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Lipid Peroxidation in Lung and Liver of Rats Given DDT and Endosulfan Intratracheally

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Lipid peroxidation is a degradative process which is associated with a variety of pathological and cytotoxic conditions in both lung and liver. Formation of lipid peroxides in lung by oxidant gases (Gilbert 1963, Thomas et al. 1968, Goldstein et al. 1969), herbicide paraquat (Bus et al. 1974, Misra and Gorsky 1981), Carbon tetrachloride (Recknagel and Glende 1973) and ethanol (DiLugio and Hartman 1969) has been reported. The lung is a target organ for inhaled xenobiotics which can cause cytotoxicity or carcinogenesis. We have evaluated the cytotoxicity of intratracheally administered endosulfan and DDT in parameters of lipid peroxidation in lung and liver, since translocation of pulmonary deposited materials have been shown in extrapulmonary organs in rats (Holt 1980).

MATERIALS AND METHODS

Male wistar strain rats (Weighing 180-200g) were used in this investigation. They were fed Hind Lever diet (Hindustan Lever Ltd. Bombay, India) ad libitum and had free access to water all the time. The rats were divided into three groups and were treated as follows:

Rats of Group A were given DDT intratracheally (5mg/100g body weight) for three consecutive days. Rats of Group B were given endosulfan intratracheally (1mg/100g body weight) for three consecutive days. Rats of Group C were given only the vehicle solution intratracheally for the same period.

The insecticides were dissolved in ground nut oil and were administered through a plastic cannula. Twenty four hours after the administration of last dose of insecticides, rats were fasted over night and anaesthetized

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lightly with ether. They were secured on a board with rubber bands and thoracic and abdominal cavities opened. The lungs were perfused through right ventricle with ice cold physiological saline. After perfusion the lungs and the liver were removed, immersed in ice cold saline, cleaned and weighed. A 20 percent homogenate was prepared in 0.01 M phosphate buffer, pH 7.4, with 1.15 percent KCl. Microsomes were isolated according to the method of Zannoni et al.(1972).Lipid peroxidation was assayed by thiobarbituric acid method of Fairhurst et al. (1983). Liver and lung microsomes (0.5 ml) were suspended in 0.5 ml of 0.15 M KCl, 5mM Hepes(N-2-hydroxyethylpiperazine-N'-2-ethane-sulfonic acid)buffer, pH 7.4; 100μM $FeCl_3$,1.7mM ADP and 0.4 mM NADPH or 0.5 mM ascorbate added and incubated with vigorous shaking in a water bath at 37°C for 30 min. The malondialdehyde produced was assayed as described by Misra and Gorsky(1981). The *in vivo* lipid peroxidation by lung and liver microsomes was studied by measuring the conjugated diens as described by Thomas et al.(1968). The protein was estimated by procedure of Lowry et al.(1951).

RESULTS AND DISCUSSION

The effects of intratracheal administration of the chlorinated hydrocarbon insecticides on organ weight and microsomal protein in rats is shown in Table 1.The results were similar to those reported earlier(Satya Narayan et al. 1984). The effect of intratracheally administered DDT and endosulfan respectively on lipid peroxidation i.e. malondialdehyde formation *in vitro* in lung and liver have been studied in two ways, viz. NADPH dependent and ascorbate induced.Except for a significant reduction in NADPH linked lipid peroxidation by DDT in lung (Table 1), neither DDT nor endosulfan significantly affected NADPH linked and ascorbate induced lipid peroxidation in lung and liver of rats. The formation of malondialdehyde in microsomes by DDT and endosulfan was measured by thiobarbituric acid method which however has recently been suggested not to give a true picture of malondialdehyde formation because of further metabolism of the latter(Plaa and Witchi,1976). Therefore, the formation of conjugated diens was also estimated.The results were identical to those observed on malondialdehyde formation(Table 1). These results show that DDT appears to inhibit lipid peroxidation in lung of rats and endosulfan does not appear to have any lipo-peroxidative effect on lung and liver membranes.The lipid peroxidation involves the formation of free radicals from unsaturated acyl chains of membrane lipids (Pryor 1978, Tappel 1980, O'Brein 1980). Antilipoperoxidative effect of DDT on lung lipids could be either due to the prevention of formation of free radicals. Recently it has been shown that DDT intercalates between fatty acyl chains of

Table 1. Effect of intratracheal administration of DDT and endosulfan on lipid peroxidation of lung and liver microsomal membranes.

Organ	Control	DDT	Endosulfan
		Organ Weight (g)	
Lung	2.13±0.14	2.43±0.19	1.73±0.11
Liver	5.57±0.31	6.37±0.16*	5.10±0.23
		Microsomal protein(mg/g tissue)	
Lung	4.75±0.26	6.88±0.64*	7.59±0.40*
Liver	17.17±0.52	24.69±1.62*	19.62±0.43*
		NADPH linked lipid peroxidation[a] (nmol malondialdehyde/min/mg protein)	
Lung	0.041±0.006	0.024±0.004*	0.047±0.002
Liver	0.191±0.008	0.142±0.012	0.186±0.014
		Ascorbate linked lipid peroxidation[a] (nmol malondialdehyde/min/mg protein)	
Lung	0.032±0.003	0.030±0.001	0.035±0.002
Liver	0.318±0.013	0.332±0.026	0.324±0.019
		Formation of conjugated diens(ABS)[b]	
Lung	0.065±0.006	0.047±0.004*	0.059±0.004
Liver	0.038±0.002	0.033±0.002	0.039±0.002

Values represent the mean±SE from six to eight animals in each group.

a, The rat lung and liver microsomes were incubated at 37°C for 30 min, under the peroxidation conditions as described in the text. Peroxidation was induced by 0.4mM NADPH or 0.5mM ascorbic acid.

b, ABS, is expressed as the absorbance of 1.0 mg microsomal lipid extract from lung and liver of control and treated animals at 235 nm through 1.0 cm light path.

* significantly different from controls ($P \leq 0.05$)

phosphatidylcholine (Buff and Burndt 1981). This may prevent the formation of free radicals and may possibly explain the antilipoperoxidative effect of DDT.

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REFERENCES

Buff K, Berndt J (1981) Interaction of DDT (1,1,1-trichloro-2,2-bis (p-chlorophenyl)-ethane with liposomal phospholipids. Biochim Biophys Acta 643:205-212.

Bus JS, Aust SD, Gibson JE (1974) superoxide-and singlet oxygen-catalyzed lipid peroxidation as a possible mechanism for paraquat (methyl viologen) toxicity.Biochem Biophys Res Commun 58:749-755.

DiLuzio NR, Hartman AD (1969) The effect of ethanol and carbon tetrachloride administration on hepatic lipid-soluble antioxidant activity. Exp Molec Path 11:38-52.

Fairhurst S, Barber DJ, Clark B, Horton AA(1983) Development of the cytosolic defence system against microsomal lipid peroxidation in rat liver. Biochim Biophys Acta 752:491-496.

Gilbert DL (1963) The role of pro-oxidants and antioxidants in oxygen toxicity. Radiat Res Suppl 3:44-53.

Goldstein BD, Lodi c, Collinson C, Balchum OJ (1969) Ozone and lipid peroxidation. Arch Environ Health 18:631-635.

Holt PE (1980) Dust elemination from pulmonary alveoli. Environ Res 23 : 224-227.

Lowry OH, Rosebrough NJ, Farr AL, Randall AJ (1951) Protein measurement with Folin-Phenol reagent. J Biol Chem 193:265-275.

Misra HP, Gorsky LD (1981)Paraquat and NADPH-dependent lipid peroxidation in lung microsomes. J Biol Chem 256:9994-9998.

O'Brein PJ (1980)in Autoxidation in Food and Biological systems. ed. MG Simic and M Karel, pp 563-587. Plenum Press, New York.

Plaa GL, Witschi H (1976) Chemicals, drugs and lipid peroxidation. Ann Rev Pharmacol Toxicol 16:125-141.

Pryor WA (1978) The formation of free radicals and the consequences of their reactions in vivo. Photochem Photobiol 28 : 787-801.

Recknagel RO, Glende EA JR. (1973).Carbon tetrachloride hepatotoxicity: an example of lethal clearage. CRC Crit Rev Toxicol 2 : 263-297.

Satya Narayan, Dani HM, Misra UK (1984) Effect of intratracheally administered DDT and endosulfan on pulmonary and hepatic respiratory cytochromes. Bull Environ contam toxicol (In press).

Tappel AL (1980) in free radicals in Biology. ed WA Pryor, 4 : 1-47, Academic Press, New York.
Thomas HV, Mueller PK, Lyman RL (1968) Lipoperoxidation of lung lipids in rats exposed to nitrogendioxide. Science 159 : 532-534.
Zannoni VG, Flynn EJ, Lynch M (1972) Ascorbic acid and drug metabolism. Biochem Pharmacol 21 : 1377-1392.

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