

*Full Length Research Paper*

# Environmental assessment of ground water pollution by heavy metals and bioaccumulation of mercury residues in chicken tissues

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The aim of this study was to investigate the relationship between the concentrations of heavy metals in well water and bioaccumulation of the most abundant metals in chicken tissues in some areas in the province of Mecca Almokaramah, Saudi Arabia. Among the heavy metals (Cd, Zn, Cr, Mn, Cu Hg, Pb and Ni) studied, mercury (Hg) revealed highest in concentration in well waters. The concentration of mercury in ground water, beside in liver, kidney, muscle and blood samples of 10 chickens from each of four poultry- production farms were estimated using atomic absorption spectrophotometer. The present results showed that the kidney followed by liver are the organs with the highest bioaccumulation of mercury in all farm samples. The level of mercury in ground water was 7.06 µg/L. There is no doubt that the relationship between mercury accumulation levels in kidney and those in liver tissues were proportionally correlated and altered with elevation in antioxidant enzyme activities such as serum enzymes aspartate aminotransferase (AST) and serum glutamate pyruvate transaminase (GPT). These elevated enzymatic activities were induced by the level of toxicity. There was a significant elevation in the level of liver and kidney malondialdehyde (MDA), while the activities of antioxidant enzymes superoxide dismutase and catalase (SOD and CAT) were significantly decreased. Biochemical observations were supplemented by histopathological examination of liver and kidney sections.

**Key words:** Environmental toxicology, ground water, heavy metals, mercury, bioaccumulation- chicken histopathology.

## INTRODUCTION

Water pollution is the contamination of water bodies (example, lakes, rivers, oceans and groundwater). Water pollution occurs when pollutants are discharged directly

or indirectly into water bodies without adequate treatment to remove harmful compounds. Water pollution affects animals and other organisms living in these bodies of water; and, in almost all cases the effect is damaging not only to individual species and populations, but also to the natural biological communities (Mapanda et al., 2005; Anne et al., 2007).

Heavy or toxic metals are trace metals whose density is at least five times that of water. As such, they are stable elements (meaning they cannot be metabolized by the body) and bio-accumulative (passed up the food chain to

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**Abbreviations:** AST, Aspartate aminotransferase; GPT, glutamate pyruvate transaminases; MDA, malondialdehyde; SOD, superoxide dismutase; CAT, catalase.

humans). These heavy metals include: mercury, nickel, lead, arsenic, cadmium, aluminum, platinum, and copper (the metallic form versus the ionic form required by the body). Heavy metals have no function in the body and can be highly toxic (Mohammad et al., 2010).

The rate of urbanization and industrialization has been in the increase for the last two decades in Saudi Arabia. Besides many problems associated with such social changes, the pollution is considered to be a major concern for the health of the nation. Among the numerous types of environmental pollutions that constitute a danger to humanity, the contamination of food chain appear to be a growing threat that requires immediate attention and action (Khan et al., 1996; Bachman et al., 2002; Anne et al., 2007; Mohammad et al., 2010).

Most heavy metals tended to be associated with sulphur in protein (Rossi and Santaroni, 1976). The heavy metals content of streams, lakes and rivers did not normally exceed 0.1 ppm although some water sources located near different heavy metals deposits may contain mixed amounts up to 80 ppm (Wershow, 1970; Bachman et al., 2002). Limited data is available for the concentration of heavy metals in rain water and snow. Manahan (1989) reported that most notorious mercury compounds (for example) in the environment are mono-methyl mercury salts and diethyl mercury salts which are water soluble.

The contaminated water with metals is the route of ill-health in human and in animals. Among various pollutants in the environment, heavy metals are directly related to diseases in humans. Although, it is difficult to classify trace metal into essential and toxic groups, yet it is a well known fact that an essential metal becomes toxic at sufficiently high intakes (Khurshid and Qureshi, 1984; Harter et al., 2002; Anne et al., 2007; Akbar et al., 2010).

Lead may enter the atmosphere during mining, smelting, refining, and manufacturing processes and by the use of lead containing products. Lead intake occurs from the consumption of whisky, fruit juices, food stored in lead lined containers, cosmetics, cigarettes and motor vehicle exhaust etc (Harter et al., 2002; Aradhi et al., 2009). Excess lead can cause serious damage to the brain, kidneys, nervous system and red blood cells. Young children, infants and fetuses are particularly vulnerable to lead poisoning than adult. US Environment Protection Agency (EPA) says that lead may be implicated in causing Leukemia (Anonymous, 2002).

Zinc is essential for normal functioning of cells including protein synthesis, carbohydrate metabolism, cell growth and cell division (Saeed, 1998). However, if Zn concentration in the air is over 15 mg/m<sup>3</sup>, "metal fume fever" may result; which causes fever, depression, malaise, cough, vomiting, salivation and headache. Cadmium replaces Zn, in many enzymes. Therefore, a higher amount of Zn is required to overcome the toxic effects of

cadmium (Khan et al., 1990).

High level of tissue concentration of iron causes increased risk of myocardial infarction (Harvey and Champe, 1994) and high or low level of magnesium causes kidney failure and heart problems. High level of calcium is responsible for thirst, increased volume of urine, muscle fatigue, poor mental concentration and formation of kidney stones (Saeed, 1998).

Water pollution with mercury is one of the major problems confronting health officials everywhere. Mercury is a widespread environmental and industrial pollutant, which induces severe alterations in the tissues (Timbrell, 1982; Manahan, 1989; Lund et al., 1993; Mahboob et al., 2001; Sener et al., 2007), causes numerous neurological abnormalities (Kingman et al., 2005; Auger et al., 2005) and produces peripheral neuropathy (Boyd et al., 2000; Chuu et al., 2007) in experimental animals and human beings. Mercury poisoning can result from inhalation, ingestion, or absorption through the skin and may be highly toxic and corrosive once absorbed into blood stream. Furthermore, it combines with proteins in the plasma or enters the red blood cells but does not readily pass into the brain or fetus; instead, they may enter other body organs (El-Shenawy and Hassan, 2008). Those authors reported that the liver is a major site of metabolism for mercury and it can accumulate in the liver resulting in severe hepatic damages. Previous studies have revealed that HgCl<sub>2</sub> caused histopathological and ultrastructural lesions in the liver evidenced by periportal fatty degeneration and cell necrosis. Schurz et al. (2000) stated that DNA was a vital molecule in the cell activities and was the main target for HgCl<sub>2</sub>-induced cell injuries.

This study is a survey of heavy metals pathway from the environment through ground water to chicken in some particular areas in the province of Mecca Almokaramah where poultry farms are established. This study will endeavor to measure these metals as environmental pollutants in ground water, and to investigate the bio-accumulation of mercury residues (as one of the major concentrated heavy metals) in chicken tissues of major poultry farms. To date no such studies have been conducted in this area where the chicken consumers are on the increase. The outcome of this study will help in taking precautionary steps in monitoring metal contamination in ground water and to advice the authorities of the health impact that might have in Jeddah population as consumers.

## MATERIALS AND METHODS

This study was carried out on four farms of poultry production located in four sites in the province of Mecca Almokaramah, Saudi Arabia (Figure 1). The first Farm (A) located in Om Al-Jood area, 150 Km east of Jeddah governorate. The second Farm (B) located in Hada El-Sham area, 80 km east north Jeddah, the third one (C) in Al-Wazeeria region, 60 km east south Jeddah and the fourth one

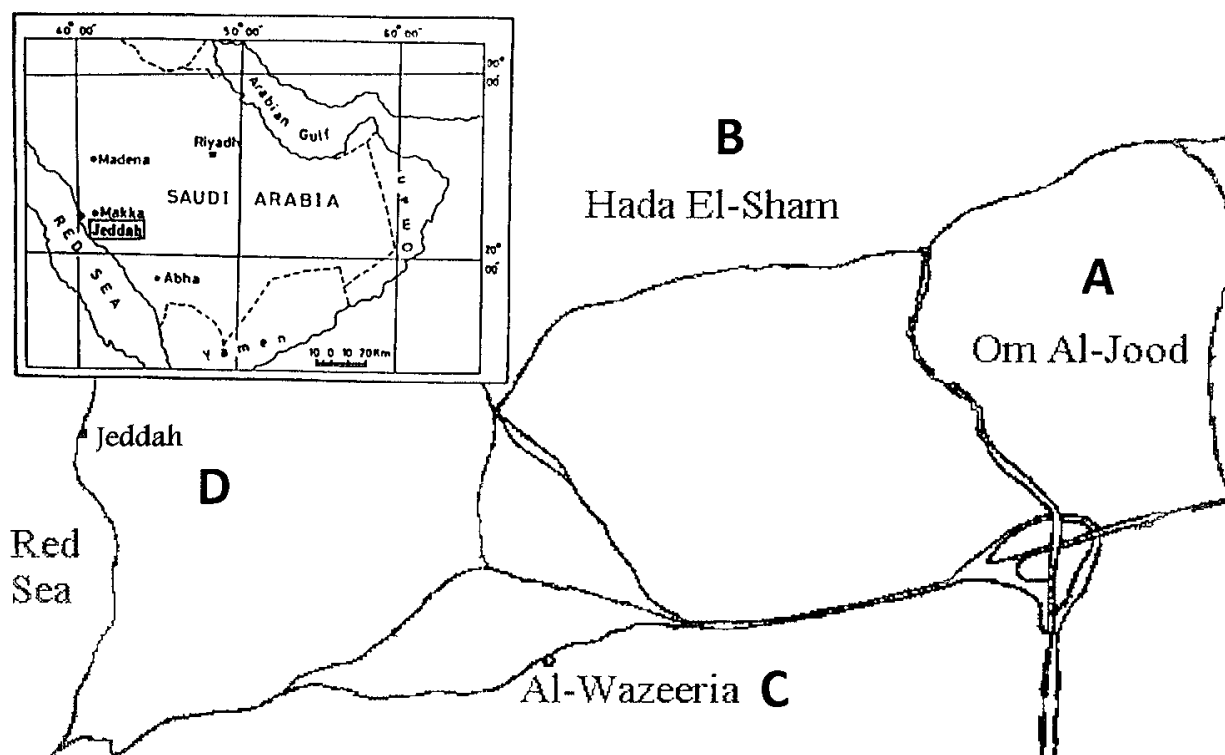


Figure 1. Location map of study sites.

is Al-Fakeih poultry Farm (D) in Jeddah governorate which act as a control group. The first three Farms, A, B and C mainly depend on ground water and regularly supplied by water from separate wells I, II and III, respectively. While the fourth Farm (D) was regularly supplied with healthy pure drinking water source (IV). Management of the four farms was identical except water source. Water samples were collected from each well and water source along three months at intervals of one month. Water samples were collected in clean glass bottles for chemical analysis according to APHA (1995).

Qualitative determination of cadmium (Cd), zinc (Zn), chromium (Cr), magnesium (Mn), copper (Cu), mercury (Hg), lead (Pb) and nickel (Ni) in wells supplied the Farms (A to C) and fresh water supplied the control Farm (D). Water analyses were carried out using Flameless Atomic Absorption Spectrophotometer (Perkin Elmer 2380, U.S.A.). The actual representative concentration (ppm) of the most abundant heavy metal was determined per source. Precautions were taken to avoid contamination during sample collection.

The domestic white farm chickens (*Gallus domesticus*) are used in this study. Ten chickens, six weeks old were chosen from each farm and slaughtered. Liver, kidneys, muscles and blood were taken to determine the mercury residues. Mercury was also determined in tissue samples by flameless atomic absorption spectrophotometer equipped with a deuterium arc background corrector.

#### Serum biochemical assay and estimation of MDA, SOD, CAT in liver and kidney tissues

Serum enzymes aspartate aminotransferase (AST) and serum glutamate pyruvate transaminase (GPT) were determined according to (Reitman and Frankel, 1957). Liver and kidney

samples were dissected out and washed immediately with ice cold saline to remove as much blood as possible. Each tested tissue homogenates (5% w/v) were prepared in cold 50 mM potassium phosphate buffer (pH 7.4) using glass homogenizer in ice. The cell debris was removed by centrifugation at 5000 rpm for 15 at 40°C using refrigerated centrifuge. The supernatant was used for the estimation of malondialdehyde (MDA) (Yagi and Rastogi, 1979), superoxide dismutase (SOD) (Kakkar et al., 1972) and catalase (CAT) (Smna, 1972) levels.

#### Histopathological studies

The target organs (liver and kidney) tissues were dissected out and fixed in 10% formalin, dehydrated in gradual ethanol (50 to 99%), cleared in xylene, and embedded in paraffin. Sections were prepared and then stained with hematoxylin and eosin dye for microscopic investigation.

#### Statistical analysis

Statistical analysis was performed on a PC using SPSS, V.13, (special package for social sciences). Data are presented as arithmetic mean  $\pm$  S.D. The difference among means was analyzed by one way ANOVA followed by student *t* test. A value of  $P < 0.05$  was considered as statistically significant.

## RESULTS

Table 1 shows the mean concentration (ppm) of

**Table 1.** Heavy metal concentrations (ppm) in the water sources supplied to the chicken farms in different study sites.

Farm	Level	Concentrations of heavy metals in farm water (ppm)							
		Cd	Zn	Cr	Mn	Cu	Hg	Pb	Ni
A	Mean	0.002	0.09	0.03	0.12	0.11	7.34	0.10	0.05
	Median	0.001	0.07	0.02	0.09	0.05	4.22	0.06	0.03
	Range	0.00–0.002	0.00-0.12	0.00-0.12	0.00–0.18	0.00–0.84	3.09–9.01	0.00-0.37	0.00-0.09
B	Mean	0.003	0.17	0.09	0.14	0.15	6.78	0.06	0.04
	Median	0.001	0.09	0.06	0.08	0.03	3.74	0.04	0.02
	Range	ND–0.004	0.02-0.25	0.00-0.10	0.00–0.28	0.00–0.44	2.99–8.22	0.00–0.39	0.00-0.07
C	Mean	0.001	0.23	0.15	0.15	0.15	1.36	0.08	0.06
	Median	0.002	0.15	0.13	0.10	0.11	1.21	0.05	0.04
	Range	ND–0.003	0.03-0.37	0.00-0.27	0.00–0.43	0.00–0.53	1.00–1.48	0.00–0.25	0.00-0.09
D	Mean	0.002	0.20	0.06	0.16	0.27	0.38	0.07	0.05
	Median	0.001	0.17	0.05	0.12	0.18	0.18	0.04	0.03
	Range	ND–0.004	0.02-0.27	0.00-0.18	0.00–0.33	0.00–0.51	0.00–0.41	0.00–0.09	0.00-0.6
Permissible limits of heavy metals (ppm) in water (WHO, 1984)		0.00–0.005	1.02-2.99	0.00-1.20	0.00–1.05	0.00–1.00	1.00–2.68	0.00–0.50	0.00-1.12

cadmium, zinc, chromium, magnesium, copper, mercury, lead and nickel in polluted well supplied Farms (A to C) and h water supplied Farm (D). Heavy metal concentrations in studied wells showed significant variations between sites ( $P < 0.05$ ). The mean concentrations (ppm) of heavy metals among the study sites ranged from 0.001 to 0.003 for Cd, 0.09 to 0.23 for Zn, 0.03 to 0.15 for Cr, 0.12 to 0.16 for Mn, 0.11 to 0.27 for Cu, 0.38 to 7.34 for Hg, 0.06 to 0.10 for Pb, and 0.04 to 0.06 for Ni (Table 1). Among the heavy metals, mercury (Hg) was highest at Farms A and B which received the well I and II which exceeds the permissible metal limits of drinking water. The two Farms (C and D) receiving the well III and healthy water source IV, Hg concentration lies in the permissible metal limits of drinking water (WHO, 1980, 1984). For this reason, the residual quantities of mercury, which were the most

abundant heavy metals in the analyzed wells water samples were estimated in different chicken tissues to find the bioaccumulation of this toxic metal in the tissues and blood of the chicken in poultry farms supplied with this water.

Table 2 shows mercury concentration in wells-water samples. The mercury concentration in wells-water I and II ranged from 6.78 to 7.34 ppm with a mean of 7.06 ppm, while wells-water III and IV had no detectable levels. The highest Hg concentrations were detected in kidney, which is considered the main target organ followed by liver. The residual quantities of mercury in 100  $\mu$ g wet weight estimated in liver tissues ranged from 110 to 179 with a mean of 143.4 in 8 cases from Farm A, while the rest revealed undetectable amount of mercury; 123 to 183 with a mean of 137.4 in 9 cases from Farm B, a mean of 132 in 4 cases from Farm C; and a mean of 117.2 in 4

cases from Farm D. The rest of the samples revealed undetectable mercury levels (Table 2).

The residual quantities of mercury, estimated in kidney tissues ranged from 351 to 734 with a mean of 542 in 9 cases from Farm A, 406 to 645 with a mean of 548.3 in 9 cases from Farm B; a mean of 499 in 4 detectable cases from Farm C; a mean of 464 in 3 detectable cases from Farm D. The rest of the samples revealed undetectable mercury levels (Table 2). All the examined blood and muscle samples showed concentrations below the detectable level.

Mercury contents in liver and kidney were significantly elevated in all samples collected from Farms A and B than from Farms C and D ( $P < 0.001$ ). The well water analysis in the four tested farms also clearly showed higher concentration of mercury in Farms A and B than those from Farms C and D. This is accompanied by a significant

**Table 2.** Relationship between the concentrations of mercury in various tissues of chickens (6 weeks old) and in the ground water in four different poultry farms in Mecca Al-Mokaramah Province.

Well number	Mean concentration of mercury in well water (ppm)	Farm	Organ	Concentration of mercury in the collected samples(µg/100 g) wet weight									
				1	2	3	4	5	6	7	8	9	10
I and II	7.06±1.4	A	Liver	110±3.37	155±4.02	126±3.89	152±3.66	ND	165±3.86	ND	179±3.42	143±2.75	129±2.13
			Kidney	734±9.55	508±5.72	351±5.09	646±7.41	491±5.33	410±4.22	---	516±4.76	485±3.71	561±4.76
			Muscle	---	---	---	---	---	---	---	---	---	---
			Blood	---	---	---	---	---	---	---	---	---	---
		B	Liver	183±3.76	129±3.40	136±3.09	134±3.33	160±2.15	ND	131±2.11	123±3.06	135±2.13	132±2.66
			Kidney	627±7.58	484±4.11	457±3.66	645±6.03	532±4.50	616±7.11	406±4.29	505±3.35	ND	645±6.55
			Muscle	---	---	---	---	---	---	---	---	---	---
			Blood	---	---	---	---	---	---	---	---	---	---
III and IV	Not detectable	C	Liver	154±3.75	130±2.66	ND	ND	141±2.34	---	---	103±2.05	---	---
			Kidney	487±4.01	542±4.64	463±3.99	504±5.22	---	---	---	---	---	
			Muscle	---	---	---	---	---	---	---	---	---	
			Blood	---	---	---	---	---	---	---	---	---	
		D	Liver	100±2.12	---	---	124±3.55	ND	127±2.15	---	118±2.11	---	---
			Kidney	---	---	---	457±4.41	498±5.77	---	---	437±4.44	---	---
			Muscle	---	---	---	---	---	---	---	---	---	
			Blood	---	---	---	---	---	---	---	---	---	

Data are expressed as µg/100 g wet tissue as mean ± S.E. of five samples from each individual, ND: Not detectable.

increase in the concentration of mercury in kidney as well as in liver (P < 0.01). The accumulation of mercury in kidney of all tested samples was found to be very highly significant (P < 0.001) than those in the liver (Table 3).

Enzyme activities of liver and kidney of chickens of the three polluted well Farms (A to C) and the control Farm (D) are illustrated in Table 4. Serum AST and GPT were significantly increased in the

three contaminated farms as compared to the control group (P < 0.001). The elevated activities of serum AST and GPT were significantly reduced in the animal groups supplied with pure water (control). Supplying with well water I and II in Farms A and B respectively showed significantly more enzyme activity (P < 0.001) than those supplied with well water III (Farm C). Results obtained also revealed an increase in the level of

liver and kidney MDA in polluted water farm chickens groups compared to the control group. The activities of SOD and CAT were significantly reduced in the first two contaminated farms (Farms A and B), while they were significantly elevated near the normal values in the third group (Farm C) or control group (Farm D) of non-detected mercury levels.

The chicken liver of Farms A and B which were

**Table 3.** Means of mercury concentration in farm water (ppm) and in different organs ( $\mu\text{g}/100\text{g}$  wet weight).

Mean conc. of mercury in farm water	7.06 $\pm$ 1.40			No detectable		
	Farm A	Farm B	Farms A+B	Farm C	Farm D	Farms C+D
Mean of all liver samples	107.5 $\pm$ 3.22	126.0 $\pm$ 3.08	116.7 $\pm$ 3.11	044.0 $\pm$ 1.42	048.0 $\pm$ 2.05	046.0 $\pm$ 1.66
Mean of all detectable samples	143.4 $\pm$ 2.45	137.4 $\pm$ 2.18	140.1 $\pm$ 3.07	132.0 $\pm$ 2.33	117.2 $\pm$ 2.75	123.7 $\pm$ 2.82
Mean of all kidney samples	406.5 $\pm$ 4.14	456.9 $\pm$ 3.67	431.7 $\pm$ 3.08	166.3 $\pm$ 2.41	116.0 $\pm$ 1.55	141.0 $\pm$ 3.72
Mean of all detectable samples	542.0 $\pm$ 3.56	548.3 $\pm$ 5.11	545.3 $\pm$ 4.26	499.0 $\pm$ 3.87	464.0 $\pm$ 4.27	484.0 $\pm$ 4.23

**T – Test data**

Farm	Organ	Mean $\pm$ S.E.	Value of t	P
(A+B)	Liver	140.1 $\pm$ 3.07	194.392	< 0.001
	Kidney	545.3 $\pm$ 4.26		
(C+D)	Liver	123.7 $\pm$ 2.82	111.013	< 0.001
	Kidney	484.0 $\pm$ 4.27		
(A+B) + (B+C)	Liver	131.9 $\pm$ 3.74	006.047	> 0.05
(A+B) + (B+C)	Kidney	514.6 $\pm$ 5.12	022.371	< 0.01

Insignificant difference ( $P > 0.05$ ); significant difference ( $P < 0.01$ ) and highly significant difference ( $P < 0.001$ ).

regularly supplied by polluted well I and II respectively showed massive fatty changes, necrosis, and broad infiltration of the lymphocytes (Figures 2A and B). The histological architecture of liver sections of the chickens supplied with undetectable mercury water well III (Figure 3C) showed more or less normal patterns, with a mild degree of necrosis and slightly lymphocyte infiltration, almost comparable to those of the control group. The histological examination of liver sections of control animals (Figure 2D) showed normal hepatic cells with well preserved cytoplasm prominent nucleus.

The chicken kidneys of Farms A and B which were regularly supplied by polluted well I and II, respectively showed tissues with tubular epithelial damage, capillary proliferation certain degenerated uriniferous tubules and dilatation of Bowman's capsule (Figures 3A and B). The above pathological changes were disappeared in chickens supplied with undetectable mercury water well III (Figure 3C). The histological examination of liver sections of control animals (Figure 3D) also showing normal renal tissues and normal uriniferous tubules and glomeruli in control chicken farm.

**DISCUSSION**

The essential and primary purpose of water supply programs is to deliver potable water which must be safe, adequate and accessible for human and animals. However, in developing countries these aims are rarely

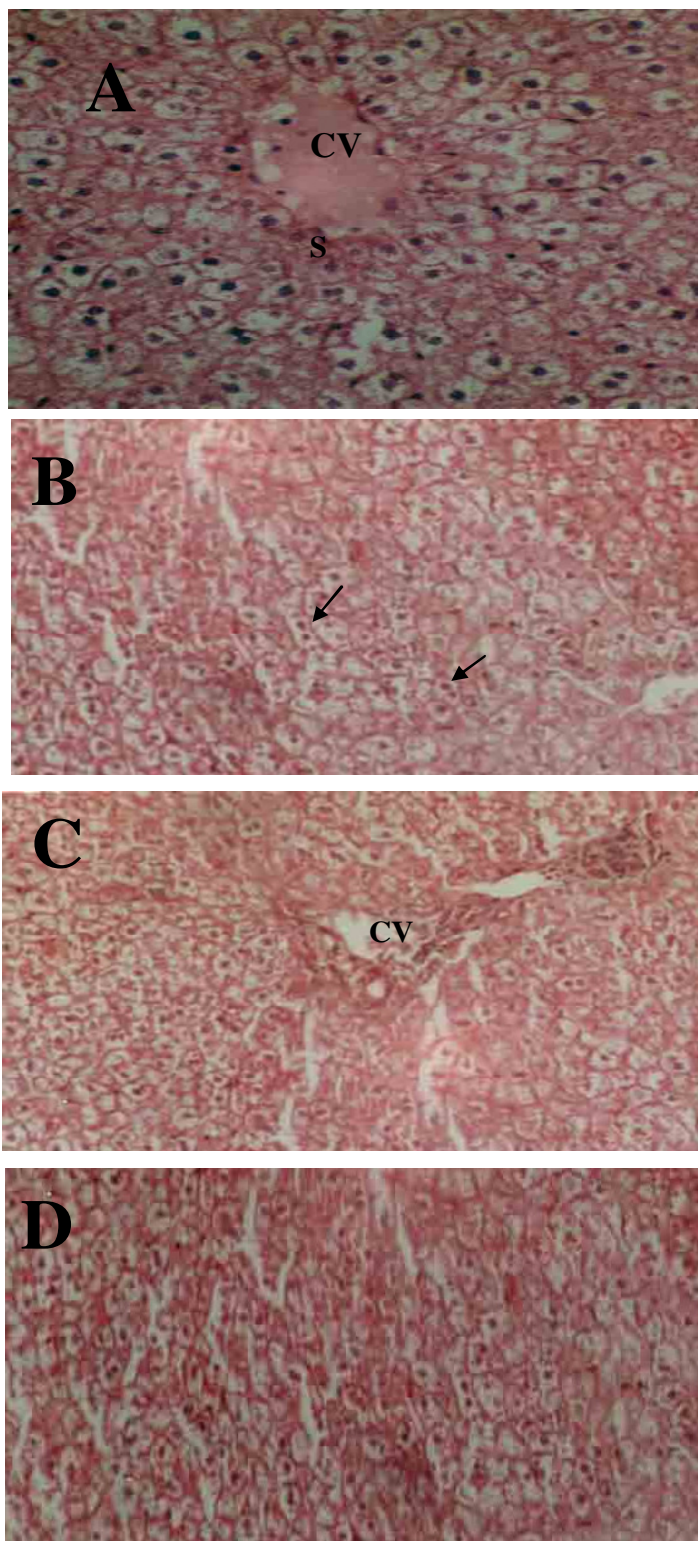
attained. Data of this investigation proved that the improper disposal of untreated ground water affected the health performance of chickens drinking from this polluted water.

Table (1) presents the mean concentration (ppm) of cadmium, zinc, chromium, magnesium, copper, mercury, lead and nickel in polluted well supplied Farms (A to C) and healthy water supplied Farm (D). Although, the concentrations of these metals in ground water and fresh tap water lie within the permissible limits recommended by WHO (1984), yet the concentrations of mercury in polluted well water was considered highly significant increased than those of control healthy water and highly elevated than the recommended values by WHO (1984). Nearly similar findings were obtained by Dall (1968), Warshaw (1970), Zaki et al. (1994) and Abd El-Nasser et al. (1996) and by Youssef and Haleem (1999). Limited data were available for concentrations of mercury in wells water. Dall (1968) measured 300 samples from natural water for mercury in Italy and found values in the range of 10 to 15 ppm. The present study showed undetectable mercury levels in 2 wells and 7.06 ppm in the other two wells. Similar results were reported by Wershaw (1970), who revealed that the mercury content of streams, lakes and rivers does not exceed 0.1 ppm but some water sources located near mercury deposits may contain mercury up to 8.0 ppm. It is obvious that the examined water samples from wells I and II exceeded the permissible limits indicative of water pollution (1.00 to 2.68 ppm) and water quality standard (WHO, 1984).

**Table 4.** Enzyme activities of liver and kidney of chickens of the control and three well polluted farms; superoxide dismutase (SOD), catalase (CAT), lipid peroxide product or Malendialdyde (MDA) and serum aminotransferase enzymes (ALT and AST) of all studied groups (Mean  $\pm$  SD).

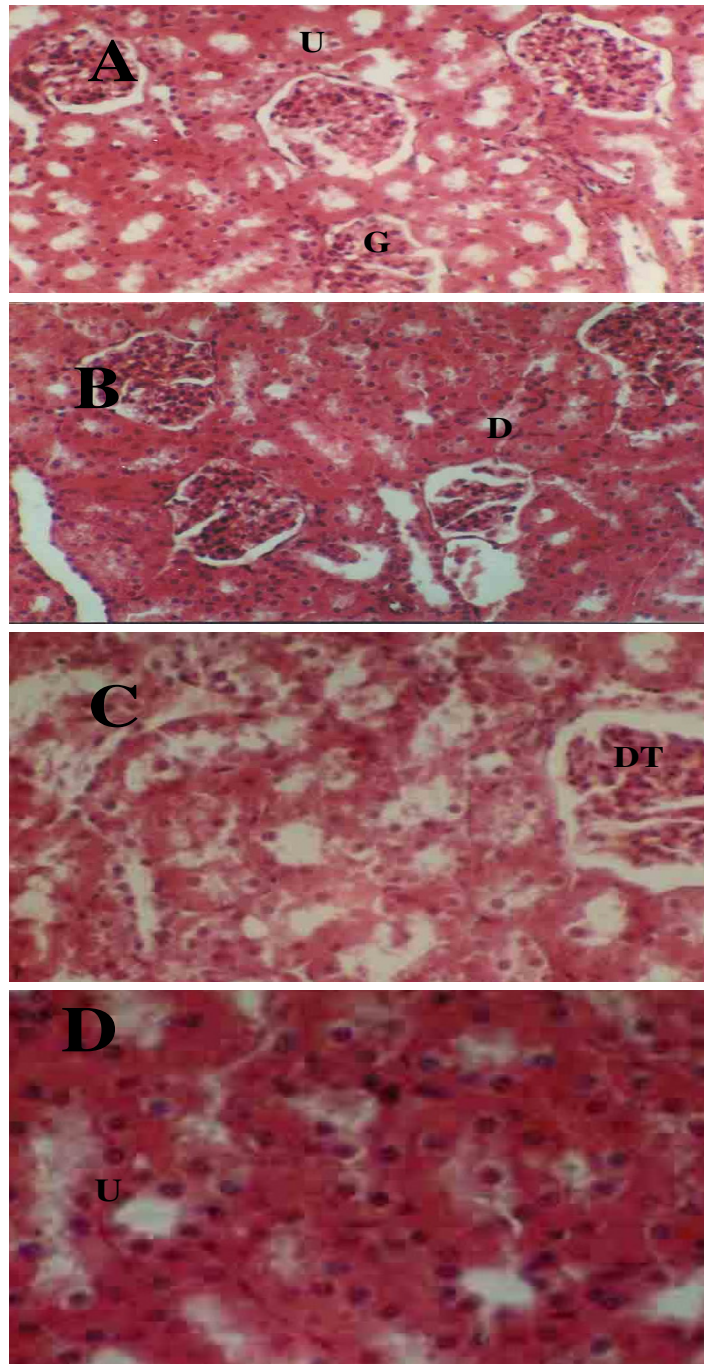
Chicken group parameter	Control (Farm D)		Farm A		Farm B		Farm C	
	Kidney	Liver	Kidney	Liver	Kidney	Liver	Kidney	Liver
Serum ALT(IU/ml)	37.1 $\pm$ 4.44	36.9 $\pm$ 4.49	56.2 $\pm$ 8.88	57.8 $\pm$ 7.91	46.0 $\pm$ 7.09	45.8 $\pm$ 6.99	40.2 $\pm$ 4.67	39.9 $\pm$ 4.99
P <sub>1</sub> value	-	-	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001
P <sub>2</sub> Value	-	-	-	-	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.05	P $\leq$ 0.05
P <sub>3</sub> Value	-	-	-	-	-	-	P $\leq$ 0.05	P $\leq$ 0.05
Serum AST(IU/ml)	41.1 $\pm$ 4.49	40.8 $\pm$ 4.55	55.2 $\pm$ 7.99	56.7 $\pm$ 8.01	49.1 $\pm$ 7.007	48.8 $\pm$ 6.69	43.9 $\pm$ 4.32	44.8 $\pm$ 4.72
P <sub>1</sub> Value	-	-	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001
P <sub>2</sub> Value	-	-	-	-	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.05	P $\leq$ 0.05
P <sub>3</sub> Value	-	-	-	-	-	-	P $\leq$ 0.05	P $\leq$ 0.05
MDA (n mol/mg protein)	2.35 $\pm$ 0.18	2.33 $\pm$ 0.16	3.28 $\pm$ 0.49	3.21 $\pm$ 0.55	3.01 $\pm$ 0.31	2.98 $\pm$ 0.23	2.61 $\pm$ 0.47	2.56 $\pm$ 0.32
P <sub>1</sub> Value	-	-	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.001
P <sub>2</sub> Value	-	-	-	-	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.01	P $\leq$ 0.01
P <sub>3</sub> Value	-	-	-	-	-	-	P $\leq$ 0.05	P $\leq$ 0.05
SOD (MU/mg protein)	224.1 $\pm$ 13.8	221.3 $\pm$ 14.1	110.2 $\pm$ 2704	106.9 $\pm$ 21.7	207.1 $\pm$ 27.7	203.3 $\pm$ 23.8	188.9 $\pm$ 24.	187.7 $\pm$ 23.3
P <sub>1</sub> value	-	-	P $\leq$ 0.001	P $\leq$ 0.001	N.S.	N.S.	P $\leq$ 0.001	P $\leq$ 0.001
P <sub>2</sub> value	-	-	-	-	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.01	P $\leq$ 0.01
P <sub>3</sub> value	-	-	-	-	-	-	P $\leq$ 0.01	P $\leq$ 0.01
CAT(n mol/min/mg protein)	9747.9 $\pm$ 142.1	9757.1 $\pm$ 122.1	2300.4 $\pm$ 144,8	2174.8 $\pm$ 139.1	8382.7 $\pm$ 137.7	8300.4 $\pm$ 144.7	4010.7 $\pm$ 146.4	3989.1 $\pm$ 177.6
P <sub>1</sub> value	-	-	P $\leq$ 0.001	P $\leq$ 0.001	N.S.	N.S.	P $\leq$ 0.001	P $\leq$ 0.001
P <sub>2</sub> value	-	-	-	-	P $\leq$ 0.001	P $\leq$ 0.001	P $\leq$ 0.01	P $\leq$ 0.01
P <sub>3</sub> value	-	-	-	-	-	-	P $\leq$ 0.05	P $\leq$ 0.05

P1: comparison to normal control, P2: comparison to Farm B group, P3: comparison to Farm C group, N.S= not significant difference.



**Figure 2.** A: Hepatic tissues showing hepatic strands with necrosis around the central vein (CV) leaving blood sinusoids (S) X 400, (B): hepatic tissues of farm 'B' group showing highly cellular necrosis (arrows) around the central veins X 250, (C): hepatic tissues of farm "C" showing degenerating central vein (CV) and (D): hepatic tissues of farm 'D' showing clear regular hepatic strands X 250 (H and E stains).





**Figure 3.** A: Renal tissues showing convoluted urineferous tubules (u) and glomeruli (G) X440, B: renal tissues of farm "B" group showing certain degenerated urineferous tubules (D) X 440, C: renal tissues of farm "C" showing dilatation of Bowman's capsule (DT) X 440, and D: renal tissues of farm "D" showing normal renal structure with regulated nuclear arrangement of urineferous tubules (u) X 500 (H&E stains).

The trace amounts of mercury detected in the liver, kidneys, muscle and blood tissues of chickens showed the current status and the background Hg concentrations

in water supply the poultry farms. The detected Hg levels in chicken organs are in accordance with those reported by other authors (Pribilincov et al., 1997; Marettova et al.,

2003; Cabanero et al., 2005).

Most mercury compounds of the contaminant water are inorganic form. Absorption of inorganic mercury compounds may be 15% or less (WHO, 1980), whereas methyl mercury is almost completely absorbed. Inorganic mercury compounds are rapidly accumulated in the kidney which is the main target organ for these compounds. Animal data indicate that the kidney accumulate the highest tissue concentration no matter what form of mercury is administered (WHO, 1976). This opinion gets along with the present results which showed that the kidney followed by liver are the organs with the highest bioaccumulation of mercury in all farm samples. Similar results were previously reported by Manahan (1989), Lund et al. (1993), Mahboob et al. (2001) and Sener et al. (2007).

Although, the kidney tissue showed the highest concentration of mercury, its residue could not be detected in all samples which may be explained by uncertain and indirect nature of relationships calculated between the intake of mercury through water and levels of mercury in the indicator organs (WHO, 1972; Manahan, 1989; Pribilincov et al., 1997; El-Shenawy and Hassan, 2008). Considerable individual variation around the average values of mercury residual have been noted, which must be taken into account in the estimation of risk in exposed populations.

Regarding to the presence of mercury in the kidney and or liver in chicken supplied by undetectable mercury level well-water, WHO (1972) reported that the rate of mercury accumulation is independent of the intake level and that of toxic level would be reached eventually, even at a very low intake level. Such conclusion is in agreement with that of Cabanero et al. (2005). It is evident therefore, that even very small concentration of mercury in the environment may constitute eventual toxicological hazard.

In the present study, alteration in the normal levels of various serum biochemical parameters accompanied by the histopathological necrosis and degenerative changes in the liver and kidney tissue were the main toxic effects observed in the chickens drinking polluted water. In fact, some physiological changes has been accompanies with mercury toxicities. Such decrease in body weight gain of chicken due to mercury exposure has been reported earlier also (Marettova et al., 2003). Furthermore, it has also been reported that mercury exposure increased the activities of AST and qualitative vascular degenerative of kidney tissues and necrotic changes were also observed in the liver of chickens (Zraly et al., 2008). These observations are clearly in agreement with the present findings. Serum AST and GPT activities were used as a marker of tissue damage. Mercury toxicity's produces tissue damage due to its toxic metabolites (Sharma et al., 2002). The toxic metabolite free radical is produced by cytochrome p450 which further reacts with oxygen to produce trichloromethyl peroxy radicals (Borg et al.,

2003). These radicals bind covalently with the macromolecule and cause peroxidative degradation of lipid membranes of the liver and kidney. Increased lipid peroxidation under pollution conditions can be due to increased oxidative stress in the cell as a result of depletion of antioxidant scavenger systems. Associated with the changes in lipid peroxidation, the affected tissues showed decreased activities of key antioxidants SOD and CAT and increase MDA which play an important role in scavenging the toxic intermediate of incomplete oxidation. SOD and CAT are the two major scavenging enzymes that remove toxic free radicals *in vivo*. Previous studies have reported that the activity of SOD is low after mercury toxicities, Sobutskii et al. (2007) who measured biochemical indexes of blood after low doses of mercury exposures come in agreement with our results.

Elevation in the activity of serum AST and GPT, the cytoplasmic enzymes, indicates for necrotic lesions in the liver and tissue degeneration of the kidney, while a decrease in serum SOD and CAT levels indicates for no congestion or cholestasis (Lysenko, 2000; Borg et al., 2003; Cabanero et al., 2005). These researchers reported that chickens treated with HgCl<sub>2</sub> showed significant elevations in serum glutamate oxaloacetate transaminase (SGOT) and serum glutamate pyruvate transaminase (SGPT) activities, whereas a significant decline in the SOD and CAT activities and also come in agreement and confirm with our results.

The present study also showed that chickens supplied with mercury polluted water have elevated levels of serum AST and SGPT, whereas a reduced level of SOD and CAT as compared to the control group, indicating clearly that our results are in agreement with other studies on chickens (Borg et al., 2003; Cabanero et al., 2005; Zraly et al., 2008).

On the other hand, in non-chicken model also (teleost fish), Sastry and Sharma (1980) reported that SOD activity decreased in acute exposure to HgCl<sub>2</sub> and increased in chronic exposure to HgCl<sub>2</sub>, however, there was elevation of both AST and SGPT either in acute or chronic exposure to HgCl<sub>2</sub>. Thus, SGPT activity in serum, could serve as a marker enzyme to evaluate functional status of liver as suggested by Sobutskii et al. (2007). Furthermore, Jagadeesan and Pillai (2007) also reported significant increase in the level of serum AST and SGPT in rats due to HgCl<sub>2</sub> treatment for longer time period (30 days). In another study, a significant rise in the serum SGPT and AST also has been reported in mercury exposed rats (Singh et al., 2007).

Altogether, the present results in the light of the above cited literature clearly indicate that increase in serum GPT and AST and decrease in serum SOD and CAT, can be used as potential enzyme biomarkers for mercury-induced hepatotoxicosis and nephrotoxicosis which ultimately affects the general health by altering the functional and structural integrity of liver and kidney and

serve as possible bio-indicators for mercury poisoning. However, in order to establish these serum enzyme levels as biomarkers for mercury poisoning, further detailed studies are required at experimental as well as clinical levels.

In conclusion, heavy metals in polluted water have a great reflection on the blood serum levels of the studied antioxidant enzymes in chickens drinking from this polluted water. Thus, ingestion of contaminated edible poultry should receive special attention in order to protect human beings from the dangerous hazards. Assessment of the employment of chicken as a sensitive biomonitor for the pollution of ground water environment may make a general conclusion if the vital activities and general health in human and wild life are adverse trends associated with environmental chemical exposure.

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