The protective role of vitamin C in reducing the pathological changes of cadmium chloride in albino rats: with special reference to thyroid gland

By

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SUMMARY

The human and animal population are exposed to several xenobiotics. One of them is cadmium, that may contaminate food, water and air. Fifty adult albino rats of both sex were divided into 5 equal groups to evaluate the adverse effects of cadmium chloride (CdCl₂) on the kidneys, liver and thyroid gland and to evaluate the protective role of vitamin C. Gp.(1) was the control, Gps. (2,3,4) served as experimental control. Gp. (5) was the main experimental group. Gp. (1) was the nontreated control. Gp.(2) was orally given 1 ml distilled water / rat once daily. Gp. (3) was orally given 10 mg vitamin C / kg B.wt once daily. Gp. (4) was orally given 10 mg Cd Cl₂ / kg B.wt once daily. Gp. (5) was given 10 mg vitamin C / kg B.wt and 10 mg Cd Cl₂ / kg B.wt once daily. The treatments were given orally by the stomach tube for 16 weeks. Necropsy was performed at the end of the experiments. All the macroscopic abnormalities were recorded. Specimens from the kidneys, liver and thyroid gland were collected and prepared for microscopical examination. Other thyroid specimens were taken and prepared for electron microscopical examination.

Gp. (4), cadmium chloride group, revealed degenerative and necrotic changes in the renal epithelia which was subsequently, invaded and replaced by fibrous connective tissue and round cell infiltrations. Hypercellularity of the glomerular tufts besides perivascular edema and congestion of the renal blood vessels were observed. The liver showed degenerative changes and coagulative necrosis of the hepatocytes. Congestion of the hepatic blood vessels and lymphocytic infiltrations in the portal areas and in the interstitial tissue were observed. The thyroid gland revealed dilated follicles with pale eosinophilic vacuolated colloid and lined with flattened epithelium. The transmission electron microscopic examination of thyroid gland revealed apoptotic follicular cells, dilation of the endoplasmic reticulum and distroted Golgi apparatus and mitochondria. Gp. (5), Cd Cl₂ and vitamin C group, showed milder lesions in comparison with gp. (4) which pointed out a partial protective role of vitamin C against CdCl₂ toxicity.

The thyroid function test of gp. (4) showed a decrease in T3 and T4 and an increase in TSH which were statistically significant when compared with
group 2 (positive control group). The presence of vitamin C in gp. (5) produced an improvement in the values of T3, T4 and TSH in comparison with gp. (4).

It can be concluded that, Cd Cl2 has adverse effects on the kidneys, liver and thyroid gland, but vitamin C has a partial protective effect against these effects.

INTRODUCTION

Humans and animals interact with their environments on a daily basis and, as a consequence, are exposed to a broad spectrum of synthesized chemicals present in the food they eat, the air they breathe and the water they drink (Wade et al., 2000 and Bolkent et al., 2007). Cadmium has been released into the environment through human activities and is routinely found as a contaminant in tissues collected from the human population throughout the world (Newsome et al., 1995). Exposure to cadmium has increased because of its presence in fertilizers and in sewage sludge and also its industrial use in cadmium nickel batteries (Ellis, 1995). Cadmium is unique among the other metals because of its toxicity at a very low dosage and long biologic half life (30 years in human) and its low rate of excretion from the body (Jones and Cherian, 1990). Although there is number of reports on occupational and environmental exposure to cadmium compounds, there is neither a safe nor a specific chelating agent for acute or chronic cadmium intoxication (Hideaki et al., 1997). Murugavel and Pari (2007) observed that, cadmium administered subcutaneously for 3 weeks to rats induced hepatic necrosis and severe infiltration of the portal areas with inflammatory cells. Renal damage and congestion of the perirenal blood vessels in guinea pigs given 1 mg cadmium / animal / day in drinking water for 12 weeks and vitamin C can be effective in the protection of cadmium-induced nephrotoxicity (Nagyova et al., 1994).

Environmental contaminants may cause alterations in the function of endocrine system, thereby disrupting developmental and physiological functions in animals and humans (Brouwer et al., 1998). Chronic cadmium administration produced a significant increase in the median thyroid follicle colloid (Wade et al., 2002), deterioration of the rough endoplasmic reticulum and marked swelling of the mitochondria in the thyroid follicular epithelium of the rat (Pilat-Marcinkiewicz et al., 2002).
Interestingly, several compounds have been shown to induce tolerance to cadmium-induced tissue injury (prevention of toxicity). These chemicals include metals (zinc and nickel), steroid hormones (androgens and progestogens), antioxidants (ascorbate and tocopherol) (Waalkes and Goering, 1990). Vitamin C is an essential antioxidant which plays a vital role in protecting cells from damage of oxidizing agents (Grosicki, 2004). Peters et al (1995) reported that, vitamin C can alter the toxicity of cadmium.

The objective of the present study was to investigate the pathologic effects of cadmium chloride on the kidneys, liver and thyroid glands with special reference to the ultrastructural feature of the thyroid gland, besides the evaluation of the thyroid function by measuring serum tri-iodothyronine (T3), thyroxin (T4) and thyroid stimulating hormone (TSH). Moreover, the role of vitamin C in protecting the kidneys, liver and thyroid gland against the lesions induced by of cadmium chloride, if found, was considered.

**MATERIALS AND METHODS**

This study was conducted on 50 normal adult albino rats of both sexes, weighing approximately 160-200 gm each. They were obtained from the breeding animal house of Faculty of Medicine, Zagazig University. The rats were divided into 5 equal groups (Table 1). Gp.(1) was the non treated control group. Gps. (2, 3 and 4) were the experimental control. Each rat of gp. (2) was orally given 1 ml distilled water once a day. Each rat of gp. (3) was orally given vitamin C, obtained from ADWIC (10 mg/kg. B.wt. dissolved in 1 ml distilled water) once a day (Sinha and Bose, 1992). Each rat of gp. (4) was orally given Cd Cl₂, obtained from El-Nasr Pharmaceutical Chemical Company (10 mg / kg. B.wt. dissolved in 1 ml of distilled water) once a day (Baranski et al., 1982). Each rat of gp.(5), experimental, was orally given vitamin C (10 mg/ kg. B.wt.) once a day, then CdCl₂ was orally given at a dose of 10 mg/kg B.wt. once a day. The treatments were orally given by the stomach tube for 16 weeks.
Table (1): Experimental design.

<table>
<thead>
<tr>
<th>Gps.</th>
<th>No. of rats</th>
<th>Design</th>
<th>Oral treatments</th>
<th>No. of sacrificed rats, 16 weeks PA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Distilled water, 1 ml/rat</td>
<td>Vitamin C, 10 mg/kg B.wt</td>
</tr>
<tr>
<td>1</td>
<td>10</td>
<td>Control</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>2</td>
<td>10</td>
<td>Experimental</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>10</td>
<td>Experimental</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>4</td>
<td>10</td>
<td>Experimental</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>10</td>
<td>Experimental</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Total</td>
<td>50</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

PA = Post administration.  
No. = Number.

A-Thyroid Function Tests:

Venous blood samples were obtained from all the animals by means of capillary glass tubes from the retro-orbital plexus, under light anaesthesia at the end of experiment according to the procedure described by Schemere (1967). The collected blood samples were incubated at 37°C until blood clotted, then centrifuged to separate the serum for the determination of triiodothyronine (T3), thyroxin (T4) and thyroid stimulating hormone (TSH) concentrations. T3 and T4 were determined using radioimmunoassay kits (RIA) according to Larsen (1981) and Lehotay et al. (1982). Immunometric assay of TSH was done according to Kwiatkowski et al. (1990).

B- Histopathological examination:

After taking the blood samples, necropsy was performed and all the macroscopic abnormalities were recorded. Specimens from the kidneys, liver and thyroid glands were obtained and fixed in 10% neutral buffered formalin solution. Paraffin sections of 5 µ thickness were prepared and stained with Haematoxylin (H) and Eosin (E) for light microscopic examination (Bancroft and Steven, 1996). Other thyroid specimens were taken and dissected by sharp blade into very small blocks for electron microscopic processes. These small blocks were fixed in 2.5% glutaraldehyde overnight. The tissue was then rinsed in 1.5% phosphate buffer (pH 7.35) and fixed in osmium tetroxide for
one hour. The tissue was dehydrated and embedded in Epon araldite resin \textit{(Hayat, 1989)}. Ultrathin sections were obtained, stained with uranyl acetate and lead citrate and then were examined by transmission electron microscope (JEOL 1000 X) in Zagazig University, Electron Microscope Centre. Photographs were taken, developed and printed.

C- Statistical Analysis:

The collected results were computerized for statistical analysis by using Epi-info (version 6.1) software statistical package \textit{(WHO, 1994)}. Comparing the means of T3, T4 and TSH was done by using t-test and analysis of variance (ANOVA). The least significant differences were calculated by using the SPSS statistical package \textit{(SPSS, 1997)}.

RESULTS

A-Pathological findings:

Group (4): (given 10 mg CdCl$_2$/kg B.wt).

The kidneys were macroscopically slightly enlarged. Microscopically, the kidneys revealed congestion (fig. 1) and thickening of the wall of the renal blood vessels with perivascular edema (Fig. 2). Periglomerular proliferation of fibrous connective tissue, infiltrated with round cells (fig. 3), was observed, which subsequently invaded and replaced the renal parenchyma (fig. 4). Hypercellularity of the glomerular tufts (fig. 5) was seen. Degenerated renal tubules in the form of cloudy swelling and hydropic degeneration were detected. Some renal tubules showed coagulative necrosis in their lining epithelium.

The liver was macroscopically slightly congested.

Microscopically, congestion of the hepatic sinusoids, associated with pressure atrophy of some hepatocytes (fig. 6) was observed. The portal areas showed congested blood vessels and were infiltrated with round cells (fig. 7) mostly lymphocytes. Focal coagulative necroses were encountered and represented by Karyolysis (fig. 8). Interstitial aggregations of lymphocytes focally replaced the hepatic parenchyma (fig. 9). Vacuolation of the hepatocytes was detected. These vacuoles were sharp empty or contained pale eosinophilic materials.

The thyroid gland, was macroscopically normal.

Microscopically, some thyroid follicles were dilated with pale eosinophilic vacuolated colloid and lined with flattened epithelium (fig. 10). Under electron microscope, the thyroid follicular cells showed apoptosis, dilatated endoplasmic reticulum and distorted Golgi apparatus and mitoch-
ondria (fig. 11). Distortion and disruption of thyroid follicular architecture were seen.

**Gp. (5): (given 10 mg CdCl₂/kg B.wt and 10 mg vitamin C /kg B.wt).**

No gross lesions were recorded in the collected organs. Microscopically, the kidneys showed mild congestion of the intertubular blood vessels, and few round cell infiltration among the renal tubules (fig. 12). The liver revealed congestion of the hepatic blood vessels (fig. 13) without inflammatory cells. The thyroid gland revealed normal thyroid follicles, lined by cuboidal epithelium and filled with evenly stained colloid (fig. 14). Milder ultrastructural changes were seen in comparison with group (4). Such changes were represented by dilated endoplasmic reticulum and less distortion of the mitochondria (Fig. 15).

**Gp. (1) (control that was left without treatment), Gp. (2): (given 1 ml distilled water / rat) and Gp. (3) (given 10 mg vitamin C /kg B.wt) showed neither gross nor microscopical lesions. Ultrastructural, the thyroid gland revealed normal nuclei, endoplasmic reticulum and mitochondria (fig. 16).**

**B- Thyroid Function Tests**

Serum T₃ (ng/dL), T₄ (µg/dL) and TSH (µU/ dL) were estimated in the 5 studied groups and the results were tabulated in (tables, 2-8). Table (2) shows no significant difference between the control and the experimental control groups, so only the results of the experimental control group will be used for comparison. Gps. (2 and 3) showed no abnormal findings, while gp. (4) displayed a significant decrease in T₃ and T₄ and a significant increase in TSH when compared with gp.(2) (P<0.001). Gp. (5) showed a significant increase in the mean values of T₃ and T₄ (77 ± 8.767, 4.194 ± 0.524) when compared with gp.(4) (70 ± 9.743, 3.671 ± 0.681) respectively with P<0.05. There was a significant decrease in the mean value of TSH in gp. (5) (1.400 ± 0.183) in comparison to group 4 (1.528 ± 0.197) with P< 0.05.

The addition of vitamin C to CdCl₂ in gp. (5) produced a significant increase in the function of the thyroid gland when compared with gp. (4). Gp. (5) was still suffering hypofunction of the thyroid gland, when compared with gp. (2).
Figs. (1-4): Gp. (4), Kidney:

1- Severely congested blood vessel (thin arrow) and cloudy swelling of the epithelial lining of some renal tubules (thick arrow), H & E., x1200.

2- Perivascular edema (thin arrow) and thickening of the wall of the blood vessel (thick arrow), H & E., x300.

3- Periglomerular proliferation of fibrous connective tissue infiltrated with round cells (arrow), H & E., x1200.

4- Focal replacement of the renal parenchyma by fibrous connective tissue and lymphocytes (arrow), H & E., x1200.
Figs. (5-8): Gp. (4):

5- **Kidney**: peritubular congestion (thin arrow) and hypercellularity of the glomerular tufts (thick arrow), H & E., x300.

6- **Liver**: congestion of hepatic sinusoids (arrow) associated with pressure atrophy of adjacent hepatocytes, H & E., x300.

7- **Liver**: portal area with congested blood vessel and few leukocytic infiltration (thin arrow) besides focal coagulative necrosis of the hepatocytes (thick arrow), H & E. x300.

8- **Liver**: high power of fig. (7) to show the coagulative necrosis of hepatocytes represented by karyolysis (arrow), H & E. x1200.
9- Liver : focal interstitial aggregation of round cells (arrow),  
H & E., x300.

10- Thyroid gland: dilated thyroid follicles with pale eosinophilic 
vacuolated colloid (arrow),  
H & E. x300

11- A transmission electron micrograph of thyroid gland: apoptosis “N”,
dilated fragmented rough endoplasmic reticulum (R) and distorted 
Golgi apparatus (G) and mitochondria (M),  
EM, x 8000.

Fig. 12- Kidney of Gp. (5): mild congestion of intertubular blood vessels 
(arrow) and few round cell infiltration among the renal tubules,  
H & E., 300.
Fig. (13-15): Gp. (5):

13- Liver: congestion of the hepatic blood vessel (arrow), H & E. x 300.

14- Thyroid gland: normal thyroid gland filled with evenly stained colloid (arrows), H & E., x 300.

15- A transmission electron micrograph of thyroid gland: dilated endoplasmic reticulum (R) and distorted mitochondria (M), EM., x 8000.

Fig. 16- A transmission electron micrograph of thyroid gland of control group: normal nuclei (N) mitochondria (M), Golgi apparatus (G) and rough endoplasmic reticulum (R), EM., x 8000.
Table (2): Means (X), and standard deviations (SD) of triiodothyronine (T3) tetraiodothyronine (T4) and thyroid stimulating hormone (TSH) in the rats of group 1 (-ve control group) and group 2 (Distilled water group).

<table>
<thead>
<tr>
<th></th>
<th>Group 1</th>
<th>Group 2</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>X</td>
<td>SD</td>
<td>X</td>
</tr>
<tr>
<td>T3 (ng/dL)</td>
<td>82.315</td>
<td>11.857</td>
<td>82.150</td>
</tr>
<tr>
<td>T4 (µg/dL)</td>
<td>5.33</td>
<td>0.989</td>
<td>5.27</td>
</tr>
<tr>
<td>TSH(µU/dL)</td>
<td>1.243</td>
<td>0.175</td>
<td>1.257</td>
</tr>
</tbody>
</table>

The number of rats in each group = 10 rats.
There is no significant difference between negative and positive control groups.

Table (3): Means (X), standard deviations (SD) and F-test of triiodothyronine (T3) in the rats of studied groups 2, 3, 4 and 5.

<table>
<thead>
<tr>
<th>Groups</th>
<th>X±SD</th>
<th>Range ng/dl</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 ( Distilled water group)</td>
<td>82.160 ± 11.965</td>
<td>61-105</td>
</tr>
<tr>
<td>3 (Vit. C group )</td>
<td>83.640 ± 9.864</td>
<td>60-101</td>
</tr>
<tr>
<td>4 ( CdCl₂ group)</td>
<td>70.0 ± 9.743</td>
<td>58-86</td>
</tr>
<tr>
<td>5 (CdCl₂ &amp; Vit. C group)</td>
<td>77.0 ± 8.767</td>
<td>59-94</td>
</tr>
<tr>
<td>F test</td>
<td>7.462</td>
<td></td>
</tr>
<tr>
<td>P value</td>
<td>0.0002</td>
<td></td>
</tr>
</tbody>
</table>
Table (4): Least significant difference (LSD) for comparison of serum triiodothyronine (T₃) in gps. (2, 3, 4 and 5).

<table>
<thead>
<tr>
<th>Groups</th>
<th>2 (Distilled water group)</th>
<th>3 (Vit.C group)</th>
<th>4 (CdCl₂ group)</th>
<th>5 (CdCl₂ &amp; Vit. C group)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 (Distilled water group) P=</td>
<td>-</td>
<td>0.154 (NS)</td>
<td>0.0003 **</td>
<td>0.098 (NS)</td>
</tr>
<tr>
<td>3 (Vit. C group ) P=</td>
<td>-</td>
<td>-</td>
<td>0.0008 **</td>
<td>0.112 (NS)</td>
</tr>
<tr>
<td>4 (CdCl₂ group ) P=</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.042 *</td>
</tr>
<tr>
<td>5 (CdCl₂ &amp; Vit. C group) P=</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

P = Probability
LSD: 6.394
*: Significant (P < 0.05)
NS: Non significant (P > 0.05)
**: Highly significant (P < 0.001)

Table (5): Means (X), standard deviations (SD) & F-test of triiodothyronine (T₄) in gps. (2, 3, 4 and 5).

<table>
<thead>
<tr>
<th>Groups</th>
<th>X±SD</th>
<th>Range µg/dl</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 ( Distilled water group)</td>
<td>5.26±0.986</td>
<td>3.5-7.5</td>
</tr>
<tr>
<td>3 (Vit. C group )</td>
<td>5.385±0.892</td>
<td>3.5-6.8</td>
</tr>
<tr>
<td>4 (CdCl₂ group)</td>
<td>3.671±0.681</td>
<td>3.0-5.5</td>
</tr>
<tr>
<td>5(CdCl₂ &amp; Vit. C group)</td>
<td>4.194±0.524</td>
<td>3.5-5.6</td>
</tr>
<tr>
<td>F test</td>
<td>22.280</td>
<td></td>
</tr>
<tr>
<td>P value</td>
<td>0.0001</td>
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</tr>
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</table>
Table (6): Least significant difference (LSD) for comparison of serum triiodothyronine (t4) in the rats of studied gps 2, 3, 4 and 5.

<table>
<thead>
<tr>
<th>Groups</th>
<th>2 (Distilled water group)</th>
<th>3 (Vit.C group)</th>
<th>4 (CdCl₂ group)</th>
<th>5 (CdCl₂ &amp; Vit. C group)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 (Distilled water group) P=</td>
<td>-</td>
<td>0.215 (NS)</td>
<td>0.0003 **</td>
<td>0.037 *</td>
</tr>
<tr>
<td>3 (Vit. C group) P=</td>
<td>-</td>
<td>-</td>
<td>0.0007 **</td>
<td>0.025 *</td>
</tr>
<tr>
<td>4 (CdCl₂ group) P=</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.041 *</td>
</tr>
<tr>
<td>5 (CdCl₂ &amp; Vit. C group) P=</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

P = Probability
LSD: 0.4999
NS: Non significant. (P > 0.05)
*: Significant (P < 0.05)
**: Highly significant (P < 0.001)
N.B: Number of rats in each group = 10 rats

Table (7): Means (X), standard deviations (SD) & F-test of thyroid stimulating hormone (TSH) in gps (2, 3, 4 and 5).

<table>
<thead>
<tr>
<th>Groups</th>
<th>X±SD</th>
<th>Range Ug/dl</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 ( Distilled water group)</td>
<td>1.258±0.176</td>
<td>0.86-1.47</td>
</tr>
<tr>
<td>3 (Vit. C group)</td>
<td>1.217±0.138</td>
<td>0.91-1.42</td>
</tr>
<tr>
<td>4 (Cd Cl 2 group)</td>
<td>1.528±0.197</td>
<td>1.10-1.76</td>
</tr>
<tr>
<td>5( Cd Cl 2 &amp; Vit. C group)</td>
<td>1.400±0.183</td>
<td>1.00-1.70</td>
</tr>
<tr>
<td>F test</td>
<td>14.01</td>
<td></td>
</tr>
<tr>
<td>P value</td>
<td>0.0001</td>
<td></td>
</tr>
</tbody>
</table>
Table (8): Least Significant Difference (LSD) for comparison of thyroid stimulating hormone (TSH) in gps (2, 3, 4 and 5).

<table>
<thead>
<tr>
<th>Groups</th>
<th>2 (Distilled water group)</th>
<th>3 (Vit. C group)</th>
<th>4 (CdCl₂ group)</th>
<th>5 (CdCl₂ &amp; Vit. C group)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>-</td>
<td>0.084</td>
<td>0.0007 **</td>
<td>0.034 *</td>
</tr>
<tr>
<td>3</td>
<td>-</td>
<td>-</td>
<td>0.0005 **</td>
<td>0.017 *</td>
</tr>
<tr>
<td>4</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.039 *</td>
</tr>
<tr>
<td>5</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
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</tbody>
</table>

P = Probability
LSD: 0.1073           NS: Non significant. (P > 0.05)
*: Significant (P < 0.05)    **: Highly significant (P < 0.001)
N.B: Number of rats in each group = 10 rats

**DISCUSSION**

Heavy metals, such as cadmium, pose a number of environmental problems in addition to being detrimental to human and animal health (Warren et al., 2000). This study showed that, the cadmium chloride revealed degenerative and necrotic changes in the renal epithelia which was subsequently replaced by fibrous connective tissue and round cells. Hypercellularity of the glomerular tufts, besides congestion of the renal blood vessels were observed. Thickening of the renal blood vessels and perivascular edema were seen. The previous lesions are in partial agreement with Horiguchi et al. (2006) and Thijssen et al. (2007) who noticed proximal tubular damage of the rat and mouse kidneys. The animals were given cadmium chloride in the drinking water and by injection respectively. Gubrelay et al. (2004) showed congestion at corticomедullary junction and necrosis of the renal tubular epithelia of kidneys in rats exposed to 0.5 and 1 mg cadmium chloride for 3 days. The liver showed focal coagulative necrosis and degenerative changes of the hepatocytes. Congestion of the hepatic blood vessels and lymphocytic infiltrations in the portal areas and in the interstitial tissue were observed. The achieved results are in partial concurrence with Gubrelay et al. (2004) and Kayu et al. (2006). They found vacuolar...
degeneration in the hepatocytes and congestion of the hepatic blood vessels. The aforementioned lesions in the kidneys and liver may be attributed to the direct effects of the cadmium chloride or its active metabolites on the renal and hepatic tissue. Newairy et al. (2007) reported that cadmium induced hepatotoxicity due to its active stress by increasing the level of free radicals and decreasing the antioxidant level. Microscopically the thyroid of the rats in gp. (4) revealed dilated thyroid follicles with pale eosinophilic vacuolated colloid which led to disruption of the thyroid follicular architecture. This result is similar to that obtained by Wade et al. (2002). They found a significant increase in the median thyroid follicle colloid cross sectional area. The transmission electron microscopic study of the thyroid gland of cadmium chloride group revealed apoptosis, dilatation of the endoplasmic reticulum and distorted Golgi apparatus and mitochondria. These findings are in agreement with Yoshizuka et al. (1991) who found that, the electron microscopic examination of the thyroid glands of rats exposed to cadmium showed deterioration of the rough-surfaced endoplasmic reticulum and marked swelling of the mitochondria of the thyroid follicular epithelium. They added that the accumulation of cadmium in the mitochondrial of the thyroid follicular epithelium may disturb the oxidative phosphorylation of this organelle and the loss of energy supply possibly caused the inhibition of the synthesis and release of thyroid hormones.

Gp. (5) showed milder lesions than gp. (4). Mild congestion of the renal blood vessels and a few leukocytic infiltration were seen in the kidneys. The liver showed mild congestion of the hepatic blood vessels, meanwhile, the thyroid gland showed nearly normal microscopic picture. The previous results coincide with Shiraishi et al. (1993) who recorded that the pre-treatment of rats with ascorbic acid decreased the hepatic toxicosis produced by cadmium. Nagyova et al. (1994) showed that vitamin C can be effective in the protection of cadmium induced nephrotoxicity. Sato et al. (1990) and Grosicki (2004) recorded that vitamin C is an excellent antioxidant. It was effective in reducing the toxic effects of cadmium, through inhibition of lipid peroxidation (Peters et al., 1995) and increase the activities of antioxidant enzymes (Newairy et al., 2007).

The thyroid function tests showed that, gp.(4) exhibited a statistically significant decrease in both the serum T3 and T4 (P<0.001) and a statistically significant increase in TSH (P<0.001). These resu-
The results are consistent with Levesque et al. (2003) who reported that the environmental exposure of fish to Cd decrease of the plasma T₄ and T₃ with structural alteration in the thyroid follicle epithelium. Pavia Junior et al. (1997) found that, T₄ and T₃ concentrations in cadmium chloride (CdCl₂)-treated rats were significantly (P<0.01) decreased. The environmental contaminants, like cadmium, have been shown to cause a significant toxicity to the hypothalamic-pituitary-thyroid axis in laboratory animal studies with significantly increased TSH (Wade et al., 2002). Lafuente et al. (1997) reported that the subchronic CdCl₂ administration for 14 days led to an increase in the level of TSH in adult male albino rats. Shupnik et al. (1986) reported that the TSH secretion by the pituitary gland is inversely related to the thyroid hormone levels that serve as the basis for feedback control of the circulating thyroid hormone.

Group (5) showed an increase in the mean values of the T₃ and T₄ and a reduction in the mean value of the TSH in comparison with group 4 (CdCl₂ only) (P<0.05). The results showed the ability of vitamin C to improve the disturbance of the thyroid function caused by CdCl₂ but not to the basal level. The results are in line with previously reported work in vivo experiments, which revealed that, the human equivalent therapeutic dose (10 mg/kg body weight / day) of vitamin C could reduce the toxin-induced damage, but failed to restore the control level (Sinha and Bose, 1992). Similar findings were observed in individuals exposed to environmental toxins and recovered faster in the presence of high vitamin C status (Kato et al., 1981). The current results are consistent with Gupta and Kar (1998) who reported that, the administration of Cd to Swiss male mice induced thyroid dysfunction and lipid peroxidation leading to a decrease in the serum concentrations of the thyroid hormones and hepatic type I iodothyronine 5-Monpdeiodinase (5 D-I) activity and an increase in the level of lipid peroxidation. The metal-induced a decrease in the hepatic 5D-I activity, but the serum T3 concentration was restored by treatment with ascorbic acid.

Finally, it could be concluded that, cadmium chloride has adverse effects on the kidneys, liver and thyroid gland. Meanwhile, vitamin C has a partial protective effect against this effect. People exposed to cadmium should be advised to take regular doses of vitamin C to decrease the metal adverse effects on the kidneys, liver and thyroid gland.
REFERENCES


Thijssen, S.; Maringwa, J.; Faes, C.; Lambrihts, and Van Keokhove, E. (2007): "Chronic exposure of mice to environmentally relevant, low doses of cadmium leads to early renal damage, not pred-
icted by blood or urine cadmium levels." Toxicology, 229: 145-156.


