BIOACCUMULATION DETECTION & COMPARISON OF HEAVY METALS IN FRESH WATER VEGETABLES WITH WASTE WATER VEGETABLES OF QUETTA CITY.

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Abstract

Heavy metals in small concentration in environment are considered to be harmful. Heavy metals are one of the environmental toxins. They have carcinogenic impact on animals, since they are not metabolized rather they accumulate and are magnified through the food chain, they cause serious damage to a living system which is irreversible and has a hazardous effect on the metabolism and life cycles of a living cell. This is more dangerous especially for secondary and tertiary consumers. These effects take time and not necessarily cause a failure in the system but may be responsible for the stress on it. This study was done on the local agricultural water supply and vegetables of Quetta. The farmers here use waste water from the main drain of the city 'Habib Nala' to grow vegetables. The water from the drain and their vegetables were screened by atomic absorption for different heavy metals and compared with the fresh water and fresh water vegetables, the heavy metals which were tested include.

Lead, copper, chromium, cadmium, iron, manganese, nickel, zinc, arsenic and aluminum selected because of their dangerous effects on the biological system. The vegetables include spinach, cauliflower, onion, pumpkin, tomato, salad, mint, eggplant, green chillis and coriander. The result in ug/g in farming vegetables by waste water were. Fe <Zn<Cr<Mn<Cu<Cd<Pb. Fe (3.04-15.31), Zn (0.49-3.11), Cr (0.23-0.49), Mn (0.19-0.49), Cu (0.14-0.49), Cd (0.05-0.38), Pb (0.0025-0.42), Ni (0.08-0.24), Ar (nil), Al (0.005-0.095).The results from fresh water vegetables were Fe (3.1-7.04), Zn (0.04-1.05), Mn (0.04-1.05), Cr (nil), Cu (nil), Cd (nil), and Pb (nil).The result showed much difference between fresh water vegetables concentration and waste water concentration of heavy metals this difference can lead to more exposure to the toxicity which becomes the part of the system, leading higher and higher bioaccumulation. This can lead to serious effects because non-essential and harmful substances may either be stored or are reacted upon by the enzyme systems; it depends upon the exposure and uptake rate of these elements. Due to the process of bioaccumulation the metals seem to pile up in the system as they go through the food chain. Theoretically speaking, plants being the producers will have the lowest concentration of heavy metals, while consumers will have more increasing concentrations. Research needs to be done to prove this fact. But if the results from the waste water vegetables are considered, it clearly implicates that the heavy metals would be high in any animal grazing on these vegetables. If the grazing animal is man, or he is the secondary consumer of an animal grazing on these plants then these metals could seriously affect his health.Biodegradation is one of the solutions which overcome this pollution, if we use the technique of Bioremediation to all the pollutant sites by applying living organisms, tiny microbes such as fungi and bacteria, to the waste site which can lead to removal of these chemicals. The only thing which we have to research is to find suitable microbes.

Key words: bioaccumulation, biomagnified, bioremediation

INTRODUCTION

"Heavy metals" are chemical elements with a specific gravity that is at least five times the specific gravity of water and is poisonous at low concentration. The specific gravity of water is 1 at 4°C (39°F). Simply stated, specific gravity is a measure of density of a given amount of a solid substance when it is compared to an equal amount of water. Heavy metals are natural components of Earths crust. They cannot be degraded or destroyed. Heavy metals are dangerous because they tend to bio-accumulate. Bioaccumulation means an increase in the concentration of a chemical in a biological organism over time, compared to the chemicals concentration in the environment. Compounds accumulate in living things any time they are taken up and stored faster than they are broken down metabolized or excreted. There are 35 metals that concern us because of occupational or residential exposure; 23 of these are the heavy elements or "heavy metals": antimony, arsenic, bismuth, cadmium, cerium, chromium, cobalt, copper, gallium, gold, iron, lead, manganese, mercury, nickel, platinum, silver, tellurium, thallium, tin, uranium, vanadium, and zinc. Heavy metal toxicity can result in damaged or reduced mental and central nervous function, lower energy levels, and damage to blood composition, lungs, kidneys, liver, and other vital organs. Long-term exposure may result in slowly progressing physical, muscular, and neurological degenerative processes that mimic Alzheimer's disease, Parkinson's disease, muscular dystrophy, and multiple sclerosis. Allergies are not uncommon and repeated long-term contact with some metals or their compounds may even cause cancer (International Occupational Safety and Health Information Centre 1999). Heavy metals may enter the human body through food, water, air, or absorption through the skin when they come in contact with humans in agriculture and in manufacturing, pharmaceutical, industrial, or residential settings. Industrial exposure accounts for a common route of exposure for adults.

According to Bowen (1979), a number of elements such as Nickel (Ni), cadmium (Cd), Cobalt (Co), Chromium (Cr), Copper (Cu) and Selenium (Se) can be harmful to plants and humans even at quite low concentrations.The intentional and incidental disposal of sewage sludge and industrial wastes to the agricultural lands causes the accumulation of heavy metals; as a result, it leads to contamination of the food chain, because vegetables absorb heavy metals from the soil and polluted air and water. In general, heavy metals are not biodegradable and therefore can accumulate in human vital organs. As a result, this situation leads to progressive toxic effects (Stoepper 1984).Although problems do not occur directly in the soil, this process occurs in water ways when pollutants are leached out of the soil. If the plants die and decay, heavy metals taken into the plants are redistributed, so the soil is enriched with the pollutants. Uptake and accumulation of elements by plants may follow two different paths, which are the root system and the foliar surface (Sawidis *et al.* 2001).The uptake of metals from the soil depends on different factors such as their soluble content in

it, soil pH, plant species, fertilizers and soil type (Lubben and Sauerberck 1991). Vegetables, especially leafy vegetables, accumulate higher amounts of heavy metals because of the fact that they absorb these metals in their leaves (Sharma and Kansal 1986). There is a strong link between micro nutrition of plants, animals and humans and the uptake and impact of contaminants in these organisms (De Leonardis et al., 2000; Yuzbasi et al., 2003; Baslar et al., 2005; Yaman et al., 2005). The content of essential elements in plants is conditional, the content being affected by the characteristics of a soil and the ability of plants to selectively accumulate some metals.Toxic trace heavy metals like cadmium and lead are known to pose a variety of health risks such as cancer, mutations or miscarriages (Weigert, 1991). High heavy metal concentrations change the ecosystem and the relationship between its components. Plants have a natural ability to extract elements from soil and to distribute them between roots and shoot depending on the biological processes in which the element is involved (Ximénez-Embún *et al* ., 2002). In addition to the uptake of nutrients, toxic compounds such as heavy metals can also be taken up by the plants. Heavy metals are defined as metals with a density >5.0 g cm − (Seaward & Richardson, 1990).Both kinds of heavy metal, nutrients and pollutants, can accumulate in excess in the plant to levels undesirably high for human or animal nutrition, and may even become toxic to plants at a certain concentration(William *et al* ., 2000). The distribution of heavy metals in certain plants sometimes may depend on the availability of other elements. An accumulation of Mn at the penetration position of *Erysyphe graminis* on wheat leaves was observed only in plants supplied with silicate, but not in comparable plants grown without silicate (Leusch & Buchenauer, 1988). Therefore, the absorption depends on many environmental factors.

MATERIALS AND METHODS

The plants were collected during the months of March and July 2006. Following care was taken during collection.

1-Plants covered with soil, dust or residue chemicals.

2-Plants damaged by insects or mechanically injured.

3-Plants under moisture or temperature stress.

Collected plants were handled carefully to ensure that no loss in dry weight occurred as it significantly affects the plant analysis result (Lockman). Before drying and storage plant samples were prepared for laboratory analysis for obtaining accurate analytical data and reliable interpretations of bioassays results. To remove contamination such as soil and dust particles, the fresh turgid plant samples were brushed briskly or washed with water quickly for period of 15 seconds. Then dried immediately to stabilize the tissue and stop enzymatic reactions. Plants were placed on clean papers, in shady dry environment for 7 days. After drying plants were ground using ordinary grinder machine and then thoroughly mixed. 1 kg of plants were taken from the ground plant material for laboratory analysis, the rest were stored in closed, airtight plastic bags in cool dry environment.

Sampling

In total 38 samples of vegetables and water were taken from different location near jinnah town. The waste water of the city that flows through an open drain called 'Habib Nala' was central as samples were taken from this drain and the vegetables that were grown in that water. Fresh water grown vegetables were also taken to avoid any error. Vegetables included cucumber, tomato, green pepper, eggplant and pumpkin. The samples of the vegetable species were collected from the different fields of the sampling sites of jinnah town during the year 2006. Vegetables were sampled by hands protected with vinyl gloves and carefully packed into the polyethylene bags (Alam et al .2003). Water samples were collected in sterile bottles for accurate results.

Moisture content

The moisture content of the leaves was determined by following formula.

$$\text{Moisture content (\%age)} = \frac{\text{Fresh weight} - \text{dry weight}}{\text{Fresh weight}} \times 100$$

Sample preparation

Vegetables were washed several times with tap water followed by distilled water.(Dilek et al . 2005). All vegetable samples were oven dried at 40C for 24h, drying of the collected materials is important because it protects the plant material from microbial decomposition and also ensures a constant reference value by determining dry weight in contrast to fresh weight, which is difficult to quantify (Markert 1993; Aksoy et al. 2005). Dried and milled samples were powdered and kept in clean polyethylene bags and stored for analysis through atomic absorption.

Analytical method

Samples of dried and milled materials were ashed in furnace at 460C for 24hrs, while the weighed ash was digested in 10ml of HNO3. All digested samples were centrifuged and then made up to volume 25ml with 1%HNO3 (Dilek et al 2005). The samples were analyzed with four different locations per samples using atomic absorption. Then the mean of these four values was used.

RESULTS

The concentrations of heavy metals in sample vary both in water and vegetables. The order of the levels of heavy metals obtained from different kinds of vegetables from waste water was Fe <Zc<Cr<Mn<Cu<Cd<Pb. The difference between the water and vegetable concentration of a metal is consistent in the case of fresh water and waste water, showing that the only reason for high levels is the waste water being used. The samples from fresh water and their vegetables result showed that the concentration is much low even nil. However the result showed low concentration of heavy metal with the standards but if it is continued for long period of time, it may lead to a hazardous situation. Heavy metal small concentration in environment is considered to be harmful.

Table1 WHO Guideline (standards).
Chemicals of health significance

Inorganic	Mg/L
Arsenic	0.01
Cadmium	0.03
Chromium	0.050
Copper	2.000
Lead	0.010
Manganese	0.500
Nickel	0.020
Iron	0.3
Zinc	3

Source: water quality status in Pakistan. Page 19
Pakistan council of research in water resources
June 2004 publication no 125_200

Table2Analysis of waste water samples (habib nala) Mean result with standard deviation. Units µg/100ml

Elements	Result obtained (Mean)	SD
Aluminum	0.07	±.06
Arsenic	Nil	Nil
Cadmium	0.0697	±.01
Chromium	2	.± 08
Copper	1.7	±.22
Lead	0.13	±.01
Manganese	7.43	±.4
Nickel	2.03	±.12
Iron	8.62	±.45
Zinc	1.29	±.22

Table3Analysis of fresh water samples (Mean result) with standard deviationUnits µg/100ml

Elements	Result obtained (Mean)	SD
Aluminum	Nil	
Arsenic	Nil	
Cadmium	Nil	
Chromium	Nil	
Copper	0.004	±0.002
Lead	Nil	
Manganese	0.29	± 0.51
Nickel	Nil	
Iron	1.187	±1.2
Zinc	0.95	±.12

Table4Waste water vegetables (Mean result) with standard deviation.

Elements	Units	Bringal	Chilli	Mint	Salad	Spinach	SD
Lead	µg/gm	0.4	0.39	0.003	0.42	0.0025	±.2
Copper	µg/gm	0.24	0.159	0.14	0.20	0.245	±.04
Chromium	µg/gm	0.65	0.24	0.23	0.35	0.95	±.3
Cadmium	µg/gm	0.065	0.05	0.35	0.095	0.16	±.12
Iron	µg/gm	7.54	9.59	6.25	8.08	15.31	±3.5
Manganese	µg/gm	0.5	0.375	0.19	0.41	0.49	±.125
Zinc	µg/gm	1.19	1.15	3.04	1.59	3.11	±.98
Arsenic	µg/gm	Nil	Nil	Nil	Nil	Nil	
Nickel	µg/gm	0.19	0.14	0.24	0.17	0.08	±.05
Aluminum	µg/gm	0.095	0.08	0.0095	0.05	0.005	±.04

Table4(cont.)waste water vegetables (Mean result) with standard deviation

Elements	Units	Cauliflower	Onion	Pumpkin	tomato	Coriander	SD
Lead	µg/gm	0.365	0.35	0.35	0.27	0.02	±.14
Copper	µg/gm	0.23	0.49	0.34	0.29	0.25	±.10
Chromium	µg/gm	0.40	0.34	0.28	0.32	0.57	± .11
Cadmium	µg/gm	0.28	0.38	0.29	0.29	0.28	±.04
Iron	µg/gm	3.04	3.54	4.12	4.54	5.64	±.99
Manganese	µg/gm	0.45	0.4	0.39	0.43	0.48	±. 03
Zinc	µg/gm	0.49	0.57	0.51	0.69	0.67	
Arsenic	µg/gm	Nil	Nil	Nil	Nil	Nil	
Nickel	µg/gm	0.14	0.14	0.12	0.16	0.09	±.02
Aluminum	µg/gm	0.07	0.09	0.095	0.03	0.09	±.02

Table5.Fresh water vegetables (Mean result) with standard deviation.

Elements	Units	Bringal	Chilli	Mint	Salad	Spinach	SD
Lead	µg/gm	0	0	0	0	0	
Copper	µg/gm	0	0	0	0	0	
Chromium	µg/gm	0	0	0	0	0	
Cadmium	µg/gm	0	0	0	0	0	
Iron	µg/gm	4.59	5.59	4.4	4.58	5.62	± 0.59
Manganese	µg/gm	0.5	0.6	0.34	0.78	0.69	± . 17
Zinc	µg/gm	0.08	0.075	0.065	0.065	0.15	±.03
Arsenic	µg/gm	0	0	0	0	0	
Nickel	µg/gm	0	0	0	0	0	
Aluminum	µg/gm	0	0	0	0	0	

Table5(cont.)Fresh water vegetables (Mean result) with standard deviation

Elements	Units	Cauliflower	Onion	Pumpkin	tomato	Coriander	SD
Lead	µg/gm	0	0	0	0	0	
Copper	µg/gm	0	0	0	0	0	
Chromium	µg/gm	0	0	0	0	0	
Cadmium	µg/gm	0	0	0	0	0	
Iron	µg/gm	3.1	3.39	4.12	7.04	5.57	± 1.6
Manganese	µg/gm	0.54	0.37	0.39	0.44	0.48	± .06
Zinc	µg/gm	0.54	0.56	0.04	0,19	1.05	± .30
Arsenic	µg/gm	0	0	0	0	0	
Nickel	µg/gm	0	0	0	0	0	
Aluminum	µg/gm	0	0	0	0	0	

DISCUSSION AND CONCLUSION

The levels of heavy metals in waste water vegetables from Quetta city were determined and compared with the vegetables from fresh water which clearly showed that fresh water vegetables have negligible concentration of heavy metals. This clearly shows that the presence of heavy metals is due to the waste water. Due to the process of bioaccumulation the metals seem to pile up in the system as they go through the food chain. Theoretically speaking, plants being the producers will have the lowest concentration of heavy metals, while consumers will have more increasing concentrations. Research needs to be done to prove this fact. But if the results from the waste water vegetables are considered, it clearly implicates that the heavy metals would be high in any animal grazing on these vegetables. If the grazing animal is man, or he is the secondary consumer of an animal grazing on these plants then these metals could seriously affect his health.Living systems, be it man, animal or a plant are based on the same mechanism of metabolism. Trying to utilize the raw material provided, to sustain and protect itself against harm. Chemicals that enter the system which cannot be metabolized are either excreted or get deposited in the storage tissues (some examples are adipose for animals and roots for plants). The system may survive the first few exposures of this harmful chemical because its concentration might not be lethal. But as the exposure is not removed the overall content of this chemical in the system may increase. The system thinks that it has solved the problem by locking the chemicals into the tissues which are not being used at that moment. This is a very important mechanism of safety and for forensic scientists a way to find out which chemical killed a person just by taking a sample of his hair. Now the system is superior as it has averted a disastrous outcome. But for how long, the system is not always in a land of bounty, always getting what it needs. Due to shortage of fuel, disease, any other incident or accident the system comes under stress. Then there is no other option but to use the stored assets which were put in for the coming winters. This would be all good if the assets were clean, in our case they are loaded with the chemicals the body cannot metabolize. Now when the stored assets are being used the chemicals come in to action. They are free once again and in higher concentration than before. The system already under stress becomes easy target for these chemicals causing more injury. If this proposed aspect is true it could explain for the example given below.

We being humans have the most chances of getting hit. Firstly, because of the vegetables we consume which have already accumulated a lot of these heavy metals. Secondly, by eating the animals which graze on these plants, they have over the period stored a lot of these heavy metals in their adipose. Finally, we also start to accumulate these within our own system. This all might pile up when we are in stress, pointing towards the fact that some individuals recover early then others from the same disease.

Although we need to investigate the levels of heavy metals in people from areas where irrigation is based on waste water specially people who have a disease but some things do point toward the fact. For example, it's a known fact that people living in high altitude like Quetta have a high level of hemoglobin due to low partial pressure of oxygen in the environment. But when I did a survey in the local areas of Quetta, I found out that many people where affected by anemia. Most of these were idiopathic without any thing being wrong, which points us towards the environment and food being the most important aspect. I think we need to investigate these things in more detail but the results that I provided, do point toward these effects. Not only anemia but recently the occurrence of cancer specially one effecting the stem cells of blood has increased in the localities of this city.

I strongly recommend the government and the citizens to reconsider their thinking that these vegetables are safe for human or animal use. I also recommend that a comprehensive research project be launched to monitor the effects of these heavy metals, from the vegetables and other animals, on humans.

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