BENHA VETERINARY MEDICAL JOURNAL, VOL. 23, NO. 2, DEC 2012: 172-179

CYTOTOGENETIC STUDIES ON THE EFFECTS OF ACUTE EXPOSURE TO FORMALDEHYDE ON MICE

Fatma E. El-gendy[a], Bakry, H. H.[b], Hemeda, SH.A.[c], Abd El-Aal, S.A.[d]

[a] Department of Animal Wealth, [b] Department of Forensic Medicine and Toxicology, Faculty of Veterinary Medicine, Benha University, [c] Department of Animal Wealth, Faculty of Veterinary Medicine, Alexandria University, [d] Department of Animal Hygiene, Faculty of Veterinary Medicine, Benha University, Egypt.

A B S T R A C T

Although formaldehyde is widely used in consumer goods to protect the products from spoilage by microbial contaminations, research has shown that it has various side effects. Therefore, the aim of this study was to investigate the cytogenetic effects of formaldehyde solution in mice and to evaluate the possible protective effect of vitamin C against the cytogenetic effects of formaldehyde. Thirty-six Swiss albino mice were exposed to various concentrations of formaldehyde. The animals were divided in to six groups (two groups received 1/10 LD_{50}, 1/5 LD_{50} of formaldehyde other two groups received vitamin C in addition to 1/10 LD_{50}, 1/5 LD_{50} of formaldehyde and two control groups). Animals were sampled at two different times (48 and 72 hrs). The results revealed that the formaldehyde increased the number of structural and numerical chromosomal aberrations, and decrease the rate of cell division (mitotic index) at either 48 or 72 hrs. There was no significance difference between the two doses of formaldehyde observed. Moreover, the use of vitamin C gave promising results against formaldehyde toxicity as it significantly decreased the frequency of chromosomal aberrations and improving rate of cell division.

KEY WORDS: Chromosomal aberrations, Formaldehyde, Mice, Mitotic index, Vitamin C.

(BVMJ 23(2): 172-179, 2012**)**

1. INTRODUCTION

Formaldehyde occurs as a natural constituent in a variety of raw fruits (pear, apples, tomatoes and white radish in amount 3.7 to 60 ppm) and raw vegetables (cabbage, carrot, green onion, and spinach in amount varying from 3.3 to 26.3 ppm) and also has been found in meat, milk, and fish. The quantity of formaldehyde is ingested via food difficult to be estimated but may range from 1.5 to 14 mg/person/ day [21].

The histopathological, cytotoxic and cell proliferation effects of formaldehyde especially on respiratory epithelium were reported by Monticello et al. [16]. Formaldehyde was demonstrated to induce mutation and genotoxic effects on a variety of experimental systems including rodent in vivo and human cells in vitro [11]. Human are exposed to formaldehyde from both direct environment sources as well as from the metabolism of xenobiotics. Everyday exposures to formaldehyde include building material (e.g. paint, plywood, cosmetics, cigarette smoke, disinfectant, pesticide and even various fruits [5]. Formaldehyde induce DNA-protein crosslink (DPCs) and chromosomal changes, expressed as chromosomal aberration (CA), sister chromatid exchange (SCEs) and micronuclei (MN). These alterations have been demonstrated by a large number of studies in vitro, in exposed animals, and to varying degrees in

the circulating lymphocytes of exposed people [2]. Formaldehyde being a very reactive compound can react with different macromolecules such as proteins and nucleic acid or with low molecular weight substance as amino acid [4]. Vitamin C (L-ascorbic acid) can promote the removal of oxidative DNA damage from the DNA and/or nucleotide pool, through up-regulation of repair enzymes [6]. Furthermore, vitamin C is an essential dietary nutrient required as a co-factor for many enzymes and a very efficient antioxidant and protecting cells against free radical-mediated damage [19].

The present study was therefore carried out to investigate the cytotoxic effects of the acute exposure to formaldehyde solution on mice and the possible protective effects of vitamin C.

2. MATERIALS AND METHODS

Formaldehyde was obtained from EL-Nasr Company as commercial preparation of formaldehyde 37%. Vitamin C obtained from Sigma pharmaceutical Company, Mubarak industrial Zone, Quisna, Egypt, as a white crystalline powder soluble in water.

Thirty-six albino mice (*Mus musculus*) were used in this study. They were 8-10 weeks old and weighted (20-25g). Mice were obtained from Lab Animal Care Centre, Faculty of Veterinary Medicine (Benha University). Pelleted ration and water were offered *ad-libitum*. Mice were divided into six experimental groups of six animals in each group; the first group was kept as control (negative control). The second group was injected intra-peritoneally with 65.2 mg/kg B.wt (Therapeutic dose of human) with vitamin C (positive control). The third group was injected intra-peritoneally with formaldehyde 20 mg /kg B.wt This dose was $1/10LD_{50}$ of formaldehyde according to Deltour et al. [8]. The fourth group was injected intra-peritoneally with formaldehyde (20 mg/kg B.wt) in addition to vitamin C (65.2 mg/kg B.wt). The fifth group was injected with formaldehyde (40 mg/kg B.wt). This dose was $1/5LD_{50}$ of formaldehyde. The sixth group was injected intra-peritoneally with formaldehyde 40 mg/kg.B.wt.in addition to 65.2 mg/kg B.wt. vitamin C. After 24 hrs the same doses were injected to each group, half of the animals half (n=3 of each group) of each group were taken for cytogenetic studies after 48 hrs from last injection. The other half were taken after 72 hrs from the first injection.

All animals were injected intra peritoneally with 4 mg/kg B.wt of aqueous solution of colchicines two hours before the time of the sacrifice. Bone marrow cells preparations were prepared for chromosome aberrations [3] with certain modifications [1]. One hundred metaphases per animal were analyzed in order to determine the frequencies of chromosomal aberration. The mitotic indices of 3000 cells per group were also analyzed. The differences in chromosomal abnormalities between different treatments were statistically tested using one way analysis of variance. The differences in mitotic indices between treatments were statistically tested using Chi-square for independence by means of 2×2 contingency table. Statistical analyses were done using SPSS software packages.

Table 1 Means of aberrant cells at 48 hrs.

Group (48hrs)	Abbreviation	Aberrant cells (means ±SE)
I	Control	26.33 ± 2.72^{b}
II	65.2 mg/kg B.wt. vitamin C	23.67 ± 2.2^{b}
III	1/10 LD_{50} of formaldehyde	51.00 ± 0.57^{a}
IV	1/10 LD_{50} of formaldehyde + 65.2 mg/kg B.wt. vitamin C	28.00 ± 2.64^{b}
V	1/5 LD_{50} of formaldehyde	55.67 ± 2.08^{a}
VI	1/5 LD_{50} of formaldehyde + 65.2 mg/kg B.wt .vitamin C	28.33 ± 4.41^{b}

3. RESULTS AND DISCUSSION

3.1. *Chromosomal aberrations*

In 48 hours group:

The results showed in table (1) revealed that there was a significant difference in the number of aberrant cells between control groups (26.33±2.72 and 23.67±2.20) and groups treated with 1/10 LD_{50} and 1/5 LD_{50} of formaldehyde (51 ± 0.57 and 55.67±2.08). Vitamin C induced significant reduction in number of aberrant cells in both 1/10 LD_{50} and 1/5 LD_{50} of formaldehyde (28.00±2.64 and 28.33± 4.41). Normal metaphase chromosome of mice bone marrow cells (fig. 1) and different types of aberrations of treated groups (fig. 2) are presented in table (2). The results showed that the highest type of chromosomal aberration at 1/10 LD_{50} of formaldehyde was fragment (11.0±2.1). While the highest types of chromosomal aberrations at 1/5 LD_{50} are fragment and chromatid deletion (12.0 ± 2.1 and 11.0±2.1).On other hand, the lowest types of chromosomal aberrations at different types of treatments are hypoploidy (2.7±0.7, 1.3±0.3, 3.3±0.3, 2.0±0.6 respectively) and chromosomal gap (1.3±0.9, 1.3±0.9, 2.3±1.2, 1.0±0.6 respectively).

Table 2 Different types of chromosomal aberrations at 48 hrs.

Group 48 hrs.	Types aberrations								
	Hhypoploidy	G-gap	Fragment	Chromatid deletion	Chromatid break	Centric fusion	Centromeric attenuation	stickiness	Ring
I	00.0±0.0^{d}	11.3±0.9^{a}	0.0±0.6^{b}	5.0±0.6^{b,c}	3.3±0.7^{b}	2.0±0.6^{c}	4.0±0.6^{a}	3.7±0.9 ^{b,c}	33.0±1.0^{a}
II	00.0±0.0^{d}	00.0±0.0^{a}	4.0±1.5^{b}	6.3±1.7^{b,c}	0.3±1.5^{a,b}	2.0±1.2^{c}	1.0±0.6^{b}	2.0±1.2^{c}	22.3±1.2^{a}
III	22.7±0.7^{a,b}	11.3±0.9^{a}	11.0±2.1^{a}	7.7±1.5^{a,b}	4.3±1.9^{a,b}	0.7±0.3^{a,b}	3.3±0.7 ^{a}	6.3±0.7 ^{a,b}	11.7±0.3^{a}
IV	11.3±0.3^{c}	11.3±0.9^{a}	6.0±0.6^{b}	3.0±0.6^{c}	3.0±0.6^{b}	3.7±0.7^{b,c}	2.3±0.3 ^{a,b}	4.3±0.7^{a,b,c}	11.0±0.6^{a}
V	33.3±0.3$^{a.}$	22.3±1.2^{a}	12.0±2.1^{a}	11.0±2.1^{a}	7.7±1.5^{a}	6.7±0.9 ^{a}	3.7±0.67^{a}	7.00±1^{a}	22.7±0.7^{a}
VI	22.0±0.6^{b,c}	11.0±0.6^{a}	7.7±0.3^{a,b}	7.7±0.3 ^{a,b}	2.0±0.6^{b}	0.0±0.6 ^{b,c}	2.3±0.3 ^{a,b}	5.0±0.6 ^{a,b}	11.7±0.3^{a}

Means having different letters are significantly different at the level of $P < 0.05$

In 72 hours group:

The results in table (3) demonstrated that there was a significant difference in the number of aberrant cells between control groups (negative and vit. C) (31.33±0.30 and 29.67±0.88) and groups treated with 1/10 LD_{50} and 1/5 LD_{50} of formaldehyde (51.33±1.45 and 53±1.15). Vit. C induced significant reduction in number of aberrant cells in both 1/10 LD_{50} and 1/5 LD_{50} of formaldehyde (27.00±1.15 and 27.67± 1.76).

Table 3 Means of aberrant cells at 72 hrs.

Groups (48hrs)	Aberrant cells (means ±SE)
I	31.33±0.30^{b}
II	29.67±0.88^{b,c}
III	51.33±1.45^{a}
IV	27.00±1.15^{c}
V	53.00±1.15^{a}
VI	27.67±1.76^{b,c}

Normal metaphase chromosome of mice bone marrow cells (fig. 1) and different types of aberrations of treated groups (fig. 2) are presented in table (4), illustrated that the highest types of chromosomal aberrations in group treated with 1/10 LD_{50} of formaldehyde were chromatid deletion and fragment (10.00±0.00 and 8.70± 0.70). On other hand, the lowest type of chromosomal aberration in 1/10 LD_{50} of formaldehyde were gap chromosome (2.00±0.60). While the highest types of chromosomal aberrations at 1/5 LD_{50} of formaldehyde were deletion and fragment (15.7±2.3 and 11.00±2.6). On other hand, the lowest types of chromosomal aberrations in 1/5 LD_{50} of formaldehyde treated group were gap and ring chromosome both of them equal (2.00±0.6).

Table 4 Difference types of chromosomal aberrations at 72 hrs.

Groups 72 hrs.	Types aberrations								
	Hypoploidy	Gap	Fragment	Chromatid deletion	Chromatid break	Centric fusion	Centromeric attenuation	stickness	RRing
I	0.0±0.00 ^{d}	22.0±0.6^{a}	4.7 ±0.9 ^{b}	7.33±0.33^{b,c}	3.67±0.33^{a,b}	3.7±0.7^{a,b}	3.7±0.9^{b}	33.0±0.6^{b,c}	33.3±0.3^{a}
II	0.00±.0^{d}	00.3±0.3 ^{a}	8.3±0.7 ^{a,b}	6.33±1.2^{b,c}	4.67±1.45^{a,b}	2.0±0.6^{b}	3.3±0.7^{b}	22.0±1.15^{c}	00.3±0.3^{b}
III	33.3±0.6^{b}	22.0±0.6^{a}	8.7±0.7^{a,b}	10.00±0.00^{b}	7.00±1.15^{a}	5.0±1.2^{a,,b}	4.0±0.6^{b}	77.3±0.33^{a}	22.3±0.7^{a}
IV	11.7±0.6 ^{c}	11.3±0.3 ^{a}	5.3±0.3^{b}	5.67±0.67^{c}	3.67±0.67^{a,b}	2.7±0.3^{b,c}	2.3±0.3^{b}	44.0±0.6^{b,c}	00.3±0.3^{b}
V	44.7±1.5^{a}	22.0±0.6^{a}	11.0±2.6^{a}	15.7±2.3 ^{a}	6.67±1.67^{a,b}	5.7±0.7^{a}	6.0±0.6^{a}	55.0±0.00^{b}	22.0±0.6^{a}
VI	11.9±0.6 ^{c}	11.0±0.6^{a}	6.0±1.2^{b}	6.67±1.20^{b,c}	3.0±1.0^{b}	4.0±1.7^{a,b}	2.7±0.3^{b}	33.3±0.9^{b,c}	00.0±0.0^{b}

Means having different letters are significantly different at the level of $P < 0.05$

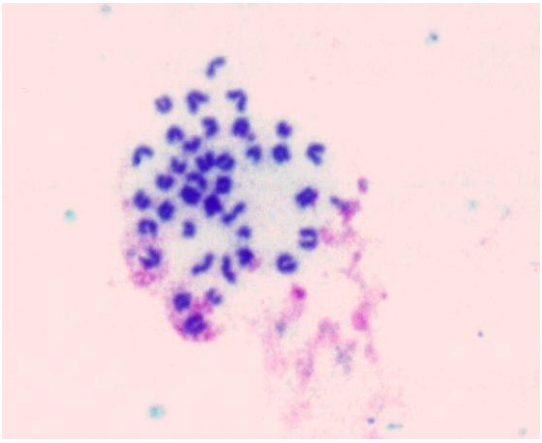

Fig. **1** Normal metaphase chromosome of mice bone marrow cells.

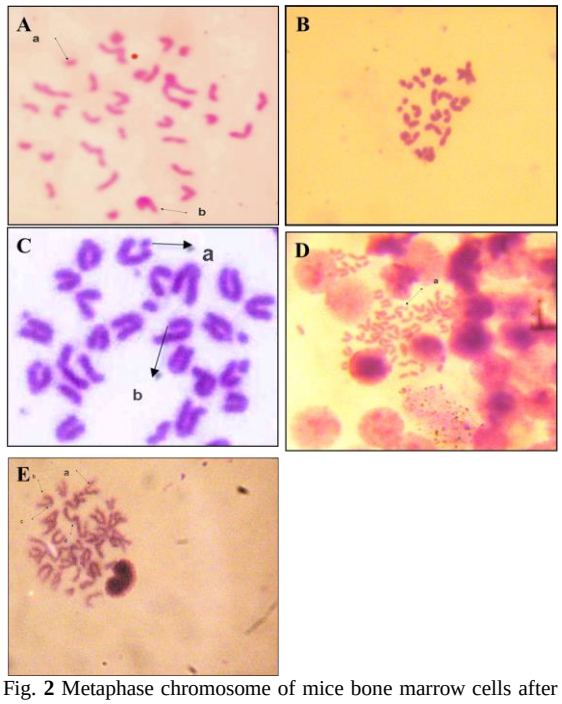

Fig. **2** Metaphase chromosome of mice bone marrow cells after formaldehyde treatment. **A**: fragment (a) and deletion (b). **B:** hypoploidy. **C:** break (a) and deletion (b). **D:** chromatid break. **E:** break (a), deletion (b), gap (c), and stickiness (d) chromosomal aberrations.

No significant differences were observed between 1/10 LD_{50} and 1/5 LD_{50} of formaldehyde at 48 and 72 hrs. The results of the acute exposure to formaldehyde indicated that the acute treatment with formaldehyde for 48 and 72 hrs caused a significant increase in the occurrence of chromosomal aberrations. The results also illustrated that vitamin C showed a protective effect and decreased the occurrence of chromosomal aberrations. These findings agree with those of Levy et al. [15], in which they reported that formaldehyde reacts with a number of cellular components such as nucleic acids and nucleoproteins giving rise to the formation of methylol and methylene derivatives. From this finding, it may be inferred that methylene bridges linking purines would occur between complementary chains of DNA actually causing interstrand cross linking with resultant induction of inactivating DNA alterations that inhibit DNA replication across altered sites unless the lesions are eliminated by repair. Inducers of inactivating DNA alterations also induce chromosome break, fragmentation and other type of chromosomal aberrations. These results also agree with the results obtained by Synder and Houten [20] who found that the formaldehyde treated synthetic templated incorporated G (guanine) and to lesser extent to C (cytosine) in adenosine (A) – thymin (T) containing polymers. Consequently, they concluded that formaldehyde does form covalent DNA modifications which are

likely lead to base mispairing so the formaldehyde was a mutagenic in prokaryotic and eukaryotic systems.

Similar results were obtained by Rithidech et al. [18] who found that the chromosomal aberrations (break, fragment, ring and others) in mouse (spleen lymphocytes) after subchronic exposure to formaldehyde. The results indicated that the formaldehyde causing hypoploidy as found by Kitaeva et al. [14], in which they found increase in cells with chromosomal aberrations from 0.7% (negative control) to 2.4% (0.42 ppm) and 4.0% (1.25 ppm) with regard to numerical chromosome aberrations, also they found increase in cells with hypoploidies from 7.0% (negative control) to 10.9% (0.42 ppm) and 13.6% (1.25 ppm). The observed a significant induction of numerical aberrations in some formaldehyde treated groups might be attributed to the ability of formaldehyde to react with proteins and interfere with proper functions of the microtubulin and microtubule dynamics in mitosis with resultant chromosome malsegregation and chromosome loss. These findings were in agreement with Zimmermann and Mohr [22]. Also Cosma and Marchok [7] detected both DNA-protein crosslink (DPC) and single-strand breaks (SSB) as a result of formaldehyde treatment, in rat tracheal epithelial cells. Conversely, a number of studies have failed to demonstrate the genotoxic potential of formaldehyde as Epstein and Shafner [9]; they performed two tests with intra-peritoneal administration of formaldehyde up to 40 mg/kg B.wt.in mice that exhibited no genotoxic effects. From the present study it was found that vitamin C had protective effect against formaldehyde genotoxicity which agreed with Reithel and West [18] who found that the vitamin C inhibits the addition of formaldehyde to the 6-amino group of L-lysine, and the possibility that the reducing effect on formaldehyde that might exert, so vitamin C decrease genotoxicity caused by formaldehyde.

3.2**.** *Mitotic index*:

Chi square values of the two control groups and treated groups showed that there were significant differences between the control and the treated groups after 72hrs treatment (Table 5&6). This indicate that the formaldehyde (1/10 LD_{50} and 1/5 LD_{50}) causing significant decrease in mitotic indices. On the other hand the addition of vitamin C significantly increased the mitotic indices.

Table 5 Mitotic indices (M.I) in animals received formaldehyde and /or vitamin C after 72 hrs.

Group	No. of divided cells	No. of non-divided cells	M.I
I	170	2830	5.60
II	180	2820	6.00
III	88	2912	2.93
IV	170	2830	5.60
V	76	2924	2.53
VI	170	2830	6.00

Table 6 Chi square analysis of mitotic indices after 72 hrs.

Groups	I	II	III	IV	V
II	0.246				
III	26.6**	32.3**			
IV	0.003	0.25	26.54**		
V	36.7**	43.3**	0.76	36.63**	
VI	0.000	0.21	26.6**	0.000	36.7**

*= Means having different letters are significantly different at the level of $P < 0.05$. **= Means having different letters are significantly different at the level of $P < 0.01$

These results are in corresponding to the results of Kitaeva et al. [14], who found that the mitotic indices significantly decreased in peripheral blood lymphocytes in human after 72 hrs of formaldehyde treatment. Also, the results agreed with findings of Inas et al. [12] who found that the acute and chronic formaldehyde treatment caused a significant inhibition in mitotic activities that led to decrease in mitotic index Hoda [10] found also that ascorbic acid at concentration of 100 ppm

activates the cells to enter mitosis and induces a high mitotic activity.

4. REFERENCES

1. Adler, I.D. 1984. Cytogenetic tests in mammals. In: Mutagenecity Testing: a practical approach. Venitt, S. and Parry, J.M. (Eds.) IRL Press, Oxford. Pp.275-306.
2. ATSDR. 1999. Agency for Toxic Substances and Disease Registry Toxicological Profile for Formaldehyde. Atlanta, GA: ATSDR.
3. Brusick, D. 1980. Principles of genetic toxicology, New York and London. Pp. 33-34
4. Cheng, G., Sturla, S.J., Jalas, J.R., Shi, Y.L., Villalta, P., Wang, M.Y., Hecht, S. S. 2003. Reactions of Formaldehyde Plus Acetaldehyde with Deoxyguanosine and DNA: Formation of Cyclic Deoxyguanosine Adducts and Formaldehyde Cross-Links. *Chem. Res. Toxicol.* **16**: 145-152.
5. Conaway, C.C., Whysner, J., Verna, L.K. and Williams, G.M. 1996. Formaldehyde mechanistic data, endogenous protection from DNA adduct formation**.** *Pharmcol. Ther.* **71**: 29-55
6. Cook, M.S., Evans, M.D., Podmore, I.D., Podmore, K.E., Mistry, N., Mistry P., Hickenbotham, PT., Hussieni, A., Griffiths, H.R. and Lunec, J. 1998. Novel repair action of vitamin C upon *in vivo* oxidative DNA damage. *FEBS Lett*. **363**: 363-367.
7. Cosma, G.N. and Marchok, A.C. 2002. Benzo[*a*]pyrene- and formaldehyde-induced DNA damage and repair in rat tracheal epithelial cells. Oak Ridge Graduate School of Biomedical Sciences, University of Tennessee, Oak Ridge National Laboratory, Oak Ridge, TN 37831.
8. Deltour, L., Foglio, M.H. and Duster, G. 1999. Metabolic deficiencies in alcohol dehydrogenase Adh1, Adh3, and Adh4 null mutant mice. Overlapping roles of Adh1 and Adh4 in ethanol clearance and metabolism of retinol to retinoic acid. *J. Biol. Chem.* **274**: 16796-16801.
9. Epstein, S.S. and Shafner, H. 1968. Chemical mutagens in the human environment. *Nature*. **219**: 385-387
10. Hoda H. Barakat 2003. Interactive Effects of Salinity and Certain Vitamins on Gene Expression and Cell Division. *Int. J. Agric. Biol*. **5:** 219–225**.**
11. IARC 1995. IARC Monographs on the Evaluation of Carcinogenic Risks to Humans. *Wood Dust and Formaldehyde* **62**: 217-362.
12. Inas, H. El-Mehallawi, Eman, M. Soliman, Zowail, M.E., Abeer, A.Al-Menshawy and Yassin, A.A. 1999. Potential mutagenecity of formaldehyde added to milk .Faculty of medicine Cairo Univ. *Int. J. Legal Med.* **7**: 91-107.
13. Kitaeva, L.V., Kitaev, E.M. and Pimenova, M.N. 1990. The cytopathic and cytogenetic sequelae of chronic inhalational exposure to formaldehyde on female germ cells and bone marrow cells in rats. *Tsitologiia* **32**:1212-1216.
14. Kitaeva, L.V., Mikheeva, E.A., Shelomova, L.F. and Schwartzman, P. 1996. Genotoxic effect of formaldehyde in somatic human cells in vivo. *Genetika*. **32**:1287-1290.
15. Levy, S., Nocentini, S. and Billadoson, C. 1983. Induction of cytogenatic effects in human fibroblast cultures after exposure to formaldehyde or x-ray. *Mutat. Res*. **119**: 309-317.
16. Monticello, T.M., Morgan, K.T. and Everett, J.I. 1989. Effects of formaldehyde gas on the respiratory tract of Rhesus monkeys. *Am. J. Pathol*. **134**: 515-527.
17. Rithidech K., Au, W.W., and Ramanujam, V.M.S. 1987. Induction of chromosome aberrations in lymphocytes of mice after subchronic exposure to formaldehyde. *Mutation Res.* **188**: 135-140.
18. Reithel, F. J. and West, E. S. 1948. Studies on the reactions between formaldehyde and enediols. *J. Am. Chem. Soc.* **70**: 898-900
19. Sanchez-Moreno C., Paniague M., Madrid A. and Martin A. 2003. Protective effect of vitamin C against the ethanol mediated toxic effects on human brain glial cells. *J. Nut. Bioch*. **14**: 606-613
20. Synder, R. and houten, B.V. 1986. Genotoxicity of formaldehyde and an

evaluation of its effect on the DNA repair process in human diploid fibroblasts. *Mutat, Res.* **165**: 21-36.

21. WHO 1987. Air quality guidelines for Europe. *WHO. Reg. Public. Eur.* Ser. No 23 World Health Organization Regional Office for Europe Copehagen. Pp. 426.
22. Zimmermann, B.M. and Mohr, A. 1992. Formaldehyde glyoxal, urethane, methyl carbamate, 2, 3 butanedione induce chromosome lose in *saccharomyces cerevisiae*. *Mutat. Res.* **270**: 151-166

مجلة بنها للعلوم الطبية البيطرية
BENHA UNIVERSITY
FACULTY OF VETERINARY MEDICINE

دراسات وراثية خلوية للتأثير الحاد للفورمالدهيد على الفئران

فاطمة عيد الجندى[1]، حاتم حسين بكرى[2]، شعبان حميدة[3]، سمير عبد اللطيف[4]

[1]قسم الرعاية وتنمية الثروة الحيوانية- كلية الطب البيطري – جامعة بنها، [2] قسم الطب الشرعى والسموم –كلية الطب البيطرى – جامعة بنها،

[3]قسم الرعاية وتنمية الثروة الحيوانية-كلية الطب البيطرى– جامعة الأسكندرية، [4] قسم الصحة وسلوكيات ورعاية الحيوان -جامعة بنها

الملخص العربي

بالرغم من أن الفورمالدهيد يتم اضافته الى المنتجات الغذائية لحمايتها من التلف إلا أن الأبحاث الحديثة أكدت أن للفورمالدهيد آثار جانبية. ولهذا أجريت هذه الدراسة لتوضيح التأثير الخلوى السام للفورمالدهيد علي الفئران. وكذلك لتوضيح مدي قدرة فيتامين ج علي حماية الخلية من التأثير السام للفورمالدهيد. صممت هذه التجربة من 36 فأر تجارب تعرضوا لتركيزات مختلفة من الفورمالدهيد و فيتامين ج والفورمالدهيد مع فيتامين ج . وتم أخذ العينات من نخاع الفئران بعد 48 و72 ساعة. أوضحت النتائج أن للفورمالدهيد قدرة علي زيادة التشوهات الكروموسومية العددية و التركيبية فى الخلية. وأيضا أوضحت النتائج أن الفورمالدهيد له تأثيرعلي معدل الإنقسام الميتوزي لخلايا نخاع عظام الفئران عند 72 ساعة.كما أوضحت النتائج أن إستخدام فيتامين ج أدي إلي نتائج جيدة ضد التأثير الضار للفورمالدهيد حيث أنه أدي إلي تقليل معدلات التغيرات الكروموسومية في الفئران.

(مجلة بنها للعلوم الطبية البيطرية: عدد 23(2)، ديسمبر 2012:172-179)