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Effect of Toxins (Microcystines, Shiga & Botulinum) on Liver Functions

Moghira Badar[1*], Qamar Mahmood K[1], Fatima Batool[2]

[1]Department of Environmental Management, National College of Business Administration and Economics, Lahore
[2]National Centre of Excellence in Molecular Biology, University of the Punjab, Lahore

Abstract— *Lfts (liver function tests) and Rfts (renal functions tests) values of patients show abnormalities and it is confirmed swelling of liver and poor filtration rate of kidneys. So, drinking water treatment is most needed.*

All the blood samples were taking under controlled environment to keeping the good quality and standards that support real data for final analysis. LFTs tests are taking by drawing blood samples from infected persons with the help of testing machines, it shows average value of ALT and SGOT is very high as 50 u/l and 53u/l that is not good sign for liver health due to high value of toxins in human blood stream which is confirmed by toxins medical laboratory tests. All these problems is mainly due to taking bio contaminated food and unsafe drinking water, so if we make assure the security and safety about our taking the food as essential component of life.

Keywords— LFTs, Toxins, Liver Functions, Blood sampling, Medical tests.

I. INTRODUCTION

Microcystins are 200 times more toxic and poison than cyanide metal. This toxin has structural variants include amino acid substitutions and alterations such as methylation and demethylation. Drinking water supplies contaminated with Cyanobacteria toxins is a main cause of a health hazard for human beings, domestic animals both large and small, and wildlife animals [1].

Pakistani population is the world's fastest increasing population and it may exceed to 180 million is observed by now; it is still growing with an alarming speed about 2.8% yearly. Current century gives a revolution for improvement in utilization of water and food. We must need to change our cultivation method and life living styles. Concurrently the water quality of underground and surface is poor, further it is deteriorating for the reason is unchecked disposal of untreated industrial and municipal wastes mix in natural sources. In this study, we are identifying the toxins in drinking water and blood and then removing from drinking water [2].

It is proved from study, the diffusion between activated carbon and toxins improve the taste and ordure. In order to test the effect of reducing raw water pH on the nature of the adsorption process value decreases. This increase in the adsorption capacity can be explained as toxins are predominately negatively charged; therefore, decreasing the pH values the negatively charged organic molecules more neutral. According to mechanism of reaction, it is less soluble the neutral molecule in water due to no charged on atoms or molecule, so increasing in pH values is very helpful to remove organic molecules from sample of drinking water [3].

The parameters like light, temperature and humidity are responsible for water quality as taste, natural and colour and their effects are showed the presence of organic matter in water reservoirs because these parameters increase with the dissociation and degradation characters of dissolved organic compound in drinking water such as toxins and other nitrogen based compounds [4].

Conversely, observations due to microcystins toxins from laboratory results presented the toxins which are created by the cyanobacteria specious as found slow growing. The major aspects in measuring for the removal of cyanobacteria toxin from water treatment which includes removing the soluble and suspended substance removes. Previous studied are showing the some like microcystins, botulinum toxins and shiga toxin resolvable in water [5].

The aim of this study is to detect the toxins in human blood samples and their harm effects on liver performing functions.

II. MATERIALS AND METHODS

Human Blood sampling

Collect the random human blood sampling from different places were with the frequency of samples (n= 116). All samples were collected by syringe in sterilized blood vessel used as container and blood sample 5 ml collected by volume and actual capacity of container was 5 ml. The temperature of the day when collect the samples was 16 ^{0}C.

Serums of samples were collected after mechanical centrifugation of the samples blood, and start the analysis of

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blood samples. It was used the Chemical reagent (in form kits) to determine concentrations of following parameters in the serum [6].

Microcystins Toxin Testing Method

The technique involved by the adding of 50 µL of negative control, each calibrator with each sample put in the wells and get 50 µL of microcystin with added assay diluent and was incubated for 30 minutes on a shaker. Next, added 100 µL of microcystins enzyme conjugate to each well and once again wells for 30 minutes were incubated, after then the wells four times with wash solution.

The step solution was added of substrate to each well and further for 30 minutes incubated. 100 µL of the stop solution was added to each well after incubation, the contents of the wells change into yellow and measured their optical density with Microplate Reader at 450 nm. The optical density values given by the reader and toxin concentration in the samples was calculated from the standard curve from the 3 Calibrators [7].

Testing Method of Botulinum Toxin

The procedure of testing is involved the following steps for investigating liquid samples for the identification of botulinum toxin, Dilution buffer of 50: 1L was added to all wells that hold a sample, Test sample of 50: L was added to the dilution buffer and incubated the plates for one hour at 25 ^{0}C temperature. Washed three times Plates with 200: 1L of ELISA washes buffer per Well. Detect antibody, was added of 100: L to each sample well and incubated the plates for one hour at 25 ^{0}C temperature [8].

Testing Method of Shiga Toxins

The concentration of residual Shiga toxins after heat treatment was determined by using ELISA, the Premier TM EHEC ELISA kit (Meridian Bioscience, Cincinnati, OH). Multi-point standard curves as well as positive and negative controls were performed to reduce variations among ELISA testing. The cut-off concentration according to the manufacturer's instructions was OD=0.18.

The Premier EHEC test kit utilizes monoclonal anti-Shiga toxin capture antibodies absorbed onto the bottom of micro wells. When testing, 100 µL samples were added to each well mixed thoroughly with the pipette and incubated at room temperature (RT) for 1 hourof the 96-well plates. Then Wells were washed according to manufacturer's instructions. Volumes of 100 µL of polyclonal anti-Shiga toxin antibodies provided by the kit were added into each well, mixed well as before, and incubated at RT for 30 min. Wells were washed again to remove unbound antibody. Aliquots of 100 µL of enzyme-conjugated anti-IgG polyclonal antibody was added into each well, mixed and incubated at RT for 30 min [9].

Liver functions Tests

Collected blood samples were homogenised by using the anticoagulant EDTA vial. Analysis the concentrations of blood parameters like, Protein, Alkaline phosphatase(ALP), Total Bilirubin, Direct bilirubin, Aspartate amino- transferase (AST), Alanine amino transferase (ALT), G-glutamyl transferase (GGT) [10].

III. RESULTS

Concentration of toxins in the blood

All the nations of world are very conscious on drinking water quality, so effective efforts in form of research and study make on this issue. E. coli related with Coliform family which is basic indicator of faecal contamination that also known as disease causing pathogen. Microbial quality is totally depending on presence of coliform group E. Coli. recognized as pathogenic microbes and their metabolise chemical known as toxins. In figure 1, it is clearly shown that all the toxins as microcystins, botulinum and shiga have high values in blood samples due to eating the contaminated food as well as drinking water linked with high absorption of toxins in blood samples.

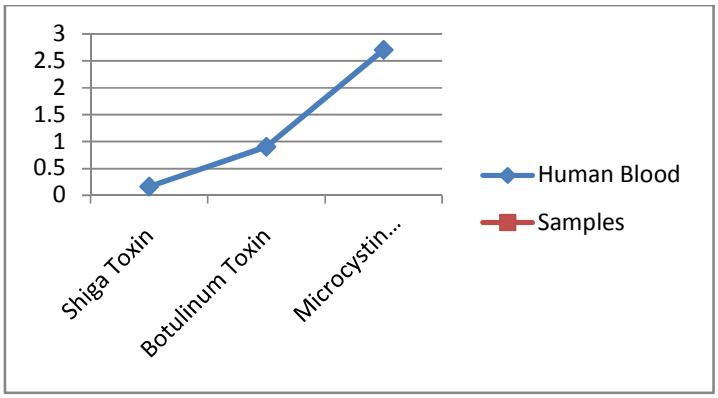

Fig.1: Chemical Analysis of Toxins in Blood (Humans) Samples

Effect of toxins on Liver Function Values

Liver damage situation is a very serious health issue across world due to drinking of bad quality water. Liver function tests are usually represent and recognized as the reliable indicator of liver performance of detoxification function. Inside the liver, enzymatic activity have been raised up, this is may be due to synthesis of enzymes, their low levels indicate that the enzymatic inhibition due to liver injury without specific regeneration. In figure-2, among liver enzymes, amylase GOT, GPT and ALT were elevated in the samples of human's blood, it was showing acute liver damage (hepatitis),

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while in samples of blood, all these enzymes were inhibited showing hypocondition or dysenzymia.

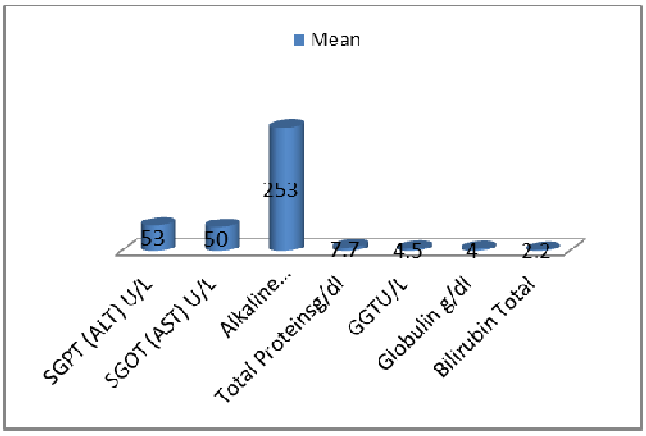

Fig.2: Live Function Clinical Test of Humans

Toxins accumulating within the blood were responsible for other end-organs being damaged. Some of these factors may be due to changes in the cellular component of blood, but many of these deleterious effects are due to changes in the humoral component of circulating blood. These toxins may arise either as a consequence of a failure of normal hepatic functions, or elsewhere in the body as an importance of simple liver disease. These toxic factors in the blood affect the function of many organ systems, such as the systemic and portal vasculature and the brain, as well as the liver itself. The exact nature of these toxins is unknown and may be different and multiple for each organ system damaged. Ammonia, aromatic amino-acids, tryptophan, indoles, mercaptans and endogenous benzodiazepines are implicated in the development of hepatic encephalopathy [11].

Effect of high values of liver functions on human health

Water sources treatment is only solution of these problems; in this case we use the coagulation process with ferric chloride solution of different doses. In this study, it has proved that microcystins are removed using the concentration of ferric chloride dose is 16 mg/l. Liver function tests were also played very important role to know the actual working positions of liver function, so values of Rfts indicated the abnormalities of liver in cows and buffaloes due to continues taking the microcystin toxins from water and food sources.

Results clear about amylase activity in the present investigation increased in both of samples of cows. Amylase is secreted by the exocrine region of pancreas in mammalians system by help of liver function. The increased activity may be due to pancreatitis or due to the damage of the amylase secretary cells inside body. There is also the possibility that greater amounts of amylase were secreted into the intestine, which causes the consequently enhanced starch digestion, and transferred itself to the degradation products into portal blood, and then into liver and hepatic cells through assimilation, which may also be caused for hyperglycemic response in animals.

IV. DISCUSSIONS

In developed countries water treatment and sanitation has removed the problem of diseases such as typhoid and cholera. These diseases, however, among other water related issues, remain a serious problem in developing countries. Modern water treatment processes control the spread of water related disease; remove numerous contaminants, such as organic chemicals and heavy metals, producing safe water. However the presence of pharmaceutical residues, disinfection by-products, and the possibility of disease causing agents as Cryptosporidium, which are unaffected by common water purification processes and so, need of new treatment technologies for this purposes. The single largest consumer issue affecting potable water under developing states is off-flavour. Off-flavour is caused by compounds in water that are known for their undesirable taste and odour characteristics. A survey conducted of more than 800 water usages in the America and Canada found the 16 % of utilities experience the serious taste and odour problems, spending approximately 4.5 % of their total budget for taste and odour control [12].

Whereas, prostanoids, inflammatory cytokines, nitric oxide and oxidative stress, are all considered to be important factors in the development of the haemodynamic and renal changes seen in liver failure. It is, however, the substances that are directly hepatotoxic that are particularly important in terms of recovery, as they may perpetuate liver injury invoking a downward spiral with further reduction in functional liver mass and increased toxin load. It is notable that many of the suggested toxins are insoluble in water and exist in the circulation bound to albumin [13].

Previous research and studies have been showing that one solution of treatment of drinking water is to measure the mass of activated carbon first and then put it into a prepared bottle, used as vessel. A thrilling bar put inside the bottle then it was sealed with cover of aluminium. On weighing machine, it was calculated the mass of activated carbon, bottle and stirring bar. Then 27 ml of ultra-pure water was added to each of bottle and boiled them for 3 minutes for remove air from the activated carbon bottle. After some time, cooling down the bottle to normal temperature and then it was measured total weigh difference after and before boiling process. To added the one litre of samples of different water and Then mass of the bottle calculated and recorded on data book, finally [14].

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Aluminium sulphate dosed at 20 mg/l without polymer addition removed about 80 percent of the toxicity from neurotoxic bloom of microcystins, Coagulation had an ability to eliminate the toxins in water samples in several studies. These studies tested the coagulant as Aluminium sulphate in different concentrations. Coagulation and clarification studies have had mixed results on cell lysis and the subsequent release of cyanobacterial algal toxins [1].

Nitrogen based biological compound inside samples of canal water can be detached by aluminium sulphate setting if the matter is based on organic compound a minor in quantity, Pietsch et al. (2001) initiate that the removal of nitrogenous matter is problematic to attain with simple coagulation in some cases and the nitrogen based compound are detached by microbial degradation and zonation processes [15, 16].

For this purpose, use the activated carbon filtering process is used in this research. It is achieved toxins removal by help with using the drinking water filtration process under filtered drinking water have normal values that given by WHO standards of drinking water guidelines.

V. CONCLUSIONS

In present study, it is investigate the toxins in drinking water samples from microbe's activities, very harmful health effect on humans and especially their liver functions disturb badly. Liver abscesses have a major economic and social impact on the health issues.

All these problems is mainly due to taking bio contaminated food and unsafe drinking water, so if we make assure the security and safety about our taking the food as essential component of life.

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