithiasis amongst patients in the study areas.

Fig. 10 shows that daily Ca consumption in 9% of subjects under study were consuming 900 mg, 19% with 1000 mg, 32% with 1050 mg, 30% with 1100 mg and 10% were taking 1200 mg Ca. Table 2 showed mean daily intake of water by nephrolithiasis affected individuals, it was evident that mean daily water intake was in range of 2.7 to 3.1 L. The data on calcium and oxalate excretion in their urine samples was also presented. Table 2 clearly showed that there was obvious difference between Ca concentration in urine samples of control and tested kidney patients. Likewise, oxalate concentrations were also much different in control and nephrolithiasis patients. Drinking water consumption per day also differ in all family members. Some family members have same level of drinking water uptake and diet but with huge difference of life style, medications, cigarette smoking, alcohol consumption, exercises and sleeping habits.

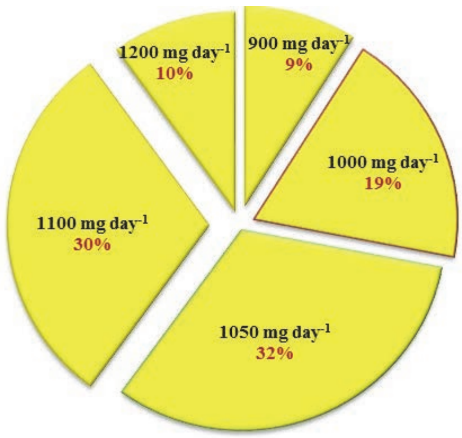


**Fig. 10.** Percentage consumption of Ca in affected subjects

## 4. Discussions

The current study on association of nephrolithiasis with drinking water quality in selected patients of Abbottabad gave deep insights into the disease. Although some positive association of drinking water quality and kidney stones has been observed among studied population, still much work is required to prove it. Merely water quality is not decisive parameter to cause kidney stones in the patients.

Other data on metabolism of patients, dietary habits, and thorough analysis of diet of patients on long term basis will also be required to produce concrete evidence. However, the impact of drinking water quality on the occurrence of nephrolithiasis cannot totally neglected. The current work will provide guidelines to carry out similar research work in other parts of the world. Water quality is important for domestic consumption, an important factor contributing to bad water quality of the study area may be mining activity, especially in Kakul region revealing reliably high rate of urolithiasis (Khan et al., 2016). The prevalence of kidney stones has been noted in the range of 1-13% in various regions of the world. Sorokin et al. (2017) mentioned various factors associated with nephrolithiasis including population growth, increases in obesity and diabetes, geography and environment etc. High Ca levels increase the risk of nephrolithiasis.

**Table 1.** Relation of water quality with prevalence of kidney stones in study area

| ***Locations*** | ***Hardness (mg $L^{-1}$)*** | ***Intake of Ca and oxalate (mg $day^{-1}$)*** | | ***Prevalence of kidney stone*** | | | |
|---|---|---|---|---|---|---|---|
| | | ***Ca*** | ***Oxalate*** | ***Ca-oxalate*** | ***Uric acid*** | ***Struvite*** | ***Mixed type*** |
| Havelian | 400 | 1070 | 214 | 6 | 0 | 1 | 0 |
| Salhad | 500 | 1180 | 212 | 9 | 3 | 0 | 0 |
| Kehal | 744 | 1000 | 249 | 18 | 0 | 0 | 0 |
| Malikpura | 600 | 1120 | 214 | 20 | 0 | 0 | 0 |
| Supply | 350 | 900 | 197 | 5 | 1 | 1 | 0 |
| Sheikhal Bandi | 450 | 1010 | 177 | 3 | 0 | 0 | 0 |
| Nawansher | 550 | 990 | 177 | 5 | 2 | 1 | 0 |
| Kakul | 513 | 900 | 179 | 6 | 2 | 1 | 1 |
| Mandian | 558 | 911 | 176 | 10 | 0 | 0 | 0 |
| Mirpur | 549 | 940 | 166 | 8 | 0 | 0 | 0 |

**Table 2**. Data on mean drinking water intake, urine calcium and oxalate of affected human subjects

| ***Average daily drinking water intake (liters)*** | ***Mean values of calcium (mg $L^{-1}$) in urine samples of kidney patients*** | ***Mean values of oxalate (mg $L^{-1}$) in urine samples of kidney patients*** | ***Mean values of calcium (mg $L^{-1}$) in urine samples of control population*** | ***Mean values of oxalate (mg $L^{-1}$)in urine samples of control population*** |
|---|---|---|---|---|
| 2.9~3.1 | 280 ~ 400 | 20 ~ 40 | 180 ~ 250 | 25 ~ 42 |

Both high Mg and Ca along with high oxalate consumption are not suggested for patients with nephrolithiasis. Therefore, Ca and Mg levels higher than suggested may affect water quality and make it unfit for drinking (Brener et al., 2011; Vasudevan et al., 2017). The prevalence of high level of total hardness in drinking water samples from study area may be attributed to its geological formation containing calcium carbonate rocks (Bilqees et al., 2012).

Dietary sources of calcium are also important contributing factors to nephrolithiasis in this region. On the basis of field data, it was inferred that the maximum mean value of 1180 mg Ca $day^{-1}$ intake was reported from Salhad. The minimum mean value of Ca intake 900 mg $day^{-1}$ was found in patients from Kakul. According to National Food Standards, healthy human adult requires 1000 mg Ca $day^{-1}$. Sorokin et al. (2017) reported that prevalence of calculi varies among different regions of the world e.g. its prevalence in Asia, Europe and North America ranges from 1-5, 5-9 and 7-13%, respectively.

Overall, the proportion of vesical calculi decreased sharply but vice versa for prevalence and incidence of urolithiasis. About 3-7 decades ago, lower urinary tract (LUT) stones were dominant categories in many countries. However, vesical stones accounted for 30-94% of all stones are found in the patients of various countries. Upper urinary tract (UUT) stones are increasing and comprising approximately 85-93% of urolithiasis (Huang et al., 2013; Liu et al., 2018). Among Asian countries, lifetime risk of recurrence rate of urolithiasis is about 60-80%, 6-17% after 1 year and 21-53% after 3-5 years (Tae et al., 2018). It implies that occurrence of nephrolithiasis partly depends upon the physiology of affected individuals. It is interesting to note that normal population is also drinking same water as kidney patients; however, kidney patients are suffering more than normal healthy individuals. Thus, main contributing factors of kidney stones also include the metabolic conditions of a person along utilizing hard water. Chronic kidney disease may result due to complicated urinary tract infections (UTIs). In most cases of kidney stones, the blood starts to absorb more Ca from water and food than actually required for normal human body due to certain inherent problems. As a result, excess Ca is deposited on the walls of the kidney during excretion processes (Brener et al., 2011). It has been found that the persons who do not drink the recommended amount of good quality water with balanced ionic concentration under kidney stones frequently.

The prevalence of kidney stones can be reduced if the relevant precautionary measures are adopted by the kidney patients.

For this very purpose, proper awareness of drinking and eating habits, precautionary measures and medical care are necessary. The information obtained from the present work will serve as an example to carry out similar studies in other regions of the world.

## 5. Conclusions

Water quality of various water samples tested from areas with reported nephrolithiasis showed that water was not safe for drinking purposes. Total hardness of the water samples exceeded the standard value and the water contained a higher Ca content. High values of hardness in the studied area partly contributed to high prevalence of Ca-oxalate kidney stones in district Abbottabad.

The current study demonstrated that major possible factors contributing to the formation of kidney stone included bad drinking water quality and high dietary oxalate consumption. About 88% of all kidney stones and 40% of patients were consuming about 300 mg oxalates daily which is too high according to National Food Standards. Therefore, dietary oxalate was also considered as a contributing factor to the nephrolithiasis.

It is recommended that along with proper medication, installation of water treatment facilities will also be important remedy to create a healthy society in Hazara region.

**Acknowledgements**
This work was financially supported by the Guangxi Major Projects of Science and Technology (Grant No. AA17202032, AA18118013) from the Guangxi Science and Technology Department of China.

## References

Abbasi A., Shaikh A.A., Abbasi S.A., Siyal, A., Mirani A., Shaikh A.B., Shaikh N.A., (2019), Hounsfield units in upper urinary tract Urolithiasis: Our experience at Chandka Medical College Hospital, Larkana, Pakistan, *Rawal Medical Journal*, **44**, 521-523.

APHA, (2005), *Standard Methods for the Examination of Water and Wastewater*, 21st Edition, American Public Health Association, Washington, DC.

Aslam M.S., Adil M., Mirza M.S., Frigon D., (2016), Sustainable community-based drinking water systems in developing countries: stakeholder perspectives, *Journal of Water Supply: Research and Technology-Aqua*, 407-416.

Bilqees R., Tahirkheli T., Pirzada N., Abbas S.M., (2012), Industrial applications of Abbottabad limestone; utilizing its chemical and engineering properties, *Journal of Himalayan Earth Sciences*, **45**, 91-96.

Brener Z.Z., Winchester J.F., Salman H., Bergman M., (2011), Nephrolithiasis evaluation and management, *Southern Medical Journal*, **104**, 133-139.

Cheng Y.Y., Huang N.C., Chang Y.T., Sung J.M., Shen K.H., Tsai C.C., Guo H.R., (2017), Associations between arsenic in drinking water and the progression of chronic kidney disease: a nationwide study in Taiwan, *Journal of Hazardous Materials*, **321**, 432-439.

Curhan G.C., Willett W.C., Rimm E.B., Stampfer M.J., (1993), A prospective study of dietary calcium and other nutrients and the risk of symptomatic kidney stones, *New England Journal of Medicine*, **328**, 833-838.

Fakhoury M.Q., Gordon B., Shorter B., Renson A., Borofsky M.S., Cohn M.R., Cabezon E., Wysock J.S., Bjurlin M.A., (2019), Perceptions of dietary factors promoting and preventing nephrolithiasis: A cross-sectional survey, *World Journal of Urology*, **37**, 1723-1731.

Guasch-Ferré M., Bulló M., Babio N., Martínez-González M.A., Estruch R., Covas M.I., Wärnberg J., Arós F., Lapetra J., Serra-Majem L., Basora J., Salas-Salvadó J., (2013), Mediterranean diet and risk of hyperuricemia in elderly participants at high cardiovascular risk, *The Journals of Gerontology Series A: Biological Sciences and Medical Sciences*, **68**, 1263-1270.

Huang W.Y., Chen Y.F., Carter S., Chang H.C., Lan C.F., Huang K.H., (2013), Epidemiology of upper urinary tract stone disease in a Taiwanese population: a nationwide, population-based study, *The Journal of Urology*, **189**, 2158-2163.

Kahlown M.A., Tahir M.A., Ashraf M., (2003), *Water quality issues and status in Pakistan*, Proc. of seminar on strategies to address the present and future water quality issues, Pakistan Council of Research in Water Resources, Islamabad.

Kale S.S., Ghole V.S., Pawar N.J., Jagtap D.V., (2014), Inter-annual variability of urolithiasis epidemic from semi-arid part of Deccan Volcanic Province, India: climatic and hydrogeochemical perspectives, *International Journal of Environmental Health Research*, **24**, 278-289.

Khan N., Hussain S.T., Saboor A., (2019), Physicochemical investigation of the drinking water sources from Mardan, Khyber Pakhtunkhwa, Pakistan, *Life Science Journal*, **16**, 64-74.

Khan S.R., Pearle M.S., Robertson W.G., Gambaro G., Canales B.K., Doizi S., Tiselius H.G., (2016), Kidney stones, *Nature Reviews Disease Primers*, **2**, 1-23.

Kozisek F., (2003), Health significance of drinking water calcium and magnesium, Czech, On line at: http://www.szu.cz/chzp/voda/pdf/hardness.pdf.

Levallois P., Villanueva C.M., (2019), Drinking water quality and human health: an editorial, *International Journal of Environmental Research and Public Health*, **16**, 631.

Liu Y., Chen Y., Liao B., Luo D., Wang K., Li H., Zeng G., (2018), Epidemiology of urolithiasis in Asia, *Asian Journal of Urology*, **5**, 205-214.

Memon A., Anwar K., Orakzai N., Ather M.H., Biyabani S.R., Nasir A.R., Paryani J.P., Ghani F., Siddiqui K.M., Abbas F., Bangash, K., (2012), *Epidemiology of Stone Disease in Pakistan*, In: *Urolithiasis*, Talati J.J., Tiselius H., Albala D.M., Ye Z. (Eds.), Springer, London, 21-38.

Mahmood Q., Pervez, A., Zeb B.S., Zaffar H., Yaqoob H., Waseem, M., Zahidullah, Afsheen, S., (2013), Natural treatment systems as sustainable ecotechnologies for the developing countries, *BioMed Research International*, **2013**, 1-19.

Menon M., Resnik M.I., (2002), *Urinary Lithiasis; Etiology, Diagnosis and Medical Management*, In: *Campbell's Urology*, Walsh P.C., Retik A.B., Vaughan E.D. (Eds.), 8th Edition, Saunders, Philadelphia, 3243-3248.

NICE, (2019), NICE Guideline, Renal and ureteric stones: assessment and management, On line at: https://www.ncbi.nlm.nih.gov/pubmed/31194311.

Pandit A.B., Kumar J.K., (2015), Clean Water for Developing Countries, *Annual Review of Chemical and Biomolecular Engineering*, **6**, 217-246.

Rodgers A., Cele P., Ravenscroft N., Edmonds-Smith C., Jackson G., (2019), Theoretical and laboratory investigations of the effects of hydroxyproline ingestion on the metabolic and physicochemical risk factors for calcium oxalate kidney stone formation in a small group of healthy subjects, *International Urology and Nephrology*, **51**, 1121-1127.

Soleimani H., Nasri O., Ojaghi B., Pasalari H., Hosseini M., Hashemzadeh B., Kavosi A., Masoumi S., Radfard M., Adibzadeh A., Feizabadi G.K., (2018), Data on drinking water quality using water quality index (WQI) and assessment of groundwater quality for irrigation purposes in Qorveh & Dehgolan, Kurdistan, Iran, *Data Brief*, **20**, 375-386.

Sorokin I., Mamoulakis C., Miyazawa K., Rodgers A., Talati J., Lotan Y., (2017), Epidemiology of stone disease across the world, *World Journal of Urology*, **35**, 1301-1320.

Tae B.S., Balpukov U., Cho S.Y., Jeong C.W., (2018), Eleven-year cumulative incidence and estimated lifetime prevalence of urolithiasis in Korea: a national health insurance service-national sample cohort-based study, *Journal of Korean Medical Science*, **33**, 1-11.

Tavasoli S., Alebouyeh M., Naji M., Shakiba majd G., Shabani Nashtaei M., Broumandnia N., Basiri A., (2019), Association of the intestinal oxalate degrading bacteria with recurrent calcium kidney stone formation and hyperoxaluria: a case-control study, *BJU International*, **125**, 133-144.

UN, (2018), SDG 6 Synthesis Report 2018 on Water and Sanitation, On line at: https://www.unwater.org/publication_categories/sdg-6-synthesis-report-2018-on-water-and-sanitation.

Vasudevan V., Samson P., Smith A.D., Okeke Z., (2017), The genetic framework for development of nephrolithiasis, *Asian Journal of Urology*, **4**, 18-26.

Vinarova L., Vinarov Z., Tcholakova S., Denkov N.D., Stoyanov S., Lips A., (2016), The mechanism of lowering cholesterol absorption by calcium studied by using an in vitro digestion model, *Food and Function*, **7**, 151-163.

Wasana H.M., Perera G.D., Gunawardena P.D.S., Fernando P.S., Bandara, J., (2017), WHO water quality standards Vs Synergic effect (s) of fluoride, heavy metals and hardness in drinking water on kidney tissues, *Scientific Reports*, **7**, 1-6.

Wilkinson H., (2001), Clinical investigation and management of patients with renal stones, *Annals of Clinical Biochemistry*, **38**, 180-187.

WHO, (2018), A global overview of national regulations and standards for drinking-water quality, World Health Organization (WHO) Geneva, On line at: https://www.who.int/water_sanitation_health/publications/national-regulations-and-standards-for-drinking-water-quality/en.

Yousefi M., Dehghani M.H., Nasab S.M., Taghavimanesh V., Nazmara S., Mohammadi A.A., (2018), Data on trend changes of drinking groundwater resources quality: a case study in Abhar, *Data Brief*, **17**, 424-430.