



Arsenic Toxicity in Animals Through Food Chain in Two Endemic Districts of West Bengal, India

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Abstract

To investigate the level of arsenic in biological samples such as grass, water, and hair/wool, samples were collected from animals of Nadia and East Bardhaman districts and estimated with the Atomic absorption spectrometer and to investigate the appearance of clinical signs in animals. A total of 145 hair and wool, 15 grass, and 10 water samples were collected. Hair/wool samples collected from goats, sheep, and cattle were 104,39 and 13, respectively. The average Arsenic concentration (ppb) in wool and hair, grasses, and water were 856.556 (0.85655 mg/kg), 1227.75 (1.22775 mg/kg), and 29.720 (0.02972), respectively through atomic absorption spectrophotometry. The arsenic concentration in hair of goats, wool, and hair of cattle were 931.4608, 899.7414, and 1181.51,7 respectively. The average arsenic concentration in grasses in Nadia and East Bardhaman was 1323.00 and 1037.25, respectively, which are higher. The arsenic concentrations in water of Nadia and East Bardhaman were 0.020 and 0.039, respectively, from deep tube wells. The affected animals with high arsenic content showed chronic diarrhea, ataxia, dehydration, loss of condition, dryness, lusterless of skin coat, weakness, emaciation, ulcerative skin, change in wool and hooves. The study revealed that plants get Arsenic from soil and water, which is transferred to animals through fodder, feed, water and to humans through animal food and also through contaminated water ingestion.

Keywords: Arsenicosis; Sheep; Goat; Cattle; Water; Fodder

Introduction

Arsenic is a naturally occurring metalloid with the symbol of 'As', atomic weight 74.92. An arsenicosis in plants and humans is worldwide. Several States of India are affected by arsenic, particularly in the Eastern states of West Bengal, Bihar, Uttar Pradesh, Jharkhand, and Assam are highly affected. In West Bengal, nine different districts, including Nadia, Murshidabad, East Bardhaman, Malda, Hoogly, Howrah, North 24 Pargana, and South 24 Pargana, including 83 blocks, are affected by arsenicosis

through water and food. Animals and humans can be contaminated through a wide range of contaminants bioaccumulation, diet/feed, soil, water, erosion of arsenic salts in hills and pebbles, industrial sources, bioaccumulate arsenic plants like Breken fern, Arum (*Colocasia antiquorum*), Kalmi sak (*Ipomoea sp*), maize, barley, ryegrass etc and sources of human food arsenic are many like crab, sea fish, mollusca, shellfish, sea algae etc. Arsenic being highly toxic metalloid affect human, animal and plant health that is why the drinking water should be minimum contaminant, it should be

maximum 10 µg/L or 0.01 mg/L as per WHO recommendation [1] as well as US Environmental Protection Agency (EPA) also recommended this standard who earlier recommended 50µg/L but due accumulation of arsenic in several organs clinical signs appears later [2]. Several countries, including India, Bangladesh, China, Vietnam, and Latin American countries where groundwater is contaminated with Arsenic. The rapid contamination Arsenic in ground water and feed affects animal health with clinical signs of dermatoses, keratoses, neurological, skin and hair pathology, incoordination, posterior paresis, inappetence, increased heart rate, respiratory rate, reduced ruminal motility, chronically loose stool, dermatosis, salivation, stiffness of gait, lameness and coffee color urine, though in animals very clear clinical signs are rare except loss of production if not severely affected [3]. Therefore, the animals maintained in an arsenic-contaminated water and feed regime hamper the productivity and require restricting the use of contaminated feed and water. Agriculture and animal farming, including fisher need to be done possibly with soft water, and public health water supply needs to be through refined surface water [4]. Therefore, studies in Arsenic contamination in underground, feed, and animal sources must be conducted to restrict arsenicosis in animals.

Materials and Methods

Selection of study area

The average Arsenic concentration of Nadia and East Bardhaman districts is reported to be higher. In these two districts, the groundwater arsenic levels are 500-1000 and 100-500 µg/L [5]. We selected the villages of Nadia district are Saguna, Kalyani, Silinda of Kalyani, and Chadha block, and the villages of East Bardhaman are Narayanpur and Yogeswarpur of Purbasthali Block -2 (Patuli).

Design of experiment

Collection of wool/hair from clinically suspected animals of goat, sheep, and cattle. Sample of grass, fodder, and tube well water was collected from the owner of the animals, farmer's field, and the public tube well from which water is fed to animals and humans.

Collection of samples

Fresh grass and fodder of 250 grams were collected from the field, which are used for animal feed, air-dried in a dark place, and stored for further study. Water samples were collected from

the tube well from different villages in 500ml bottles with a few drops of 0.1% HCl. Hair and wool samples were collected 20g from each animal in polyethylene bags and stored for further analysis. The selection of animal hairs/wool to be collected with suspected clinical signs of chronic arenicolids.

Clinical observations of affected animals

Animals show some clinical symptoms that are suffering from chronic exposure to arsenic contamination through the feed and water chain. Clinical signs like weakness, health condition, skin lesions, appetite, growth condition, etc. were noted in different species of animals.

Emaciation, weakness, and skin lesions like ulcers and hair coat abnormalities. Digestive issues like loss of appetite, diarrhea, and inflammation of the gastrointestinal tract are also common. In some cases, animals may exhibit neurological signs, such as incoordination or paralysis

Digestion of samples for arsenic estimation

One gram of cool air-dried samples of grass, fodder, wool, hair, etc. was triturated. The samples were digested by a wet oxidative process, i.e., oxidizing with acid such as concentrated hydrochloric acid in a larger test tube of 50 mL, again 5ml 30% hydrogen peroxide was added that creates frothing. After the froth is diminished, added 5 mL of 30% H₂O₂ is added again and kept as such for half an hour. Again, 2 mL of conc. nitric acid was added to the test tube and kept at 60°C on a hot plate for 45 minutes. Here, the solution changed to a light straw color. The aliquot was filtered with Whatman filter paper No.1 with triple-distilled water. The volume was made up to 25 mL with water if necessary. The samples were ready for estimation with the atomic absorption Spectrophotometer model Varian AA240. In case of water analysis, 100 mL of collected water was treated with 7-8 drops of concentrated Nitric acid. The water was then filtered with Whatman filter paper No.1 and ready for analysis [6,17].

The AAS Varian AA240 attached to the MHS-10 Mercury/Hydrite System and HGA 2100 graphite furnace was utilized. The measurement was made with deuterium background correction. To estimate arsenic samples, 1 mL of 10% potassium Iodide solution was added to 10 mL of the aliquot. After 60 minutes, arsenic was determined by the hydride evolution method with Sodium borohydride and the MHS-10 Mercury/Hydrite system.

Statistical analysis

Statistical analysis of data was carried out in SPSS (version 20.0). Analysis of variance (ANOVA) was employed to see the effect of fixed factors (District, Species, village, sample type, etc.) on the dependent variable (Arsenic content expressed in ppb). DMRT was carried out to test the differences in the mean values of various experimental/observational groups/units. An independent samples t-test was performed when the sample size was less than 30. For comparing the arsenic concentrations in grass and water, independent sample t rest (assuming unequal variance) was carried out.

Results

From sample analysis of hair, wool, fodder/grass and water it was observed that arsenic content in hair/wool samples was observed higher (1123.667 ± 118.357 , in ppb) in cattle, intermediate (934.257 ± 40.398 , in ppb) in goat and while lower (869.750 ± 74.855 , in ppb) in sheep (Table 1, Figure 1). Arsenic content of cattle was found to be significantly ($p < 0.05$) higher than that of sheep. While the arsenic content of goat did not differ significantly from both cattle and sheep.

Among species		Cattle (n = 12)	Goat (n = 103)	Sheep (n = 30)	Overall (Mean $\pm$ SE)	Level of significance
Arsenic content in hair/wool in ppb (Means)		1123.667 ± 118.357^a	934.257 ± 40.398^{ab}	869.750 ± 74.855^b	915.344 ± 31.962	0.05
Between districts			East Bardhaman (n = 89)	Nadia (n = 63)	Overall (Mean $\pm$ SE)	Level of significance
Arsenic content in hair/wool ppb (Mean $\pm$ SE)			1097.062 ± 41.154^a	733.627 ± 48.915^b	915.344 ± 31.962	0.01
Arsenic content in water ppb (Mean $\pm$ SE)			39.00 ± 6.258	20.20 ± 10.318	-	NS
Arsenic content in grass ppb (Mean $\pm$ SE)			1078.80 ± 65.974	1323.00 ± 151.654	-	NS
Among villages/ areas	Silinda (n = 25)	Kalyani Farm (n = 23)	Dulepara, Kalyani (n = 15)	Patuli (n = 89)	Overall (Mean $\pm$ SE)	Level of significance
Arsenic content in hair/wool in ppb (Mean $\pm$ SE)	724.680 ± 78.023^{bc}	777.326 ± 81.344^b	$681.533 \pm 100.727^{bcd}$	1097.062 ± 41.352^a	820.150 ± 39.179	0.01
Between water and grass sample			Grass (n = 15)	Water (n = 10)	Overall (Mean $\pm$ SE)	Level of significance
Arsenic content in ppb (Mean $\pm$ SE)			1241.600 ± 105.916^a	29.60 ± 6.494^b	635.600 ± 136.462	0.01

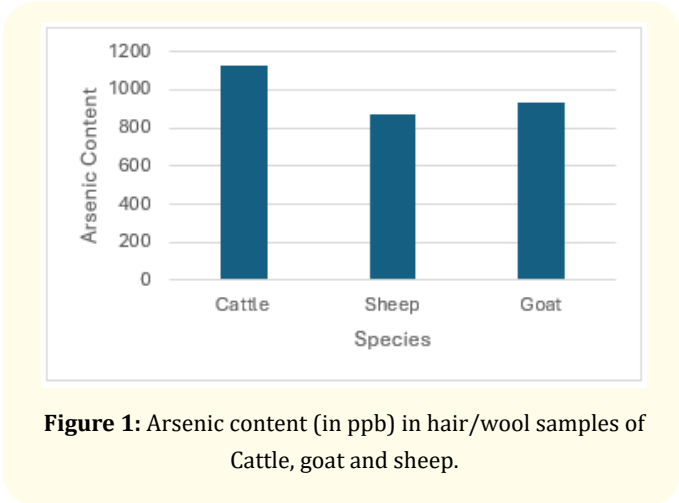
Table 1: Arsenic content in species, district, villages with mean and standard error (in ppb).

*Different superscripts in a row indicates that the means differ significantly.

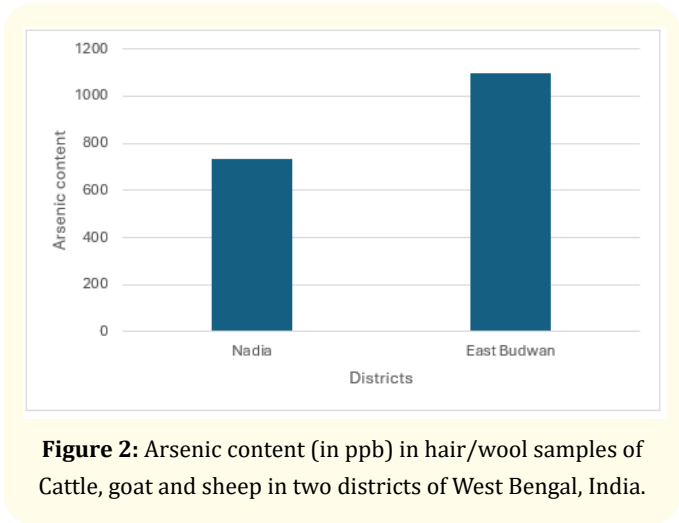
Clinical parameter	Goat n=103	Cattle n=12	Sheep n=30	Total n=145	% Affected
Weakness	25	4	6	35	24.14
Inappetence	36	3	8	47	33.79
Emaciation	9	1	4	14	9.65
Alopecia	33	4	10	47	34.48
Skin ulcer	2	1	0	3	2.06

Diarrhoea	14	2	4	20	13.79
Incoordination/paresis	3	0	1	4	2.75
Change in wool and hooves	3	0	2	5	7.25

Table 2: Clinical manifestations of arsenic toxicity in affected animal species.

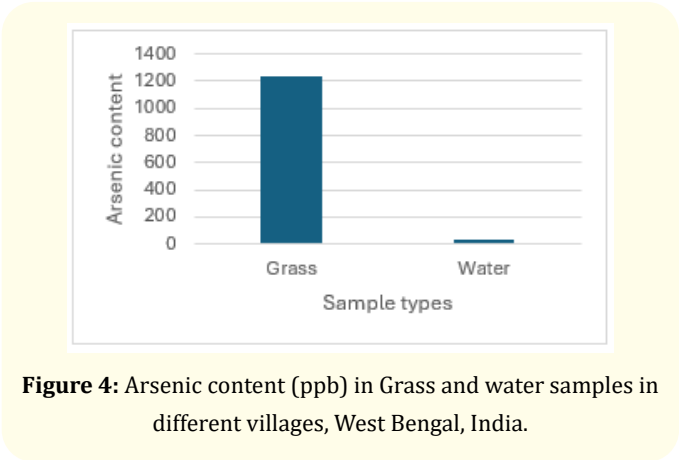
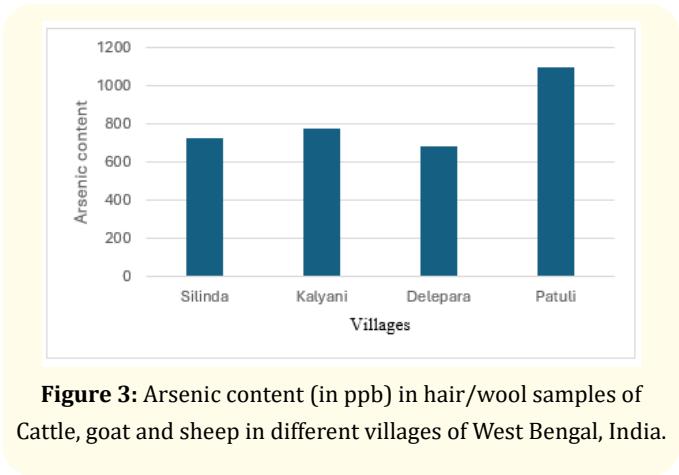


While comparing the arsenic content in hair/wool samples of all the species under the investigation, it was observed that arsenic content in East Badhaman district (1097.062 ± 41.154 , ppb) was significantly ($p < 0.01$) higher than that in Nadia district (733.627 ± 48.915 , ppb). The data is presented in Table 1 and Figure 2.



It was found that arsenic content in grass (1241.600 ± 105.916 ppb) is much higher (significant, $p < 0.01$) than in water samples (29.60 ± 6.494 ppb) collected from different villages. Data is presented in Table 1 and Figure 4. The higher concentration in in grass might be due to bioaccumulation of water-borne arsenic in the plant and grass while water arsenic contamination comparatively remains stable.

There was no significant difference in arsenic content in water samples between Nadia and East Bardhaman district, though arsenic content in East Bardhaman district (39.00 ± 6.258 ppb) was higher than that of Nadia district (20.20 ± 10.318 ppb). This is probably due to the small sample size ($n = 5$ from each district, Table-1, Figure 5).



There was no significant difference in arsenic content in grass samples between Nadia and East Bardhaman district, though arsenic content in Nadia district (1323.00 ± 151.654 ppb) was higher than that of East Bardhaman district (1078.80 ± 65.974 ppb) in table-1, Figure 6. This is probably due to the small sample size ($n = 5$ for East Bardhaman and $n = 10$ from Nadia).

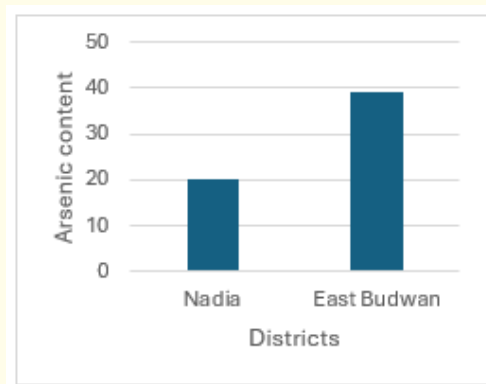


Figure 5: Arsenic content (in ppb) in water samples in two districts.

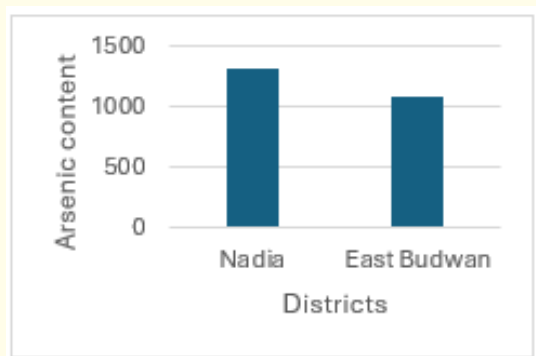


Figure 6: Arsenic content (in ppb) in grass samples in two districts.

Clinical findings of suffering animals in sheep, goat, and cattle remain wide. In cattle usually there no death due to chronic arsenicosis, but certain clinical signs like chronic diarrhea, ataxia, dehydration, loss of condition, and dry, lusterless skin coat. In sheep, clinical signs of weakness, emaciation, ulcerative skin, change in wool and hooves, and diarrhoea were observed. In goat most visible clinical signs reported were ataxia, mild posterior paresis, reduced ruminal motility, diarrhea, hind leg paresis,

hyperkeratosis, lameness, dark urine, and palor mucous membrane of the eyes (Photo 1-2). All the clinical signs are not persistence to each animal examined; some animals one or two clinical signs, some showed many of the above clinical signs.



Photo 1: Goat showing paresis, lameness.



Photo 2: Goat showing alopecia, hyperkeratosis.

Discussions

The investigation of the level of Arsenic in different sources of feed, fodder, water, hair, and wool found to be much higher than the recommended level [6-8]. However, the WHO recommended a reduced concentration of arsenic (5 ppb) in drinking water [9]. Of the three species of animal cattle found highly susceptible than sheep, a similar observation was also reported [10]. Of the two districts East Bardhaman animals have higher arsenic content than Nadia. The higher arsenic in animal products could be the variations of animal rearing and feed, and the source of water used. The arsenic content water of East Bardhan was found to be higher (39.0 ± 6.25 ppb) than Nadia groundwater (20.20 ± 10 ppb) but not

significant may be to a small sample size. On the other hand, grass samples of Nadia contain non-significant higher arsenic (1323 ± 151 ppb) than East Bardhaman (1078 ± 65.97 ppb).

The higher arsenic concentration in water in East Bardhaman district in comparison to Nadia is due to geological factors of Iron pyrite concentration, aquifer composition, and depth of aquifer. Other factors for higher water arsenic concentration in East Bardhaman are local land use patterns, high soil microbial activity, high Iron oxides and hydroxides dissolved in the ground water reported [11]. It was also observed that among the villages in Nadia district, the arsenic content in hair/wool samples of animals does not differ significantly (Table 1, Figure 3).

The clinical signs in chronic exposure to arsenic through feed and water reveal more or less similar with differences in species involvement. Sheep and goat show much effects than cattle, particularly on locomotion, skin, and anaemic condition with weight loss. The arsenic toxicity in different species of animals has been reported [12-14] corroborating our observation.

The clinical signs exerted were very slow growth and chronic in nature. Arsenic damage capillaries of gut wall due to chemical irritation secrets mucous and blood in it, this causes diarrhea, reduction in ruminal motility, malabsorption of nutrients leads to chronic weakness and emaciation [15]. As chemical irritant arsenic capillary damage in high keratin content tissue of skin and hooves, this causes skin necrosis and cut off cell growth of high keratin content tissues that hamper the hoof sloughing and haemorrhages. Arsenic also attacks on peripheral and spinal nerve causing demyelination that leads to ataxia and posterior paresis. Pigmentation with hyperkeratosis occurs due to binding of arsenic with keratin particularly of skin that cause over production of keratinocytes that produces hard keratin layers. Pallor of mucous membrane due to destruction of RBCs and suppression of bone marrow with arsenic element [16].

Arsenic toxicity is a chronic exposure of arsenic in animals and human through food chain. Shallow ground water is the main source of arsenic exposure in animals. Ground of several districts of West Bengal are affected with arsenic. Grass, fodder, feed, soil, drinking water and agricultural irrigation disperse the toxic arsenic in animals and humans [17].

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