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Ameliorative effect of *Triticum aestivum* Linn against experimentally induced arsenic toxicity in male albino rats

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ABSTRACT

The present study evaluated the effect of Wheatgrass extract against arsenic induced toxicity in male albino rats. Wheatgrass extract (200 and 400mg/kg) was administered orally to rats for 20 consecutive days before oral administration of sodium arsenite (10 mg/kg) for 8 days. Then the body weights, organ weights, haematological profiles, serum biochemical profile; hepatic and renal antioxidative parameters viz. lipid peroxidation, reduced glutathione, glutathione reductase and catalase were evaluated. Pretreatment with Wheatgrass extract markedly and significantly normalized body weights, organ weights, haematological profiles, serum biochemical profile and significantly modulated all the hepatic and renal biochemical parameters. Cystic degeneration in the tubular region and mild tubular hemorrhage in kidney, hepatic degeneration and disturbance of sinusoids in the liver was observed after exposure to arsenic. Treatment with Wheat grass extract ameliorated the changes induced by arsenic. The present findings conclude that Wheat grass extract possessed remarkable effect against arsenic induced organ toxicity in male albino rats mediated by alleviation of arsenic induced oxidative stress by enhancing the anti-oxidant defense mechanism and also by detoxification of free radicals generated in the body.

Keywords: Arsenic toxicity, Wheat grass extract, Oxidative stress, Sodium arsenite, Liver, Kidney

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INTRODUCTION

Arsenic is the twentieth most abundant element, natural and ubiquitous in earth's crust and introduced into soil and groundwater during weathering of rocks followed by subsequent leaching and runoff. It can also be introduced into soil and groundwater from anthropogenic activities [1]. Humans may receive arsenic exposure predominantly through contaminated drinking water, whereas inhalation and skin absorption can be the minor entry routes [2]. Besides the sources of arsenic contaminated drinking water, use of arsenic containing pesticides, herbicides and rodenticides are also potential origins of arsenic toxicity.

According to the World Health Organization (WHO) the consumption of drinking water containing arsenic more than 10g/L is harmful to the human body [3]. Chronic ingestion of arsenic above this level precipitates chronic arsenic poisoning (arsenicosis). Chronic arsenic exposure through drinking water to humans leads to carcinogenesis of almost all organs, skin diseases including hyper pigmentation, hyperkeratosis leading to cancers of skin and epithelial tissues; hepatic, renal, cardiovascular, respiratory, central nervous system (CNS), gastrointestinal, reproductive complications and impairment of children's intellectual development, thus increasing morbidity and mortality [4, 5].

Groundwater with elevated concentrations of arsenic has been recognized as a problem of global concern. Incidences of chronic arsenic poisoning by long term consumption of arsenic contaminated groundwater have been reported in several countries throughout the world. Out of them the most affected countries are India, Bangladesh, China and Taiwan [6]. Arsenic contamination in groundwater in the Ganga-Brahmaputra fluvial plains in India and Padma-Meghna fluvial plains in Bangladesh and its consequences to the human health have been regarded as one of the world's biggest natural groundwater calamities to the mankind [7].

Arsenic is known to exert its toxicity by binding to cellular sulfhydryl groups, accounting for its ability to interfere with energy generation [8]. Recent studies suggest that arsenicals during biotransformation in cells generate several reactive oxygen species (ROS) that cause organ toxicity, mediated either by oxidative damage of cellular biomolecules like lipids proteins and DNA, or through ROS signalling cascades associated with carcinogenesis pathways [9]. Arsenic toxicity is considered as a serious problem worldwide, as still now there is no specific, safe and efficacious therapeutic management of arsenicosis. Nowadays, the management of chronic arsenic toxicity is mainly restricted to the use of a handful of sulfhydryl containing chelating agents which have been reported to exhibit certain adverse effects [10]. Therefore, the current research trend is to exploit the therapeutic potential of medicinal plants to combat arsenic induced tissue damage.

Shoot of *Triticum aestivum* Linn. (Hindi Name- gehun, kanak, and Sanskrit name- godhuma) is called as a Wheat grass, belonging to family: Gramineae, which possess high chlorophyll content and essential vitamins, minerals, vital enzymes, amino acids, dietary fibers [11]. Wheat grass extract has been shown to possess anti-cancer activity [12], anti-ulcer activity [13], antioxidant activity [14], anti-arthritic activity [15], and blood building activity in Thalassemia Major [16]. It has been argued that Wheat grass extract helps blood flow, digestion and general detoxification of the body. Wheatgrass extract contains minerals and trace elements including calcium, iodine, magnesium, selenium, zinc, chromium, antioxidants like vitamin C, vitamin E, β-carotene, vitamin B_1, antianemic factors like vitamin B_{12}, iron, folic acid, pyridoxine, abscissic acid, ferulic acid, and vanilic acid the concentrations of which increase with the germination period [17]. The major clinical utility of Wheat grass extract in diseased conditions might be due to the presence of these biologically active compounds and minerals in it and due to its antioxidant potential which is derived from its high content of bioflavonoids such as apigenin, quercitin, luteoline. Furthermore, indole compounds namely choline and laetrile present in it might be also responsible for its therapeutic potential. The presence of 70% chlorophyll, which is almost chemically identical to hemoglobin, in Wheat grass extract makes it more useful in various clinical conditions involving hemoglobin deficiency and other chronic disorders.

Considering the rich antioxidant and vitamin contents of Wheatgrass extract, this study investigated status of Wheatgrass extract in modifying the arsenic induced biochemical alterations in blood of albino rats.

MATERIALS AND METHODS

2.1.1. Growing of Wheat grass extract The grass of *T. aestivum* used in this study was grown under indoor conditions. Over-night soaked *T. aestivum* seeds were used to cultivate. Little quantities of water were sprinkled evenly over soil and 3-4 hours indirect sunlight was allowed daily for growth of grass. On the Seventh day, grass is harvested and used for further studies.

2.1.2. Preparation of plant extract The harvested wheatgrass (*T. aestivum) is* washed dried at room temperature. The dried grass was subjected to size reduction to a coarse powder by using dry grinder and passed through sieve. This powder was packed into soxhlet apparatus and made to extract using ethanol. The extract was then filtered and concentrated under reduced pressure using a rotator evaporator at 40°C until the solvent completely dried. The yield of the ethanolic extract was 30%. The extract obtained was then dissolved in 2% Gum acacia for the pharmacological studies.

2.2. Drugs and chemicals

Bovine serum albumin, Bradford reagent, quercetin from Sigma–Aldrich Chemical Co., St.Louis, Mo, USA; trichloroacetic acid (TCA) and 1-dichloro-2,4-dinitrobenezene (CDNB) from Merck Ltd., Mumbai, India; thiobarbituric acid (TBA), nitroblue tetrazolium chloride (NBT) from Loba Chemie, Mumbai, India; 5,5_-dithio *bis*-2-nitro benzoic acid (DTNB), phenazonium methosulphate (PMS), reduced nicotinamide adenine dinucleotide (NADH) reduced glutathione (GSH) sodium arsenite (NaAsO2) trichloroacetic acid (TCA) from SISCO Research

Laboratory, Mumbai, India. All the other reagents used were of analytical reagent grade obtained commercially. Doubled distilled water from all-glass still was employed throughout the study.

2.3. Experimental animals

Thirty male Wistar Albino rats weighing 150gms-250gms were obtained from National Institute of Nutrition, Hyderabad. The rats were housed in polypropylene cages and maintained under standard conditions (12 h light and dark cycles, at 25±3° C and 35-60% humidity). Standard pelletized feed and tap water were provided *ad libitum*. All the pharmacological experimental protocols were approved by the Institutional Animal Ethics Committee (Reg no: MRCP/CPCSEA/IAEC/2012-13/MPCOL/07). Acute toxicity study of wheatgrass extract was carried out for determination of LD_{50} by adopting fixed dose method of CPCSEA, OECD guideline no.423. A group of albino mice was used for this study. Acute toxicity studies were conducted and no mortality was observed till the dose of 2000mg/kg. Hence 1/5th, 1/10th of the dose 2000mg/kg i.e. 200mg/kg and 400mg/kg has been fixed for the study.

2.4. Experimental animals design:

Thirty Wistar Albino male rats of weight 150gms-250gms were selected for this study. Animals were divided into five groups of six animals each.

Group 1: Control group (1% Gum acacia 1ml)
Group 2: Sodium arsenite10mg/kg bodyweight p.o.
Group 3: Quercetin 10mg/kg body weight p.o.+ Sodium arsenite 10mg/kg body weight p.o.
Group 4: Wheatgrass extract extract 200mg/kg body weight p.o.+ Sodium arsenite 10mg/kg body weight p.o.
Group 5: Wheatgrass extract extract 400mg/kg body weight p.o.+ Sodium arsenite 10mg/kg body weight p.o.

Animals were grouped into five groups as explained above. The control group animals were given 1% Gum acacia 1ml for 28 days. Group 2 animals were given normal saline water until 20th day and doses of Sodium arsenite 10mg/kg body weight p.o. was given from 21st day to 28th day of the experiment. Group 3 animals were given Quercetin 10 mg/kg body weight p.o. until 28th day and doses of Sodium arsenite 10mg/kg body weight p.o. were given from 21st day to 28th day of the experiment. Group 4 animals were given Wheatgrass extract 200mg/kg body weight p.o. until 28th day and doses of Sodium arsenite 10mg/kg body weight p.o. were given from 21st day to 28th day of the experiment. Group 5 animals were given Wheatgrass extract 400mg/kg body weight p.o. until 28th day and doses of Sodium arsenite 10mg/kg body weight p.o. were given from 21st day to 28th day of the experiment.

Twenty-four hours after the last dose and after 18 h of fasting, blood was collected from rats of each group, by cardiac puncture for the estimation of hematological parameters and then organs were isolated for the study of hepatic and renal biochemical parameters.

2.5. Body weight and organ weights

The body weights of rats of each group were measured just before and after completion of the treatments. Liver and kidney weights of all rats were measured after post treatment sacrifice.

2.6. Evaluation of haematological parameters

Collected blood was used for the estimation of haemoglobin (Hb) content; red blood cell (RBC) and white blood cell (WBC) counts [18, 19].

2.7. Evaluation of serum biochemical parameters

Collected blood was used for the estimation of serum biochemical parameters *viz*. SGPT, SGOT [20], alkaline phosphatase (ALP) [21], serum bilirubin [22] and total protein [23].

2.8. Evaluation of hepatic and renal biochemical parameters

Lipid peroxidation, *i.e.,* thiobarbituric acid reactive substances (TBARS) was measured as per reported method [24]. Reduced glutathione (GSH) was assessed by the method of Hissin and Hilf (1973) [25]. The enzymatic activity of glutathione reductase (GR) was evaluated by the reported method (Smith et al., 1988) [26]. The activity of catalase (CAT) was assayed following previously reported method (Sinha, 1972) [27].

2.9. Statistical analysis

All data are presented as the mean ±standard error of mean (SEM). The results were analysed for statistical significance by one-way analysis of variance (ANOVA) followed by Dunnett's *post hoc* test of significance. *P* values less than 0.05 ($p \leq 0.05$) were considered as statistically significant.

RESULTS

3.1. Preliminary phytochemical screening of Wheatgrass extract:

The main chemical constituents that are found in the extract of Wheatgrass extract are Saponins, Carbohydrates, alkaloids, aminoacids, flavonoids, fixed oils, fats and proteins. Glycosides, sterols, triterpenoids are completely absent.

3.2. Effect of Wheatgrass extract on Body weights and organ weights:

The bodyweight, liver and kidney weights of rats from toxin control group were significantly ($p<0.05$) decreased when compared with normal control group. Treatment with Wheatgrass extract prior to arsenic intoxication significantly ($p<0.05$) maintained the bodyweight, liver and kidney weights towards normal in a dose related manner as compared to toxin control group (Figure 1, 2). The effect of Wheatgrass extract at higher dose was comparable to those of quercetin.

3.3. Effect of Wheatgrass extract on Hematological parameters:

Hematological parameters of toxin control rats were found to be significantly altered compared to those of the normal control group. The leucocytes (WBC) count was found to be increased; RBC and haemoglobin count was significantly decreased in toxin control animals significantly when compared to the normal control group. Treatment with Wheatgrass extract produced a significant increase in Hb level, RBC count. Whereas, in sodium arsenite intoxicated animals treatment with Wheatgrass extract prevented the hemolysis and significantly increased the Hb content and RBC count. WBC count was also brought back towards the normal levels when compared to toxin control animals. The effect of Wheatgrass extract was comparable to those of Quercetin treated animals (Figure 3).

3.4. Effect of Wheatgrass extract on Serum biochemical parameters:

Serum biochemical parameters like AST, ALT, ALP and serum bilirubin in the toxin control group were significantly ($p < 0.05$) elevated as compared to those of normal control group. Treatment with Wheatgrass extract before arsenic challenge significantly ($p < 0.05$) reduced the AST, ALT, ALP and serum bilirubin levels towards the normal values in a dose dependant mannner. The total protein was found to be significantly decreased in the toxin control group as compared with the normal control group ($p < 0.05$). Administration of Wheat grass extract prior to arsenic induction in rats significantly ($p < 0.05$) increased the total protein content as compared with the toxin control group. The effect of Wheatgrass extract at higher dose was comparable to those of reference compound Quercetin (Table 1).

3.5. Effect of Wheatgrass extract on renal and hepatic antioxidant parameters:

Treatment with Wheatgrass extract prior to arsenic intoxication dose dependently and significantly ($p < 0.05$) reduced the MDA level when compared with toxin control rats. In the toxin control group, GSH content was found to be significantly ($p < 0.05$) lowered. Wheatgrass extract pre-administration dose dependently and significantly ($p < 0.05$) normalized their contents in both hepatic and renal tissues. Due to arsenic intoxication the activities of GR were found to be significantly ($p < 0.05$) diminished in the toxin control animals when compared with normal control ones. In both liver and kidney, Wheatgrass extract treatment before arsenic induction significantly ($p < 0.05$) brought back their activities towards normal in a dose related way as compared with the toxin control group (Table 2, 3).

There was significant ($p < 0.05$) reduction in CAT activities in toxin control group compared with the normal control group. Treatment with Wheatgrass extract prior to arsenic challenge significantly ($p < 0.05$) and dose dependently recovered their activities near to normal values when compared with toxin control animals.

DISCUSSION

The present study was focused to evaluate the protective effect of Wheat grass extract in preventing Arsenic induced damage in Wistar Albino rats. The results of the present study revealed that Wheat grass extract treatment at the doses of 200 and 400 mg/kg prior to arsenic intoxication exerted marked amelioration from arsenic toxicity as evidenced by significantly restoring the altered body weights, organ weights, haematological and serum biochemical parameters towards normal values. The Wheat grass extract pretreatment significantly recuperated the perturbed hepatic and renal antioxidative parameters implying its antioxidant role.

Body weight is regarded as a nonspecific indicator of general wellbeing of animals. Reduction in body weight is an indicator of decline in general health condition of arsenic induced experimental rats. The results of the current study indicated that arsenic intoxication markedly decreased the body weight, liver and kidney weights of experimental rats implying impaired animal growth and organ functions. Treatment with Wheat grass extract prior to arsenic intoxication significantly maintained the body weight, liver and kidney weights of experimental rats towards normal after arsenic exposure.

It is known that arsenic mediates its toxicity through reaction with cellular sulfhydryl (SH) groups [28]. Arsenic intoxication resulted in depletion in haemoglobin and RBC and on the other hand proliferation of WBC, indicating its toxicity towards the blood and hematopoietic system. WBC is regarded as one of the important sources of ROS generation [29]. Results of the present study indicated that Wheatgrass extract pretreatment dose dependently and significantly brought the reduced hemoglobin content back towards normal, raised the erythrocyte count and reduced the leucocyte count significantly as compared with those of toxin control rats. These indicating parameters revealed that Wheatgrass extract exerted less toxic effect to the blood and hematopoietic system and thus could maintain the normal hematological profile in arsenic induced rats.

It is well known that liver is the important target organ of arsenic toxicity [30]. Several enzymes of blood serum are considered as indicators of hepatic dysfunction, damage and the leakage of hepatic enzymes such as AST, ALT and ALP into blood are routinely used as a reliable biochemical index for hepatocellular damage [31]. It was also found that hepatocellular damage exhibited good correlation with the enzyme leakage to bloodstream [32]. In the present findings, arsenic intoxication caused a significant increase in the activities of AST, ALT and ALP probably due to the leakage of these enzymes from the damaged hepatocytes into the blood, and/or liver dysfunction and disturbance in the biosynthesis of these enzymes. In the present investigation, the increase in serum bilirubin concentrations in toxin control rats was observed. It was found that increase in serum bilirubin is associated with free radical production [33]. Raise in bilirubin level can be either due to increased RBC and hemoglobin breakdown evidenced form the hematological alterations, or due to hepatic damage evidenced from the anomalous serum biochemistry. In the present study, significant decrease in serum total protein was recorded in arsenic induced rats. This decrease might be due to alterations in protein synthesis and/or metabolism [34]. The observed decrease in serum proteins could also be attributed in part, to the damaging effect of arsenic on hepatocytes as evidenced and confirmed by the abnormal increase in activities of fore said serum enzymes [35]. Treatment with Wheat grass extract prior to arsenic intoxication significantly reduced the AST, ALT, ALP and bilirubin levels and increased the total protein content towards normal in arsenic treated experimental rats.

Recent studies demonstrated that arsenic compounds during their metabolism generate excessive amount of ROS leading to oxidative stress impairing endogenous antioxidant defense mechanisms and simultaneously damaging the cellular macromolecules such as lipids, proteins and DNA, resulting in disruption of cell structure and functions [36, 37]. In the present finding, arsenic administration induced oxidative stress in rat liver and kidney as evidenced by perturbations in various hepatic and renal antioxidative parameters,

which can be suppressed by treatment with Wheatgrass extract prior to arsenic administration. Lipid peroxidation is considered as a molecular mechanism of oxidation of cellular lipid based macromolecules. Overproduction of ROS enhances the lipid peroxidation and subsequently increases the lipid peroxidation products like malondialdehyde (MDA) and other MDA levels which lead to degradation of cellular macromolecules. Increased MDA is regarded as a biomarker of enhanced lipid peroxidation [38]. Enhanced tissue lipid peroxidation is one of the characteristic features of arsenic toxicity induced oxidative insult [7]. The present results showed that MDA levels in the toxin control liver and kidney tissues were much higher than those in normal control tissues. Oral treatment with Wheatgrass extract prior to arsenic challenge significantly prevented tissue lipid peroxidation as revealed by reduction of MDA levels in hepatic and renal tissues towards normal.

The cellular enzyme-based antioxidative mechanism serves the first line of endogenous defense against oxidative impact. Superoxide dismutase (SOD) prevents the deleterious consequences of superoxide ion by accelerating its dismutation into comparatively less toxic hydrogen peroxide, which can be regarded as primary defense, as it prevents further generation of reactive free radicals; hydrogen peroxide subsequently converts into non toxic water and oxygen molecules by the action of catalase (CAT). In our present study, 28 days oral Wheatgrass extract treatment prior to arsenic administration significantly modulated the activities of CAT and GR in hepatic and renal tissues of arsenic treated rats, indicating the boosting of enzymatic antioxidant defense mechanisms by which Wheatgrass extract may have mediated amelioration of arsenic induced organ toxicity.

The non-enzymatic thiol based antioxidative mechanism serves as the second line of endogenous defense against oxidative damage. Glutathione, the most abundant tripeptide thiol, exists as GSH (reduced form) in cells and participates in several physiological processes, including detoxification of xenobiotics. Primarily it serves as a catalyst in disulphide exchange reactions. It functions as reducing agent by scavenging the free radicals as well as detoxifying xenobiotics and itself gets oxidized to GSSG [39]. Besides its involvement in the detoxification process, GSH probably also plays an important role in lymphocyte function and depletion in GSH content were found to be associated with impaired immune response and increased risk of malignancy [40]. As previously discussed, arsenic has got a high affinity for tissue sulfydryl groups and hence it actively binds with reduced glutathione causing its depletion in tissues. Arsenic is detoxified by methylation by S-adenosyl methionine leading to formation of methylated end metabolites which are readily excreted in urine. Glutathione and possibly other thiols serve as reducing agents. When GSH levels are depleted, the inorganic arsenic accumulates in the system and thus increases toxicity [41]. In the present study, Wheatgrass extract pretreatment resulted in significant recovery of GSH contents in arsenic intoxicated rats. The improved levels of GSH might protected the sulfydryl groups form binding with arsenic and promoted the detoxification of arsenic by modulating arsenic methylation reactions which favoured its excretion form the system [42].

Natural antioxidant quercetin was also pretreated prior to arsenic challenge in the current study similarly as Wheatgrass extract. This putative antioxidant is known to ameliorate arsenic toxicity [7, 43]. Quercetin is more effective than Wheat grass extract.

The protective efficacy of Wheatgrass extract may be due to presence of several active components. The active component found in Wheatgrass extract may provoke the activity of free radical scavenging enzyme systems and renders protection against Arsenic-induced damages. The metalloprotective role of Wheatgrass extract may be attributed to the presence of chlorophyll, saponins and SOD.

Cystic degeneration in the tubular region and mild tubular hemorrhage in kidney, hepatic degeneration and disturbance of sinusoids in the liver was observed after exposure to arsenic. Treatment with Wheat grass extract ameliorated the changes induced by arsenic.

Chlorophyll: A 70-83% increase in red blood cells and hemoglobin concentration was noted within 10-16 days of regular administration of chlorophyll derivatives. It was reported that chlorophyll enhanced the formation of blood cells in anemic animals. Chlorophyll is soluble in fat particles, which are absorbed directly into blood via the lymphatic system. Chlorophyll present in Wheatgrass extract can protect us from carcinogens; it strengthens the cells, detoxifies the liver and blood stream, and chemically neutralizes the polluting elements.

It has superoxide dismutase and Cytochrome oxidase [6], which neutralize free radicals and prevent them from causing the cell damage. Because it is also high in saponins, Wheatgrass extract helps to detoxify the body by increasing the elimination of hardened mucous, crystallized acids and solidified, decaying fecal matter and thus, provides an optimum nutritional environment inside the body system. Wheat grass extract is an extremely effective way of boosting the immune system to fight against diseases.

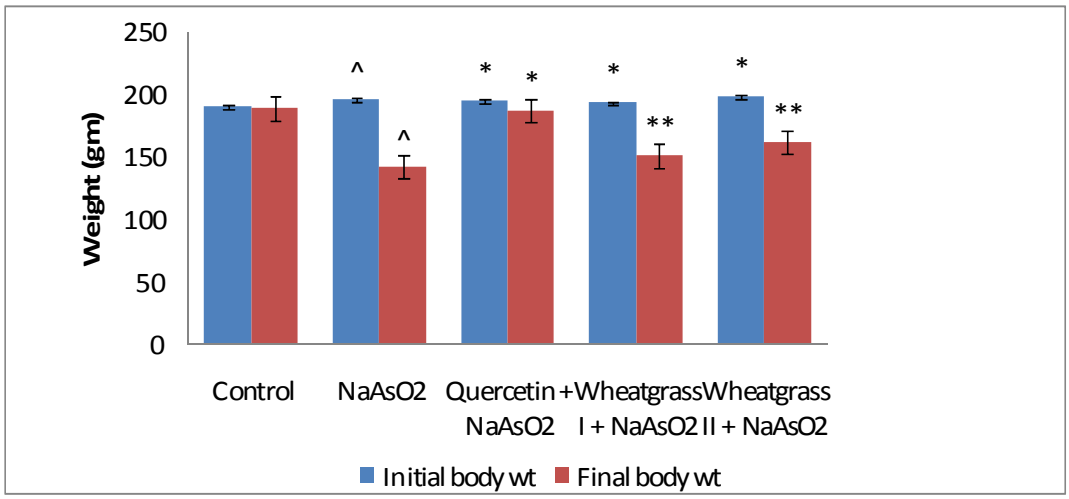

Figure 1: Effect of Wheatgrass on body weight in normal and arsenic intoxicated rats
*Values are expressed as mean ±SEM., Data analyzed by one way ANOVA followed by Dunnet's Multiple Comparison Test. **p<0.01 and *p<0.05 as compared with arsenic group, ^p<0.05 as compared with control group.*

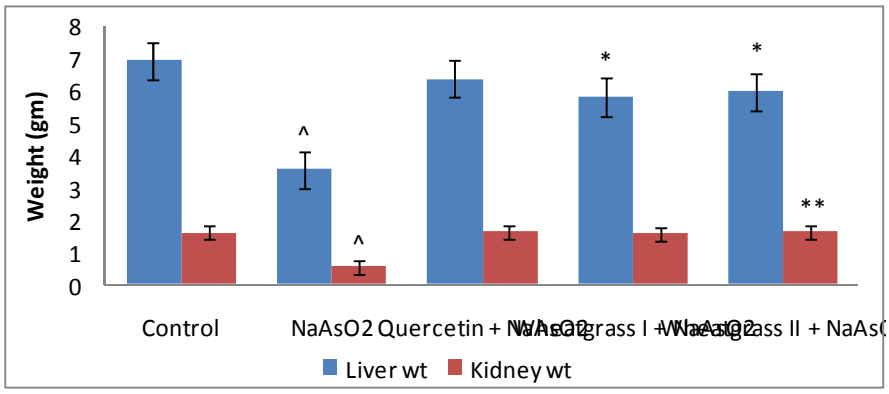

Figure 2: Effect of Wheatgrass on organ weight in normal and arsenic intoxicated rats.
*Values are expressed as mean ±SEM., Data analyzed by one way ANOVA followed by Dunnet's Multiple Comparison Test. **p<0.01and *p<0.05 as compared with arsenic group, ^p<0.05 as compared with control group*

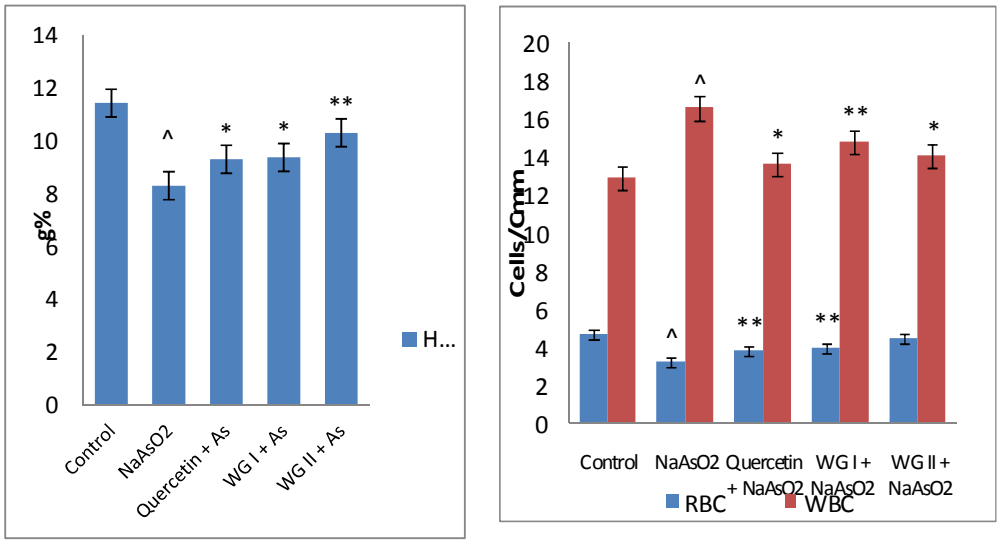

Figure 3: Effect of Wheatgrass on Hematological Parameters in normal and arsenic intoxicated rats

*Values are expressed as mean ±SEM., Data analyzed by one way ANOVA followed by Dunnet's Multiple Comparison Test. **p<0.01and *p<0.05 as compared with arsenic group, ^p<0.05 as compared with control group.*

Table 1: Effect of Wheatgrass extract on serum biochemical parameters in normal and arsenic intoxicated rats

Parameters/Groups	SGPT IU/L	SGOT IU/L	ALP IU/L	Total Protein g/dl	Bilirubin mg/dl	Creatinine mg/dl
Normal Control	60.5±6.95	96.88±o.44	178.73±0.75	9.846±0.008	1.496±0.03	6.86±0.39
$NaAsO_2$ 10mg/kg	173±4.97^	186.7±0.28^	342.63±0.49^	6.875±0.035^	3.75±0.145^	4.55±0.19^
Quercetin+$NaAsO_2$ 10mg/kg + 10mg/kg	115.3±8.68*	108.9±0.4*	318.53±0.318*	8.95±0.02*	1.9±0.006*	5.61±0.13*
WGE I + $NaAsO_2$ 200mg/kg+10mg/kg	162±5.30**	143.36±0.53**	303.11±0.1*	5.77±0.025*	2.1033±0.12*	6.78±0.199**
WGE II + $NaAsO_2$ 400mg/kg+10mg/kg	141.2±8.07*	123.9±0.46**	293.55±0.5**	8.22±0.02*	1.79±0.01*	5.03±0.2*

*Values are expressed as mean ±SEM., Data analyzed by one way ANOVA followed by Dunnet's Multiple Comparison Test. **p<0.01and *p<0.05 as compared with arsenic group, ^p<0.05 as compared with control group.*

Table 2: Effect of Wheatgrass extract on MDA and CAT in normal and arsenic intoxicated rats

Groups (N=6)	MDA(nm/g)		CATALASE(k/min)	
	LIVER	KIDNEY	LIVER	KIDNEY
Normal Control	76.86±0.23	35.5±0.95	56.34±0.43	36.7±0.39
$NaAsO_2$ 10mg/kg	44.55±0.56^	53.96±1.97^	40.94±0.81^	18.6±0.19^
Quercetin + $NaAsO_2$ 10mg/kg + 10mg/kg	69.61±0.76**	39.5±0.68	64±0.54**	32.4±0.13
WGE I + $NaAsO_2$ 200mg/kg + 10mg/kg	56.78±0.43**	47.3±0.30*	51±0.32 *	24.5±0.1
WGE II + $NaAsO_2$ 400mg/kg + 10mg/kg	65.03±0.65**	42.5±0.07*	62.56±0.38**	28.7±0.2**

*Values are expressed as mean ±SEM., Data analyzed by one way ANOVA followed by Dunnet's Multiple Comparison Test. **p<0.01and *p<0.05 as compared with arsenic group, ^p<0.05 as compared with control group.*

Table 3: Effect of wheatgrass extract on GR and GSH in normal and arsenic intoxicated rats

Groups (N=6)	GR(nmol/min/mg of protein)		GSH(mcg/ml)	
	LIVER	KIDNEY	LIVER	KIDNEY
Normal Control	35.5±0.87	56.86±0.39	49.1±0.60	35.34±0.73
$NaAsO_2$ 10mg/kg	53.96±0.66^	34.55±0.19^	34.6±0.22 ^	20.94±0.60^
Quercetin + $NaAsO_2$ 10mg/kg + 10mg/kg	39.5±0.26*	54.61±0.13*	47.8±0.43 *	32.57±0.499
WGE I + $NaAsO_2$ 200mg/kg + 10mg/kg	47.3±0.32*	42.78±0.19**	39.7±0.65 **	27.04±0.90**
WGE II + $NaAsO_2$ 400mg/kg + 10mg/kg	42.5±0.11**	53.03±0.3	42.3±0.23 *	31.56±0.08

*Values are expressed as mean ±SEM., Data analyzed by one way ANOVA followed by Dunnet's Multiple Comparison Test. **p<0.01and *p<0.05 as compared with arsenic group, ^p<0.05 as compared with control group.*

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Abbreviations:

Fig. - Figure
i.p. - intra peritoneal
kg - kilogram
mg - milligram
ml - millilitre
p.o. - Per oral
w/w - weight/ weight
w/v - weight/ volume
LD_{50} - Lethal dose
rpm - rotations per minute
GSH - Glutathione
CAT - Catalase
LPO - Lipid peroxidation.
MDA - malondialdehyde
SGPT – serum glutamate pyruvate transaminase
SGOT – serum glutamate oxaloacetate transaminase
ALP - alkaline phosphatase
DNA - Deoxy ribo nucleic acid
ROS - Reactive oxygen species
GR - Glutathione reductase

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