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Assessment of Body Index Values in Albino rat after Intoxication with Arsenic trioxide

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ABSTRACT
Inorganic arsenic compounds were predominantly used as pesticides, primarily on cotton fields and in orchards. Inorganic arsenic compounds can no longer be used in agriculture. The greatest use of arsenic in alloys is in lead-acid batteries for automobiles. Presently, arsenic is widely used in the electronics industry in the form of gallium arsenide and arsine gas as components in semiconductor devices. Keeping these points in view, the present study is undertaken to study the amount of arsenic in brain, liver and kidney of albino rats after acute and subacute treatments based on median lethal dose in terms of body weight, organ weight and their ratio.
Keywords: Body Wegiht, Oragn Weight, Arsenic, albino rats

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INTRODUCTION

Arsenic is one of the most dangerous occupational and environmental toxins. Both natural and anthropogenic sources are responsible for the distribution of many toxicants, mainly heavy metals throughout the environment. Arsenic is ubiquitous in earth's crust and biosphere. It is found in food, soil, minerals, surface and ground water. The major cause of human arsenic toxicity is from contamination of drinking water as well as from natural geological sources rather than from mining, smelting or agricultural sources [1].

Inorganic arsenic compounds were predominantly used as pesticides, primarily on cotton fields and in orchards. Inorganic arsenic compounds can no longer be used in agriculture. Some organic arsenic compounds are used as additives in animal feed. Small quantities of elemental arsenic are added to other metals to form metal mixtures or alloys with improved properties. The greatest use of arsenic in alloys is in lead-acid batteries for automobiles. Presently, arsenic is widely used in the electronics industry in the form of gallium arsenide and arsine gas as components in semiconductor devices [4, 5, 2].

Inhalation major sources of inhaled arsenic may come from air emissions from burning of fossil fuels that contain arsenic, cotton gins, glass manufacturing operations, pesticide manufacturing facilities, smelters, and tobacco smoke [7]. Airborne arsenic in the workplace is generally in the form of arsenic trioxide. Dermal contact when handling preserved wood products containing arsenic could result in arsenic exposure.

MATERIAL AND METHODS

EXPERIMENTAL ANIMAL: Albino rats were reared in the animal house of Zoology Department, Dr. B. R. Ambedkar University, Agra. The male albino rats, (*Rattus norvegicus,*Berkenhout) were selected for the present investigation.

MAINTENANCE AND FEEDING OF EXPERIMENTAL ANIMALS: Healthy and adult male albino rats weighting 70 to 140 gm were kept in polypropylene cages measuring 45 x 27 x 15 cm. The top of cages was made of galvanized steel mesh and cleaned regularly to avoid any undesirable odor in the laboratory. Each cage was equipped with a metallic plate and a water bottle. The rats were fed on standard laboratory feed and water *ad libitum*. The experimental rats were housed under standard husbandry condition at temperature 25±5°C, relative humidity 60±5% and 12 hour light/dark cycle. Animal experiments were designed and conducted in accordance with the guidelines of Institutional Animal Ethical Committee. The experimental rats were acclimated for twenty days prior to experimentation.

EXPERIMENTAL CHEMICAL: The experimental compound Arsenic trioxide, procured from Central Drug House, New Delhi, India,was dissolved in distilled water.

EXPERIMENTAL DESIGN:

Assessment of median lethal dose (MLD): Total twenty five albino rats were used for assessment of lethal dose, they were divided into five groups with five rats in each. The test chemical (As_2O_3) was dissolved in distilled water and the different doses were administered orally with the help of gavages tube.The number of dead and survived rats was recorded after fourteen days. The data were analyzed statistically by log-dose probit regression line method [3]. On the basis of log-dose and empirical probit, the regression line was drawn on the ordinary graph paper. The regression line was used to determine the expected probit necessary for LD_{50} determination.

Selection of Doses : The sub-lethal dose of As_2O_3 for acute (3 hrs, 6 hrs, 12 hrs and 1 d) treatment LD_{50} /10^{th}, while for sub-acute treatment (7, 14 and 21 days) $1/7^{th}$, $1/14^{th}$ and $1/21^{th}$ of sub-lethal dose respectively.To determine the absorption and distribution of As_2O_3, in the albino rats, they were classified into eight groups, the first, the control, the second, the acute (3 hrs, 6 hrs, 12 hrs and 1 d) and the third, sub-acute (7, 14 and 21 ds) with three rats in each. The first sub-group was given distilled water and served as control. The second sub-group was exposed to As_2O_3. Peripheral blood and the vital organs (Brain, Kidney and Liver) selected in the present investigation were assayed for arsenic quantity after 3 hrs, 6 hrs, 12 hrs, 1 d, 7 ds, 14 ds and 21 ds.

ASSESSMENT OF BODY WEIGHT: The rats were weighted before and after acute and sub acute treatments of each group in order to assess the body weight.

ASSESSMENT OF TISSUES (BRAIN, LIVER, KIDENY) WEIGHT AND TISSUES (BRAIN, LIVER, KIDNEY) WEIGHT / BODY WEIGHT RATIO: The tissues (brain, liver, kidney) were excised out from the experimental rats after necropsy, put in physiological saline (pH 7.4) and blotted off between ash free filter paper. They were weighted at predetermined time intervals as per protocol after 3 hours, 6 hours, 12 hours, 1day, 7days, 14days and 21days treatments respectively.

The relative tissues (brain, liver, kidney) weight of acute (3 hrs, 6 hrs, 12 hrs and 1 d), sub-acute (7 ds, 14 ds and 21 ds) and of control was calculated *vide infra*.

$$\text{tissues (brain, liver, kidney) weight / body weight ratio} = \frac{\text{tissues(brain, liver, kidney) weight}}{\text{body weight}}$$

RESULTS AND DISCUSSION

Table I : Percent changes in body weight after As_2O_3 intoxication

TREATMENT DAYS	NO. OF RATS	BODY WEIGHT (g)		% CHANGE IN BODY WT.
		Initial wt.	Final wt.	
		Mean ± S.E.m	Mean ± S.E.m	
Acute (3 hrs)	3	106 ± 0.0	106 ± 3.79	0.00 %
Acute (6 hrs)	3	87 ± 4.1	86 ± 5.1	1.15 %
Acute (12 hrs)	3	122 ± 3.0	120 ± 2.89	1.64 %
Acute (1 d)	3	79 ± 1.5	78 ± 3.79	1.26 %
Sub-acute (7 ds)	3	92 ± 1.1	90 ± 2.1	2.17 %
Sub-acute (14 ds)	3	97 ± 3.1	95 ± 1.16	2.17 %
Sub-acute (21 ds)	3	82 ± 4.1	79 ± 4.17	3.65 %
Control	3	69 ± 3.0	75 ± 5.5	8.70%

Table II : Body weight of male albino rats ater As_2O_3 intoxication

TREATMENT DAYS	NO. OF RATS	BODY WEIGHT (g)		SIGNIFICANCE LEVEL
		CONTROL	As_2O_3	
		Mean ± S.E.m	Mean ± S.E.m	
Acute (3 hrs)	3	75 ± 5.5	106 ± 3.79	P < 0.05
Acute (6 hrs)	3	75 ± 5.5	86 ± 5.1	P > 0.05
Acute (12 hrs)	3	75 ± 5.5	120 ± 2.89	P > 0.05
Acute (1 d)	3	75 ± 5.5	78 ± 3.79	P > 0.05
Sub-acute (7 ds)	3	75 ± 5.5	90 ± 2.1	P < 0.05
Sub-acute (14 ds)	3	75 ± 5.5	95 ± 1.16	P < 0.05
Sub-acute (21 ds)	3	75 ± 5.5	79 ± 4.17	P > 0.05

Table III : Organs weight (brain, kidney and liver) of male albino rats after As_2O_3 intoxication

TREATMENT DAYS	NO. OF RATS	ORGANS WEIGHT (g)						SIGNIFICANCE LEVEL
		CONTROL			As_2O_3			
		LIVER	KIDNEY	BRAIN	LIVER	KIDNEY	BRAIN	
		Mean ± S.E.m	Mean ± S.E.m	Mean ± S.E.m	Mean ± S.E.m	Mean ± S.E.m	Mean ± S.E.m	
Acute (3 hrs)	3	4.52 ± 0.76	0.64 ± 0.067	1.25 ± 0.066	4.1 ± 0.25	0.79 ± 0.049	1.58 ± 0.009	P < 0.05
Acute (6 hrs)	3	4.52 ± 0.76	0.64 ± 0.067	1.25 ± 0.066	3.28 ± 0.27	0.72 ± 0.055	1.46 ± 0.05	P < 0.05
Acute (12 hrs)	3	4.52 ± 0.76	0.64 ± 0.067	1.25 ± 0.066	6.23 ± 1.53	1.3 ± 0.039	1.67 ± 0.075	P < 0.05
Acute (1 d)	3	4.52 ± 0.76	0.64 ± 0.067	1.25 ± 0.066	3.3 ± 0.49	0.7 ± 0.039	1.42 ± 0.066	P < 0.05
Sub-acute (7 ds)	3	4.52 ± 0.76	0.64 ± 0.067	1.25 ± 0.066	3.55 ± 0.25	0.67 ± 0.017	1.39 ± 0.085	P < 0.05
Sub-acute (14 ds)	3	4.52 ± 0.76	0.64 ± 0.067	1.25 ± 0.066	3.68 ± 0.297	0.75 ± 0.03	1.47 ± 0.017	P < 0.05
Sub-acute (21 ds)	3	4.52 ± 0.76	0.64 ± 0.067	1.25 ± 0.066	3.29 ± 0.27	0.65 ± 0.036	1.29 ± 0.038	P < 0.05

Table IV : Organs weight (brain, kidney and liver) / body weight ratio of male albino rats after As_2O_3 intoxication

TREATMENT DAYS	NO. OF RATS	ORGANS WEIGHT(BRAIN, KIDNEY AND LIVER) / BODY WEIGHT RATIO						SIGNIFICANCE LEVEL
		CONTROL			As2O3			
		LIVER	KIDNEY	BRAIN	LIVER	KIDNEY	BRAIN	
		MEAN ± S.E.M	MEAN ± S.E.M	MEAN ± S.E.M	MEAN ± S.E.M	MEAN ± S.E.M	MEAN ± S.E.M	
ACUTE (3 HRS)	3	0.06 ± 0.007	0.008 ± 0.0005	0.017 ± 0.001	0.04 ± 0.001	0.007 ± 0.0003	0.014 ± 0.0005	P < 0.001
ACUTE (6 HRS)	3	0.06 ± 0.007	0.008 ± 0.0005	0.017 ± 0.001	0.04 ± 0.001	0.008 ± 0.0002	0.017 ± 0.0005	P < 0.001
ACUTE (12 HRS)	3	0.06 ± 0.007	0.008 ± 0.0005	0.017 ± 0.001	0.05 ± 0.01	0.01 ± 0.003	0.014 ± 0.0003	P < 0.001
ACUTE (1 D)	3	0.06 ± 0.007	0.008 ± 0.0005	0.017 ± 0.001	0.04 ± 0.002	0.009 ± 0.0001	0.018 ± 0.0001	P < 0.001
SUB-ACUTE (7 DS)	3	0.06 ± 0.007	0.008 ± 0.0005	0.017 ± 0.001	0.04 ± 0.003	0.007 ± 0.0001	0.015 ± 0.0006	P < 0.001
SUB-ACUTE (14 DS)	3	0.06 ± 0.007	0.008 ± 0.0005	0.017 ± 0.001	0.04 ± 0.004	0.008 ± 0.0004	0.015 ± 0.0002	P < 0.001
SUB-ACUTE (21 DS)	3	0.06 ± 0.007	0.008 ± 0.0005	0.017 ± 0.001	0.04 ± 0.002	0.008 ± 0.0004	0.016 ± 0.0014	P < 0.001

Inorganic arsenic is more hazardous than organic arsenic exposure in term of blood, bone marrow, cardiac, central nervous system (CNS), gastrointestinal, gonad, kidney, liver, pancreatic and skin tissues. Inorganic arsenic exhibits affinity with sulfur and its toxicity in living organism is perhaps due to the interference in enzymatic disruption in protein synthesis by binding with sulfhydryl groups of amino acid throughout the body [2].

The MLD of Arsenic trioxide has been found to be 34.36 mg/kg body weight and is in contrast to Saxena *et al.,* [6] whoreported the medial lethal dose of arsenic trioxide to be 14.98 mg/kg body weight; it may be changed due to environmental, sex and weathering factors. The present study highlights significant reduction in body weight after acute and sub-acute intoxication of arsenic trioxide and is in agreement to who Saxena *et al.* [6] observed reduction in body weight after intoxication of arsenic trioxide. Reduction in body weight perhaps is due to reduction of food and water intake under influence of arsenic trioxide. Alterations in the functioning of CNS leads to regulatory disturbances which are evident in terms of changed behavioral patterns and feeding is one such outcome which gets regulated by feeding center present in hypothalamus. CNS being the primary target of arsenic insult, results in neuropathy and neurodegeneration which have been demonstrated under stress of arsenic trioxide [1].

Significant increase in the weight of vital organs (Brain, Kidney, and Liver) and organs weight / body weight ratio compared to control, Due to formation of Reactive oxygen species (free radicals) which lead to neuron, renal and hepatic damage. Enhancements in liver and liver wt. / body wt. ratio have also been reported Saxena *et al.*, [6].

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