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PREVENTION OF ARSENIC TOXICITY WITH SPIRULINA AND MENTHA IN RATS

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Abstract

Mitigation of Arsenic toxicity urgently needed in human being. Present research designed to evaluation of the individual efficacy of spirulina and mentha on arsenic toxicity in rats. Forty-eight female rats of about 3 months of age were used in this experiment. All the 48 rats were obtained by breeding rats in the laboratory animal section of the ICDDRB, Dhaka. At first all 48 rats were randomly grouped into 4 and each group consisted of 12 rats. The experimental trial was conducted for 30 days. Rats of Group T_0 were maintained with only normal feed and water *ad libitum* as control. Other three Group T_1, T_2 and T_3 were treated with sodium arsenite, sodium arsenite with spirulin and sodium arsenite with mentha, respectively. Rats were closely observed daily for 3 times (morning, afternoon and evening) for clinical signs if any of them, during the entire experimental period (from day 0 to day 30) and the findings were recorded. On Day 10, As concentration in lung was found highest in rats of T_1 and lowest in control group, and As content was increased sharply in all treated groups and the increments were highly significant ($p<0.01$) compared to control. The highest As content was observed in liver of T_2 group rats and the lowest in control group rats and the As values were sharply and significantly ($p<0.01$) increased in all treated group rats compared to control group. The values of SGPT, SGOT and creatinine on day 10, 20 and 30 were the highest in T_1 and T_2 group rats and lowest in control group. All treated group rats showed statistically significant ($p<0.05$) increased values compared to control rats. Our study suggests that the combination of spirulina and mentha were found more effective in prevention of arsenic toxicity in rat.

Keywords: Sodium arsenite, poisoning, mitigation, herbal, human.

Introduction

Arsenic (As) is a ubiquitous trace element that is widely distributed throughout the earth's crust. Dissolution of arsenic from the soil can easily contaminate ground water with up to µg/L levels of inorganic arsenic (iAs). Therefore, one of the most serious worldwide environmental problems, now a day, is drinking water polluted by arsenic (WHO, 2001).

Bangladesh is currently facing a serious threat to public health, with 85 million people at risk from arsenic (As) in drinking water and in food crops. In Bangladesh, the groundwater As contamination problem is the worst in the world. Ninety-seven percent of the population in the country uses groundwater for drinking and domestic purposes as surfacewater is mismanaged. High levels of As in groundwater are causing widespread

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poisoning in Bangladesh. Different studies have addressed various aspects of the As issue in Bangladesh (Hossain, 2006).

Chronic arsenic toxicity is a global health issue at present (Yoshida *et al.,* 2004). It is also a major health problem of Bangladesh and surrounding regions (Kalia, 2005; Khalequzzaman *et al*., 2005). In Bangladesh, it is reported that about 2 lakhs people are suffering from arsenicosis ranging from melanosis to skin cancer (Das, 2000). Chronic As poisoning can cause serious health problems including cancers, hyperkeratosis, restrictive lung disease, and ischaemic heart disease (Rossman, 2003; Mandal and Suzuki, 2002) and increases the risk for As-induced diseases such as noncancerous skin lesions, bronchitis, hepatomegaly, neuropathy, peripheral vascular diseases, cardiovascular disease, skin cancer, lung cancer, and bladder cancer (Mazumder, 2005).

There is no specific curative treatment against arsenicosis. Immediate stoppage of drinking arsenic contaminated water and consumption of arsenic free drinking water are the mainstay of therapy (Dey, 2002). Vitamin A, E, C regimen with arsenic free water was proved to be effective in improving arsenic induced melanosis and keratosis (Ahmad *et al.,* 1998). Spirulina was found to be beneficial in goats of chronic arsenic poisoning (Halim, 2007) and Spirulina extract plus zinc was found to be beneficial in patients of chronic arsenic poisoning (Misbahuddin *et al*., 2006). Chelation therapy for arsenic toxicity is thought to be the specific therapy for relief of systemic clinical manifestations and reduction of arsenic stores in the body, reducing subsequent cancer risk (Bhattacharya, 2017).

In Bangladesh, elaborate data is available for arsenic only on tube-well water; however, data on the specific treatment in prevention of arsenic toxicity in both human and animals is very scare. Therefore, data on the effective prevention of arsenicosis with spirulina and mentha and their comparative efficacy will be evaluated. So considering all the above factors, this work has been taken to evaluation of the individual effects of spirulina and mentha on arsenic toxicity in rats.

Materials and Methods

This experiment was carried out in the Arsenic Detection and Mitigation Laboratory (ADM Lab), Department of Pharmacology, Bangladesh Agricultural University, Mymensingh in collaboration with the Department of Pathobiology, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur during the period of 01 June 2017 to December 2017.

Experimental Animals

Forty eight female rats of about 3 months of age were used in this experiment. All the 48 rats were obtained by breeding rats in the laboratory animal section of the ADM Lab. At first all 48 rats were randomly grouped into 4 and each group consisted of 12 rats, then they were individually marked using different color on their tail tips for identification. Groups were identified as T_0 for control, T_1 for arsenic ($NaAsO_2$; MERCK, E.Merck, Darmstadt, Germany) treated group, T_2 for arsenic plus spirulina treated group and T_3 for arsenic plus Mentha extract treated group. Rats of each group were kept in pre-disinfected separate steel wire cages in a well mechanical ventilated

and controlled room temperature with natural relative humidity. Rats were maintained under normal pellet feed and drinking water with admissible levels of arsenic. The excreta of the animals were cleaned in the every morning and the animal room was managed with standard bio-security.

Body weight (BW)

The rats were individually weighed firstly on Day 0 (Day 0= immediate previous day of starting treatment) after grouping and marking, Day 10, Day 20 and finally on Day 30 and the results were recorded.

Clinical signs

Rats were closely observed daily for 3 times (morning, afternoon and evening) for clinical signs if any in them, during the entire experimental period (from day 0 to d ay 30) and the findings were recorded.

Experimental trial

The experimental trial was conducted for 30 days. Rats of Group T_0 were maintained with only normal feed and water *ad libitum* as control, that of Group T_1 were treated with Sodium arsenite ($NaAsO_2$; MERCK, E. Merck, Darmstadt, Germany) at a dose of 5 mg/kg body weight in 10 ml drinking water daily, rats of Group T_2 were treated with $NaAsO_2$ at 5 mg/kg body weight in drinking water daily and spirulina (*Spirulina platensis*) simultaneously at a dose of 50 mg/kg body weight with feed daily. The animals of Group T_3 were treated with $NaAsO_2$ at 5mg/kg body weight in 10 ml drinking water daily and aqueous extract of Mentha (*Mentha arvensis*) simultaneously at a dose of 100 mg/kg body weight in 10 ml drinking water daily.

Sampling

At every 10 days interval (on day 10, day 20 and day 30) 4 rats from each group were randomly selected and individually submitted them into general anesthesia using chloroform and then about five milliliters of blood was collected directly from hearts of each rats by using disposable syringe for determination of biochemical parameters (SGOT, SGPT and serum creatinine) after giving midline incision up to thorax. The blood sample was taken into pre-marked centrifuge glass test tubes without anti-coagulant immediately after collection and kept at room temperature for 1 hour without agitation for clotting with a view to collect serum. Lungs, liver, and kidneys were collected and washed with physiologic saline and were taken into the respectively pre-marked zipper polythene bags. All the organs were preserved in a deep freeze at -20°C.

Determination of SGOT and SGPT

Following thawing, the test tubes containing blood clot were centrifuged in centrifuge machine (EBA 20, Hettich, ZENTRIFUGEN, Germany) at 1500 rpm for 15 minutes. The separated supernatant serum was collected gently from each test tube into the correspondingly marked screw capped sterile eppendorf tubes with separate sterile Pasteur pipette and were tested followed by standard procedure (Bhattacharya, 2107). Serum glutamate oxaloacetate transaminase (SGOT) level was displayed on the monitor in 75 seconds in Unit/Liter (U/L) and Serum glutamate pyruvate transaminase (SGPT) level was displayed on the monitor in 75 seconds in Unit/L.

Serum creatinine

Serum creatinine is determined following the same procedure as done in case of SGOT and SGPT by Reflotron® Plus. Briefly, serum of the sample was diluted in PBS. 25 µl of diluted serum was placed on the centre of the red application zone of the creatinine test strip with the help of micropipette after opening the sliding cover of the test strip. The strip was then placed on to the strip guide within 15 seconds from placing of serum on the strip and the slide was forwarded until it locks into place. The sliding cover was closed. The creatinine level was displayed on the monitor in 75 seconds in mg/dl.

Digestion of tissue samples and arsenic detection

The digestion of organ samples was carried out using concentrated nitric (69%; VWR International limited, poole, BH 15 1TD, England) and perchloric acid (70%; Merck, KGaA, Darmstadt, Germany) at the ratio of 9:4 (nitric acid : perchloric acid). Laboratory chemicals used in the digestion and detection of As were trace analytical grade. The digestion of tissues (lung, liver and kidney) was done followed by standard procedure (Kim *et al.,* 2015).

The As in the sample was first ionized into arsine and then atomized. Reading was taken with the help of the computer connected to the HG-AAS by using manufacturer supplied 'AAwin software' (Atomic Absorption Spectrophotometer PC-Software). The reading of the tested sample was displayed on the computer monitor in a pre-customized Microsoft excel sheet provided by the AAWin software as numerical number with giving a peak of As concentration on the respective part of the software displayed sheet on the computer monitor. Readings of As concentrations of the samples were taken in ppb.

Statistical analysis

The experimental data were designed in CRD and analyzed statistically using one way ANOVA with the help of the SPSS 11.5 software. Mean comparisons of the treatments were made by the Duncan's Multiple Range Test (DMRT) (Steel and Torrie, 1980). The values were considered significant and highly significant when the P is <0.05 and <0.01, respectively.

Results

Clinical signs

No clinical signs of arsenic toxicity were observed in trial rats during the entire period of the experimental trial.

Body weight of the rats

The highest body weight in rats of control (T_0) group and the lowest in T_2 group were found on day 0 but there was no significant difference in body weight between the rats of all groups on day 0. On day 10 body weights were found highest in rats of control group and lowest in that of T_3 group. No significant difference was observed between the rats of T_2 and T_3 group on day 10 but they were found significantly different compared to the control group ($p<0.01$). (Table 1).

On day 20, the body weight of all groups showed an increasing trend where rats of T_0 group gained the highest weight and that of T_3 group gained the lowest weight compared

Table 1. Effect of different treatment on body weight in rats

Treatment (Mean±SE)	Day 0 (g)	Day 10 (g)	Day20 (g)	Day30 (g)
Control (T_0)	168.40±5.56[a]	174.70±5.90[a]	185.57±7.53[a]	199.75±11.95[a]
Arsenic (T_1)	163.90±4.14[b]	165.80±4.06[b]	174.57±4.81[ab]	176.50±6.09[ab]
Arsenic+Spirulina (T_2)	149.30±2.47[c]	150.90±3.02[c]	160.14±3.49[bc]	166.50±5.91[b]
Arsenic+ Mentha (T_3)	151.40±4.88[c]	147.90±4.82[c]	148.86±8.84[c]	144.00±12.24[c]
Level of significance	*	**	**	**

*Significant ($p<0.05$), **Highly significant ($p<0.01$); Figures with similar superscripts mean did not differ significantly among respective figures, but figures with dissimilar superscripts mean differed significantly as per DMRT.

day 10. The differences in body weight were statistically significant *($p<0.01$)* in rats of T_2 and T_3 groups compared to control group but it was not statistically significant between that of T_2 and T_3 groups. The body weight changes in T_2 and T_3 group were significantly ($p<0.01$) different in respect to the control group (Table 1).

Arsenic content in different organs

Arsenic in lung

On day 10, As content in lung was found highest (18.25±1.50) in rats of T_1 and lowest (0.28±.022) in control group and As content was increased sharply in all treated groups and the increments were highly significant ($p<0.01$) compared to control. On the other hand, on day 20, the highest As content (18.28±0.64) was observed in lungs of As treated rats and lowest in that of control group rats. On day 30, arsenic concentration was significantly ($p<0.01$) lowered in lungs of T_2 and T_3 group rats compared to As treated group and that was significantly ($p<0.01$) decreased in T_3 group rats in respect to T_2 group (Table 2).

Arsenic in liver

The highest As content (17.69±4.64) was observed in liver of T_2 group rats and the lowest in control group rats and the As values were sharply and significantly ($p<0.01$) increased in all treated group rats compared to control group but there was no significant difference in arsenic concentration in liver among the treatment groups on day 10. In T_2 and T_3 were statistically significant ($p<0.01$) compared to T_1 group rats and the difference in As content between T_2 and T_3 group rats were not significant on day 20. The difference was significant *($p<0.01$)* in T_2 group and that was not significant in T_3 group compared to T_1 group and the difference between T_2 and T_3 group was not statistically significant on day 30 (Table 3).

Arsenic in kidneys

Arsenic content in kidneys was found highest in arsenic treated group and the lowest in control group rats and the As values were sharply and significantly *($p<0.01$)* increased in all treated group rats compared to control but there was no significant difference in arsenic concentration in kidneys among the treatment groups on day 10. The T_2 and T_3 group rats showed significant decrease in As content compared to T_1 group but between T_2 and T_3 group rats the difference was not statistically

Table 2. Effect of different treatment on arsenic content of lung in rats

Treatment (Mean±SE)	Day 10 (ppb)	Day 20 (ppb)	Day 30 (ppb)
Control (T_0)	0.28±.022^{c}	0.33±0.09^{c}	0.31±0.06^{c}
Arsenic (T_1)	18.25±1.50^{a}	18.28±0.64^{a}	29.05±1.16^{a}
Arsenic+Spirulina (T_2)	17.69±4.64^{b}	14.77±2.27^{b}	17.92±1.89^{b}
Arsenic+ Mentha (T_3)	13.89±1.04^{b}	14.80±0.70^{b}	14.69±1.90^{bc}
Level of significance	**	**	**

**Highly significant ($p<0.01$); Figures with similar superscripts mean did not differ significantly among respective figures, but figures with dissimilar superscripts mean differed significantly as per DMRT.

Table 3. Effect of different treatment on arsenic content of liver in rats

Treatment (Mean±SE)	Day 10 (ppb)	Day 20 (ppb)	Day 30 (ppb)
Control (T_0)	0.12±0.01^{c}	0.40±0.25^{c}	0.05±.011^{d}
Arsenic (T_1)	10.69±0.47^{ab}	17.26±2.06^{a}	19.35±1.51^{a}
Arsenic+Spirulina (T_2)	11.73±0.47^{a}	10.81±1.26^{b}	8.85±1.50^{c}
Arsenic+ Mentha (T_3)	8.96±0.87^{b}	9.57±2.27^{bc}	10.75±1.74^{a}
Level of significance	**	**	**

** Highly significant ($p<0.01$); Figures with similar superscripts mean did not differ significantly among respective figures, but figures with dissimilar superscripts mean differed significantly as per DMRT.

significant. The findings of As content in kidneys on day 30 were almost similar to that found on day 20 but only difference was observed in T_2 and T_3 group rats and that was the increment of As content in kidneys on day 30 compared to day 20, whereas on day 20 that was decreased in T_2 and T_3 group rats compared to that on day 10 (Table 4).

Biochemical parameters

Serum glutamate oxaloacetate transaminase (SGOT)

The values of SGOT on day 10, 20 and 30 were the highest in arsenic treated rats and lowest in control group and the differences among the treated group rats were statistically significant *($p<0.01$)* and, that in T_1 and T_2 were significant ($p<0.01$) but in T_3 group rats that was not significant compared to control (Table 5).

Serum glutamate pyruvate transaminase (SGPT)

The values of SGPT on day 10, 20 and 30 were the highest in T_1 and T_2 group rats and lowest in control group and all treated group rats showed statistically significant *($p<0.05$)* increased values compared to control rats, but the differences of the values in all treated group rats were not significant among themselves (Table 6).

Serum creatinine

The values of serum creatinine on day 10 was observed highest in control rats and lowest in T_1 and T_3 groups but the differences were not statistically significant among themselves. The values were increasing in trends on day 20 in all group rats compared to that in day 10 except in control group where that was decreased but the differences were not statistically significant

Table 4. Effect of different treatment on arsenic content of kidney in rats

Treatment (Mean±SE)	Day 10 (ppb)	Day 20 (ppb)	Day 30 (ppb)
Control (T_0)	0.25±0.018^{d}	1.71±1.08^{d}	0.11±0.08^{d}
Arsenic (T_1)	23.61±3.89^{a}	37.97±2.11^{a}	44.70±3.65^{a}
Arsenic+Spirulina (T_2)	19.34±7.69^{b}	16.65±8.20^{b}	19.06±4.24^{b}
Arsenic+ Mentha (T_3)	13.48±7.36^{ac}	12.58±1.91^{c}	12.35±4.25^{c}
Level of significance	**	**	**

**Highly significant ($p<0.01$); Figures with similar superscripts mean did not differ significantly among respective figures, but figures with dissimilar superscripts mean differed significantly as per DMRT.

Table 5. Effects of different treatment on SGOT values of rats

Treatment (Mean±SE)	Day 10 (U/L)	Day 20 (U/L)	Day 30 (U/L)
Control (T_0)	50.25±4.13^{b}	54.25±13.30^{cd}	58.00±13.60^{d}
Arsenic (T_1)	125.00±6.82^{a}	129.25±7.78^{a}	134.25±1.93^{a}
Arsenic+Spirulina (T_2)	73.75±2.56^{b}	85.50±0.67^{b}	101.00±3.19^{b}
Arsenic+ Mentha (T_3)	62.50±0.67^{b}	65.67±0.48^{c}	69.75±0.54^{c}
Level of significance	**	**	**

**Highly significant ($p<0.01$); Figures with similar superscripts mean did not differ significantly among respective figures, but figures with dissimilar superscripts mean differed significantly as per DMRT.

Table 6. Effects of different treatment on SGPT values of rats

Treatment (Mean±SE)	Day 10 (U/L)	Day 20 (U/L)	Day 30 (U/L)
Control (T_0)	30.00±1.29^{b}	28.75±0.85	27.00±0.58^{b}
Arsenic (T_1)	38.00±0.70^{a}	32.00±1.96	26.00±0.41^{bc}
Arsenic+Spirulina (T_2)	38.01±0.70^{a}	35.33±0.48	30.75±0.98^{a}
Arsenic+ Mentha (T_3)	34.33±0.80^{b}	32.00±1.78	27.00±2.58^{b}
Level of significance	*	NS	**

*Significant ($p<0.05$); ** highly significant ($p<0.01$); NS stands for figures that not significant; Figures with similar superscripts mean did not differ significantly among respective figures, but figures with dissimilar superscripts mean differed significantly as per DMRT.

among themselves. On day 30, the serum creatinine values were increased in all group of rats compared to that in day 20 but the differences were not statistically significant among themselves (Table 7).

Discussion

Induction of arsenic toxicity in rats increased arsenic concentrations in lung, liver and kidneys; spirulina and Mentha treatment lowered arsenic contents in organs and found effective against As induced toxicity in rats. Chronic arsenic toxicity results in multisystem disease. Apart from advising avoiding arsenic contaminated drinking water and certain symptomatic treatments, there are no evidence-based definitive treatment regimens to treat chronic arsenic toxicity

Table 7. Effects of different treatment on serum creatinine values of rats

Treatment (Mean±SE)	Day 10 (mg/dl)	Day 20 (mg/dl)	Day 30 (mg/dl)
Control (T_0)	0.93±0.02	0.89±0.04	0.94±0.02
Arsenic (T_1)	0.82±0.03	0.88±0.03	0.90±0.03
Arsenic+Spirulina (T_2)	0.86±0.06	0.91±0.02	0.93±0.02
Arsenic+ Mentha (T_3)	0.84±0.02	0.86±0.02	0.89±0.02
Level of significance	NS	NS	NS

NS stands for figures that not significant; Figures with similar superscripts mean did not differ significantly among respective figures, but figures with dissimilar superscripts mean differed significantly as per DMRT.

in humans. Nevertheless, antioxidants have been advocated (Ratnaike, 2003; Mazumder, 2008); since the elicitation of oxidative stress by generation of free radicals during the metabolism of arsenic in body is considered to be involved in arsenic toxicity (Shi *et al.,* 2004; Kim *et al.,* 2015).

Significantly increased *($p<0.01$)* levels of As in the lung, liver and kidney following feeding of $NaAsO_2$ (5mg/kg BW) to the rats of arsenic treated group compared to control during the whole period of the study, which was increased with the length of exposure period and agreed with the findings of Nasir *et al.* (2002) and Kamaludin and Misbahuddin (2007).

Arsenic loads detected in lung, liver and kidney of which highest accumulation was found in kidney followed by lung and liver of all groups. These data did not agree with the findings of Marafante (1982) who showed that the highest accumulation of arsenic in the spleen followed by lung, liver, kidney, skin and lowest accumulation was in intestine. The raised levels of As in the kidney may be due to the excretion process of arsenic metabolites through kidneys.

Spirulina reduced As level significantly *($p<0.01$)* in lung, liver and kidney of spirulina plus arsenic treated rats compared to arsenic treated group on all sampling days. However, spirulina effectively reduced As loads from lung, liver and kidney of rats with induced arsenicosis, which was the agreement with Fariduddin *et al.* (2001), who stated that spirulina was effective in the removal of arsenic from the arsenic loaded tissues in rats, and partially with Halim *(*2007) who stated that spirulina was effective in the removal of arsenic from blood of induced arsenicosis in goats. Spirulina and riboflavin has significant effect on hematological and biochemical parameters and increase body weight (Akter *et al*., 2018).

Mentha reduced As level significantly *($p<0.01$)* in lung, liver and kidney of Mentha plus arsenic treated rats compared to arsenic treated rats on Day 10, day 20 and Day 30 (Table 2, 3 and 4). Hence, Mentha effectively reduced As loads from lung, liver and kidney of rats with induced arsenicosis by binding methods. However, arsenic in tissues could be lowered by treating animals with spirulina or Mentha might be established by this study.

Hence, it could be said that spirulina might more efficacious than Mentha against arsenic induced toxicity in rats. This findings supported by other findings (Khan *et al.,* 2001), who stated that spirulina was effective in the treatment of chronic arsenic poisoning

in human; and spirulina extract plus zinc twice daily might be significantly *(p<0.01)* useful for the treatment of chronic arsenic poisoning in man (Misbauddin *et al.,* 2006).

The values of SGOT were increased significantly *(p<0.01)* in all samples of arsenic treated rats compared to control. Although, this finding did not agree with the findings that SGOT was reduced by As alone (Mahaffey *et al.,* 1981). But the values were decreased significantly *(p<0.05)* in arsenic plus spirulina and arsenic plus Mentha treated group compared to arsenic treated rats. However, the arsenic toxicity caused hepatic insufficiency and spirulina and Mentha treatment improved the hepatic functions (Sharma *et al*., 2007), which was in support of that spirulina reduced hepatic damage due to drug abuse and heavy metal exposure (Mazumder, 2001).

Overall SGPT values have decreasing trend with the progress of time in all groups. No change in SGPT values was observed with As supplementation for 0, 45 and 90 days (Kaur *et al.,* 2005).

There is a relationship between the As level and the degree of chronic renal insufficiency (Zhang *et al.,* 1995). The results of this study agreed with this finding, but did not agree with the finding of Nandi *et al.* (2006), which showed that the patients of arsenicosis had significantly lower levels of serum creatinine compared to the control.

From previous works it is well documented that micronutrients and antioxidants has significant role in the treatment of chronic arsenic poisoning. Selenium (Spallholz *et al.,* 2004), zinc, iron, spirulina (Saha *et al.,* 2005; Halim, 2007), lipoic acid, ascorbic acid and á-tocopherol all has got ameliorating role against chronic arsenic poisoning (Ramanathan *et al.,* 2005; Rabbani *et al.,* 2003). Arsenic toxicity following feeding of sodium arsenite (4 mg/kg BW) in rats increased day by day. Spirulina and thankuni treatment lowered arsenic toxicity where the Spirulina found more effective in reducing arsenic content in the tissues (Hasan *et al*., 2015).

Study of arsenic toxicity and its treatment with spirulina and Mentha in animals for a longer period could be able to establish unequivocal decisions about the impacts of arsenic, spirulina, and enthe on hematological and biochemical parameters, as well as about strong evidence of efficacy of spirulina and enthe treatment in arsenic toxicities. The putative dietary supplement spirulina alone and in combination with zinc were found to be beneficial in patients of chronic arsenic poisoning (Bhattacharya, 2017)

Conclusion

Arsenic toxicity in rats increased arsenic concentrations in lung, liver and kidney with the progress of time. Separate treatment with spirulina and enthe lowered arsenic contents in lung, liver and kidney. Arsenic toxicity caused hepatic and renal dysfunction and both spirulina and mentha treatment might improve the hepatic function in rats. In conclusion, spirulina and mentha were found effective in prevention of chronic arsenic toxicity in rats.

References

Ahmad, S. A., M. H. Faruquee, M. H. S. U. Sayed, M. H. Khan, M. A. Jalil, R. Ahmed and S. A. Hadi. 1998. Chronic arsenicosis: Management by vitamin A, E, C regimen. JOPSOM. 17: 19-26.

Akter, S., M. M. Hasan, R. Islam, F. B. Aziz, M. M. M. Parvez, S. A. Ripa, T. Akter and M. H. Ali. 2018. Effect of Spirulina (Spirulina platensis) And Riboflavin against Arsenic Toxicity. *Int. J. Sci. Busi.* 2(3): 447-458.

Bhattacharya, S. 2017. Medicinal plants and natural products in amelioration of arsenic toxicity: a short review, *Pharmqa. Biol.* 55(1): 349-354.

Das, H. K., D. A. Chowdhury, S. Rahman, M. U. Miah, P. Sengupta and F. Islam. 2000. Arsenic Contamination and Related Bio-hazards in Bangladesh. DPHE-BGS Groundwater studies for arsenic contamination in Bangladesh. Department of Public Health and Engineering and British Geological Survey. Pp. 11-49.

Dey, R. 2002. Management protocol for arsenicosis cases. Report of a Regional Consultation of World Health Organization on Arsenicosis. Case-Detection, Management and Surveillance, India, 5-9 November, 2002.

Fariduddin, A. K. M., M. Misbauddin, M. I. R. Mamun and N. Nahar. 2001. Alcohol extract and residue of spirulina in the prevention of accumulation of arsenic in rats. *Bang. J. Physiol. Pharmacol*. 17: 15-17.

Halim, M. A. 2007. Effects of spirulina on arsenic toxicity in goats. M.S. Thesis. Department of Pharmacology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh, Bangladesh.

Hasan, M., M. A. Awal, A. Ara, M. B. Rashid, M. G. Azam and M. H. Ali. 2015. Study on Prevention of Induced Arsenic Toxicity in Rats by using Spirulina and Thankuni Sch. *J. App. Med. Sci.* 3(2): 828-832.

Hossain, M. F. 2006 Arsenic Centaminatim in Bangladesh**.** Agricult. Ecosyst. & Environ. 113(1-4): 1-16.

Hou-Mai, H., Y. Zhu-Wen, Z. Lu-Xin, Hou-MH, Zhu-WY and Lu-XZ. 2003. Effects of vitamin C and arsenic trioxide on proliferation and apoptosis of mouse melanoma cloundman S91. Department of Dermatology, First Affiliated Hospital, Nanjing Medical University, Nanjing 210029, china. *J. C. Dermatol.* 32(12): 700-702.

Kalia, K. and S. J. Flora. 2005. Strategies for safe and effective therapeutic measures for chronic arsenic and lead poisoning. *J. Occup. Health.* 47: 1-21.

Kamaluddin, M. and M. Misbahuddin. 2007. Zinc supplement reduces tissue arsenic concentration in rats. *Bangla Med. Res. Coun.Bull.* Pp. 11-16.

Kaur, H., V. Mani and C. S. Mishra. 2005. Effect of arsenic on immunity, oxidative enzyme and various haematological parameters in crossbred calves. Asian-Australas. *J. Anim. Sci.* 18 (4): 497-501.

Khalequzzaman, M., F. S. Faruque and A. K. Mitra. 2005. Assessment of arsenic contamination of groundwater and health problems in Bangladesh. *Int. J. Environ. Res. Pub. Health*. 2: 204-13.

Kim, H. S., Y. J. Kim and Y. R. Seo. 2015. An overview of carcinogenic heavy metal: molecular toxicity mechanism and prevention. *J Cancer, Prev*. 20: 232–240.

Mahaffey, K. R., S. G. Capar, B. C. Gladen and B. A. Fowler. 1981. Concurrent exposure to lead, cadmium, and arsenic. Effects on toxicity and tissue metal concentrations in the rat. *J. Lab. Clin. Med.* 98 (4): 463-81.

Mandal, B. K. and K. T. Suzuki. 2002. Arsenic round the world: a review. *Talanta.* 58: 201-235.

Marafante, E., F. Bertolero, J. Edel, R. Pietra and E. Sabbioni. 1982. Intracellular interaction and biotransformation of arsenite in rats and rabbits. *Sci. Total. Environ.* 24:27-39.

Mazumder, D. N. 2005. Effect of chronic intake of arsenic-contaminated water on liver. Toxicol. *Appl. Pharm.* 206(2): 169-75.

Mazumder, D. N. G. 2008. Chronic arsenic toxicity and human health. *Indian J. Med. Res*. 128:436-447.

Mazumder, G., D. N. De, B. K. Santra, A. Ghosh, N. Das, S. Lahiri and T. Das. 2001. Randomized placebocontrolled trial of 2, 3-dimercapto-1-propanesulfonate (DMPS) in therapy of chronic arsenicosis due to drinking arsenic-contaminated water. *J. Clin. Toxicol.* 39(7): 665-74.

Misbauddin, M., A. Z. Islam, S. Khandker, I. A. Mahmud, N. Islam and A. Anjumanara. 2006. Efficacy of spirulina extract plus zinc in patient of chronic arsenic poisoning: a randomized placebo- controlled study. *Clin. Toxicol.* 44(2): 135-141.

Nandi, D., R. C. Patra and D. Swarup. 2006. Oxidative stress indices and plasma biochemical parameters during oral exposure to arsenic in rats. *Food Chem. Toxicol.* 16: 774-805.

Rabbani, G. H., S. K. Saha, M. Akhtar, F. Marni, A. K. Mitra, S. Ahmed, M. Alauddin, M. Bhattacharjee, S. Sultana and A. K. A. Chowdhury. 2003. Antioxidants in detoxification of arsenic-induced oxidative injury in rabbits: Preliminary results. *J. Environ. Sci. Health*., Part A: Tox Hazard Subst Environ Eng., 38(1): 273-87.

Ramanathan, K., S. Shila, S. Kumaran and C. Panneerselvam. 2005. Protective role of ascorbic acid and α-tocopherol on arsenic-induced microsomal dysfunctions. *Hum. Exp. Toxicol.*, 22(3): 129-36.

Ratnaike, R. N. 2003. Acute and chronic arsenic toxicity. *Postgrad. Med. J.,* 79: 391-396.

Roy, A., A. Das and B. Saha. 2006. Effect of ascorbic acid on reduced glutathione level in arsenic-loaded isolated liver tissues of rat. *Bangla. J. Pharmacol,* 1 (3): 68-71.

Sharma, A., M. K. Sharma and M. Kumar. 2007. Protective effect of Mentha piperita against arsenic-induced toxicity in liver of Swiss albino mice, *Basic-and-Clinical-Pharmacol. Toxicol.* 100(4): 249-257.

Shi, H., X. Shi and K. J. Liu. 2004. Oxidative mechanism of arsenic toxicity and carcinogenesis. *Mol. Cell. Biochem*. 255(1-2): 67-78.

Steel, R. G. D. and J. H. Torrie. 1980. Principles and procedures in statistics. Mc. Graw-Hill Book Company Inc. New York, USA.

WHO. 2001. Arsenic and Arsenic Compounds. 2nd ed. Environmental Health Criteria 224. Geneva, World Health Organization.

Yoshida, T., H. Yamauchi and G. F. Sun. 2004. Chronic health effects in people exposed to arsenic via the drinking water: dose-response relationship in review. *Toxicol. Appl. Phrmacol*., 198(3): 243-52.

Zhang, X., R. Cornelis, J. DeKimpe, L. Mees, V. Vanderbiesen and R. Vanholder. 1995. Determination of total arsenic in serum and packed cells of patients with renal insufficiency. Fresenius'. *J. Analytical Chem*. 353(2): 143-147.