J. Agric. Sci. Mansoura Univ., 29 (12): 7511 - 7522, 2004

TOXICOLOGICAL STUDIES OF MANCOZEB WITH AND WITHOUT WATERCRESS IN RATS.

Hasan, Nahed E.*; Samaa M.E. Bakr and Nora F. Ghanem****
* Pesticides Dept., Fac. of Agric., Kafr El-Sheikh, Tanta Univ., Egypt.
** Biology Dept., Fac. Of Education, Kafr El-Sheikh, Tanta Univ. Egypt.

ABSTRACT

Some toxicological parameters of mancozeb (50 and 100 mg/kg b.w.) were studied with and without watercress on rats. Activities of some enzymes are determined in rats after treatment such as Acetylcholinesterase (AChE) in the brain, liver acid and alkaline phosphatases (ACP and ALP), glutathione-s-transferase (GST),glutamate oxaloacetate transaminase and glutamate pyruvate transaminase (GOT and GPT) and cytochrome P450 monooxygenase. Mancozeb alone reduced significantly the activities of both AChE and GST but elevated the activities of ACP and ALP, GOT, GPT and cyto. P450 monooxygenase. Mancozeb with watercress reduced AChE, phosphatases and transaminases activities significantly in the two doses than mancozeb treatment alone. On the other hand mancozeb with watercress elevated GST activity but reduced cytochrome P450 monooxygenase activities at two doses respectively. Also, mancozeb induced chromosomal aberrations in spreads of bone marrow cells of rats which consisted mainly of stickiness, fragmentation, centromeric fusion, ring, break and end to end association in two doses of mancozeb. The mitotic activity was increased at all durations in rats bone marrow cells if compared with the control. Watercress given three weeks prior to mancozeb showed inhibition of chromosomal aberrations in bone marrow cells at all intervals and also inhibition in mitotic activity when compared with animals given mancozeb only.

Keywords: Watercress, mancozeb, enzymes changes, chromosomal aberrations, rats.

INTRODUCTION

Ethylenebisdithiocarbamates (EBDC) are an important class of fungicides used to control crop diseases and prevent mold. Ethylenethiourea (ETU), reported to be their main degradation and metabolic product in animals and man, may have teratogenic and carcinogenic properties (Pastoelli, *et al.*, 1995). Ethylenethiourea is a carcinogenic degradation product of major ethylene bis(dithiocarbamate), fungicides (mancozeb, maneb, metiram, nabam and zineb) with biological activity attributed to poorly characterized oxidation products (James, *et al.*, 1995 and Zena *et al.*, 1999) recognized carcinogenic toxicity to humans, accurate and reliable determination of the residue level of ETU in food and agricultural products has become of great importance.

Mancozeb, a polymeric complex of ethylene bis (dithiocarbamate) manganese with zinc salt, is reported (Shukla and Arora, 2001) to possess carcinogenic and co-carcinogenic activity in various tumor models.

Desager *et al.*, 2002 found that, watercress, a cruciferous vegetable, inhibited the metabolism of several cyp2E1, substrates such as paracetamol and chlorzoxazone. Since ethanol and its metabolite, acetaldehyde, are

Cyp2E1 substrates, the influence of watercress on ethanol and acetaldehyde was investigated in healthy human volunteers . Ethanol absorption was also delayed by single ingestion of watercress immediately preceding ethanol consumption.

Pereira *et al.*, (2001) determined the fatty acid content of 11 green vegetables that are commonly available in Australia. They found that, the total fatty acid concentrations of the vegetables under study ranged from 44 mg/100gm in wet weight in Chinese cabbage to 372 mg/100mg watercress. There were here omeg-3-polyunsturated fatty acids (PUFAs) in all vegetables analyzed, these were 16:3n-3, 18:2n-6 and 18:3n-3 fatty acids. All 11 green vegetables contained a high proportion of PUFAs, ranging from 59 to 72% of total fatty acids. Consumption of green vegetables could contribute to 18:3n-3 PUFA intake, especially for vegetarian populations.

Stuart, 1986 reported that, watercress contains several vitamins, gluconolates and a volatile oil comprising aromatic isothiocyanates and minerals. Amer and Farah 1974 reported that, exposure to genotoxic chemicals may lead to increased risk of heritable diseases and cancer. The genotoxic effect of carbamates was reported by Evans, (1983). Also, the chromosomal aberration can be used as an indicator of DNA damage (Natarojan and Obe, 1978).

Several derivatives of N-methyl carbonic acid including maneb, mancozeb and carbendazim could metabolically result in the corresponding N-nitrose compound; a large percentage of such compound are alkylating agents which can react with deoxyribonucleic acid and other molecules (Fishibein, 1979 and Saffhull *et al.*, 1985) leading to mutagenesis, clastogenesis and carcinogenesis.

MATERIALS AND METHODS

Animals

120 adult rats (Sprague Dawley), 8 weeks of age and 80-100 gm in weight were used and the rats were obtained from the Egyptian Organization For Vaccines and Medical Products, Cairo , Egypt. The animals were housed in stainless steal cages containing hard wood chips and a temperature controlled room with a 14 hr. light, 10 hr. dark cycles and given a standard diet composed of 60% ground corn meal, 15% ground beans, 10% wheat bran, 10% corn oil, 3% casein, 1% mineral mixture and 1% vitamin mixture, water was ad. Libitum. The animals were kept at room temperature.

Pesticides

A technical mancozeb 99% (zinc ethylene bisdithiocarbamates) is produced by Rohn and Haas Co., in Philadelphia U.S.A.

Experimental design

The rats were divided into five groups, 24 rats per group in the 1st four groups and 6 rats in group five were used as a control group. In each group half of the animals were used for examining the enzymes changes and the other half for chromosomal aberrations. Animals of groups (1and 2) were

given intraperitoneally a daily dose of (50 and 100 mg/kg b.w. of mancozed respectively) dissolved in dimethyl sulphoxide for two months (Subramoniam *et al.*, 1991). Animals of groups (3 and 4) were fed on a basal diet containing 20% of watercress (Eruca-stativa) for 3 weeks prior to mancozeb (50 and 100 mg/kg b.w. respectively) for two months. Group (5) 6 rats served as controls were given a basal diet and injected intraperitonealy with 0.5 ml dimethyl sulphoxide. Three animals of each group were sacrificed at the intervals one week, two weeks, one month and two months and assigned to the chromosomal studies. The rats were sacrificed by cervical dislocation.

Enzymes changes

Brain and liver samples were homogenized and centrifuged at 5000 rpm for 15 min. Supernatant for brain sample used to determined AChE, according to the method described by (Ellman *et al.*, 1961). Supernatant for Liver samples used to determined phosphatases and transaminases (using Commerical kits) spectrophotometric, Acid phosphatases (Hillman, 1971) Alkaline phosphatases (Wilkinson, *et al.*, 1969), transaminases GOT and GPT (Reitman and Frankel, 1957). Liver sampeles were homogenized and centrifuged at 4000 rpm. for 10 min. Supernatant were ultracentrifuged under cooling 20,000 rpm for 1hr. Pellets were suspended as a source of cytochrome P450 monooxygenase (Hansen and Hodgson, 1971) and GST (Vessy and Boyer, 1984) enzyme.

Chromosomal preparation

The animals received colchicines (1 mg/kg b.w) 2hr before sacrifice by cervical dislocation. The frmur marrow from each was used for metaphase preparations, stained and coded for analysis. Metaphase preparations were stained in buffered Giemsa ph (6.8). Structural aberrations such as, ring, fragmentation, break, deletion, centromeric fusion, end to end association and stickiness were mentioned in each metaphase spread. The mutagenic effect was estimated from the frequency (%) of cell containing chromosomal aberration (Malashenko *et al,*. 1997)

RESULTS AND DISCUSSION

Effect on AChE activity

Results in table (1) showed that the treatment with mancozeb (50 and 100 mg/kg b.w.) with or without watercress reduced significantly the specific activity of AChE in rats brain. The activities for mancozeb alone were (61.39 and 49.03%) and with watercress (52.16 and 38.02%) of control respectively. Many authors recorded the reduction on AChE activity of the rats by organophosphorus pesticides such as Pope and Chakraborti, 1992; Osman, 1994 and Abbassy *et al.*, 2000. Results showed that, the inhibitory effect for mancozeb with watercress was more pronounced than mancozeb alone at two doses respectively. Mancozeb is non-specific to inhibit AchE (e.g organophosphorus compound), there fore the activity may be affect by any factor such as watercress.

Table (1): Effect of Mancozeb with or without watercress on some enzymes activities in white albino rats.

Enzymes / Treatment	ACHE		GST		MFO		GOT		GPT		ACP		ALP	
	Mean	%C	Mean	%C	Mean	%C	Mean	%C	Mean	%C	Mean	%C	Mean	%C
Control	1.18	100	1.55^{a}	100	1.45^{a}	100	130.83^{a}	100	13.91^{a}	100	1.31^{a}	100	10.50^{a}	100
Mancozeb 50 mg/kg	0.61^{bc}	52.16	0.61^{b}	39.35	0.49c	34.02	145.6^{a}	111.3	15.05^{a}	108.2	1.96^{b}	149.6	16.41^{ab}	156.3
Mancozeb 100 mg/kg Watercress	0.45^{d}	38.02	0.59^{b}	38.06	0.69^{bc}	47.25	163.20^{ab}	124.8	18.53^{ab}	133.3	2.27^{bc}	173.3	17.70b	168.6
Mancozeb 50 mg/kg + Antioxidant	0.73^{b}	61.39	0.43^{b}	27.74	0.82^{bc}	56.27	201.50^{ab}	154.1	20.72^{b}	149.0	2.71^{cd}	206.9	24.65^{c}	234.8
Mancozeb 100 mg/kg + Antioxidant	0.58^{cd}	49.03	0.59^{b}	38.06	0.69^{bc}	47.25	163.20^{ab}	124.8	18.53^{ab}	133.3	2.27^{bc}	173.3	17.70^{b}	168.6
$LSD_{0.05}$	0.114		0.199		0.497		60.173		4.167		0.523		5.857	

AChE (Specific activity) = µ mole Ach/min/mg protein, GST (Specific activity) = Activity/min/mg protein,
Cytochrom P450 (Specific activity) = µ mole -p-nitrophenol/min/mg protein, GOT and GPT = IU/L, ACP and ALP = U/L

$$\%C\ (\%\ of\ control) = \frac{Activity\ of\ treatment}{Activity\ of\ control} \times 100$$

Effect on liver function

The effect of the treatments on liver function of rats was investigated in Table1. Activities of ACP and ALP were significantly increased in all treatment. The activities percentage for acid phosphatases at two doses of mancozeb were 206.9 and 240.9%, mancozeb with watercress were 149.6 and 173.3% of control, respectively.

The activity percentage for alkaline phosphatase at two doses of mancozeb were 234.8 and 274.2%, mancozeb with watercress 156.3 and 168.6% of control respectively.

Similarly, liver GOT and GPT activities were significantly increased in two doses of mancozeb, the activities of GOT with mancozed were 154.1 and 166.2% mancozeb with watercress 111.3 and 124.8% of control respectively. The activities of GPT with mancozeb 149.0 and 169.2%, mancozeb with watercress 108.2 and 133.3% respectively.

According to these results, it would be concluded that, significant rise in liver ACP, ALP, GOT and GPT activities at two doses of mancozeb and in the present of watercress respectively.

Bogusz, (1968) reported that, the activity of serum alkaline phosphatase and serum GOT significantly higher values in persons handling the organophosphorus insecticides when compared to the control. Menrath *et al.*, (1973) found that, the determination of the organophosphorus insecticides, bunamidine, caused liver damage in dogs which revealed by the significant increase in serum GOT and alkaline phosphatase.

Kackar *et al.*, (1999) assessment of toxicological effect of mancozeb in male rats after chronic exposure. They found that, decrease in gonadal ACP and increase in ALP. Mancozeb produced significant enzymatic changes in the activities of GOT and GPT in the rat that exposed to 1000 and 1500 mg/kg/day for 180 and 360 dayes.

Fejes *et al.*, (2002) determined the toxicity of mancozeb containing fungicide formulation (Dithan M-45 (mancozeb)) and copper sulphate to chicken embryos after administration as single compounds or in combination. They found that, the activities of GPT and GOT significantly increased with the combined copper sulphate and Dithan M-45 treatment.

The high acid phosphatase activity in rat liver indicates possible in vivo liability of lysosomal membrane with the release of this enzyme. Mancozeb may cause liver damage which in turn would lead to release of acid phosphatase, this mechanisms in agreement with finding of Ntiforo and Stein (1957) who found that malathion released arylsulfatase from rat liver isosomes

The liver is often the primary target for the toxic effects of xenobiotics, therefore it can be used as an index for toxicity of various toxicants. The assessment of liver enzymes in the blood is generally a more sensitive measure of hepta toxicity than histopathologic changes and can be assessed within a shorter time (Corelius *et al.*, 1959 and Gradwahl, 1956). Also transaminases are important critical enzymes in the biological processes. They play a role in amino acid biosynthesis. Consequently, they are considered as specific indicators of liver damage. The possible mechanism involved in the elevation of transaminases may be due to tissue damage

(Rouitter, 1964 and Korsrud *et al.*, 1972).

The activities of liver enzymes at two doses of mancozeb were more pronounced than in the present of watercress respectively. Watercress may be contains any traps conjugated with mancozeb due to stoped the action and prevent it to arrive the target. The reduction of concentration in the target reduced the relleas of liver enzymes and liver damage. Stuart, 1986 reported that, watercress contains several vitamins, gluconolates and a volatile oil comprising aromatic isothiocyanates and minerals.

It was observed that, the mancozeb at two doses significantly decreased GST activity to 27.8% and 23.5% of control more than those in the present of watercress 39.69% and 37.88% of control respectively (Table 1). These results were obtained by Scrponi *et al.*, 1991 and Hasan, 2002. They recorded that, some pesticides decreased GST in the rats.

The cytochrome P450 monooxygenase activities were significantly increase for two mancozeb doses 56.27% and 87.81% of control, than those in the present of watercress 34.02% and 47.25% of control respectively (Table 1). It is in agree with Lavrijsen *et al.*, 1986 and 1990, they studies the effect of fungicide imazolil on hepatic microsomal cytocrome P-450 and cytochrom P450 dependent activities in the Bobwhite Quail. They stated that, no significant induction of cytochrom P450 or NADPH-cytochrom c-reductase activity. Pesticides inhibited P-450 activity (Moror *et al.*, 1976 and Ritter and Franklin 1987), while other pesticide increased P-450 activity (Lavrijsen *et al.*, 1986 ; 1990 and Hasan, 2002),

The results showed that, mancozeb with watercress elevated GST activities, but resuced cytochrome P450 monooxygenase activities at two doses respectively (Chasseaud, 1979; Stuart, 1986 and Lam and Zheng 1991).

Such parameters (enzymes) are not good indicator as a biomarkers for evaluation effect of mancozeb. Since any damage of liver tissue led to release and increase the enzymes.

Effect on Chromosomes:-

Chromosomal aberrations were observed in bone marrow cells of rats which treated with two doses of mancozeb. Structural types of chromosomal aberrations were identified and quantitated relative to non treated control, the strucutural chromosomal aberrations include ring, fragmentation, break, centromeric fusion, end to end association and sticky chromosomes. Stickiness may give rise to sticky adhesions between two or more chromosomes and to formation to sticky bridge metaphase (Fig 1).

Table (2) shows numbers and frequency (%) of cells containing chromosomal aberrations and mitotic index (MI) in rats bone marrow after two doses of mancozeb for one week, two weeks, one month and two months. The linearly of mutagenic effect dose and time dependant, the frequency of cells with chromosomal aberrations significantly exceeded the control level even at minimum dose of 50 mg/kg b.w of mancozeb. The high frequency of stickiness, which followed by centromeric fusion, fragmentation, ring, break and end to end association.

Table (2): Numbers and frequency (%) of cells containing chromosomal aberrations and mitotic index in rats bone marrow cells after treatment with mancozeb and watercress with different times.

Group dose mg/kg	Sampling time	Cell number analyzed	Cell with aberrations								MI
			Ring	Centro-meric fusion	Break	End to end asso-ciation	Fragm-entation	Stickin-ess	Total (%)		
									Structured aberrations	Multiple aberrations	
Control	-	500	0.0	1(0.2)	0.0	1(0.2)	2(0.4)	3(0.6)	7(1.4)	0.0	62.8 ± 13.53
50 mg/kg	1 W	500	5(1)	8(1.6)	3(0.6)	4(0.8)	7(1.4)	10(2)	37(7.4)	5(1)	65.5 ± 15.24
	2 W	500	13(2.6)	15(3)	9(1.8)	8(1.6)	16(3.2)	18(3.8)	79(15.8)	4(0.8)	69.8 ± 12.3
	1 M	500	16(3.2)	18(3.6)	7(1.4)	10(2)	19(3.8)	23(4.6)	93(18.6)	7(1.4)	70.0 ± 14.5*
	2 M	500	20 (4)	25(5)	15(3)	13(2.6)	24(4.8)	27(5.4)	124(24.8)	10(2)	78.13 ± 17.9*
50 mg/kg + Watercress	1 W	500	2(0.4)	5(1)	2(0.4)	3(0.6)	5(1)	4(0.8)	21(4.2)	8(1.6)	60.34 ± 11.95
	2 W	500	11(2.2)	8(1.6)	4(0.8)	5(1)	10(2)	15(3)	53(10.6)	3(0.6)	65.2 ± 10.73*
	1 M	500	9(1.8)	10(2)	9(1.8)	7(1.4)	13(2.6)	18(3.6)	66(13.2)	2(0.4)	68.54 ± 13.2*
	2 M	500	15(3)	12(2.4)	13(2.6)	9(1.8)	17(3.4)	20(4)	86(17.2)	5(1)	63.8 ± 15.5
100 mg/kg	1 W	500	8(1.6)	14(2.8)	14(2.8)	18(3.6)	28(5.6)	22(4.4)	104(20.8)	12(2.4)	80.2 ± 20.25**
	2 W	500	20(4)	25(5)	17(3.4)	20(4)	35(7)	33(6.6)	150(30)	18(3.6)	84.56 ± 23.53**
	1 M	500	31(6.2)	30(6)	21(4.2)	25(5)	40(8)	46(9.2)	193(38.6)	20(4)	90.70 ± 25.27**
	2 M	500	45(9)	53(10.6)	27(5.2)	38(7.6)	66(13)	50(10)	279(55.8)	21(4.2)	108.91 ± 31.18**
100 mg/kg + Watercress	1 W	500	6(1.2)	8(1.6)	5(1)	7(1.4)	20(4)	19(3.8)	65(13)	11(2.2)	73.85 ± 19.9*
	2 W	500	18(3.6)	15(3)	16(3.2)	11(2.2)	24(4.8)	25(5)	109(21.8)	17(3.4)	71.37 ± 17.23
	1 M	500	20(4)	18(3.6)	20(4)	16(3.2)	31(6.2)	32(6.4)	137(27.4)	10(2)	75.63 ± 21.54*
	2 M	500	27(5.4)	29(5.8)	23(4.6)	20(4)	39(7.8)	38(7.6)	176(35.2)	9(1.8)	77.24 ± 18.27*

W= weak, M= month, * significant different, ** highly significant different.

A = Structured aberrations, B = Multiple aberrations C = Mean of individual data (metaphase 3000 cells ± SD)

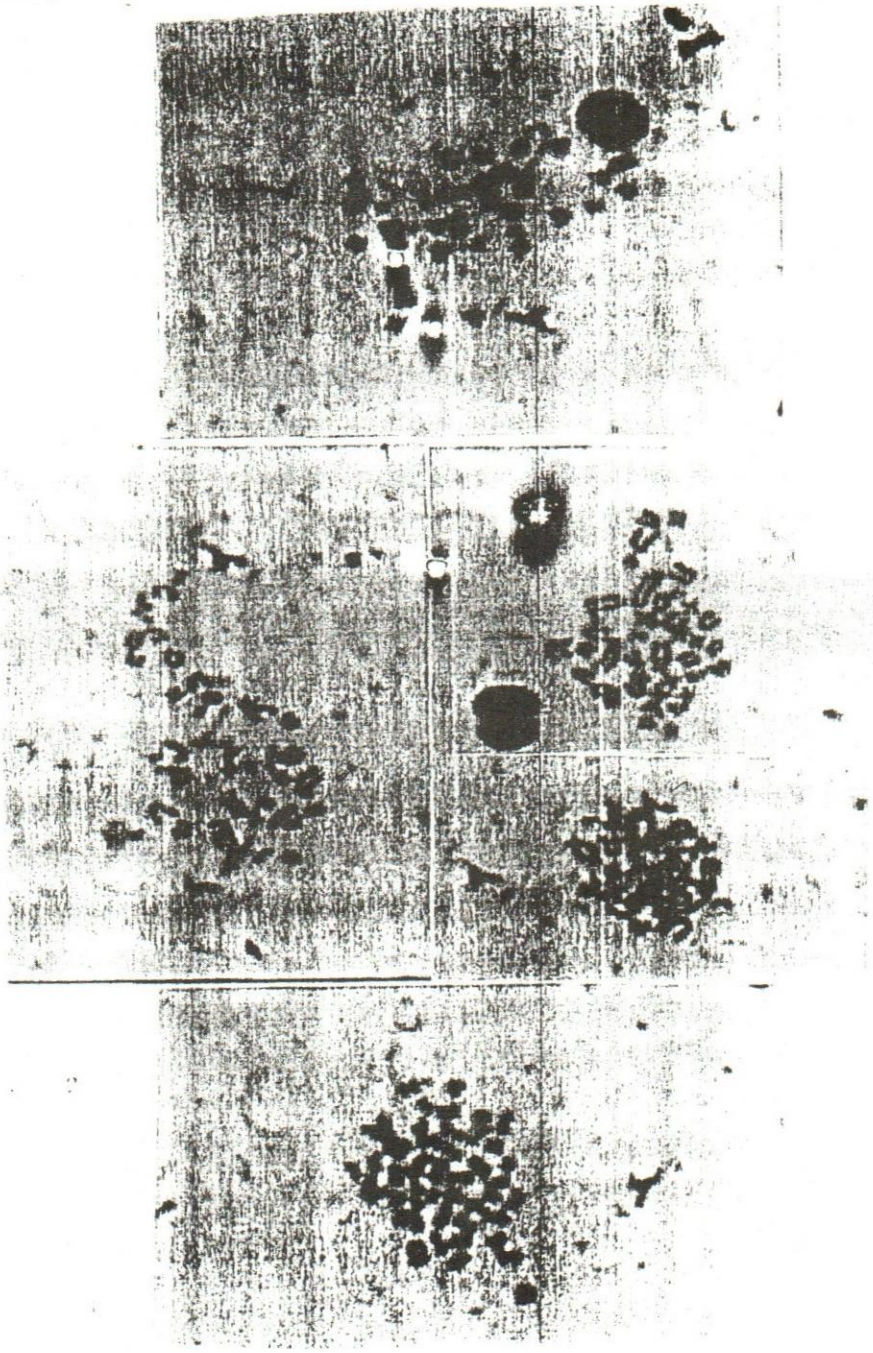

Fig. (1): Metaphase chromosomes of bone marrow cells of control and treated rats with mancozeb showing break(b), end to end association(ee), ring (r), stickiness(s), fragmentation(f) and centromeric fusion(cf).

At high dose 100 mg/kg b.w of mancozeb, there was a significantly increase in the frequency of chromosomal aberrations of fragmentation followed by centromeric fusion, stickiness, ring, end to end association and break. This table observed that, the frequency of chromosomal aberrations increase with increasing of the time which compared with the control. Also the mancozeb increased of the mitotic index after being used at both doses as compared with the control, and increasing with dose and time. Rats given watercress supplement to diet for 3 weeks prior to mancozeb administration and until the end of the experiment may displayed a considerably suppressed incidence of mancozeb under the same condition of this experiment.

Fungicides used to control crop diseases and prevent mold. They enter the organism mainly through the respiratory tract, skin and mucous membranes and the digestive tract and have as their final main metabolite ethylenethiourea (ETU). ETU has been shown to be both embryotoxic, cytogentic (Castro *et al.*, 1999)

The show that, the administration of mancozeb into rats potently induced chromosomal aberrations in their bone marrow cells. Mancozeb induced chromosomal aberrations in rats mainly ring, centromeric fusion, break, end to end association, fragmentation and stickiness. The incidence of total aberrant cells increased progressively after mancozeb treatment and reached a maximum level at the end of the experiment duration. These results showing that, the fungicide mancozeb has a potent activity to induce chromosomal aberrations are consistent with the well-known fact that mancozeb is a strong mutagen and carcinogen (Hassieb, *et al.*, 2004). Lentza-Rizos (1990) observed that etylenebisdithio carbamates (EBDC) are an important class of fungicides used to control crop diseases and prevent mold. Ethylenethiourea (ETU), reported to be their main degradation and metabolic product in animals and may have teratogenic and carcinogenic properties. These study presented that, mancozeb induced chromosomal aberrations at all times after treatment, the watercress were supplement for three weeks prior to mancozeb administration and until the end of the experiment considerably, showed inhibition effect in rats. Also, the mitotic activity was decrease in rats given watercress when comparing with mancozeb. It is an interesting point that the effect of watercress result from any component in mancozeb and this similar to Castro *et al.*, (1999) reported that, the bone marrow cells test is a sensitive and reliable method for assessing the mutagenic potential of various chemical agents.

Watercress contains several vitamins, glucosinolates and volatile oil comprising isothiocynates and minerals (Stuart, 1986). Chasseaud, 1979 and Lam and Zheng, 1991, reported that, isothiocynates increased activity of the detoxifying enzyme glutathione-S-transferase which catalyze the conjuction of glutathione with highly reactive and potently mutagenic metabolite of mancozeb in the liver to form non-mutagenic, water soluble conjugate that is readily excreted and prevent chromosomal aberrations.

REFERENCES

Abbassy, M.A.; A.Y. Keratum. and Sh. El-Hamady (2000). Detrimental effect of chlorpyrifos, pirimipos-methyl and cyfluthrin in rats fed insecticide-treated wheat grains. J.Agic. Res., Tanta Univ., 26(2):404-417.

Amer, S.M.and Farah, R.O.(1974).Cytological effects of pesticides.VIII: Mitotic effect of isopropyl carbamate and Duphar. Cytologia, 40(1): 21-29.

Bogusz, M. (1968). Activity of certain enzymic system in agricultural workers exposed to organophosphorus insecticides.Pol. Typ. Lek.,23(21):781-783.

Castro, V.L.; A.J. Tambasco; L.C. Paraiba and D.D. Tambasco (1999). Cytogenetic and teratological effect of mancozeb prenatal exposure on rats. Brazilian Archives Biology and Technology, 42(1):127- 134.

Chasseaud, L.F. (1979). The role of glutathione and glutathione-S-transferase in the metabolism of chemical carcinogens and other electrophilic agents. Adv. Cancer Res. 29: 175-184.

Corelius, C. E.; W. Charles and E. Arhode (1959). Serum and tissue transaminase activities in domestic animals. Cornell Vet., 49(l): 116-121.

Desager, J.P.; J.L. Golnez; C. Debluck and Y. Horsmans (2002). Watercress has no importance for elimination of CYP2E1 inhibition. Pharmacol. Toxical., 91(3): 103-105.

Ellman, G.L.; K.D. Courtney; V.J.R. Anders and R.M. Featberstone (1961).A new and rapid colorimetric, determination of acetylcholinesterase activity. Biochem. Pharmacology., 7(l):8895.

Evans, H. J. (1983). Effect on chromosomes of carcinogenic rays and chemicals. In chromosome mutation and neoplasia, Liss., New York p.p. 253-279.

Fejes,s.; P. Budai; L. Varnagy; T. Molnar; R.S. zabo and T. Fancsi (2002). Toxicity of mancozeb containing fungicide formulation and Cu-sulphate to chicken embryos after administration as a single compounds or in combination. Med. Fac. Wetenschappen Univ. 67(2)105-109.

Fishibein, L. (1979). Overview on potential mutagenic problems posed by some pesticides and their trace impurities. Environ. Health PersPect., 27(1): 125-131.

Gradwohl, R.B.H. (1956). Clinical laboratory methods and diagnosis. S. th C.V. Mosby Co. St. Louis.

Hansen, LO.G. and Hodgson. (1971). Biochemical characteristics of insect microsomes (N-and O-Demethylathion). Biochem. pharmacol., 20(3):1569-1578

Hasan, N.E. (2002). Toxicological studies on some pesticides with respect to the surrounding environment. Ph.D. Thesis. Fac. Agric. Tanta. Univ.

Hassieb, M.M., N.F., Ghanem and S.M.E., Bak (2004). Protective effect of piroxicam aganist the induced lung damage and chromosomal aberrations due to the fungicide maned administration in mice and rats. J. Egypt Ger. Soc. Zool. Vol (452) 255-273.

Hillman, G., (1971). Fortlaufende photometrische messung der sauren phosphatase-activitat. Z. klin. Chem. Klin. Biochem. 3 (1) 273-277.

James, J.P.; G.B. Qustad and J.E. Casida (1995). Ethylenethiourea S-oxidation products: Preparation, degradation and reaction with proteins. J.Agric. Food Chem., 43(9): 2530-2535.

Kackar, R.; M.K.Srivastave and R.B. Raizada (1999). Assessment of toxicological effects of mancozeb in male rats after chronic exposure. Indian J. Exp. Biol. ,37(6): 553-559.

Korsrud, G.D.; H.C. Grice and J.M. Mclanghlan (1972). Sensitivity of several serum enzymes in detecting carbontetrachloride induced liver damage in rats. Toxicol. Appl. Pharmacol. (22), 474 – 483.

Lam, LKT and Zheng, B. (1991). Effects of essential oils on glutathione-S-transferase activity in mice. J. Agric. Food Chem. 39:660-662.

Lavrijsen, K.; J. Van Houdt ; D. Thijs; W. Meuldermans and J. Heykants (1986). Induction potential of antifungals containing an imidazole or triazole moiety. Biochem. Pharmacol. 35:1867-1878.

Lavrijsen, K.; J. Van Houdt; D. Van Dyck; W. Meuldermans and J. Heykants. (1990). Effect of the fungicide imazalil on hepatic nlicrosornal cytochrome P-450 ind cytochronle P-450 dependet activities in the bobwhite (Colinus virginaus), Pestic. Sci. 29:47-56.

Lentza-Rizos, C. (1990). Ethylenethiourea (ETU) in relation to use of ethylene-bis-dithiocarbamate (EBDC) fungicides. Rev. Environ. Contam. Toxicol., 115-:1-37

Malashenko, A.M.; T.B. Beskova and S.V. Vasileva, (1997). Mutagenic effect of dimethylnitrosourea in male mice. Induction of dominant to lethal mutations and chromosome aberrations in germ cells; Genotype influence on the clustogenic effect in bone marrow cells. Russian Journal of Genetics, Vol. 33 (4) 429-435.

Menrath, R.L.; A. Shared; F.W. Gray and C.W. Cameron (1973). Toxicity of bunamidine 2-metabolic effects. N.Z. Vet. J. 21(10):212-219.

Moror, K.; U. Shizuko; S. Tetsuo and K. Tetsuro (1976). Enzyme induction by repeated administratioi, (if tetrachlorvinphos in rats. Toxiel. Appl. Pharmacol., 37 (2): 377 - 386.

Natarojan, A.T. and G. Obe, (1978). Molecular mechanisms involved in the production of chromosomal aberrations. Mutat. Res., 52 (1) 137-149.

Ntiforo, C. and M. Stein (1957). Labilization of lysosomes as an aspect of biochemical toxicology of anticholinesterase pesticides. Biochem. J. 102(1)44-55.

Osman, K.A. (1994). Interaction of glyphosate with some mammalian biochemitarget. Alex.J.Agric.Res. 39(I): 373 - 392.

Pastorelli, R.; R. Allevi; S. Romagnano; G. Meli; R. Fanelli and L. Airoldi (1995). Gas chromatography-mass-spectrometry determination of Ethylenethiourea hemoglobin adducts: a possible indicator of exposure to ethylene bis dithiocarbamate pesticides. Arch. Toxicol. J. 69(6): 306-311.

Pereira, C.; D. Li and A.J. Sinclair (2001). The alpha-linolenic acid content of green vegetables commomly available in Australia. Int. J. Vitam. Nutr. Res. 71(4): 223-228.

Pope, C.N. and T.K. Chakraborti (1992). Dose-related inhibition of brain and plasma cholinesterase in neonatal and adult rats following sublethal organophosphate exposures. Toxicology.,73 (1): 35- 43.

Reitman, S.M.D. and Frankel (1957). A colorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transarninase. Am. J. Clin. Path. 28: 56-64.

Ritter, J. K. and M.R. Franklin (1987). High magnitude hepatic cytochrorm P450 induction by an N-Substituted imidazole antimycotic clotrimazole. Biochem. Pharmacol. 36:2783-2787.

Rouitter, C. (1964). The liver morphology, biochemistry and physiology. New york. Acadameic Press. P. 335-376.

Saffhill, R.; G. Margison; P. Oconnor (1985). Mechanism of carcinogenesis induced by alkylanting agents. Biochem. Biophys. Act,823(1): 111-145.

Scarponi, L.; P. Perucei and L. Martinett (1991). Conjugation of 2-chloroacetanilide herbicides with glutathion: Role of molecular structures and of glutathion-S-transferase enzymes. J. Agric. Food. Chem., 39 (5): 2010-2013.

Shukla, Y. and A. Arora, (2001). Environmental carcinogenesis division. Industrial Toxicology Research Centre, Lucknow, India.

Stuart, M. (1986). The encyclopedia of herbs and herbalism. Orbis Book publ. crop, London.

Subramoniam, A.; D. Agrawal and S.P. Srivastava, (1991). Influence of mancozeb on mitogenically responsive lipids in rat cerebrum and liver. Indian J. Exp. Biol. 29(4): 943-945.

Vessey, D.A. and T.D. Boyer (1984). Differential activation and inhibition of different forms of rat liver glutathion-S-transferase by the herbicides 2,4,5-trichlorophenoxy acetate (2,4,5-T). Toxicol. Appl. Pharmacol., 73 (3): 492-499.

Wattenberg, L.W. (1983). Inhibition of carcinogenesis by minor dietary constituents. Cancer Res. 52(9): 2085-2093.

Wilkinson, J.H., H.B. Joseph and W. Seymour (1969). Evaluation of a new system for the kinetic measurement of serum alkaline phosphatase. Ciin. Chem. 15 (6): 487-495.

Zena, A.;P. Conte and A. Piccolo (1999). GC/ECD -determenatiion of ethylenethioure residues in tobacco leaves. Fresenius Environ. Bulletin, 8(3): 116-123.

دراسات تكسوكولوجية لمبيد المانكوزيب فى وجود وعدم وجود نبات الجرجير فى الفئران البيضاء

ناهد السيد حسن*، سماء محمد السعيد بكر، نورا فتح الله غانم****

***قسم المبيدات ـــ كلية الزراعة ـــ بكفر الشيخ ـــ جامعة طنطا**

****قسم العلوم البيولوجية ـــ كلية التربية ـــ بكفر الشيخ ـــ جامعة طنطا**

تم دراسة تاثير المبيد الفطرى مانكوزيب باستخدام تركيزين مختلفين هما ٥٠ و ١٠٠مليجرام/كجم/وزن الجسم و كذلك الوجبه العادية بالاضافة الى ٢٠% من اوراق الجرجير الطازجة على بعض الانزيمات والكروموسومات فى الفئران البيضاء. وأظهرت الدراسة ان هناك انخفاض ملحوظ مع المبيد مفرد فى كلا التركيزين فى نشاط انزيمات الاسيتيل كولين استريز فى مخ الفئران و انزيم جلوتاثيون ـــــ س ـــ ترانسفيراز فى كبد الفئران بينما ارتفع نشاط انزيمات الفوسفاتيز الحامضى و القاعدى وانزيمات الترانس امينيز و انزيم السيتوكروم ب ٤٥٠ مونواكسيجنيز فى كبد الفئران. اما فى حالة وجود الجرجير مع المبيد المانكوزيب فانه يحدث انخفاض معنوى فى انزيمات الاسيتيل كولين استريز و الفوسفاتيزالحامضى و القاعدى و الترانس امينيز عن حالة المانكوزيب بمفردة فى كلا التركيزين. بالنسبة لانزيم جلوتاثيون ـــــ س ـــ ترانسفيراز فان وجود الجرجير مع المبيد أدى الى ارتفاع معنوى فى نشاط الانزيم بينما ادى الى انخفاض نشاط انزيم السيتوكروم ب ٤٥٠ مونواكسيجنيز فى كلا التركيزين عن حالة المانكوزيب بمفردة.

وبفحص عينات نخاع العظام فى الفئران على مدار الشهرين لوحظ ارتفاع فى معدل التشوهات الكروموسومية فى مجموعة الحيوانات المعاملة بالمانكوزيب فقط بالمقارنة بالمجموعة الاخرى المعاملة والتى تناولت الوجبة العادية مضافا اليها ٢٠% من اوراق الجرجير قبل واثناء التجربة. كما وجد ان الجرجير يعمل على تنشيط زيادة الانقسام الخلوى فى الفئران طول فترة التجربة بالمقارنة بالمجموعة المعاملة بالمانكوزيب فقط وايضا مقارنة بالكنترول.