Assessment of disease burden in the arsenic exposed population of Chapar village of Samastipur District, Bihar, India and related mitigation initiative

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Abstract

Fast growing arsenic menace is causing serious health hazards in Bihar, India, with an estimated 10 million people at risk. The exposed population is often unaware of the problem, which only amplifies the burden of arsenic health effects. In the present study, we have assessed the current situation of arsenic exposure in Chapar village of Samastipur district, Bihar. The health of the inhabitants was assessed and correlated with (1) arsenic concentrations in the groundwater of individual wells and (2) arsenic concentration found in their hair and urine. Altogether, 113 inhabitants were assessed, and 113 hair, urine and groundwater samples were collected. The health study reveals that the exposure to arsenic has caused serious health hazard amongst the exposed population with pronounced skin manifestations, loss of appetite, anaemia, constipation, diarrhoea, general body weakness, raised blood pressure, breathlessness, diabetes, mental disabilities, diabetes, lumps in the body and few cancer incidences. It was found that 52% of the total collected groundwater samples had arsenic levels higher than the WHO limit of 10μg/L (with a maximum arsenic concentration of 1212 μg/L) and the reduced arsenite was the predominant form in samples tested for speciation (N=19). In the case of hair samples, 29% of the samples had arsenic concentrations higher than the permissible limit of 0.2mg/Kg, with a maximum arsenic concentration of 46μg/L, while in 20% exposed population, there was significant arsenic contamination in urine samples >50μg/L. In Chapar village, the probability of carcinogenic related risk in the exposed population consuming arsenic contaminated water is 100% for children, 99.1% for females and 97.3% for male subjects. The assessment report shared to the Government, enabled the village population to receive two arsenic filter units. These units are currently operational and catering 250 households providing arsenic free water through piped water scheme. This study therefore identified a significant solution for this arsenic-exposed population.

Keywords: Arsenic exposure, Chapar village, Disease burden, Gangetic plains, Health assessment, Cancer risk, Hazard quotient (HQ), mitigation initiative.

Introduction

It is estimated that arsenic poisoning of groundwater is affecting more than 300 million people worldwide, 70 million of them being in India alone (Hassan 2018). India is a vast country, with two major river basins – Ganga- Meghna- Brahmaputra (GMB) (Acharyya et al., 2000; Chakraborti et al., 2004; Gupta et al., 2015; Mahanta et al., 2015; Mukherjee et al., 2006; Saha et al. 2011; Shah, 2012; Singh et al., 2014). The Himalayan bound rivers are the major carrier of arsenic laden sediments in these river basins, which are contaminating the groundwater (Chakraborty et al., 2015; Guillot and Charlet, 2007; Mukherjee et al., 2014) with high levels of arsenic. In India, the Ganga river basin is highly populated. The upper Gangetic plains consists of states – Uttrakhand and Uttar Pradesh, the middle Ganga plains - Bihar and the lower Ganga plains – West Bengal (Ahamed et al., 2006; Chakraborti et al., 2003, 2016; Katiyar and Singh, 2014; Rahman et al., 2019[a]; Sanz et al., 2007). In the state of Bihar alone, with a total population of c.a. 105 million (Census, 2011), about 10 million people are exposed to arsenic poisoning (Hassan, 2018).

Bihar is adequately represented by the Gangetic plain and is under moderate to thick cover of alluvium. Out of the 38 districts, 18 of them are affected by high arsenic levels. Although the WHO permissible arsenic limit is 10μg/L for drinking water worldwide, the exposed population in the Gangetic plains of the state are drinking water with arsenic concentrations as high as 2 mg/L, i.e. 500 times higher than the recommended limit (Kumar et al., 2016, 2021[a]). This high intake of arsenic over prolonged periods is causing skin manifestations (melanosis, keratosis, rain drop pigmentation, etc.) and a suite of health problems (anaemia, loss of appetite, asthma, constipation, diarrhoea, muscular dystrophy, hormonal imbalances, neurological disorders, loss of mental health etc.) possibly leading to cancer and cancer related death (Abhinav et al., 2016; Kumar et al., 2015, 2016, 2019[a], 2020, 2021[a], 2021[b], 2021[c], 2021[d]; Rahman et al., 2019[b]).

The present study investigates the village Chapar of Samastipur district (Bihar state) where arsenic contamination is high, and the overall health condition of the exposed population is poor. This study evaluates arsenic levels in groundwater with the arsenic concentration in hair samples of the exposed population. This study also investigates the health and cancer risk assessment of the entire village.

Materials and Methods

Location and sample size

The present study was undertaken at Chapar village of Samastipur district, Bihar, India (25°33'3.90"N 85°39'53.25"E). The village is in the vicinity of river Ganga with a distance of mere 6 Kms. In 2011, the village consisted of 250 households and 3,600 inhabitants, 1,982 (55%) being male and 1,618 (45%) female (Census, 2011). In many cases, the households were close to each other with their hand-pumps being less than 10 feet apart. Hence, it was decided in this study to select households that were sufficiently distanced from each other; an approximate distance of 50 m was applied. In total, 113 households (c.a. 45%) were selected, 113 hair, urine and 113 groundwaters samples were collected.

Determination of arsenic concentration and speciation in groundwater samples

The water samples were collected in 500 ml polypropylene bottles that were thoroughly cleaned with distilled water and pre-treated with 2% hydrochloric acid. The depth of each handpump was obtained from the handpump owner. Arsenic total concentrations were measured by Graphite Furnace Atomic Absorption Spectrophotometer (Pinnacle 900T, Perkin Elmer, Singapore) at Mahavir Cancer Institute and Research Centre, Patna, Bihar. Speciation was carried out on 19 of the 113 samples; it consisted in the determination of arsenite As(III) and total inorganic arsenic (As(III) + As(V)) by the analytical technique of voltammetry at a gold wire electrode.

Water samples used for speciation analysis were collected on 28th May 2019 from 19 different hand pumps of the most severely affected part of the village. Most samples (13/19) were filtered (Millipore syringe, 0.2 um filter) but the remaining 6 were not (all collected solutions were clear) due to a lack of filters on the fieldwork day. All samples were acidified to pH 2 (addition of 100 uL 6 M HCl in 50 mL solution) immediately after collection to prevent any precipitation. Voltametric analysis was carried out using the portable PDV6000 potentiostat (Modern Water, UK) along with similar methods as used previously (Bullen et al., 2020), albeit slightly different. Briefly, a three-electrode cell was used with a Ag/AgCl/KCl (3M) reference electrode, a platinum counter electrode and a manufactured gold microwire electrode (6 mm long, 30 um diameter). Determination of both As(III) and total arsenic was made by anodic stripping voltammetry using linear scan voltammetry (8 V/s, 2 mV step) and the method of standard additions. For total arsenic, the sample was diluted in 0.1 M HCl and a deposition potential of -1.2 V was used. Dilution factor was between 10 and 200 times while deposition time was between 5 and 30s. As(III) was determined in a similar manner (dilution in 0.1 M HCl) but using a deposition potential of -500 mV and in presence of 5 ppm ascorbic acid (Sigma-Aldrich, UK). The latter is an anti-oxidant efficient in preventing As(III) oxidation by any chlorine possibly generated during the electrochemical measurement, (Salaün et al., 2007) similarly to hydrazine that was previously used (Salaün et al., 2012). For As(III) determination, dilution factors were between 10 and 200 and deposition times were low, either 3 or 6s. For both total arsenic and arsenite determination, triplicate scans were made for the blank (i.e. diluted original sample) and after each standard addition (2 additions). Using those conditions (LSV, 8 V.s-1, 0.1 M HCl), the arsenic peak potential was at c.a. +150 mV. The arsenic peak height was used for quantification. Samples were all analysed within 48h of collection and no differences in the overall shape of the voltammograms were observed between filtered and non-filtered samples.

Determination of arsenic concentration in hair samples

The hair samples of 113 subjects (one from each household) were collected in zipper polyethylene bags. Arsenic determination was done as per the protocol of (NIOSH, 1994). Briefly, 100mg of hair samples were first weighed in a 50ml acid washed glass beaker before the addition of 15ml of 0.1% sodium dodecyl sulphate (SDS) solution; the mixture was sonicated for 10 min and 15ml of acetone was added for the final dehydration step. The hair samples were then transferred to a 50ml glass beaker before addition of 10ml of Conc.HNO_3; the mixture was left for overnight reaction. The following day, the samples were digested on a hot plate (at 90°C -120°C) and the volume was adjusted to ~3ml. To this solution, 1ml of H_2O_2 was added and redigested on the hotplate at 90°C- 120°C, until the volume reached to 2.5ml. The solution was finally adjusted with distilled water to make a 10ml final solution and filtered it on Whatman filter paper no.41. The final reading was taken on Graphite Furnace Atomic Absorption Spectrophotometer (GF-AAS). The normal range of arsenic contamination in hair is up to 0.2mg/Kg (Chakraborti et al., 2016).

Determination of arsenic concentration in Urine samples

Urine samples from 113 individuals (one per household) were collected in polypropylene urine containers. The determination of arsenic was conducted in accordance with the protocol of (NIOSH, 1994). After collection, all urine samples were digested using HNO_3 concentrate (Merck analytic quality 69%) on a hot plate, and arsenic was estimated by Graphite Furnace Atomic Absorption Spectrophotometer (GF-AAS) (Pinnacle 900T, Perkin Elmer, Singapore) at Mahavir Cancer Sansthan and Research Centre, Patna, Bihar. Urine samples were collected as 0.5 ml aliquots in a 30 ml conical flask and 5 ml of HNO_3 were added and left for overnight reaction. The samples were then digested on hotplate at 90°C - 120°C until the sample reached to 3 ml. Then 5ml mixture of HNO_3:$HClO_4$ (6:1) ($HClO_4$ - Merck analytical grade 70%) was added to the solution and again re-digested on hotplate until the volume reached to about 2ml. Final volume was adjusted to 10ml with 1% HNO_3. The solutions were filtered through Whatman filter paper no.41 and analysed through GF-AAS. The normal range of arsenic contamination in human urine ≤ 50 µg/L as above this level it might have high health risks (ACGIH, 2001).

Quality control

The arsenic standard (1000µg/L) was from PerkinElmer Singapore (CAS no. As 7440-38-2; Lot no. 20-85ASX1; PE no. N9300102) and standard stock solutions were prepared by appropriate dilution. The calibration correlation coefficient was maintained at 0.999 throughout the analysis period. The detection limit of arsenic in water samples, urine samples and hair samples were 0.04µg/L, 0.09µg/L and 0.08µg/Kg respectively.

Health survey

The health assessment survey of the exposed population was done through a health survey typed questionnaire proforma. A total of 2,816 household members available during survey were interviewed. Data related to age, sex, health problems, number of members in the family, number of children, age of the family members suffering from any disease and photographs of the disease manifestations were recorded. Other data related to their handpump depth, duration of usage of contaminated water by them were also recorded. To determine the exact location of the handpump, the handheld Global Positioning System (GPS) receivers (Garmin etrex10, USA) with an estimated accuracy of ≈ 10m was utilised.

Health Risk Assessment

The magnitude and duration of human exposure to any toxic metal are typically reported as average daily dose (ADD, $mg_{As}/(kg_{bodyweight}.day)$) (USEPA, 2014) as expressed in equation (1),

$$\mathbf{ADD = (C \times IR \times EF \times ED)/ (BW \times AT)} \quad (1)$$

where, C is the Arsenic (As) concentration in the groundwater (mg/L)

IR is the daily water intake rate (L/day),

EF is the exposure frequency (365 days/year),

ED is the duration of exposure in years, BW is the body weight of exposed individuals (kg), and

AT is the average time (in days).

Health risk for **non-carcinogenic** exposure is calculated as a hazard quotient (HQ). HQ is defined as the ratio of the potential exposure to a level at which no adverse effects are expected. It is mathematically expressed as follows:

$$\mathbf{HQ = ADD / RfD} \quad (2)$$

In this equation, RfD is the oral reference dose for arsenic (0.3 μg/kg/day) (USEPA, 2012). A non-carcinogenic effect is considered to be possible if the HQ is calculated to be greater than 1. If the calculated HQ is1 or below, then no hostile health effects are predictable as a result of exposure.

The following equation (USEPA, 2012) was employed to calculate **carcinogenic risk** (CR):

$$\mathbf{CR = ADD \times CSF} \quad (3)$$

In above equation, CSF is the arsenic cancer slope factor (1.5 mg/ kg/day) (US-EPA 2012). CR values $\leq 1 \times 10^{-4}$are tolerable menace levels.

Spatial analysis

GPS coordinates were superimposed using QGIS software (version 3.10.1-A Coruna), shape file was created, and Google map was used as base map. Groundwater arsenic contamination was grouped into 3 classes: ≤10, 10-50 and >50μg/L, whereas, the hair arsenic concentration data was grouped into 2 classes: ≤0.2 and >0.2mg/Kg. Simple A3 landscape layout was chosen for the thematic map output. Moreover, kernel density maps of the symptoms were created with R version 4.0.3 (R Core Team, 2020) with the "density.ppp" function of the package "spatstat" version 2.1.0 (Baddeley et al., 2015). The symptom occurrence locations were used to derive the density of the symptom for a given area based on the kernel smoother. The estimation was implemented using an isotropic Gaussian kernel and the selected bandwidth by the Stoyan's rule of thumb, which determines the smoothness of the density function.

Statistical analysis

All the data were analysed using the statistical software GraphPad Prism 5.0 and the values were expressed as Mean ± SEM. Differences between the groups were statistically analysed by one-way analysis of variance (ANOVA) using the Dunnett's test while scattered graphs were plotted through another statistical software IBM SPSS- 25.0 using linear regression analysis.

Results

Groundwater arsenic assessment in Chapar village

Out of the 113 water samples, 52% of them had arsenic levels above 10µg/L and 32% above 50µg/L, while 48% samples had arsenic concentration with in the normal range i.e below 10µg/L. The maximum arsenic concentration was found at 1212 µg/L (Figure 1A).

The inorganic arsenic speciation determined in 19 water samples revealed that the proportion of As(III) varied between 41 and 110% of total arsenic, with an average of 75 ± 18% (average ± standard deviation). This proportion of As(III) was not dependent on total concentrations: for samples with total As in the range 10-50 ppb (N =3), As(III) represented 79 ± 18% of the total arsenic; for 50-100 ppb (N=4), 86 ± 24%; for As 300-600 ppb (N = 4), 78 ± 13% and for total As > 600 ppb (N=8), As(III) represented 66 ± 18%. Arsenite was thus the predominant species in the groundwater, similar to other studies carried out e.g. in West Bengal and consistent with the reducing conditions thought to occur in these regions (Bullen et al., 2020). It was also found that there was no differences in As(III) levels obtained in filtered and non-filtered samples (N=2).

Total Arsenic concentrations in hair samples of village people

Similar to water samples, arsenic concentrations in the 113 hair samples varied significantly: 71% of the samples had concentrations below the permissible limit of 0.2mg/Kg (and therefore 29% hair samples >0.2mg/Kg). The maximum hair arsenic concentration was extremely high at 46.40mg/Kg (Figure 1B).

Correlation coefficient Study

1. ***Correlation coefficient between groundwater arsenic concentration and depth of the handpumps:*** A marginal declining trend was observed between arsenic levels and handpump depths, suggesting higher arsenic contamination levels in the shallower aquifers (r=0.005, P <0.05; Figure 2A).
2. ***Correlation coefficient between hair arsenic levels and age of the subjects*:** The correlation coefficient was observed between the subject's age and their hair arsenic contamination level. The subjects aged between 35 to 60 years retained higher hair arsenic contamination values representing the age-related hair arsenic deposition (r = 0.010, P <0.05; Figure 2B).
3. ***Correlation coefficient between hair arsenic level and groundwater arsenic concentration:*** A very significant positive and increasing trend was observed

between the subject's hair arsenic level and their drinking water arsenic contamination (r= 0.172, P<0.05; Figure 2C).

4. ***Correlation coefficient between hair arsenic level and duration of arsenic exposure:*** An increasing trend was observed between the subject's hair arsenic level and duration of arsenic exposure through contaminated drinking water (r= 0.013, P<0.05, Figure 2D).
5. ***Correlation coefficient between urine arsenic contamination and groundwater arsenic concentration:*** An increasing significant trend was observed between the subject's urine arsenic concentration and their drinking water arsenic concentration (r= 0.673, P<0.05, Figure 2E).

Spatial Analysis

The **Figure 3A** shows a synoptic view of arsenic levels detected in the groundwater and hair samples across Chapar village. The map clearly highlights a strong spatial correlation between these 2 parameters.

The **Figure 3B** shows the kernel density maps of the symptoms of 1,307 samples from the 113 households. The colour represents the degree of the intensity per area unit, which is the expected number of patients with each symptom per 10,000 square meters in this case. The kernel density maps show that several symptoms (keratosis, melanosis, loss of appetite, anaemia, and cardiovascular disorders) had a similar spatial pattern as that obtained for arsenic levels in groundwater, suggesting a strong causal link.

Clinical Observations

The village population exhibited typical visible symptoms of arsenicosis. The most astonishing finding observed was that, one household had all the family members with symptoms of arsenicosis- including an only 07-year-old boy (Figure 4A), his elder brother of 14 years old (Figure 4B) and his mother of 32 years old (Figure 4C). Usually, arsenicosis symptoms are not observed in children below 10 years but, in this exposed household, members were drinking arsenic contaminated water (655µg/L was detected in their well) possibly for more than 30 years in the case of the mother.

The other symptoms observed in the exposed subjects were a cervical lump in a 6 year old girl, melanosis in a 42 year old man, mentally disabled boy 25 years old with keratosis in palm sole and melanosis on the trunk, palmoplantar keratosis in a 36 year old woman, rain drop pigmentation in a 16 year old boy. All these subjects were drinking arsenic contaminated water >500µg/L for a significant amount of time (Figure 4D).

Health Assessment

In the present study, all the 250 households representatives (2,816 household members who were present during the study period) were queried about their health. In the households where the arsenicosis symptoms or health related problems were severe, household members were interviewed extensively. The **Figure 5A** shows the associations between the symptoms. The arsenicosis symptoms, especially the keratosis in the palm and sole, were observed in 11.2%, melanosis (8.6%), other skin manifestations (45.2%), loss of appetite (14.6%), anaemia (13.2%), diarrhoea (9.7%), general body weakness (57.4%), raised blood pressure (6.4%), breathlessness (7.1%), diabetes (1.88%), mental disability (0.14%), lumps in the body (0.24%), cancer incidences (0.39%) and other health problems (14.4%) respectively. Table shows the health assessment result of the population of Chapar village (n=2,816) from a total of 250 households (**Figure 5B**).

Cancer cases

In the present village population, there were 11 cancer cases reported out of which 7 patients had died mostly due to liver cancer, gall bladder cancer and skin cancer. However, few cases of liver cancer, breast cancer and colon cancer patients were having treatment in cancer centres at the time the sampling took place. Moreover, 20% of the study population had a significantly high concentration of arsenic in urine (>50 μg/L), indicating their susceptibility to cancer disease in the future.

Health Risk Assessment

Non-carcinogenic risk of arsenic exposure was more in the child's consumers as compared with the male and female population. Higher risk for children population may be owed to their lower body weight (Kumar et al., 2019[b]). However, each sample surpassed the threshold value of 1×10^{-4} for Carcinogenic Risk (CR) in children and was found below the threshold value in female and male population group, respectively (Figure 6A & 6B).

Mitigation Intervention

The data report of the present study area was shared to the Health Department of Ministry of Health and Family welfare, Government of India, through which the state government was government was advised to intervene with mitigation measures. The state government installed two arsenic filter units in which unit-I caters 150 households while the other unit-II caters 100 households by supplying arsenic free water through the piped water scheme – *"Har Ghar Nal Ka Jal"*- means 'every household getting tapwater'. These filters were installed in August

2020. It is hoped that the exposed population (n=250) will strongly benefit from this intervention. The health review shall be conducted after the operation of 18 months of this installed arsenic filter to determine the impact (Figure 7A & 7B.).

Geological Aspect

The study shows significant correlation of arsenic contamination in the groundwater in the village due to geogenic changes (1985- 2020) (Figure 8).

Discussion

The groundwater arsenic poisoning in the plains of river Ganga has become a major problem for the exposed population. The exposed subjects are knowingly or unknowingly consuming this water for long duration, causing serious health hazards to them. The arsenic contaminated water initially reaches the gastrointestinal tract from where the arsenic is absorbed into the blood. Liver is the site where As (III) is converted to As (V) or even to the less toxic compound dimethyl arsenic acid (DMA). But this compound is also considered a carcinogen as they dysregulate the functions of various gene functions including the tumour suppressor genes such as p53, p16, NFKappa B, etc. (Agusa et al., 2015; Nava-Rivera et al., 2021; Reichard and Puga, 2010; Roy et al., 2020; Suzuki and Nohara, 2013). The trivalent inorganic arsenic binds with the sulfhydryl groups of dihydrolipoamide inhibiting the pyruvate dehydrogenase. This causes a significant reduction in the conversion of pyruvate to acetyl coenzyme A (CoA) which causes reduced ATP synthesis (Bergquist et al., 2009; Sherwood et al., 2011). Furthermore, arsenic binding to sulfhydryl groups causes significant reduction in the functions of gluconeogenesis, fatty acid oxidation and production of acetyl CoA (Intarasunanont et al., 2012; Miller et al., 2002; Ren et al., 2011; Wang et al., 2015).

According to (Mishra et.al., 2000), the riparian districts on either banks of river Ganga in Bihar are facing the problem of arsenic contamination. The present study area also falls in a zone of oscillation of river Ganga where it not only flows from west to east but also oscillates in north south direction. The river shows meandering nature within this alluvial fill deposit which constitutes the zone of miscibility of inflow of sediments from contrasting sources which is active flood plain (Holocene- Recent age) and older flood plain of (Holocene). This sinuosity likely contributes for high anomalous value of arsenic (Figure 8).

The temporal variation along the flow path of river Ganga clearly indicates change in the nearly straighter course of the river with marginal bulge or convexity southwards in 1985, to a conspicuous U-shaped meandering in 1995. This has probably resulted from enhanced sedimentation along the periphery thus guiding flow path to assume a bend as observed in the image. Ultimately the creation of river island in the middle path of the river has pushed the active flow path further southwards and northwards thus increasing the sinuosity of the river. This has almost resulted in the creation of an Oxbow-lake which disables the arsenic mobilisation from one-point bar to the other point bar, leading to secondary arsenic enrichment to enhanced levels. This water locked oxbow lakes are the major source of arsenic contamination in the ground water and habitations residing in these areas are at high risk of arsenic-laced water. The studied village population of Chapar village is also located on the point bars of oxbow lake of river Ganges, which is the major cause of the severe exposure of arsenic (Donselaar et al., 2017; Kumar et al., 2021[e]; McArthur et al., 2001, 2004, 2011; Nickson et al., 2000; Postma et al., 2007; Ravenscroft et al., 2009). The higher concentration almost follows a line of demarcation around Chapar village which also defines a probable zone of levee delimiting the concentration to the south (Figure 3A). Presumably the separation of high and low values defines the sinuosity regime of river Ganga with time.

In the present study, the maximum arsenic concentration in groundwater observed was 1212 μg/L, which is extremely high, 120 times higher than the WHO permissible limit of 10 μg/L. Fifty-two per cent of the handpump water samples had arsenic levels higher than 10μg/L and the speciation study carried out for the first time in this region suggests the predominance of arsenite which is, the more toxic inorganic form. The study also clearly shows the spatial correlation between arsenic levels in water and a suite of health symptoms, such as hyperkeratosis, melanosis, loss of appetite, anaemia, rain drop pigmentation etc. (Figure 3B). Moreover, the exposed population also displayed other symptoms such as constipation, diarrhoea, general body weakness, raised blood pressure, breathlessness, diabetes, mental disabilities, diabetes, lumps in the body and few cancer incidences. From the symptom's association analysis, keratosis was found significantly highly associated with breathlessness whereas the association was not strong for melanosis and other skin disease patients. The long exposure to arsenic generally weakens the immunity of the body which, if left untreated, leads to serious health hazards. The association between low-level exposures to arsenic in drinking water (approximately <150μg/L) and cancer risks is still not well elucidated (Tsuji et al., 2019). Our results suggest that once it reaches to develop keratosis, the patient's respiratory function is compromised. This may not eventually lead to cancer in the end but may certainly degrade the patient's quality of life and lower his/her self-esteem, social and economic status. The exposed population of the village were from socially and economically weaker section who had limited resources of expenses. Hence, they did not have any preventive measures or information related to arsenic exposure. Similar studies in the Gangetic plains of Bihar have been carried out by other researchers as well (Berg et al., 2001; Chakraborti et al., 2016, 2018; Chen et al., 1999; Chowdhury et al., 2000; Ferreccio et al., 2000; GuhaMazumdar et al., 1998; Kumar et al., 2015, 2016, 2020, 2021[a], 2021[b], 2021[c]; Rahman et al., 2019[a], 2019[b]).

The present study infers through its findings for the first time the assessment of the non-carcinogenic health risk vs carcinogenic health risk by using the hazard quotient (HQ) formula of the exposed population of the Chapar village. We show that the probability of carcinogenic risk is almost maximal for all groups (100% in children, 99.1% in females and 97.3% in male subjects). Similar studies have been carried out which correlates the present study (Ali et al., 2021; Fernández-Macías et al., 2020; Joardar et al., 2021; Kumar et al., 2019[a]; Kumari et al., 2021; Li et al., 2018; Liang et al., 2016; Sheikhi et al., 2021; Ulniković and Kurilić, 2020; Yang et al., 2015).

Urine, nail, and hair are considered as the most significant biomarkers (Hays et al., 2010; LaKind et al., 2008; Orloff et al., 2009; Rasheed et al., 2019). In the present study, urine and hair samples were used as the biological indicator for long-term exposure of arsenic, as they reflect the human exposure during the last period of 2-5 months. WHO has also recommended the use of urine and hair as environmental arsenic monitoring agent (WHO, 2001; Tseng et al., 2005; Valenzuela et al., 2005; Druyan et al., 1998; Gebel, 2000; Morton et al., 2002). The arsenic usually binds with the sulfhydryl groups of keratin and remains in the shafts and hair roots for a long duration which reflects the long term exposure (Hindmarsh, 2002). In the present study, the maximum arsenic concentration detected in hair samples was 46.04mg/Kg, while in urine as 367.2μg/L, an extremely high dose denoting the severity of the long-term exposure. About 20% of the studied population had very significant ($p<0.05$) high urine arsenic concentration >50μg/L. Very high contamination of arsenic in hair and urine samples suggest that the exposed subjects were drinking arsenic laden water for several months continuously and the skin manifestations observed all over their body is corroborating the high exposure level. The exposed population is at very high risk that they are vulnerable to get several diseases. Similar studies have correlated arsenic exposure with the contamination in the biological samples (Kumar et al., 2021[b], 2021[c]; Nowak and Kozlowski, 1998; Yoshinaga, et al., 1990).

Conclusions:

In the present study, there was significant arsenic contamination observed in the water samples of the assayed handpumps. Out of total 113 analysed handpumps, 52% had very high arsenic concentration above the WHO permissible limit i.e 10µg/L. Moreover, there was significant arsenic contamination observed in the hair samples of the exposed population as 29% of the individuals had the concentration >0.2mg/Kg, while 20% of the population had urine arsenic concentration >50µg/L Prevalence of many visible symptoms of arsenic poisoning like hyperkeratosis, melanosis, rain drop pigmentation etc. and a suite of health problems (anaemia, loss of appetite, asthma, constipation, diarrhoea, muscular dystrophy, hormonal imbalances, neurological disorders, loss of mental health etc.) possibly leading to cancer and cancer related death were observed in the exposed individuals. There was significant correlation coefficient observed between hair arsenic concentration of the individual with ground water arsenic concentration and the duration of the exposure through it. The health status of the population of the Chapar village in the district of Samastipur (Bihar) was dramatically high, with a cancer risk of 100% in children and only slightly lower for the male and female population. The report was shared with the concerned government agencies highlighting the temporal and spatial distribution of this affected area for the mitigation measures to be undertaken for the arsenic exposed population of the study area. This study report enabled the government to install two arsenic removal filter units in this affected village which caters to 250 households providing them arsenic free water through piped water system. This study is a turning point for the severely exposed village population who had no access to safe drinking water since last 20 years. It is expected that in the coming years the disease burden will significantly decrease in the exposed population of the village.

Conflict of interest: The authors declare that they have no conflicts of interest concerning this article.

Author Contributions: A.K and A.K.G. conceptualized the entire work. A.K. is the principal author and had the major contributions in writing the manuscript but support was also provided by A.K.G., P.S, M.S, A.B., M.A, R.K. M.S.R., literature search was done by M.S.R, M.K. and P.K.N., experimental work and data analysis were done by PS, P.K.N, M.S.R, S.D., K.M., A.G., geospatial mapping figures were designed by S.S. and A.B., heatmap figures were designed by M.S., arsenic risk assessment study was done by S.K., final data interpretation was done by A.K., A.K.G., P.S., M.S., M.A, R.K, D.K., K.M., S.D., S.K., and A.B. All authors read and approved the final manuscript.

Ethical Statement: Ethical approval was obtained from the Institutional Ethics Committee (IEC) of Mahavir Cancer Sansthan and Research Centre with IEC No. MCS/IEC/2019, dated 10/01/2019 (agenda no.07). The study was carried out between January 2019 and December 2019. Informed consent was obtained before the collection of the biological samples from the studied subjects.

Acknowledgement: The authors acknowledge support extended by Mahavir Cancer Sansthan and Research Centre, Patna for providing laboratory and other infrastructural facilities for this study. The authors also thank the State Government for installing this arsenic filter units in this severely arsenic exposed village.

Data Availability: The study related raw data has been submitted at Editorial desk for the validation of the present work.

Consent to participate: For any of the clarification related to the publication of the article, the authors provide the consent.

Consent for publication: The authors give the consent to publish our research data.

References:

Abhinav, S., Navin, S., Verma, S.K., Kumar, R., Ali, M., Kumar, A., Ghosh, A.K., 2016. Groundwater and blood samples assessment for arsenic toxicity in rural population of Darbhanga district of Bihar, India. In: Proceedings of the 6th International congress on arsenic in the environment, Stockholm, Sweden, 19–23 June 2016. Arsenic Research and Global Sustainability As 2016, Taylor & Francis group, London. 418-420.

Acharyya, S.K., Lahiri, S., Raymahashay, B.C., Bhowmik, A., 2000. Arsenic toxicity of groundwater in parts of the Bengal Basin in India and Bangladesh: The role of Quaternary stratigraphy and Holocene sea-level fluctuation. Environ Geol. 39:1127–1137.

Agusa, T., Kunito, T., Minh Tue, N., Thi Mai Lan, V., Binh Minh, T., Thi Kim Trang, P., Fujihara, J., Takeshita, H., Takahashi, S., Hung Viet, P., Tanabe, S., Iwata, H., 2015. Nihon eiseigakuzasshi. Japanese journal of hygiene. 70(3):186–196. https://doi.org/10.1265/jjh.70.186.

Ahamed, S., Sengupta, M.K., Mukherjee, A., Hossain, M.A., Das, B., Nayak, B., Pal, A., Mukherjee, S.C., Pati, S., Dutta, R.N., Chatterjee, G., Mukherjee, A., Srivastava, R., Chakraborti, D., 2006. Arsenic groundwater contamination and its health effects in the state of Uttar Pradesh (UP) in upper and middle Ganga plain, India: a severe danger. Sci Total Environ. 370(2-3):310-322.

Ali, N., Eqani, S., Nazar, E., Alhakamy, N.A., Rashid, M.I., Shahzad, K., Zeb, J., Shen, H., Ismail, I., Albar, H., 2021. Arsenic and lead in the indoor residential settings of different socio-economic status; assessment of human health risk via dust exposure. Environmental science and pollution research international. 28(11):13288–13299. https://doi.org/10.1007/s11356-020-11546-w.

ACGIH (American Conference of Government Industrial Hygienists) (2001) Documentation of Biological Exposure Indices, 7th edn. ACGIH Worldwide, Cincinnati.

Baddeley, A., Rubak, E., Turner, R., 2015. Spatial Point Patterns: Methodology and Applications with R. London: Chapman and Hall/CRC Press.

https://www.routledge.com/Spatial-Point-Patterns-Methodology-and-Applications-with-R/Baddeley-Rubak-Turner/9781482210200.

Berg, M., Tran, H.C., Nguyen, T.C., Pham, H.V., Schertenleib, R., Giger, W., 2001. Arsenic contamination of groundwater and drinking water in Vietnam: a human health threat. Environmental science & technology. 35(13):2621–2626. https://doi.org/10.1021/es010027y.

Bergquist, E.R., Fischer, R.J., Sugden, K.D., Martin, B.D., 2009. Inhibition by methylated organo-arsenicals of the respiratory 2-oxo-acid dehydrogenases. Journal of organometallic chemistry. 694(6):973–980. https://doi.org/10.1016/j.jorganchem.2008.12.028.

Bullen, J.C., Torres-Huerta, A., Salaün, P., Watson, J.S., Majumdar, S., Vilar, R., Weiss, D.J., 2020. Portable and rapid arsenic speciation in synthetic and natural waters by an As(V)-selective chemisorbent, validated against anodic stripping voltammetry. Water research. 175:115650. https://doi.org/10.1016/j.watres.2020.115650.

Census., 2011. Interim Report of Population Census of India. (http://www.censusindia.com).

Chakraborti, D., Mukherjee, S.C., Pati, S., Sengupta, M.K., Rahman, M.M., Chowdhury, U.K., Lodh, D., Chanda, C.R., Chakraborti, A.K., Basu, G.K., 2003. Arsenic groundwater contamination in Middle Ganga Plain, Bihar, India: a future danger?. Environmental health perspectives. 111(9):1194–1201. https://doi.org/10.1289/ehp.5966

Chakraborti D, Sengupta MK, Rahman MM, Ahamed S, Chowdhury UK, Hossain MA, Mukherjee SC, Pati S, Saha KC, Dutta RN, Quamruzzaman Q (2004) Groundwater arsenic contamination and its health effects in the Ganga-Meghna-Brahmaputra plain. J Environ Monit 6(6):74N-83N

Chakraborty, M., Mukherjee, A., Ahmed, K.M., 2015. A Review of Groundwater Arsenic in the Bengal Basin, Bangladesh and India: from Source to Sink. Curr Pollution Rep. 1:220–247. https://doi.org/10.1007/s40726-015-0022-0.

Chakraborti, D., Rahman, M.M., Ahamed, S., Dutta, R.N., Pati, S., Mukherjee, S.C., 2016. Arsenic groundwater contamination and its health effects in Patna district (capital of Bihar) in the middle Ganga plain, India. Chemosphere. 152:520–529.

Chakraborti, D., Singh, S.K., Rahman, M.M., Dutta, R.N., Mukherjee, S.C., Pati, S., Kar, P.B., 2018. Groundwater Arsenic Contamination in the Ganga River Basin: A Future Health Danger. International journal of environmental research and public health. 15(2):180. https://doi.org/10.3390/ijerph15020180.

Chen, K.L., Amarasiriwardena, C.J., Christiani, D.C., 1999. Determination of total arsenic concentrations in nails by inductively coupled plasma mass spectrometry. Biological trace element research. 67(2):109–125. https://doi.org/10.1007/BF02784067.

Chowdhury, U.K., Biswas, B.K., Chowdhury, T.R., Samanta, G., Mandal, B.K., Basu, G.C., Chanda, C.R., Lodh, D., Saha, K.C., Mukherjee, S.K., Roy, S., Kabir, S., Quamruzzaman, Q., Chakraborti, D., 2000. Groundwater arsenic contamination in

Bangladesh and West Bengal, India. Environmental health perspectives. 108(5):393–397. https://doi.org/10.1289/ehp.00108393.

Donselaar, M.E., Bhatt, A.G., Ghosh, A.K., 2017. On the relation between fluvio-deltaic flood basin geomorphology and the wide-spread occurrence of arsenic pollution in shallow aquifers. The Science of the total environment. 574:901–913. https://doi.org/10.1016/j.scitotenv.2016.09.074.

Druyan, M.E., Bass, D., Puchyr, R., Urek, K., Quig, D., Harmon, E., Marquardt, W., 1998. Determination of reference ranges for elements in human scalp hair. Biological trace element research. 62(3):183–197. https://doi.org/10.1007/BF02783970

Fernández-Macías, J.C., González-Mille, D.J., García-Arreola, M.E., Cruz-Santiago, O., Rivero-Pérez, N.E., Pérez-Vázquez, F., Ilizaliturri-Hernández, C.A., 2020. Integrated probabilistic risk assessment in sites contaminated with arsenic and lead by long-term mining liabilities in San Luis Potosi, Mexico. Ecotoxicology and environmental safety. 197:110568. https://doi.org/10.1016/j.ecoenv.2020.110568.

Ferreccio C, González C, Milosavjlevic V, Marshall G, Sancha AM, Smith AH (2000) Lung cancer and arsenic concentrations in drinking water in Chile. Epidemiology. 11(6):673–679. https://doi.org/10.1097/00001648-200011000-00010.

Gebel, T., 2000. Confounding variables in the environmental toxicology of arsenic. Toxicology. 144(1-3):155–162. https://doi.org/10.1016/s0300-483x(99)00202-4.

GuhaMazumder, D.N., Haque, R., Ghosh, N., De, B.K., Santra, A., Chakraborty, D., Smith, A.H., 1998. Arsenic levels in drinking water and the prevalence of skin lesions in West Bengal, India. International journal of epidemiology. 27(5):871–877. https://doi.org/10.1093/ije/27.5.871.

Guillot S, Charlet L (2007) Bengal arsenic, an archive of Himalaya orogeny and paleohydrology. J Environ Sci Health Part A. 42(12):1785–1794. https://doi.org/10.1080/10934520701566702.

Gupta, A., Bhattacharjee, D., Borah, P., Debkanungo, T., Paulchoudhury, C., 2015. Arsenic contamination of groundwater in Barak Valley, Assam, India: Topography-based analysis and risk assessment. In: Ramanathan, A.L., Johnston, S., Mukherjee, A., Nath, B., (ed) Safe and Sustainable Use of Arsenic-Contaminated Aquifers in the Gangetic Plain: A Multidisciplinary Approach. Springer Publishing Company, Cham, Switzerland, with Capital Publishing Company, New Delhi, India. 81–96.

Hassan, M.M., 2018. Arsenic in Groundwater: Poisoning and Risk Assessment, 1st Edition. Boca Raton: Taylor & Francis, CRC title. ISBN 9781439839270.

Hays SM, Aylward LL, Gagné M, Nong A, Krishnan K (2010) Biomonitoring equivalents for inorganic arsenic. Regul Toxicol Pharmacol. 58(1):1–9. https://doi.org/10.1016/j.yrtph.2010.06.002.

Hindmarsh, J.T., 2002. Caveats in hair analysis in chronic arsenic poisoning. Clinical biochemistry. 35(1):1–11. https://doi.org/10.1016/s0009-9120(01)00282-x.

ICMR (Indian Council of Medical Research), 2009. Nutrient requirements and recommended dietary allowances for Indians. Indians National Institute of Nutrition, Hyderabad. 334.

Intarasunanont P, Navasumrit P, Waraprasit S, Chaisatra K, Suk WA, Mahidol C, Ruchirawat M (2012) Effects of arsenic exposure on DNA methylation in cord blood samples from newborn babies and in a human lymphoblast cell line. Environ Health.11:31. https://doi.org/10.1186/1476-069X-11-31.

Joardar, M., Das, A., Chowdhury, N.R., Mridha, D., De, A., Majumdar, K.K., Roychowdhury, T., 2021. Health effect and risk assessment of the populations exposed to different arsenic levels in drinking water and foodstuffs from four villages in arsenic endemic Gaighata block, West Bengal, India. Environmental geochemistry and health. Advance online publication. https://doi.org/10.1007/s10653-021-00823-3.

Katiyar, S., Singh, D., 2014. Prevalence of arsenic exposure in population of Ballia district from drinking water and its correlation with blood arsenic level. Journal of environmental biology. 35(3):589–594.

Kumar, A., Ali, Md., Rahman, S.Md., Iqubal, A.Md., Anand, G., Niraj, P.K., Srivastava, A., Kumar, R., Gahlot, V., 2015. Ground Water Arsenic Poisoning in “Tilak Rai Ka Hatta” Village of Buxar District, Bihar, India Causing Severe Health Hazards and Hormonal Imbalance. J Environ Anal Toxicol. 5:290.

Kumar, A., Rahman, M.S., Iqubal, M.A., Ali, M., Niraj, P.K., Anand, G., Kumar, P., Abhinav, Ghosh, A.K., 2016. Ground Water Arsenic Contamination: A Local Survey in India. International journal of preventive medicine. 7:100. https://doi.org/10.4103/2008-7802.188085.

Kumar, A., Rahman, M.S., Kumar, R., Ali, M., Niraj, P.K., Srivastava, A., Singh, S.K., Ghosh, A.K., 2019[a]. Arsenic contamination in groundwater causing impaired memory and intelligence in school children of Simri village of Buxar district of Bihar. J Mental Health Hum Behav. 24:132–138. https://doi.org/10.4103/jmhhb.jmhhb_31_18.

Kumar, S., Singh, R., Venkatesh, A.S., Udayabhanu, G., Sahoo, P.R., 2019[b]. Medical Geological assessment of fluoride contaminated groundwater in parts of Indo-Gangetic Alluvial plains. Scientific reports. 9(1):1-16.

Kumar, A., Ali, M., Kumar, R., Rahman, M.S., Sivastava, A., Chayal, N.K., Sagar, V., Kumari, R., Parween, S., Kumar, R., Niraj, P.K., Anand, G., Singh, S.K., Ghosh, A.K., 2020. High arsenic concentration in blood samples of people of village Gyaspur Mahaji, Patna, Bihar drinking arsenic contaminated water. Springer Nature J Expos Health. 12:131–140. https://doi.org/10.1007/s12403-018-00294-5.

Kumar, A., Ali, M., Kumar, R., Kumar, M., Sagar, P., Pandey, R.K., Akhouri, V., Kumar, V., Anand, G., Niraj, P.K., Rani, R., Kumar, S., Kumar, D., Bishwapriya, A., Ghosh, A.K., 2021[a]. Arsenic exposure in Indo Gangetic plains of Bihar causing increased cancer risk. Scientific reports. 11(1):2376. https://doi.org/10.1038/s41598-021-81579-9.

Kumar, A., Kumar, R., Rahman, M.S., Ali, M., Kumar, R., Nupur, N., Gaurav, A., Raj, V., Anand, G., Niraj, P. K., Kumar, N., Srivastava, A., Biswapriya, A., Chand, G.B., Kumar, D., Rashmi, T., Kumar, S., Sakamoto, M., Ghosh, A.K., 2021[b]. Assessment of arsenic exposure in the population of Sabalpur village of Saran District of Bihar with mitigation approach. Environmental science and pollution research international, 10.1007/s11356-021-13521-5. Advance online publication. https://doi.org/10.1007/s11356-021-13521-5.

Kumar, A., Rahman, M.S., Ali, M., Kumar, R., Niraj, P.K., Akhouri, V., Singh, S.K., Kumar, D., Rashmi, T., Bishwapriya, A., Chand G.B., Sakamoto, M., Ghosh, A.K., 2021[c]. Assessment of arsenic exposure and its mitigation intervention in severely exposed population of Buxar district of Bihar, India. Toxicol. Environ. Health Sci. https://doi.org/10.1007/s13530-021-00086-6.

Kumar, A., Ghosh, A.K., 2021[d]. Assessment of arsenic contamination in groundwater and affected population of Bihar. N. Kumar (ed.), Arsenic Toxicity: Challenges and Solutions. [ISBN 978-981-33-6067-9; ISBN 978-981-33-6068-6 (eBook)] https://doi.org/10.1007/978-981-33-6068-6_7.

Kumar, S., Ghosh, D., Donselaar, M.E., Burgers, F., Ghosh, A.K., 2021[e]. Clay-plug sediment as the locus of arsenic pollution in Holocene alluvial-plain aquifers. CATENA. 202:105255. https://doi.org/10.1016/j.catena.2021.105255.

Kumari, P.B., Singh, Y.K., Mandal, J., Shambhavi, S., Sadhu, S.K., Kumar, R., Ghosh, M., Raj, A., Singh, M., 2021. Determination of safe limit for arsenic contaminated irrigation water using solubility free ion activity model (FIAM) and Tobit Regression Model. Chemosphere. 270:128630. https://doi.org/10.1016/j.chemosphere.2020.128630.

LaKind, J.S., Barraj, L., Tran, N., Aylward, L.L., 2008. Environmental chemicals in people: challenges in interpreting biomonitoring information. Journal of environmental health. 70(9):61–64.

Li, R., Kuo, Y.M., Liu, W.W., Jang, C.S., Zhao, E., Yao, L., 2018. Potential health risk assessment through ingestion and dermal contact arsenic-contaminated groundwater in Jianghan Plain, China. Environmental geochemistry and health. 40(4):1585–1599. https://doi.org/10.1007/s10653-018-0073-4.

Liang, C.P., Wang, S.W., Kao, Y.H., Chen, J.S., 2016. Health risk assessment of groundwater arsenic pollution in southern Taiwan. Environmental geochemistry and health. 38(6):1271–1281. https://doi.org/10.1007/s10653-016-9794-4.

Mahanta C, Enmark G, Nordborg D, Sracek O, Nath B, Nickson RT, Herbert R, Jacks G, Mukherjee A, Ramanathan AL, Choudhury R, Bhattacharya P (2015) Hydrogeochemical controls on mobilization of arsenic in groundwater of a part of Brahmaputra river floodplain, India. J Hydrol. 4:154–171.

McArthur, J.M., Ravenscroft, P., Safiulla, S., Thirlwall, M.F., 2001. Arsenic in groundwater: testing pollution mechanisms for sedimentary aquifers in Bangladesh. Water Resour. Res. 37:109–117. http://dx.doi.org/10.1029/2000WR900270.

McArthur, J.M., Banerjee, D.M., Hudson-Edwards, K.A., Mishra, R., Purohit, R., Ravenscroft, P., Cronin, A., Howarth, R.J., Chatterjee, A., Talkdar, T., Lowry, D., Houghton, S., Chadha, D.K., 2004. Natural organic matter in sedimentary basins and its relation to arsenic in anoxic groundwater: the example of West Bengal and its worldwide implications. Appl. Geochem. 19:1255–1293.

McArthur, J.M., Nath, B., Banerjee, D.M., Purohit, R., Grassineau, N., 2011. Palaeosol control on groundwater flow and pollutant distribution: the example of arsenic. Environ. Sci. Technol. 45:1376–1383. http://dx.doi.org/10.1021/es1032376.

Miller, W.H., Jr, Schipper, H.M., Lee, J.S., Singer, J., Waxman, S., 2002. Mechanisms of action of arsenic trioxide. Cancer research. 62(14):3893–3903.

Morton, J., Carolan, V.A., Gardiner, P.H.E., 2002. Removal of exogenously bound elements from human hair by various washing procedures and determination by inductively coupled plasma mass spectrometry. Anal Chim Acta. 455:23–34.

Mukherjee, A., Sengupta, M.K., Hossain, M.A., Ahamed, S., Das, B., Nayak, B., Lodh, D., Rahman, M.M., Chakraborti, D., 2006. Arsenic contamination in groundwater: a global perspective with emphasis on the Asian scenario. Journal of health, population, and nutrition. 24(2):142–163.

Mukherjee, A., Verma, S., Gupta, S., Henke, K.R., Bhattacharya, P., 2014. Influence of tectonics, sedimentation and aqueous flow cycles on the origin of global groundwater arsenic: paradigms from three continents. J Hydrol. 518:284–99.

Narsimha, A., Rajitha, S., 2018. Spatial distribution and seasonal variation in fluoride enrichment in groundwater and its associated human health risk assessment in Telangana State, South India. 24:2119–2132. https://doi.org/10.1080/10807039.2018.1438176

Nava-Rivera, L.E., Betancourt-Martínez, N.D., Lozoya-Martínez, R., Carranza-Rosales, P., Guzmán-Delgado, N.E., Carranza-Torres, I.E., Delgado-Aguirre, H., Zambrano-Ortíz, J.O., Morán-Martínez, J., 2021. Transgenerational effects in DNA methylation, genotoxicity and reproductive phenotype by chronic arsenic exposure. Scientific reports. 11(1):8276. https://doi.org/10.1038/s41598-021-87677-y.

Nickson, R.T., McArthur, J.M., Ravenscroft, P., Burgess, W.G., Ahmed, K.M., 2000. Mechanism of arsenic release to groundwater, Bangladesh and West Bengal. Appl. Geochem. 15:403–413. http://dx.doi.org/10.1016/S0883-2927(99)00086-4.

NIOSH (National Institute for Occupational Safety and Health), 1994. Elements in blood or tissue. Method: 8005. In: NIOSH manual of analytical methods. 4th Edition. O.H. Cincinnati: Department of Health and Human Service, National Institute of Occupational Safety and Health. DHHS publication. 94-113.

Nowak, B., Kozłowski, H., 1998. Heavy metals in human hair and teeth: the correlation with metal concentration in the environment. Biological trace element research. 62(3):213–228. https://doi.org/10.1007/BF02783972.

Orloff K, Mistry K, Metcalf S (2009) Biomonitoring for environmental exposures to arsenic. J Toxicol Environ Health Part B. 12(7):509–524. https://doi.org/10.1080/10937400903358934.

Postma, D., Larsen, F., Minh Hue, N.T., Thanh Duc, M., Viet, P.H., Nhan, P.Q., Jessen, S., 2007. Arsenic in groundwater of the Red River floodplain, Vietnam: controlling geochemical processes and reactive transport modeling. Geochim. Cosmochim. Acta. 71:5054–5071.

R Core Team, 2020. R: A Language and Environment for Statistical Computing; R Foundation for Statistical Computing: Vienna, Austria. https://www.R-project.org.

Rahman, M.S., Kumar, A., Kumar, R., Ali, M., Ghosh, A.K., Singh, S.K., 2019[a]. Comparative Quantification Study of Arsenic in the Groundwater and Biological Samples of Simri Village of Buxar District, Bihar, India. Indian journal of occupational and environmental medicine. 23(3):126–132. https://doi.org/10.4103/ijoem.IJOEM_240_18.

Rahman, M.S., Kumar, A., Kumar, R., Ali, M., Ghosh, A.K., Singh, S.K., 2019[b] Hematological and Free Radicals Changes among People of Arsenic Endemic Region of Buxar District of Bihar, India. Int. J. Pub. Health Safe. 4:178.

Rasheed, H., Kay, P., Slack, R., Gong, Y.Y., 2019. Assessment of arsenic species in human hair, toenail and urine and their association with water and staple food. Journal of exposure science & environmental epidemiology. 29(5):624–632. https://doi.org/10.1038/s41370-018-0056-7.

Ravenscroft, P., Brammer, H., Richards, K., 2009. Arsenic in Asia. In: Ravenscroft, P., Brammer, H., Richards, K. (Eds.), Arsenic Contamination: A Global Synthesis. Wiley Blackwell, Oxford, UK. 318–386.

Reichard, J.F., Puga, A., 2010. Effects of arsenic exposure on DNA methylation and epigenetic gene regulation. Epigenomics. 2(1):87–104. https://doi.org/10.2217/epi.09.45.

Ren, X., McHale, C.M., Skibola, C.F., Smith, A.H., Smith, M.T., Zhang, L., 2011. An emerging role for epigenetic dysregulation in arsenic toxicity and carcinogenesis. Environmental health perspectives. 119(1):11–19. https://doi.org/10.1289/ehp.1002114.

Roy, N.K., Murphy, A., Costa, M., 2020. Arsenic Methyltransferase and Methylation of Inorganic Arsenic. Biomolecules. 10(9):1351. https://doi.org/10.3390/biom10091351.

Saha, D., Sahu, S., Chandra, P.C., 2011. Arsenic-safe alternate aquifers and their hydraulic characteristics in contaminated areas of Middle Ganga Plain, Eastern India. Environmental monitoring and assessment. 175(1-4):331–348. https://doi.org/10.1007/s10661-010-1535-z.

Salaün, P., Planer-Friedrich, B., van den Berg, C.M., 2007. Inorganic arsenic speciation in water and seawater by anodic stripping voltammetry with a gold microelectrode. Analytica chimica acta. 585(2):312–322. https://doi.org/10.1016/j.aca.2006.12.048.

Salaün, P., Gibbon-Walsh, K.B., Alves, G.M., Soares, H.M., van den Berg, C.M., 2012. Determination of arsenic and antimony in seawater by voltammetric and chronopotentiometric stripping using a vibrated gold microwire electrode. Analytica chimica acta. 746:53–62. https://doi.org/10.1016/j.aca.2012.08.013.

Sanz E, Muñoz-Olivas R, Cámara C, Sengupta MK, Ahamed S (2007) Arsenic speciation in rice, straw, soil, hair and nails samples from the arsenic-affected areas of Middle and Lower Ganga plain. J Environ Sci Health Part A. 42(12):1695–1705. https://doi.org/10.1080/10934520701564178.

Shah, B.A., 2012. Role of Quaternary stratigraphy on arsenic contaminated groundwater from parts of Barak Valley, Assam, North-East India. Environ Earth Sci. 66:2491–250.

Sheikhi, S., Faraji, Z., Aslani, H., 2021. Arsenic health risk assessment and the evaluation of groundwater quality using GWQI and multivariate statistical analysis in rural areas, Hashtroud, Iran. Environ Sci Pollut Res Int. 28(3):3617-3631. doi:10.1007/s11356-020-10710-6.

Sherwood CL, Lantz RC, Burgess JL, Boitano S (2011) Arsenic alters ATP-dependent Ca2+ signaling in human airway epithelial cell wound response. Toxicol Sci: 121(1):191–206. https://doi.org/10.1093/toxsci/kfr044.

Singh, S.K., Ghosh, A.K., Kumar, A., Kislay, K., Kumar, C., Tiwari, R.R., Parwez, R., Kumar, N., Imam, MD., 2014. Groundwater Arsenic contamination and associated health risks in Bihar, India. Int. J. Environ Res. 8(1):49-60.

Suzuki T, Nohara K (2013) Long-term arsenic exposure induces histone H3 Lys9 dimethylation without altering DNA methylation in the promoter region of p16(INK4a) and down-regulates its expression in the liver of mice. J Appl Toxicol: 33(9):951–958. https://doi.org/10.1002/jat.2765.

Tseng, C. H., Huang, Y. K., Huang, Y. L., Chung, C. J., Yang, M. H., Chen, C. J., & Hsueh, Y. M. 2005. Arsenic exposure, urinary arsenic speciation, and peripheral vascular disease in blackfoot disease-hyperendemic villages in Taiwan. Toxicology and applied pharmacology, 206(3), 299–308. https://doi.org/10.1016/j.taap.2004.11.022.

Tsuji, J.S., Chang, E.T., Gentry, P.R., Clewell, H.J., Boffetta, P., Cohen, S.M., 2019. Dose-Response for Assessing the Cancer Risk of Inorganic Arsenic in Drinking Water: The Scientific Basis for Use of a Threshold Approach. Crit. Rev. Toxicol. 49(1):36–84. https://doi.org/10.1080/10408444.2019.1573804.

Ulniković, V.P., Kurilić, S.M., 2020. Heavy metal and metalloid contamination and health risk assessment in spring water on the territory of Belgrade City, Serbia. Environmental geochemistry and health. 42(11):3731–3751. https://doi.org/10.1007/s10653-020-00617-z.

USEPA (United States Environmental Protection Agency), 2012. Integrated Risk Information System. http://cfpub.epa.gov/ncea/iris/index.cfm? fuseaction_iris. show Substance List.

USEPA (United States Environmental Protection Agency), 2014. Human Health Evaluation Manual, Supplemental Guidance: Update of Standard Default Exposure Factors-OSWER Directive 9200.1-120. PP.6

Valenzuela, O. L., Borja-Aburto, V. H., Garcia-Vargas, G. G., Cruz-Gonzalez, M. B., Garcia-Montalvo, E. A., Calderon-Aranda, E. S., & Del Razo, L. M. 2005. Urinary trivalent methylated arsenic species in a population chronically exposed to inorganic arsenic. Environmental health perspectives, 113(3), 250–254. https://doi.org/10.1289/ehp.7519.

Wang X, Mu X, Zhang J, Huang Q, Alamdar A, Tian M, Liu L, Shen H (2015) Serum metabolomics reveals that arsenic exposure disrupted lipid and amino acid metabolism in rats: a step forward in understanding chronic arsenic toxicity. Metallomics: 7(3):544–552. https://doi.org/10.1039/c5mt00002e.

World Health Organization (WHO). 2001. Arsenic and Arsenic Compounds, 2nd edn. Environmental Health Criteria 224. Geneva Available at http://www.inchem.org/documents/ehc/ehc/ehc224.htm accessed April 2 2008.

Wei, T., Simko, V., 2021. R package "corrplot": Visualization of a Correlation Matrix (Version 0.88). https://github.com/taiyun/corrplot.

Yang, X., Duan, J., Wang, L., Li, W., Guan, J., Beecham, S., Mulcahy, D., 2015. Heavy metal pollution and health risk assessment in the Wei River in China. Environmental monitoring and assessment. 187(3):111. https://doi.org/10.1007/s10661-014-4202-y.

Yoshinaga, J., Imai, H., Nakazawa, M., Suzuki, T., Morita, M., 1990. Lack of significantly positive correlations between elemental concentrations in hair and in organs. The Science of the total environment. 99(1-2):125–135. https://doi.org/10.1016/0048-9697(90)90217-i.

FIGURES AND TABLES

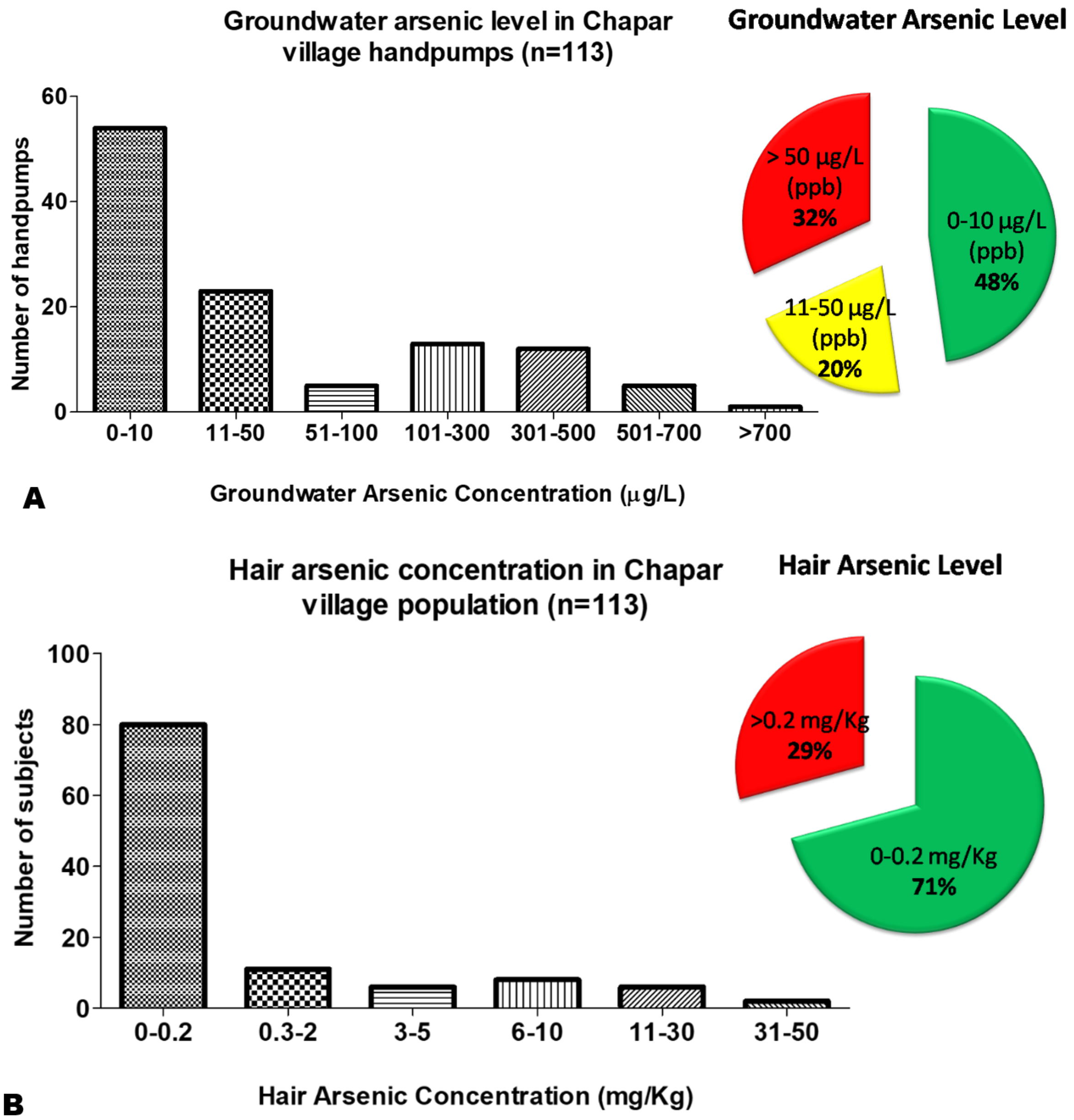


Figure 1. [A]. *Groundwater arsenic concentration in handpumps.* **[B].** *Hair arsenic concentration of Chapar village analysed through GF-AAS (ANOVA- Dunnett's Test, P<0.05).*

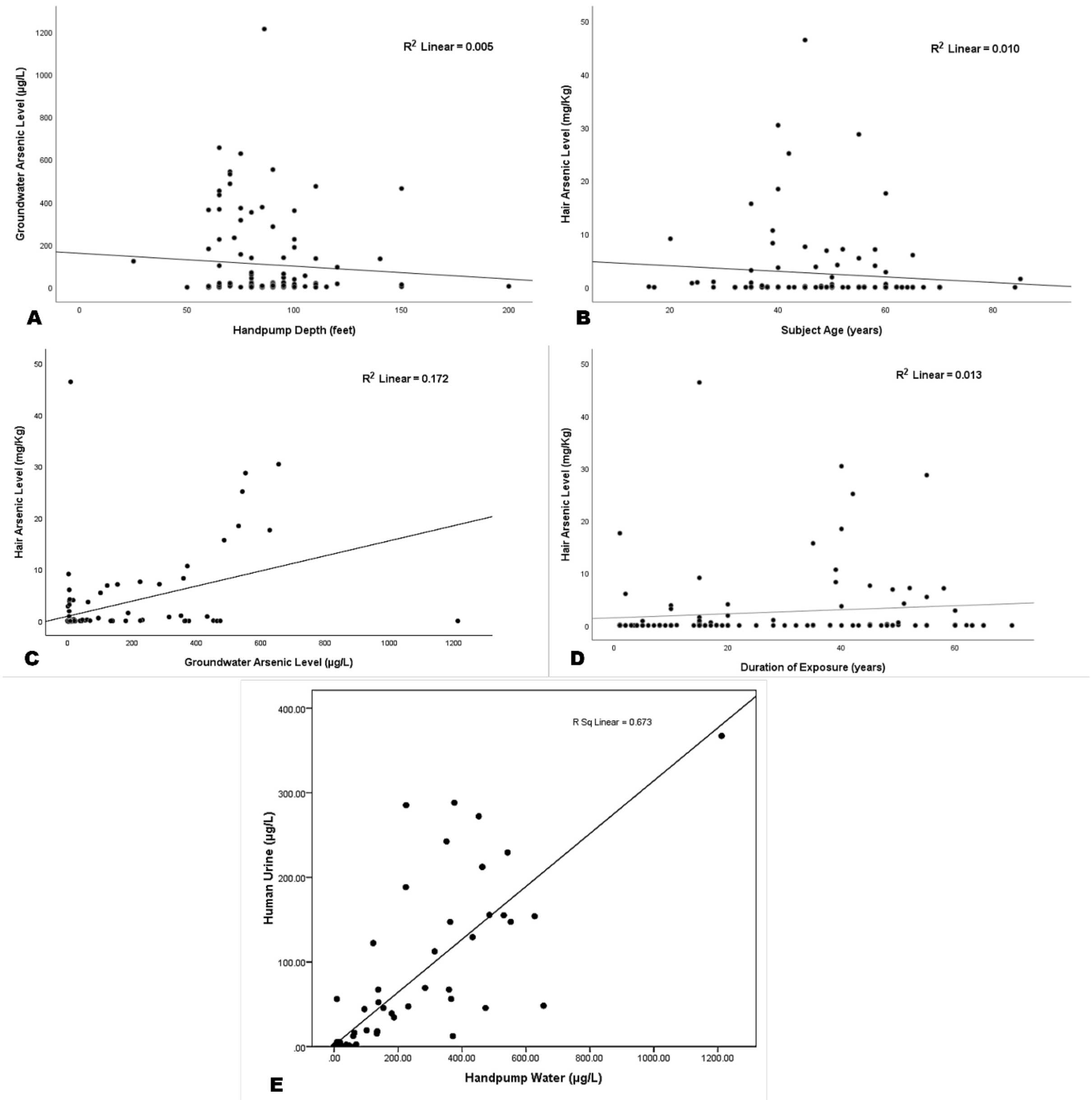


Figure 2. *The scatterplot graph of* **[A]** *Groundwater arsenic concentration and depth of the handpumps (r=0.005 & P<0.05)* **[B]** *Hair arsenic concentrations and age of the subjects (r=0.010 & P<0.05)* **[C]** *Hair arsenic level and groundwater arsenic concentration (r=0.172 & P<0.05)* **[D]** *Hair arsenic level and duration of arsenic exposure (r=0.013 & P<0.05).* **[E]** *Urine arsenic level and groundwater arsenic concentration (r=0.673 & P<0.05).*

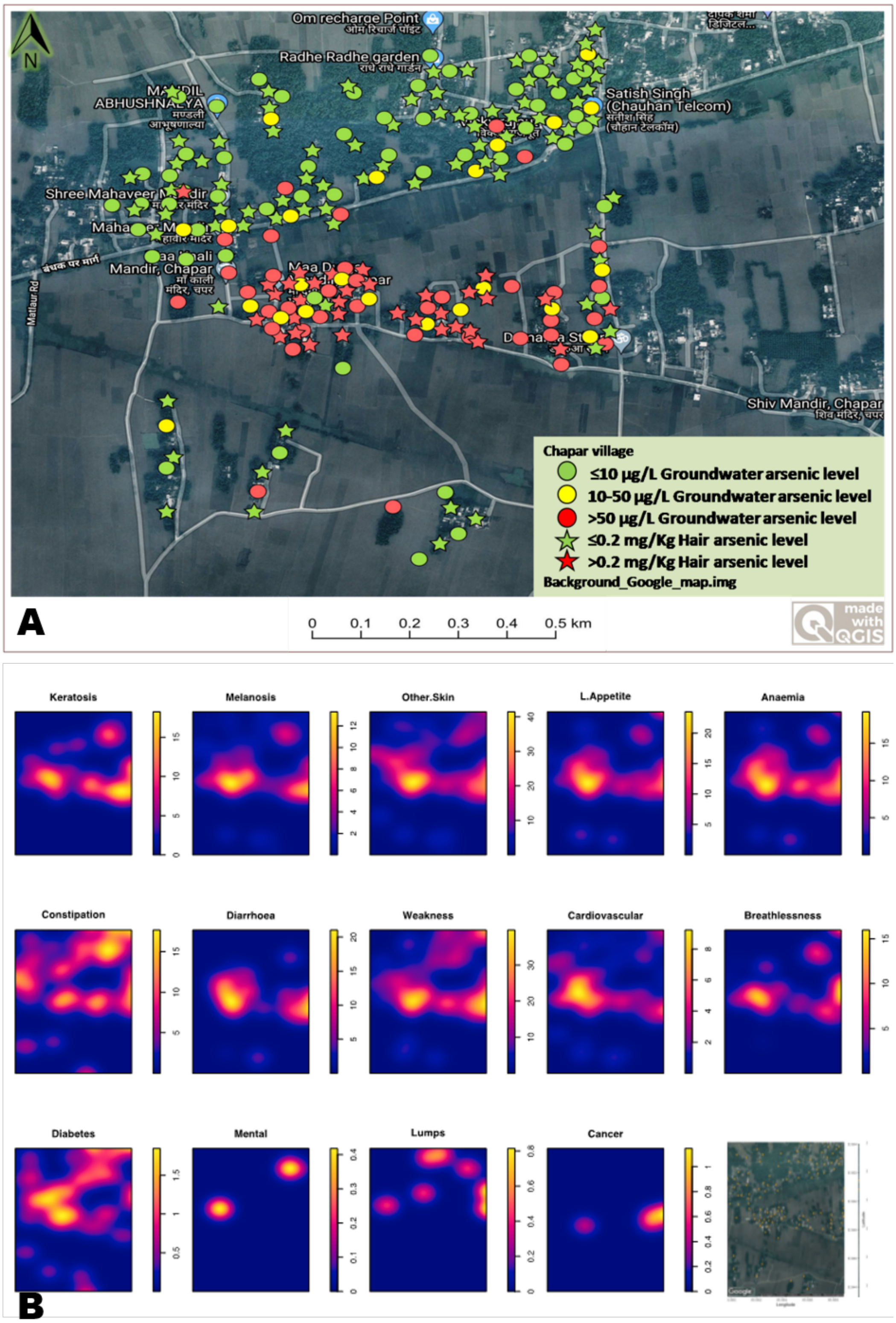
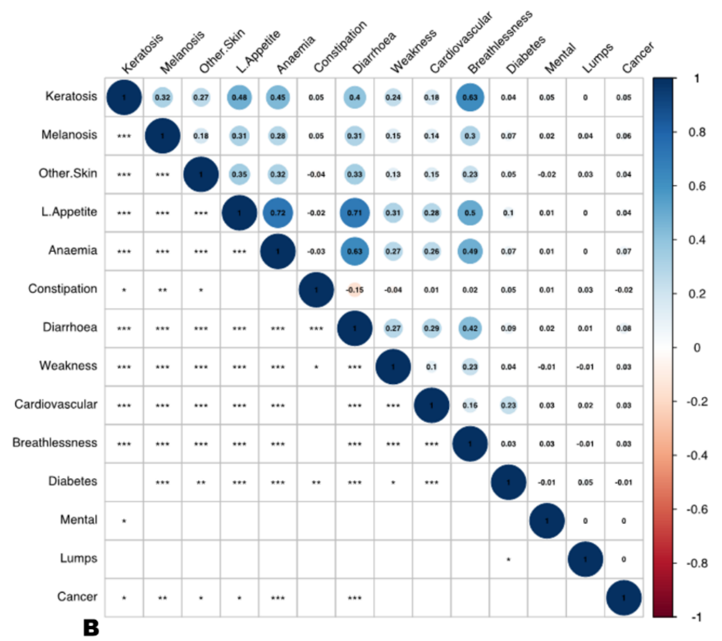


Figure 3. **[A]** *Thematic map showing synoptic view of arsenic level in groundwater and hair samples superimposed using QGIS software (version 3.10.1-A Coruna).* **[B]** *Kernel density maps of the symptoms. The colour indicates the expected number of patients with each symptom per 10,000 square metres. The right-bottom map shows the household locations.*

Figure 4. [A, B, & C] *Arsenicosis symptoms in members from a single household* **[D]** *Arsenicosis symptoms in exposed village population.*

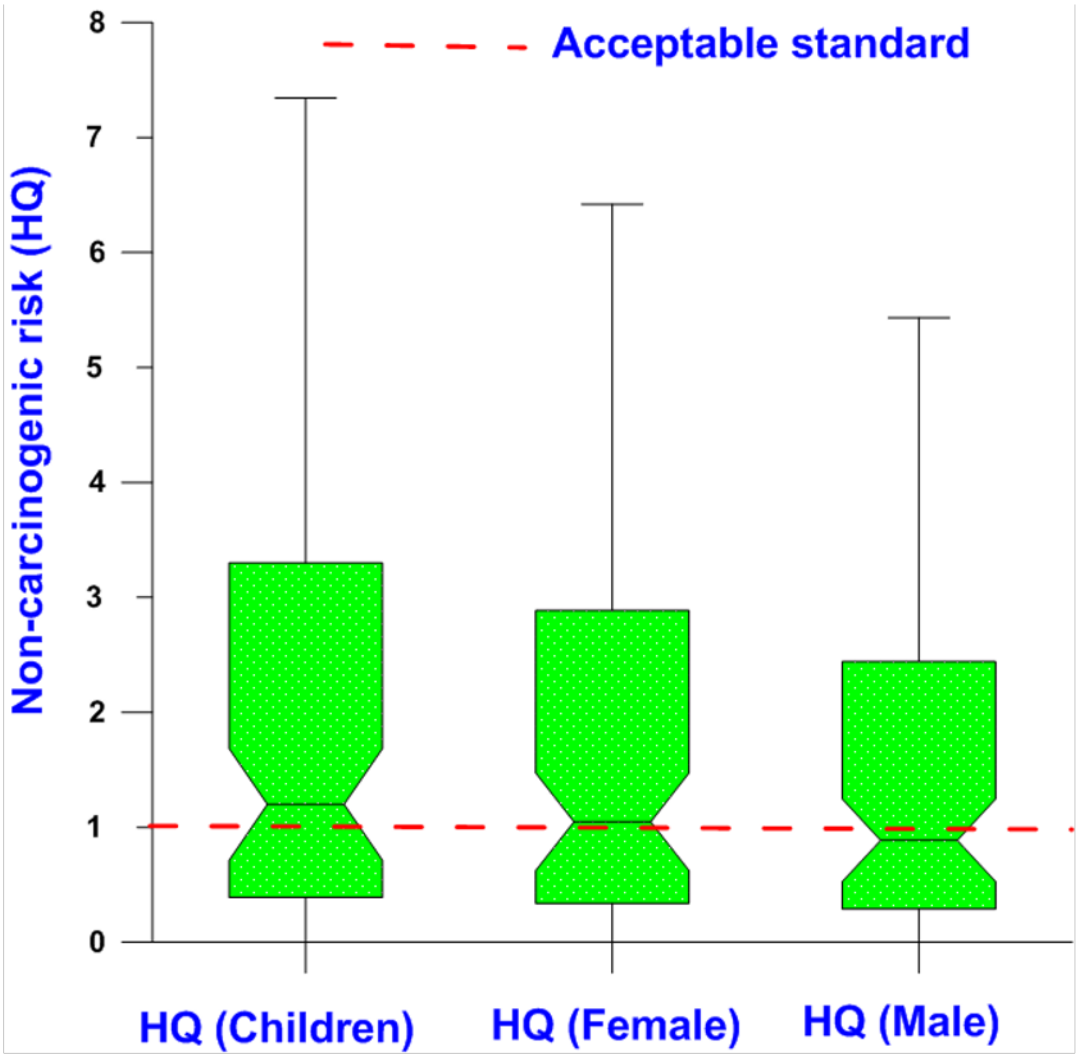
Symptoms	Problems present in the population	Number of Problems Observed	Total Cases	P Value
Keratosis in palm and sole	315 (11.2%)	2501 (88.8%)	2816	< 0.001
Melanosis in sole, palm and trunk	242 (8.6%)	2574 (91.4%)	2816	< 0.001
Other skin manifestations	1272 (45.2%)	1544 (54.8%)	2816	< 0.001
Loss of appetite	411 (14.6%)	2405 (85.4%)	2816	
Anaemia	371(13.2%)	2445 (86.8%)	2816	< 0.001
Constipation	513 (18.23%)	2303 (81.77)	2816	< 0.001
Diarrhoea	273 (9.7%)	2543 (90.3%)	2816	< 0.001
General Body Weakness	1616 (57.4%)	1200 (42.6%)	2816	< 0.0001
Cardiovascular disorders (raised blood pressure)	180 (6.4%)	2636 (93.6%)	2816	< 0.001
Breathlessness	199 (7.1%)	2617 (92.9%)	2816	< 0.001
Diabetes	53 (1.88%)	2763 (98.2%)	2816	< 0.001
Mental Disability	04 (0.14%)	2812 (99.86%)	2816	< 0.0001
Lumps in the body	07 (0.24%)	2809 (99.76%)	2816	< 0.001
Cancer	11 (0.39%)	2805 (99.61%)	2816	< 0.001
Other Health Problems	405 (14.4%)	2411 (85.6%)	2816	< 0.001

A



B

Figure 5. [A] *Table showing health assessment report of arsenic exposed population of the village Chapar (n=2,816) from households (n=250)* **[B]** *Symptoms association heatmap- The numbers represent the Spearman rank correlation coefficients and the asterisks indicate the significance levels (* p<0.05, ** p<0.01, *** p<0.001). R version 4.0.3 (R Core Team, 2020) was used for the statistical calculation, and the result was visualized with the R package "corrplot" version 0.88 (Wei and Simko, 2021).*



A

B

Parameters	Male	Female	Children	References
IR	2.5	2.5	0.78	USEPA, 2014
ED	64	67	12	Narsimha and Rajitha, 2018
EF	356	365	365	USEPA, 2014
BW	65	55	15	ICMR, 2009
ET	23360	24455	4380	USEPA, 2014
C(mg/L)				Present study

Figure 6. [A] *Human health risk assessment non-carcinogenic vs. carcinogenic risk* **[B]** *Table showing human health risk assessment.*

Installed Filter Units	Depth (Feet)	Tank Capacity (Litres)	Household catering capacity through piped water distribution	GPS Location	Arsenic Concentration in the Inlet water of the filter as tested on 31.07.2021 (µg/L)	Arsenic Concentration in the Outlet water of the filter as tested on 31.07.2021 (µg/L)
Arsenic Filter Unit - I	350	10,000	150	N25.54921, E85.66608	15.03	5.115
Arsenic Filter Unit - II	350	10,000	100	N25.54724, E85.67088	13.82	8.387

Figure 7. [A] *Showing 02 mitigation units (Unit-I with the caretaker) installed for the arsenic exposed population of Chapar village, Harail Panchayat of Samastipur district of Bihar, India.* **[B]** *Figure showing present data of the arsenic content in the ground water samples of the arsenic filter units.*

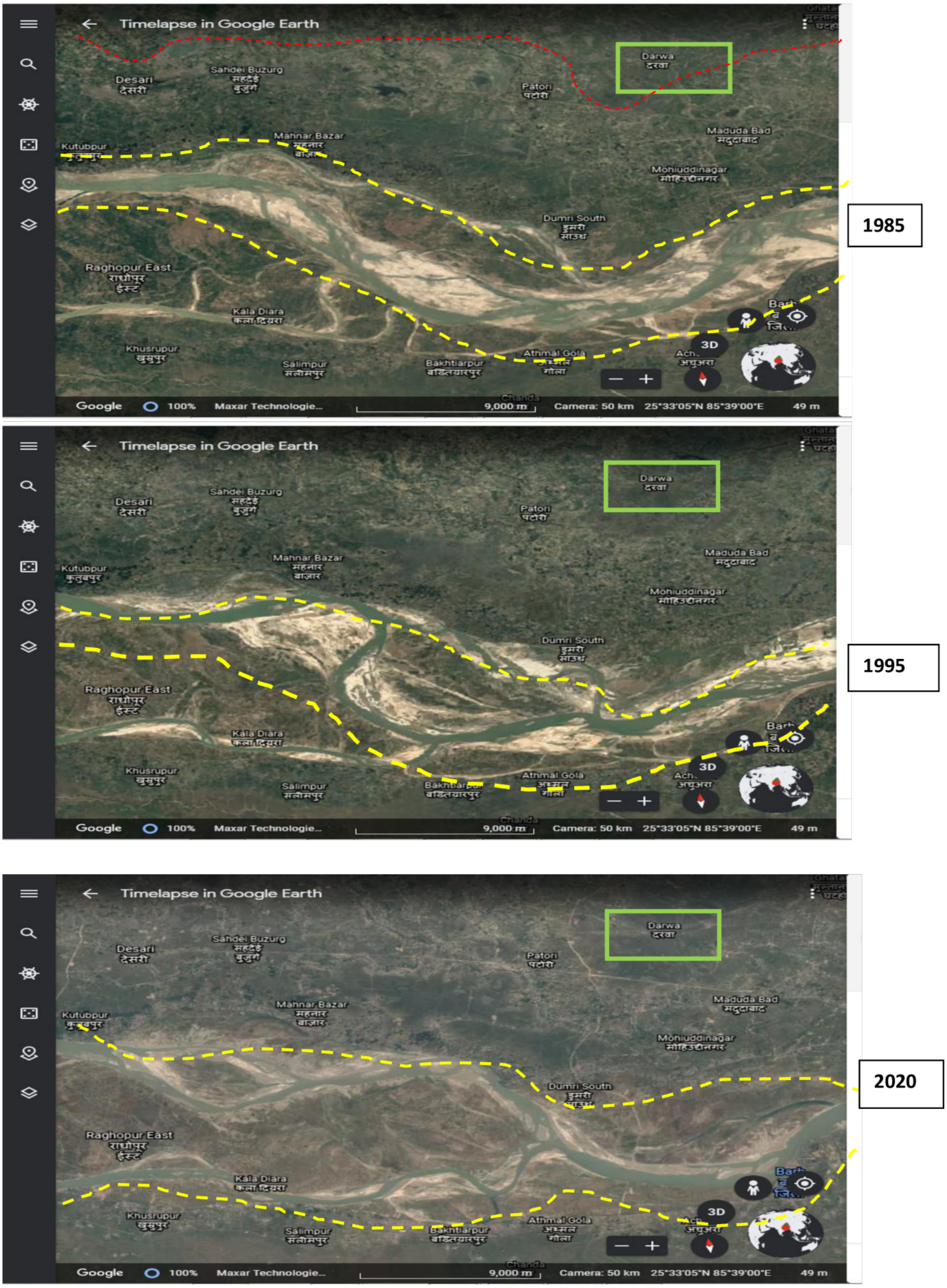


Figure 8. Google earth imagery showing temporal variation along the oscillation zone of river Ganga (*Using Google Earth software)*