

# Identifying a Less Time Consuming and Better Technique Extracting Arsenic from Human Milk Samples

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**ABSTRACT:** In this article, the arsenic levels in drinking water were studied. Hair and nail arsenic is highly linked, however, according to principle component analysis (PCA). According to our results, lactating women with hypertension greater levels. Arsenic levels in babies' blood, nails, and urine were investigated, and the results showed a significantly high body burden of arsenic in areas. The hair and nail arsenic levels were found to be age-dependent using PCA. The techniques for extracting arsenic from human milk samples are successful in both literatures. These findings would assist in the development of a quicker and more efficient method for eliminating arsenic from milk samples. Although the process will take longer than expected, it does not surpass 5-6 hours. Furthermore, such investigations show the degree of arsenic exposure in people living in complex environments. People are exposed to less arsenic as a consequence of this, which may lead to severe instances of chronic arsenic poisoning. Prenatal exposure to infants may potentially induce genetic abnormalities as well as physical and mental defects.

**KEYWORDS:** Arsenic Levels, Drinking Water, Human Milk, Samples, Skin Lesions, West Bengal.

## 1. INTRODUCTION

Arsenic was employed as a therapeutic agent by the ancient Greeks and Romans because it was regarded a type of Sulphur. Arsenic trioxide has all of the qualities of a perfect homicidal poison, giving it the nickname "King of Poisons". Women were believed to purchase arsenic to kill rats, with the rodents representing their spouses. In 1836, a Marsh investigation ruled out the deadly toxin that Arsenic is employed for. Arsenic trioxide is harmful to living species, although metallic arsenic is not dangerous since it is insoluble in liquids. Arsenic is a naturally occurring toxin that may be found in both soil and water. Humans in the Indian Subcontinent are primarily exposed to arsenic through their drinking water. The majority of people in Southeast Asia are at high risk of arsenic poisoning [1]. A number of epidemiological studies have revealed that inorganic arsenic is a powerful carcinogen, having effects on the urinary bladder, lungs, skin, as well as possibly the kidneys as well. Hyperpigmentation, hypopigmentation, and keratosis indicate early poisoning. In the globe, arsenic may be utilized in a numerous ways. A list of them is given below. Exposure of both the toxin and the carcinogen has severe health effects in humans (see Table 1).

**Table 1: Arsenic Can Be Used In A Variety Of Ways In The World. The Following Is A List of Them. In Humans, Exposure To This Toxin As Well As The Carcinogen Has Serious Health Consequences.**

Arsenic trioxide	$As_2O_3$
Arsenite	$AsO_2^-$
Arsenate	$H_2AsO_4^-$
Arsenic trisulphide	$As_2S_3$
Gallium arsenide	$GaAs$
Arsine	$AsH_3$
Methylarsonic acid	$CH_3AsO(OH)_2$
Dimethylarsinic acid	$(CH_3)_2AsO(OH)$
Trimethylarsenine	$(CH_3)_3As$
Trimethylarsenine oxide	$(CH_3)_3As=O$
Arsinobetaine	$(CH_3)_3As^+CH_2COO^-$
Arsinocholine	$(CH_3)_3As^+CH_2CH_2OHX^-$

Between April 1995 and March 1996, a cross-sectional study was conducted in West Bengal, India, to investigate arsenic associated skin lesions of keratosis and hyperpigmentation and their connection to arsenic water levels. In all, 7683 individuals were tested and questioned, and the amounts of arsenic in their drinking water were determined. Despite the fact that arsenic contents in the water varied up to 3400 g/l, over 80 percent of the subjects drank water with less than 500 g/l. The age-adjusted incidence strongly related, rising category drinking water containing > 800 g/l) exposed to arsenic levels of 100 g/l in their drinking water. Hyperpigmentation findings were close, with excellent dose-response correlations. 29 instances of hyperpigmentation were subjected to drinking water with a concentration of 100 g/l. Men had approximately rate dosage, according to estimates based on dose per body weight. The prevalence of keratoses was 1.6 times greater in individuals who were under 80 percent of the normal, indicating that starvation may play a small increase. The surprising finding linked with individuals who had a modest exposure to arsenic in childhood verified in trials of more comprehensive exposure evaluations. More study on susceptibility is greatly required and have been done [2].

### 1.1. Toxic Salts of Arsenic:

#### 1.1.1. Arsenious Oxide[As<sub>2</sub>O<sub>3</sub>]

- Occurs in powder as well as porcelain forms.
- White coloured, tasteless and odourless
- Soluble in water

#### 1.1.2. Arsenic acid[H<sub>3</sub>AsO<sub>4</sub>]

#### 1.1.3. Arsenic trichloride [AsCl<sub>3</sub>]

- Butter of arsenic
- Colorless poisonous oil

#### 1.1.4. Arsenic trisulphide[As<sub>2</sub>S<sub>3</sub>]- mineral orpiment

#### 1.1.5. Arsine[AsH<sub>3</sub>]- toxic gas

#### 1.1.6. Arsenobetaine

#### 1.1.7. Arsphenamine

- Used to treat syphilis and trypanosomiasis
- First chemotherapeutic agent

#### 1.1.8. Arsenalic acid

- Colourless solid
- Organoarsenic compound
- Conjugated with ovalbumin to treat arsenic poisoning

#### 1.1.9. Copper acetoarsenite-insecticide and pigment

#### 1.1.10. Copper arsenite

#### 1.1.11. Melarsoprol-organic arsenical used to treat trypanosomiasis

#### 1.1.12. Tetraarsenic tetrasulphide[As<sub>4</sub>S<sub>4</sub>]-mineral realgar (manseel)

Arsenic levels in groundwater are over the World Health Organization's maximum permitted limit of 50 g/L in nine districts in West Bengal, India, and 42 districts in Bangladesh. The experts observed area and population of Bangladesh's 42 districts and West Bengal's 9 districts. Researcher discovered throughout during our preliminary examination. Researcher discovered throughout in Bangladesh. Researcher's collected 10,991 water samples for research from 42 arsenic-affected districts in Bangladesh, and 58,166 water samples from nine arsenic-affected districts in West Bengal. Arsenic levels over 50 g/L were detected in 59 and 34 percent of the water samples were examined, respectively. Thousands of scalp, examined to date; on average, 93 and 77 percent of samples had arsenic over the normal/toxic threshold in Bangladesh and West Bengal,

respectively. Researcher searched for arsenic patients in 27 of Bangladesh's 42 districts, and researcher discovered individuals with arsenical skin lesions in 25 of them. Researcher randomly tested; 24.47 and 15.02 percent, respectively. Researcher feel that they have only touched the top of the iceberg after 10 years of study in Bangladesh and five years in West Bengal [3].

Despite early survey studies pollution findings in the Terai region of Nepal, the pandemic of arsenic poisoning due to contaminated was thought to be confined to the Ganges Delta. Researcher examined arsenic toxicity showed that 56.8 percent of them above 50 g/L. Upon health assessment, 13 percent of adults and 6.3 percent of children in a self-selected sample of 550 showed normal skin lesions, an exceptionally high of, even in acute exposures linked. Arsenic concentrations in urine, blood, and nails were revealed to be significantly linked with detected in 63 percent of the adults on neurologic evaluation, prevalence previously only seen in severe. Researcher has observed an increase in fetal death born in the mothers who had levels in their drinking water. Other regions in the Middle and Upper Ganga Plain should be examined for the danger of contaminated groundwater [4].

Mass arsenic poisoning instances have been recorded in the past 2-3 decades which have proven catastrophic to the society in certain regions. Reports of such instances have been highest in the South East Asian nations and the Indian Subcontinent. Arsenic poisoning is found frequently in Bangladesh and other poorly developed nations. World Health Organisation (WHO) estimates that the exposure of Arsenic to Bangladeshi people ranges from 28-35 million people to 77 million people which is nearly half the entire population. This is not just confined to poor nations. In Western regions of United States of America (USA), 13 million people consume arsenic contaminated water. Though this is not a large amount as compared to Bangladesh, it is nonetheless a rising mortality [5].

Also, it is a well-known fact that the quantity of arsenic poisoning observed in poorly developed nations and particularly regions near the dumping sites of businesses is the highest. Even if the amount of arsenic in the soil, water or any other consumable biological material may be minimal on examination, the frequent intake of the same can lead to severe poisoning. This in turn impairs the general health of the people living in and around the region and also poses a danger to the future generations' entirely [6].

## 2. LITERATURE REVIEW

G. Samanta *et al.* presented in the article those two hundred samples of collected from lactating mothers in three West Bengal's arsenic-affected areas. Just 39 of the 226 samples tested positive for arsenic. The arsenic body load of nursing mothers was evaluated using urine, scalp, and nail samples. Arsenic levels in drinking water were also studied. Hair and nail arsenic is highly linked, however, according to principle component analysis (PCA) (PCA). According to our results, lactating mothers with high arsenic body pressure higher levels. Arsenic levels in babies' blood, analyzed, and the results showed a significantly increased body burden of arsenic in those areas. The hair and nail arsenic levels of the mothers and their infants were found to be age dependent utilizing PCA [7].

M. R. Islam *et al.* stated in the article that inorganic arsenic poisoning during infancy may be related to significant health consequences later in adulthood. However, there is a paucity of evidence on postnatal arsenic poisoning via human milk. The objective of this study was to determine the amounts of arsenic in human milk and the connection between arsenic in human milk and arsenic in the urine of mothers and infants. This prospective research identified 120 new mother baby couples in Kashiani (subdistrict), Bangladesh, between March 2011 and March 2012. Thirty women were selected at random for human milk samples at one, six, and nine months after delivery, and chosen one and six months. For arsenic speciation, 12 selected at random. The amount of arsenic in human milk was low and irregularly distributed. For all three phase phases, stood at 1/2 gram/Liter. Arsenic concentration dropped by 0.035 g/L between 1 and 6 months and between 6 and 9 months, according to mixed model estimations. The amount of arsenic observed in an infant's urine increased by 0.13 g/L over time, which was not statistically significant (95 percent) (95 percent). Arsenic in foetal milk was not related with arsenic in the 6 months ( $r = 0.09$  at 6 months). Total urinary arsenic was made up of arsenite (As III), arsenate (As V), monomethyl arsonic acid (MMA), and arsenobetaine (AsB); dimethyl arsinic acid (DMA) was the most prevalent arsenic metabolite in kid urine. Arsenic levels in human milk were determined to be low. The concentration was lower than the limit established by the World Health Organization (WHO) (WHO). Also in arsenic-contaminated settings, our findings confirm the safety of breastfeeding [8].

P. Verma *et al.* presented in the article that arsenic is a radioactive metal that is present in many natural settings. It is a human carcinogen in water across a wide pH range, and even at low quantities, it has severe

impacts on both human health and the environment. To safeguard consumers served by municipal water systems, WHO as well as the US Environmental Protection Agency (USEPA) established the arsenic limit for drinking water at 0.010 particles per million. Arsenic-contaminated water has been reported in many nations, including Bangladesh having the highest susceptible population, followed by India (West Bengal) (West Bengal). For arsenic poisoning, there is no suitable treatment. And by eliminating arsenic from the water can the poisoning be prevented. While considerable research has been done in recent decades to decrease arsenic levels in drinking water, there is still a need to develop low-cost viable methods. Oxidation, adsorption, absorption, coagulation, and membrane separation are some of the major arsenic reduction methods presently in use. Recent research in the area of arsenic recovery from contaminated water, as well as a review of all available methods, with an emphasis on adsorption [9].

H. R. Lohokare *et al.* stated in the article that to our knowledge, this is the first time has been proven to be efficient in eliminating arsenic. By utilizing the Donnan exclusion principle, the surfaces by NaOH resulted in the formation of carboxylate (single bond COO) capacity, resulting in As-V rejection capacity. The decrease-increase polyethylene glycol (PEG) rejection revealed a drop in pore capacity. Fourier Transform Infrared – Attenuated Total Reflection (FTIR-ATR) research showed that NaOH treatment resulted in the development of a carboxylate group on the membrane surface, while contact angle measurements revealed enhanced hydrophilicity. SEM and AFM measurements showed that this approach resulted in smoothing of the membrane interface. After the NaOH process, the molecular weight limit was found to be approximately 6 kDa. With various feed concentrations, rate, strain, the rejection of pentavalent arsenic studied. Experiments with 50 ppb As-V in feed showed almost 100 percent and remained constant for up to 6 hours. The rejection coefficient was not influenced by strain for arsenic concentrations less than 50 ppm in the diet. As the concentration polarization was substantial, the rejection for 1000 ppm As-V concentration varied from 40 to 65 percent depending on strain [10].

### 3. DISCUSSION

The main objective of the study is to look at the different methods for removing arsenic from human milk. The study also includes an in-depth assessment of the existing extraction methods available today, as well as potential enhancements and modifications that might be made to the processes. Additional stages are designed in the procedures since the goal of the study is to develop a novel pattern in arsenic extraction while keeping laboratory facilities in mind. When adding the stages, the time spent is frequently taken into account, because it is important in the given situation.

#### 3.1. Mechanism of Action:

- Absorbed via all ports of entry (oral, nasal, cutaneous) (oral, nasal, cutaneous)
- Trivalent Arsenic (As<sup>3+</sup>) possesses affinity to sulfhydryl groups and bind to the mitochondrial membrane SH groups and destroys them. Cytochrome c is released which activates caspases and leads in apoptosis.
- Downregulates gene expression of B-Cell Lymphoma 2 (BCL2), a pro-survival protein.
- Inhibits pyruvate dehydrogenase complex resulting to impairment of oxidative phosphorylation.
- Replaces phosphorous in the bone where it stays for extended period of time.

Monitor samples were collected from two communities with arsenic-free groundwater. Table 2 shows the arsenic concentrations in the control samples. Both water samples showed arsenic concentrations level, and the (maximum 36 g/L). Just 5 of the 10 contained arsenic, with a mean arsenic content of 5.0 g/L.

**Table 2: Water, Urine, and Breast Milk Samples Were Obtained From Non-Affected West Bengal Residents [Village: Padmatamli, And Srirampur, District: Medinipur].**

Volunteer	Age (years)	Arsenic in breast milk (µg/L)	Arsenic in wine (µg/L)	Arsenic in drinking water (µg/L)
1	30	Not detected	15	<3
2	36	Not detected	10	<3
3	20	3.5	17	<3
4	34	5	30	<3
5	30	4	34	<3
6	33	Not detected	18	<3
7	21	3	12	<3
8	27	Not detected	8	<3
9	30	2	22	<3
10	26	Not detected	36	<3

Two major scientific literatures are of vital importance from the viewpoint of a research. The following are the details. The study's goal was to isolate and analyze the quantity of arsenic in milk, as well as the consequences on infants and nursing women. Previous investigations of the presence of arsenic in ground water in Kashiani Sub District in Gopalganj, Bangladesh, led to the selection of the study site. It lasted 13 months, and 120 mother-infant couples were selected to participate. Thirty couples were selected at random from the 120 to collect milk samples. Each mother's milk was collected ethically and kept in an ice box after being securely sealed with paraffin and appropriately marked. Ultrex high purity Nitric Acid HNO<sub>3</sub> was utilized in the extraction procedure. 2 mL of extremely pure H<sub>2</sub>O<sub>2</sub> was added to the tubes, which were then heated for 20 minutes in a microwave oven at 1600W power and 180°C. The combination was then allowed to cool for 10 minutes before being done twice more. Using ultra-pure (18.2M) water, the volume of each sample was adjusted to 5mL. Arsenic was identified in the samples utilizing the Graphite Furnace-Atomic Absorption Spectroscopy (AAS).

The extraction procedure includes three phases of wet digestion, followed by volume adjustments for the analytical technique to be employed. Because of the heavy metal toxicity of the region's ground water, it's not surprising if additional metals like Mercury, Lead, or Cadmium are found in the tests owing to long-term exposure. Since a consequence, thorough digestion and analysis are required for Arsenic determination, as preliminary testing will result in false positive findings when performed. The study focuses on the extraction and analysis of arsenic in milk and water in the North-24 Paraganas area of West Bengal, where 20 of the 22 blocks had arsenic contamination in drinking water above 50 µg/L, and individuals in 16 of the blocks have acquired arsenical skin lesions. A total of 226 lactating women were selected for the study, which took place between 1996 and 2006. Arsenical skin lesions had developed in 15 of the 226 mothers. Every mother's milk was collected in polypropylene containers and kept at -20°C in the laboratory. The samples were defrosted overnight at 28-30°C before being homogenised for 10 minutes in a vortex the following day. In 2-3mL of pre-washed Teflon Vessel, 1mL Supra-pure H<sub>2</sub>O<sub>2</sub> was added. The mixture's hue changed from yellow to colourless. After that, the containers were heated to 110°C for 24 hours. The sample volume was reduced to approximately 0.5 mL, and was then filled with pure deionized water. The presence of arsenic was detected in the samples.

In this study, the extraction of arsenic from milk took approximately three days in total. This is also without taking into consideration the time required to maintain the samples in the laboratory prior to extraction. The area under examination has high levels of arsenic poisoning, which requires a comprehensive study. There is a considerable difference in the time periods utilized in both experiments. The extraction of arsenic from milk samples takes approximately 3 hours in the first analysis. The extraction of the same from the samples in the second analysis takes approximately three days and is a time intensive process. The materials utilized in both methods are different. The materials and chemicals employed in the first study suggest that the extraction was

conducted in a high-performance, well-equipped laboratory. The second study, on the other hand, uses more conventional and easy-to-handle materials. The technique employed in the first analysis is a well-established approach for extracting arsenic. However, the second study advises utilizing conventional methods for the same. In the first sample, arsenic was found in very tiny quantities as well. The identification of arsenic was not detected in any samples in the second study, despite high levels of arsenic toxicity in the regions where the samples were collected.

#### 4. CONCLUSION

This article demonstrates that the techniques for removing arsenic from human milk samples are effective. These results assist in the development of a less time-consuming and more efficient technique for extracting arsenic from milk samples. A few modifications to the processes described may also be implemented if required. Employing a water bath instead of a microwave oven and using more nitric acid and hydrogen peroxide as a more conventional but less time-consuming technique of extraction may be beneficial. This therapy will take longer than expected, but it should not take more than 5-6 hours. Furthermore, studies of this type reveal the amount of arsenic concentration in people living in particular settings. This tends to minimize the amount of arsenic individuals are exposed to, which may result in severe instances of chronic arsenic poisoning. Furthermore, prenatal exposure to infants may result in a range of genetic abnormalities as well as physical and mental impairments. Scalp, and nails were high, and as they got older, the amounts of arsenic rose. This may be linked to an increase in arsenic-contaminated water or food consumption as individuals grow older. As a consequence, more study is required to assess causes larger cohort of infants.

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