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The Different Responses of Some Adult Albino Rats' Body Organs to the Genotoxic Effect of Cadmium

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Abstract Cadmium (Cd) is one of the most toxic environmental pollutants affecting the DNA. Cadmium interferes with DNA repair process and induces oxidative stress leading to DNA damage. Since the most recent molecular cancer genetic and cytogenetic data have provided strong evidence that genetic damage can be the initial step of the malignant process, the early detection of DNA-damaging effect of Cd in different organs is very important to avoid its carcinogenic potential .Hence, this study was undertaken to determine the response of some different rat organs to the genotoxic effect of Cd. The study was conducted on 48 adult male albino rats divided into three main groups; group I (negative control group) consisted of 15 rats, group II (positive control group) consisted of 15 rats, received 1 ml distilled water daily by gavage and group III (cadmium group) consisted of 18 rats, received cadmium chloride in a dose of 5 mg/kg body weight daily by gavage. Each one of these three main groups was subdivided according to the experimental period into three equal subgroups; (a), (b), and (c). The experimental period was two weeks for subgroups Ia, IIa, and IIIa, four weeks for subgroups Ib, IIb, and IIIb and eight weeks for subgroups Ic, IIc, and IIIc. Determination of possible DNA damage using comet assay in lymphocytes, bone marrow (BM), lung, liver, and kidney along with malondialdehyde and Cd levels were carried out. The results of this study revealed a heterogeneous response of the studied organs to the genotoxic effect of Cd. Lymphocytes and BM were early affected while the liver showed the highest genetic damage with longer exposure period. These findings suggest that lymphocytes and BM are more sensitive to Cd genotoxicity than the lung, liver, and kidney. So lymphocytes and BM can be used as early determinants of Cd-induced genetic damage. Considering these issues, health educational programs should be implemented to highlight the dangerous impact of Cd on health. Moreover, persistence of this metal in the environment requires a long-term move towards minimizing human exposure through environmental management and preservation of lower Cd levels wherever possible. Also, further studies are needed to detect the genotoxicity of Cd in other organs.

Introduction

Cadmium (Cd) is a worthy study toxic environmental heavy metal. Generally, about 13,000 tons of Cd are produced yearly worldwide, mainly for nickel-Cd batteries, pigments, chemical stabilizers, metal coatings and alloys (Flora et al., 2008; Yilmaz et al., 2012).

In 2009, Cd has emerged as a major media topic due to a flurry of product recalls triggered by Cd in jewelry, toys, paints, and other common items. In spring 2010, companies recalled necklaces, earrings, and bracelets after discovering the products contained substantial levels of Cd. Then, in June, McDonald's recalled 12 million drinking glasses due to concerns over Cd levels in paint pigments used on the glassware (Mead, 2010).

Due to its low permissible exposure limit, overexposure may occur even in situations where trace quantities of Cd are found in the parent ore or smelter dust (Lampe et al., 2008).

Contamination of water and soil by Cd may result in the uptake of the metal by aquatic and terrestrial organisms. Cadmium is a contaminant of most human foodstuffs because of its high rates of soil-to-plant transfer, rendering diet a primary source of exposure (Asagba, 2010). A recent study in Sweden demonstrated that among foodstuffs, potatoes and

wheat flour were the most important food categories, contributing with 40-50% to the exposure (Sand and Becker, 2012). Moreover, cigarette smoking significantly adds to the body burden of Cd (Klaassen et al., 2009).

Because of its diverse toxic effects; its long biological half-life (20-30 years in humans), low rate of excretion from the body (1-2 µg/day), and its predominant long-term accumulation in soft tissues, such as liver and kidney, Agency for Toxic Substances and Disease Registry (ATSDR) has listed Cd as the 6th most hazardous substance to human health (Yadav and Khandelwal, 2006).

Cadmium alone is genotoxic and enhances the genotoxicity of the other DNA-damaging agents (Filipic et al., 2006). Because of its carcinogenic properties, Cd has been classified as a human carcinogen by the International Agency for Research on Cancer and US National Toxicology program (IARC, 1993; National Toxicology Program, 2000).

The induction of genotoxicity by environmental agents as Cd is very important aspect of modern environmental research. Most recent molecular cancer genetic and cytogenetic data have provided strong evidence that genotoxic events can be the initial step of the malignant process and metals could evoke the first event (Joseph, 2009). Therefore early detection of DNA-damaging effect of Cd in different organs is very important to avoid its carcinogenic potential.

Among the methods for detection of DNA damage, the single cell gel electrophoresis (comet) assay is a well-established, highly-sensitive technique that has been used to detect a broad spectrum of DNA damage (Rozga et al., 2005). In alkaline version of this test, DNA strand breaks and alkali-labile sites are detected and the extent of DNA migration indicates the amount of DNA damage in the cell (Singh et al., 1988).

Hence, this study was undertaken to determine the response of some different rat organs to the genotoxic effect of Cd.

Material and methods

Chemicals

Cadmium chloride ($CdCl_2$) was obtained from Sigma-Aldrich Chemicals, Egypt as white crystalline powder which was dissolved in distilled water before administration to rats by gavage. Also, Fetal Bovine Serum (FBS), normal- and low-melting-point agarose, and sodium sarcosinate, were provided from Sigma-Aldrich Chemicals. All the other chemicals are of analytical grade and were obtained from commercial sources.

Animals

This study was conducted on adult male albino rats weighing 150-170 gm. All approved conditions used for animals housing and handling were considered. Standard laboratory animal feed and water were given *ad libitum*. In addition, the protocol used followed the standard regulations for administration in experimental animals. The animals were acclimatized to experimental conditions for a period of one week prior to the start of dosing.

A number of 48 rats were divided into three main groups; group I (negative control) consisted of 15 rats, group II (positive control group) consisted of 15 rats, received 1 ml distilled water daily by gavage and group III (Cd group), consisted of 18 rats, received $CdCl_2$ in a dose of 5 mg/kg BW/day by gavage to induce DNA damage according to Ohba et al. (2007). Each one of these three main groups was subdivided according to the experimental period into three equal subgroups; (a), (b) and (c). The experimental period was two weeks for subgroups Ia, IIa, and IIIa; four weeks for subgroups Ib, IIb and IIIb and eight weeks for subgroups Ic, IIc and IIIc.

At the end of each experimental period, animals were sacrificed under sodium thiopental anesthesia. Blood samples were obtained from abdominal aorta into heparinized tubes and separated into two portions; one for comet assay and the other one was used for the preparation of plasma to determine Cd and malondialdehyde levels. Livers, lungs and kidneys were extracted, washed with cold PBS (pH 7.4), and then were cut into two parts; One part of tissues was homogenized in PBS, centrifuged and the resultant supernatants were used for determination of Cd and malondialdehyde levels. The other part (2 gm) was quickly minced, suspended in 2 ml of pre-chilled homogenization buffer (20 mM. EDTA, and 10% DMSO), and then homogenized gently. Cells in 1.5 ml homogenate were used for comet assay.

Bone marrow (BM) was obtained through exposing both femurs of each animal, just above the knee, and the bone marrow was aspirated into tubes containing Fetal Bovine Serum (FBS).

Malondialdehyde assay

Determination of malondialdehyde (MDA) level in plasma, BM, liver, lung, and kidney was carried out using the double heating method of Draper and Hadley (1990).

Cadmium analysis

Cd concentration in plasma, BM, liver, lung and kidney samples was determined by atomic absorption spectrophotometer according to the method of Mukherjee et al. (1985).

Comet assay

The comet assay was carried out under alkaline conditions, as described by Singh et al. (1988). Ordinary microscope slides were pretreated with of 0.6 % standard-melting-point agarose, solidified at 4°C. A second layer containing the sample mixed with of 0.5% low-melting-point (LMP) agarose was pipetted onto the lower agarose layer. Ten minutes later, after

solidification on ice, slides were covered with a third layer of 0.5% LMP agarose and allowed to be solidified. Slides were placed for 1 h in ice-cold freshly prepared lysis solution [2.5 M NaCl, 100 mM disodium EDTA, 10 mM Tris–HCl, 1% sodium sarcosinate, pH 10 with 1% Triton X-100 and 10% DMSO were added fresh] to lyse cells and allow DNA unfolding. The slides were then placed on a horizontal gel electrophoresis tank, facing the anode. The unit was filled with fresh electrophoresis buffer (300 mM NaOH, 1 mM disodium EDTA, pH 13.0) and the slides were placed in alkaline buffer for 30 min to allow DNA unwinding prior to electrophoresis for 40 min at 25 V (300 mA). Denaturation and electrophoresis were performed at 4°C. Slides were washed gently three times with neutralization buffer (0.4 M Tris-HCl, pH 7.5) to remove excess alkali and detergents. Each slide was stained with ethidium bromide (20 µg /ml) and covered with a cover slip. Slides were stored at 4°C in dark sealed boxes until being scored.

Evaluation of DNA damage

After application, the cover slips were removed. Slides were coded and examined using a fluorescence microscope equipped with appropriate filters. Images of 200 randomly selected cells from each organ (100 cells from each of two slides) were analyzed from each rat. According to the method described by Kumaravel and Jha (2006), cells were graded by eye into five categories, based on the distance of migration and the perceived proportion of DNA in the tail, and given a value of 0, 1, 2, 3 or 4 ; from undamaged (0) to maximally damaged (4). The percentage of cells with each level of damage was calculated. In this way the total score could range from 0 (all undamaged) to 4 (all maximally damaged).

Statistical analysis

The data were expressed as mean ± SD and analyzed using one-way analysis of variance (ANOVA). Statistical Package for Social Sciences (SPSS 17) was used and significance was set at $P < 0.05$.

Results

Tables (1, 2, and 3) show that there was no significant difference ($p>0.05$) in the score of DNA damage in lymphocytes (fig.1), BM, liver, lung and kidney; Cd concentration and MDA level in plasma, BM, liver, lung and kidney between negative control subgroups (Ia, Ib, Ic) and distilled water subgroups (IIa, IIb, IIc).

Table (4) and fig. (2) show that Cd administration resulted in significant increase in the score of DNA damage in lymphocytes in subgroup IIIa (Cd for 2 weeks) when compared to subgroup Ia (control). This increase continues to appear in subsequent subgroups (Cd for 4 and 8 weeks).

Regarding BM, Cd administration resulted in significant increase in the score of DNA damage in all Cd subgroups when compared to subgroup Ia (control). On the other hand, no significant difference was observed between the three studied Cd subgroups.

In the same table, there was no significant difference in the score of DNA damage in liver, lung and kidney in subgroups IIIa (Cd for 2 weeks) when compared with subgroup Ia (control). However, a significant increase in the score of DNA damage was only observed in the liver in subgroup IIIb (Cd for 4 weeks) when compared with subgroups Ia and IIIa (control and Cd for 2 weeks, respectively). This damage increased with Cd administration for 8 weeks. Also there was significant increase in the score of DNA damage in subgroup IIIc (Cd for 8 weeks) in lung and kidney when compared with subgroup Ia (control).

Table (5) also shows a significant increase in Cd tissue concentration in plasma, BM, liver, lung and kidney in two week treated subgroup (IIIa) when compared with subgroup Ia (control). In addition, this increase continues to appear in the 4 and 8 week treated subgroups (IIIb and IIIc) being more in plasma and liver.

Table (6) shows that MDA level increased in plasma, BM and liver in all Cd subgroups (IIIa, IIIb, IIIc) when compared with subgroup Ia (control). Also, MDA level significantly increased in the liver in subgroup IIIc (Cd for 8 weeks) when compared with subgroup IIIa (Cd for 2 weeks). As regards kidney and lung , a significant increase in MDA level was only observed in subgroups IIIb and IIIc (Cd for 4 & 8 weeks) when compared to subgroup Ia (control group).

Table (1): ANOVA one way statistical analysis of the score of DNA damage in lymphocytes, BM, liver, lung and kidney for subgroups Ia, Ib and Ic (negative control) and subgroups IIa, IIb, and IIc (distilled water).

Groups / Tissues	Ia N=5	Ib N=5	Ic N=5	IIa N=5	IIb N=5	IIc N=5	Fc
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	Mean±SD	Mean±SD	
Lymphocytes	90.5±18.1	93.2±23.4	95.5±18.2	91.6±9.8	89.34±11.6	88.15±19.5	1.3
BM	100.0±19.3	105.2±16.9	98.1±10.5	103.5±31.7	110.4±14.2	97.2±20.1	1.6
Liver	108.4±25.2	113.1±31.6	101.9±18.8	106.1±23.1	110.2±14.2	100.3±20.6	1.5
Lung	85.0±19.4	90.3±12.2	92.6±9.7	88.1±10.5	91.4±7.9	89.5±11.3	1.0
Kidney	126.6±26.5	121.9±16.5	130.4±30.4	120.7±21.3	125.0±18.1	131.5±20.7	1.8

BM: bone marrow. Fc, F calculated. Tabulated F at $p < 0.05 = 2.89$

Table (2): ANOVA one way statistical analysis of Cd concentration in plasma, BM, liver, lung and kidney for subgroups Ia, Ib and Ic (negative control) and subgroups IIa, IIb and IIc (distilled water).

Groups / Tissues	Ia N=5	Ib N=5	Ic N=5	IIa N=5	IIb N=5	IIc N=5	Fc
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	Mean±SD	Mean±SD	
Plasma (µg /L)	2.48± 0.23	2.10±0.60	2.69±0.33	2.41±0.16	2.93±0.74	2.64±0.11	0.72
BM (µg/g wet tissue)	1.2±0.3	1.0±0.11	0.95±0.2	1.13±0.2	1.1±0.1	1.25±0.26	0.83
Liver (µg/g wet tissue)	15.2±3.2	14.5±3.6	15.4±3.1	15.9±3.8	14.8±3.2	15.2±3.3	1.29
Lung (µg/g wet tissue)	1.29±0.07	1.37±0.04	1.47±0.08	1.37±0.07	1.13±0.02	1.22±0.05	0.63
Kidney (µg/g wet tissue)	13.5±2.5	13.6±2.8	14.1±2.7	12.6±2.5	13.7±2.4	13.9±2.4	1.72

Cd: Cadmium, BM: bone marrow. Fc, F calculated. Tabulated F at p < 0.05 = 2.89.

Table (3): ANOVA one way statistical analysis of MDA level in plasma, BM, liver, lung and kidney for subgroups Ia, Ib and Ic (negative control) and subgroups IIa, IIb and IIc (distilled water).

Groups / Tissues	Ia N=5	Ib N=5	Ic N=5	IIa N=5	IIb N=5	IIc N=5	Fc
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	Mean±SD	Mean±SD	
Plasma (µmol/ml)	3.6±0.84	3.1±0.51	3.9±0.29	3.5±0.75	4.1±1.1	3.2±0.32	0.893
BM (µmol/g prot)	0.84±0.11	0.81±0.04	0.78±0.08	0.80±0.01	0.88±0.09	0.75±0.04	0.27
Liver (µmol/g prot)	5.37±0.32	5.09±0.40	5.21±0.34	5.6±0.39	5.31±0.22	5.51±0.27	0.552
Lung (µmol/g prot)	7.43±0.26	7.1±1.3	7.5±2.4	7.3±2.7	6.9±2.5	7.2±3.1	0.311
Kidney (µmol/g prot)	10.05±0.19	9.4±2.1	10.2±1.5	9.8±0.6	10.1±0.8	10.5±1.1	0.192

MDA: malondialdehyde, BM: bone marrow. Fc,F calculated. Tabulated F at p < 0.05 = 2.89,

Table (4): ANOVA one way statistical analysis of the score of DNA damage in lymphocytes , BM, liver, lung and kidney for control subgroup Ia and Cd subgroups IIIa, IIIb, and IIIc (Cd for 2, 4, and 8 weeks).

Groups / Tissues	Ia N=5	IIIa N=6	IIIb N=6	IIIc N=6	LSD
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	
Lymphocytes	90.5±18.1	116.5±21.7 [a]	143.2±13.6 [ab]	171.6±33.5 [abc]	15.3
BM	100.0±19.3	155.2±26.0 [a]	177.8±19.1[a]	184.0±22.1[a]	42.9
Liver	108.4±25.2	112.6±30.5	156.8±20.0 [ab]	206.8±18.4 [abc]	31.6
Lung	85.0±19.4	87.3±9.5	90.1±14.9	108.4±14.3 [a]	22.1
Kidney	126.6±26.5	132.3±20.6	130.5±28.4	163.8±17.2 [a]	35.1

Cd: cadmium, BM: bone marrow, [a]p< 0.05: significant with control, [b] p< 0.05: significant with subgroup IIIa. [c]p< 0.05: significant with subgroup IIIb. LSD, least significant difference. Tabulated F at p < 0.05 = 2.65.

Table (5): ANOVA one way statistical analysis of Cd concentration in plasma, BM, liver, lung and kidney for control subgroup Ia and Cd subgroups IIIa, IIIb, and IIIc (Cd for 2, 4, and 8 weeks).

Groups / Tissue	Ia N=5	IIIa N=6	IIIb N=6	IIIc N=6	LSD
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	
Plasma (µg /L)	2.48± 0.23	7.29±1.4 [a]	16.52±3.7 [ab]	21.66±9.6 [abc]	4.1
BM (µg/g wet tissue)	1.2±0.3	4.01±1.8 [a]	7.2±1.5 [ab]	9.1±2.3 [ab]	2.5
Liver (µg/g wet tissue)	15.2±3.2	45.5±4.6 [a]	78.7±9.6 [ab]	116.5±11.5 [abc]	25.2
Lung (µg/g wet tissue)	1.29±0.5	6.43±0.7 [a]	8.61±3.5 [a]	11.30±1.9 [ab]	3.4
Kidney (µg/g wet tissue)	13.5±2.5	24.9±4.6 [a]	50.4±5.5 [ab]	54.7±7.5 [ab]	6.2

Cd: Cadmium, BM: bone marrow [a]p< 0.05: significant with control, [b] p< 0.05: significant with subgroup IIIa. [c]p< 0.05: significant with subgroup IIIb. LSD, least significant difference. Tabulated F at p < 0.05 = 2.65.

Table (6): ANOVA one way statistical analysis of MDA level in plasma, BM, liver, lung and kidney for control subgroup Ia and Cd subgroups IIIa, IIIb, and IIIc (Cd for 2,4,8weeks).

Groups / Tissue	Ia N=5	IIIa N=6	IIIb N=6	IIIc N=6	LSD
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	
Plasma (µmol/ml)	3.6±0.84	8.32±0.9 [a]	9.51±0.4 [a]	9.97±0.6 [a]	4.53
BM (µmol/g prot)	0.84±0.11	1.9±0.40 [a]	2.1±0.43 [a]	2.5±0.15 [a]	1.0
Liver (µmol/g prot)	5.37±0.32	13.3±2.7 [a]	17.19±4.6 [a]	21.36±9.5 [ab]	4.29
Lung (µmol/g prot)	7.43±0.26	9.24±1.4	10.75±2.6 [a]	12.08±2.9 [a]	2.11
Kidney (µmol/g prot)	10.05±0.19	11.65±0.7	15.64±0.3 [a]	18.53±5.4 [a]	5.2

MDA: malondialdehyde, BM: bone marrow, Cd: cadmium, [a]p< 0.05: significant with control, [b]p< 0.05: significant with subgroup IIIa. LSD, least significant difference. Tabulated F at p < 0.05 = 2.65.

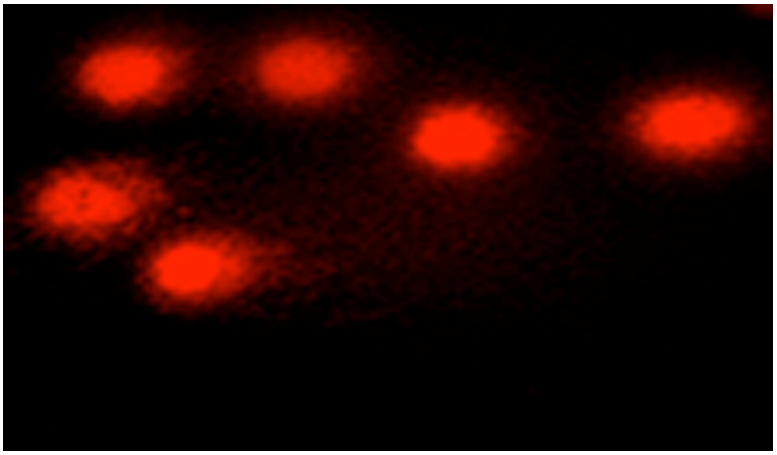

Fig (1): Photomicrographs showing the DNA migration pattern in rat lymphocytes of control group (Ethidium bromide dye ×200).

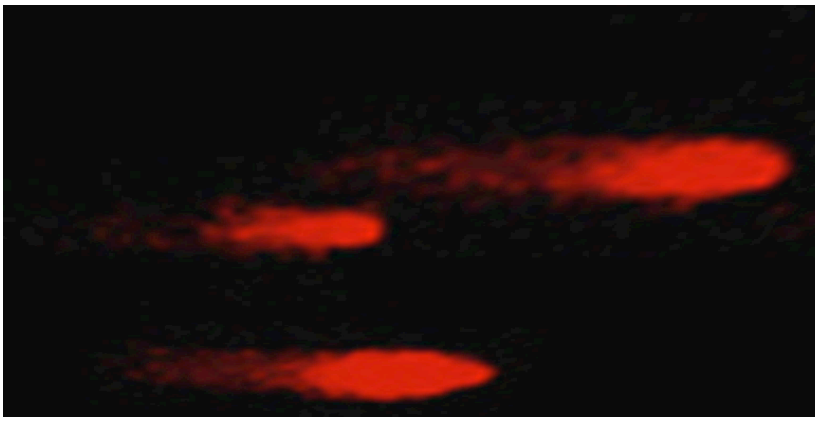

Fig (2): Photomicrographs showing the DNA migration pattern in rat lymphocytes after treatment with $CdCl_2$ (Ethidium bromide dye ×200).

Discussion

Cadmium (Cd) is one of the most toxic environmental pollutants affecting the DNA. It is often referred to as the metal of the 20th century. Cadmium contamination is very persistent. It enters into the food chain and accumulates over time in soft tissues such as liver and kidney (Schwerdtle et al., 2010).

Genotoxicity of Cd is well established and was mainly attributed to its ability to induce oxidative DNA damage and to interfere with the DNA repair process (Hartwig , 2010).

This experimental study was carried out to determine the response of different rat organs to the genotoxic effect of Cd. This may help in early detection of DNA damage and allow early interference before occurrence of malignancy.

In vivo comet assay used in this study is a very sensitive method to detect single DNA strand break in mammalian cells exposed to chemicals. In addition, this assay is simple, requires a small number of cells, can evaluate DNA damage in non-proliferating cells, and since the DNA migration data are obtained on a cell by cell basis, it is possible to measure the intracellular distribution of DNA damage (Betancourt et al., 2005).

The present study revealed heterogeneous responses of the studied organs to the genotoxic effect of Cd in the form of variation in the score of DNA damage in lymphocytes, bone marrow (BM), liver, lung and kidney with increased Cd concentration in these tissues. Also, there was an increase in MDA level suggesting oxidative stress as indirect cause of Cd-induced DNA damage. The lymphocytes and BM were early affected whereas the liver showed the highest genetic damage with longer administration period.

DNA damage observed in this study coincides with the results of previous studies on human lymphocytes that demonstrated an increase in DNA migration pattern and single strand breaks at different non cytotoxic concentrations of Cd (Mouron et al., 2001; Depault et al., 2006). Moreover, Celik et al. (2009) found that Cd chloride administration induced genetic damage in both BM and peripheral blood with increase in DNA single strand breaks indicating an interaction between DNA and Cd chloride. Recently, the study of Abrahim et al. (2011) revealed the presence of various chromosomal aberrations such as chromosome breaks, chromatid breaks, acentric fragment, dicenetic chromosome and tetraploidy in Cd-exposed workers .

In the current study, BM was highly susceptible to Cd genotoxicity as it showed an early induction of DNA damage following two weeks of exposure. This sensitivity to Cd could be due to the high level of proliferation of BM cells as this metal interferes with a number of enzymes involved in DNA repair and replication (Celik et al., 2009).

Regarding lymphocytes, they showed an increase in the score of DNA damage with Cd administration for two weeks. This increase continued to appear in subsequent subgroups IIIb and IIIc (Cd for 4 and 8 weeks, respectively). Early affection of lymphocytes could be attributed to the initial transport of Cd in plasma following its absorption through binding to albumin and other large proteins (Klaassen et al., 2009).This reflects high levels of metal – tissue interaction and could also explain the accumulation of damage throughout the study period.

Liver, lung, and kidney were not affected early by Cd as they showed no significant difference in the score of DNA damage in subgroup III a (Cd for 2 weeks). By increasing the time of exposure to 4 weeks (IIIb), only the liver was affected whereas the extended exposure to Cd (8 weeks) resulted in a remarkable increase in DNA damage in liver and kidney, being more marked in the liver.

The early negative response of these organs in the present study could be due to induction of metallothioneins (MTs) which are cysteine- rich small metal-binding proteins present in mammalian tissues. In fact, Cd exposure activates transcription of MTs gene, resulting in marked increase in the cellular content of this high affinity cadmium-binding protein (Filipic et al., 2006). Metallothioneins function in Cd detoxication primarily through the high binding affinity of the metal to MTs forming Cd-MT complex. Cadmium - MT complex is mainly formed in the liver to sequestrate Cd in the cytosol, with concomitant reduction of the amount of Cd available for other critical organelles. Other proposed functions of MTs, such as maintaining essential metal (zinc) homeostasis, scavenging reactive oxygen species, regulating gene expression and tissue regeneration could all contribute to MTs protection against Cd toxicity (Cherian and Kang, 2006).

Cadmium exposure for longer duration might saturate the capacity for detoxification .Thus Cd-MT complex is released into the blood stream from damaged hepatocytes. Then it is filtered by the kidney and taken up into proximal tubule cells, where it is degraded, releasing locally high levels of “free” Cd to produce tubular injury (Klaassen et al., 2009). This could explain the high level of DNA damage observed in the liver and kidney with longer exposure period.

The results of the present study are in agreement with those of Valverde et al. (2001) who found increased DNA fragmentation and MDA level in lung and liver following Cd inhalation. Recently, Schwerdtle et al. (2010) demonstrated that Cd increased the baseline level of oxidative DNA damage with inhibition of the nucleotide excision repair in cultured human cells in a dose-dependent manner.

The accumulation of DNA damage, observed in this study could be attributed to inhibition of the polymerization or ligation step in excision repair. One reason for this inhibition could be the competition with zinc ions which are essential in DNA polymerases and other DNA-binding proteins. Besides a direct interaction with repair enzymes, Cd ions might also interfere with calcium-regulated processes involved in the regulation of DNA replication and repair (Giaginis et al., 2006).

Furthermore, Cd, unlike other heavy metals, is unable to generate free radicals by itself, however, reports have indicated that superoxide, hydroxyl, and nitric oxide radicals could be generated indirectly. In addition, Watanabe et al. (2003) have shown that Cd-induced generation of the non-radical hydrogen peroxide became a significant source of free radicals via the Fenton reaction. Cadmium could replace iron and copper from a number of cytoplasmic and membrane proteins like ferritin, which in turn would release and increase the concentration of unbound iron or copper ions. These free ions participate in causing oxidative stress via the Fenton reactions (Waisberg et al., 2003). A subsequent study of Watjen and Beyersmann (2004) provided evidence in support of the proposed mechanism. They showed that copper and iron ions displaced by Cd, were able to catalyze the breakdown of hydrogen peroxide via the Fenton reaction.

In addition, Casalino et al. (2002) proposed that Cd binds to the imidazole group of the His-74 in superoxide dismutase (SOD) which is vital for the breakdown of hydrogen peroxide, thus causing its toxic effects. Cadmium could also substitute for manganese in the liver mitochondrial Mn-SOD causing suppression of its activity (Flora et al., 2008).

Finally, Hartwig (2010) reported that with respect to DNA repair processes, Cd has been shown to disturb nucleotide excision repair, base excision repair

and mismatch repair; consequences are increased susceptibility towards other DNA-damaging agents. Furthermore, cadmium induces cell proliferation, inactivates negative growth stimuli, such as the tumor suppressor protein p53, and provokes resistance towards apoptosis. Particularly the combination of these multiple mechanisms may give rise to a high degree of genomic instability in cadmium-adapted cells, relevant not only for tumor initiation, but also for later steps in tumor development.

In conclusion, the present study revealed that Cd-induced genetic damage in BM, lymphocytes, liver, lung and kidney. Lymphocyte and BM were early affected by Cd genotoxic effect. The liver showed the highest genetic damage with longer exposure period. As there is strong evidence that genetic damage can be the initial step of the malignant process, lymphocyte, and BM can be used as early determinants of cadmium-induced genetic damage to avoid its carcinogenic potential. Considering these issues, workers exposed to Cd should be withdrawn from exposure when they have lymphocyte genetic damage. Also, health educational should be implemented to highlight the dangerous impact of Cd on health. Moreover, persistence of this metal in the environment requires a long-term move toward minimizing human exposure through environmental management and preservation of lower cadmium levels wherever possible. Strictly enforced limits of Cd in food are also necessary for overall decrease in exposure. Also, further studies are needed to detect genotoxicity of Cd in other organs.

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الملخص العربي

الاستجابات المختلفة لبعض أعضاء جسم الفئران البيضاء البالغة للتأثير السمي الجيني للكادميوم

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يعد الكادميوم أحد الملوثات البيئية الأكثر سمية و الذي يؤثر على الحامض النووي (د ن أ). يتداخل الكادميوم مع عملية إصلاح الحامض النووي (دن أ) ويحدث شدة الأكسدة مما يؤدي إلى تلف الحامض النووي (دن أ). وحيث أن أحدث بيانات السرطان الجزيئية الجينية و الوراثة الخلوية قدمت أدلة قوية على أن الضرر الجيني يمكن أن يكون الخطوة الأولى في عملية السرطان الخبيث فإنه من المهم جدا الكشف المبكر عن التأثير الضار للكادميوم على الحامض النووي (د ن أ) في مختلف الأعضاء لتجنب احتمالية إحداثه للسرطان. وبالتالي فقد أجريت هذه الدراسة لتحديد استجابة بعض أعضاء الفئران المختلفة للتأثير السمي الجيني للكادميوم. أجريت هذه الدراسة على 48 فأر أبيض ذكر بالغ تم تقسيمهم إلى ثلاث مجموعات رئيسية: المجموعة الأولى (الضابطة السالبة) وتألفت من 15 فأرا، والمجموعة الثانية (الضابطة الموجبة) تألفت من 15 فأرا وحقنت 1 مل من الماء المقطر عن طريق الفم والمجموعة الثالثة (مجموعة الكادميوم) تألفت من 18 فأرا وحقنت كلوريد الكادميوم بجرعة قدرها 5 ملجم/كجم من وزن الجسم عن طريق الفم. وتم تقسيم كل واحدة من هذه المجموعات الثلاث الرئيسية، وفقا للفترة التجريبية، لثلاث مجموعات فرعية؛ (أ) و(ب) و(ج). وكانت الفترة التجريبية لمدة أسبوعين للمجموعات الفرعية (أ)، وأربع أسابيع للمجموعات الفرعية (ب)، و ثمان أسابيع للمجموعات الفرعية (ج). وقد أجري تحديد للضرر المحتمل بالحامض النووي (د ن أ) باستخدام فحص كومت في الخلايا اللمفاوية و نخاع العظام و الرئتين والكبد و الكلى جنبا إلى جنب مع مستويات المالوندياالهيد والكادميوم. و أظهرت النتائج وجود استجابة غير متجانسة في الأعضاء محل الدراسة للتأثير السمي الجيني للكادميوم. وقد تأثرت الخلايا اللمفاوية و نخاع العظام مبكرا بالسمية الجينية للكادميوم. و أظهر الكبد أعلى ضرر وراثي مع التعرض لفترة أطول. وتشير هذه النتائج إلى أن الخلايا اللمفاوية و نخاع العظام هي الأكثر حساسية للسمية الجينية للكادميوم أكثر من الرئتين و الكبد و الكلى و لذلك يمكن استخدامهما كمحددات مبكرة للضرر الجيني الذي يسببه الكادميوم. وبالنظر في هذه المسائل ينبغي أن يتم تنفيذ برامج تثقيفية صحية لتسليط الضوء على التأثير الخطير للكادميوم على الصحة و علاوة على ذلك فإن ثبات هذا المعدن في البيئة يتطلب التحرك على المدى الطويل للتقليل من التعرض البشري من خلال التدابير البيئية والحفاظ على مستويات أقل من الكادميوم بقدر الإمكان. أيضا هناك حاجة لإجراء المزيد من الدراسات للكشف عن السمية الجينية للكادميوم في الأعضاء الأخرى.

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