http://www.pjbs.org

PJBS

ISSN 1028-8880

Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Pakistan Journal of Biological Sciences 11 (1): 127-130, 2008
ISSN 1028-8880

The Potential Toxicity of Diazinon on Physiological Factors in Male Rat

P. Alahyary, M. Ilkhani Poor, F. Fathy Azarbaijani and V. Nejati
Department of Biology, Faculty of Science, Urmia University, Iran

Abstract: Diazinon is an Organophosphate Insecticide (OPI) is commonly used in agriculture to protect of crops and to control pests in home gardens and farms. Many alterations observed by diazinon have been described, such as; alterations in blood factors (RBC, Hb and Hct), plasma testosterone and glucose levels. We selected 12 albino Wistar rats weighting between 220-280 g were divided into two experimental groups, as follow, control group and diazinon treated group. The effects of diazinon, on rat interstitial cell testosterone production, blood factors and plasma glucose levels were evaluated. Male rats were treated orally with a single dose of 1/4 LD_{50} of diazinon. Animals received treatment for 28 days. Present results indicated that in diazinon treated group, plasma glucose and testosterone levels increased compared to control. Also in diazinon group, reduce of blood factors were observed than control. In conclusion, diazinon disturbs the synthesis of testosterone and glucose release from liver into blood and it led to anemia.

Key words: Diazinon, organophosphate insecticides, blood factors, testosterone

INTRODUCTION

The control of insect pests relies heavily on the use of synthetic insecticides. But, their widespread use has led to some serious problems including toxic residues on grass and toxicity to non-target organisms such as mammals, birds and fishes (Zettler and Cuperus, 1990; White, 1995; Riebeiro *et al*., 2003).

Diazinon is an organophosphorus compound with an anticholinesterase mode of action. It is used extensively to control flies, lice, insect pests of ornamental plants and food crops, as well as nematodes and soil insects in lawns and croplands. Diazinon degrades rapidly in the environment, with half-time persistence usually less than 14 days. But under conditions of low temperature, low moisture, high alkalinity and lack of suitable microbial degraders, diazinon may remain biologically active in soils for 6 months or longer.

At recommended treatment levels, diazinon-related kills have been noted for songbirds, honeybees and especially waterfowl that consume diazinon-treated grass; however, incidents involving agricultural applications may be under reported. Accidental deaths through misapplication of diazinon have also been recorded in domestic poultry, monkeys and humans. It has been suggested, but not yet verified, that wildlife partially disabled in the field as a result of diazinon poisoning would be more likely to die of exposure, predation, starvation, or dehydration, or face behavioral modifications, learning impairments and reproductive declines than would similarly treated domestic or laboratory animals. There have been increasing concerns about the effects of various organophosphate insecticides in humans and experimental animals. These include cholinergic and noncholinergic biological disturbances (Ali and Abdalla, 1992; Quistad *et al*., 2001; Bomser *et al*., 2002; Quistad *et al*., 2002; Quistad and Casida, 2002; Gordon and Mack, 2003). Some reports have been published with respect to the organophosphate insecticide, diazinon (McGill *et al*., 1981).

Dikshith *et al*. (1975) observed mild structural and functional changes in the liver as well as in the testis of rats after a single intraperitoneal administration of diazinon. Ansari and Kumar (1988) reported that the exposure of zebrafish to diazinon for up to 168 h has significantly reduced DNA, RNA and the total protein in the liver, but significantly increased the amino acid content in a dose and time-dependent response. These included Glutamic Oxaloacetic Transaminase (GOT), Glutamic Pyruvic Transaminase (GTP), Glutamyltransferase (GT) and Lactate Dehydrogenase (LDH). This inhibition was enhanced by the addition of ascorbic acid into the diet. Matin *et al*. (1990) showed that the administration of diazinon into rats resulted in carbohydrate metabolism changes that were abolished by adrenalectomy, suggesting a possible involvement of the adrenals in the induced changes in diazinon-treated animals. Contreras *et al*. (2006) measured alteration of plasma testosterone level in treated rats with parathion (an organophosphate pesticide). Jyotsana *et al*. (2003) investigated effect of pesticides on some blood factors level. Fatemeh *et al*. (2006) evaluation of alteration of hepatic cells glucose metabolism under effect of diazinon.

Corresponding Author: P. Alahyary, Department of Biology, Faculty of Science, Urmia University, Iran

The purpose of the present study was to evaluate the effects of diazinon on alterations in blood factors, plasma testosterone and glucose levels in male rats.

MATERIALS AND METHODS

Animals and treatments: Twelve male Wistar-albino rats weighting 220 to 280 g were divided into two experimental groups, each with six rats, as follows: control group and diazinon treated group (diazinon).

Diazinon [o, o-diethyl o-(2-isopropyl-6-methyl-4-pyrimidinyl) phosphorothioate] was obtained from Sigma, USA. All experiments were administered in animal physiology Lab., Urmia University, Iran, in July 2006. Diazinon group was treated orally with a single dose of 1/4 LD_{50} (75 mg kg^{-1}). Diazinon in 4 mL corn oil. Corn oil was given in the same way to the control.

All groups received the above treatments for 28 days. The animals were fasted overnight for 12 h before the blood was collected. The rats were anaesthetized with ether and venous blood samples were collected by direct heart puncture and were divided to two parts, one was maintained in EDTA bulb and plain tube for assay of blood factors, other was centrifuged and serum was discarded and then was kept at 21°C until the tests for evaluation of plasma glucose and testosterone levels.

Statistical analysis: The statistical analysis was performed by SPSS Continuous data are expressed as mean±SD. Data were compared using one-way ANOVA. P value <0.05 was considered to be statistically significant.

RESULTS AND DISCUSSION

The results show that blood factors level (RBC, Hb and Hct) decrease in diazinon group (Table 1). Administration of diazinon at doses of 75 mg kg^{-1} increased plasma glucose concentration, so that glucose level in the diazinon group was higher than control. As results indicated, 28 days after treatment, there was a significant increase in blood testosterone level in rat treated with diazinon compared to control (Table 2).

The results of the present study confirmed that acute exposure to diazinon increases plasma glucose and testosterone also reduces levels of some blood factors.

In this study, the hematological parameters like RBC, Hb and Hct, were significantly decreased in diazinon treated group as compared to control. The effect of Hb on human exposed to organophosphorus pesticides has been observed by several workers (Bhatnagar, 1980; Ray, 1992). The decrease in the Hb along with the decrease in the RBC might be due to the effect of pesticides on blood forming organ in rats. Many steps in heme biosynthesis are inhibited by pesticidal residues. The poisoning by pesticide residues is the development of anemia due to interference of Hb biosyrithesis and shortening of the life span of circulating erythrocytes. The finding of our study agrees to reports of study of Jyotsana *et al.* (2003) that showed pesticides decrease some blood factors level. Our results showed that diazinon led to increase plasma glucose level that it is similar to observations of Fatemeh *et al.* (2006), they observed that some of alteration of hepatic cells glucose due to pest insecticides. Can be concluded that liver, the most important organ of glucose homeostasis in the body, is a target organ for diazinon toxicity. The liver plays a major role in blood glucose homeostasis by keeping a balance between the uptake and storage of glucose via glycogenesis and the release of glucose by way of glycogenolysis and gluconeogenesis. Thus the first idea that comes to mind is that hepatic glycogenolysis and gluconeogenesis pathways are stimulated to provide more glucose by effects of diazinon. Moreover, altered glucose level may be considered for increase glucose release from liver into blood through activation of glycogenolysis and gluconeogenesis as a detoxication mechanism to overwhelm diazinon-induced toxic stress.

In present experiments increased plasma testosterone level that it confirms report of Contreras *et al.* (2006) that measure hormone level in treated rats with an organophosphate pesticides (parathion) and reported that plasma testosterone level increase after 40 days. Testicular Leydig cell is the main site of testosterone synthesis. This steroidal hormone plays a key role in maintenance of spermatogenesis, male sex characteristics and fertility. A wide range of agents is known to alter Leydig cells in rat, including numerous chemicals, some of them agropesticides of the organochlorine type (Cooke, 1998).

Table 1: Change in blood factors levels of rat 28 days after administer of diazinon. Values are expressed as mean±SE of six animals, p<0.05

	Treatments	
Blood factors	Control	Diazinon
RBC (Million μL^{-1})	0.18±5.66	0.02±4.96
Hb (g dL^{-1})	0.22±11.65	0.19±10.63
Hct (%)	0.49±40.3	0.12±36.63

Table 2: Effects of diazinon on rat plasma glucose and testosterone levels. Values are expressed as mean±SE of six animals, p<0.05

	Treatments	
Serum factors	Control	Diazinon
Glucose (mg dL^{-1})	0.82±71	1.60±100.8
Testosterone hormone	0.36±2.01	0.90±4.73

The sites of action of testicular toxicants acting on Leydig cells might be many, since the normal pathway of testosterone production involves hypothalamic secretion of GnRH, stimulation of the adenohypophysis by GnRH to produce LH and the interaction of LH with Leydig cells to stimulate testosterone production, which in addition has a paracrine regulation by Sertoli cell derived estradiol and GnRH (Bustos-Obregón and Gonzalez-Hormazabal, 2003). Moreover, testosterone secretion is also influenced by plasma prelatic levels and cytokines produced by macrophages (Morris, 1996).

Present observations on blood testosterone levels Following treated organophosphoric pesticide indicate that Leydig cell steroidogenesis is acutely and deeply damaged by, diazinon. Increasing production of testosterone, with an overshooting over control levels is obtained only 28 days for diazinon treated animals. Present results is adverse to reports of Naqvi and Vaishnavi (1993), they show that plasma testosterone level decreased in non-target animals treated with insecticide. Effects of organophosphate pesticides on steroidogenesis were unclear (Contreras *et al*., 2006). These results suggest that the dramatic effects observed *in vivo* might be due to interference of the toxicant with the hypophysis modifying the release of LH, or altering the response of Leydig cells to LH. The cascade of events may be as complex as those reported for the effect of dopamine agonists on Leydig cell function.

To answer satisfactorily this hypothesis more experiments are necessary (Cooke, 1996). Further insight on the way agropesticides act on Leydig cell function requires *in vitro* analysis of steroidogenesis and the effect that these toxicants have on enzymatic pathway of testosterone synthesis as well as on general detoxifying mechanisms of the cells, such as the cytochrome P-450 complex (Butler and Murray, 1993).

Results obtained from this study and previous researches show that organophosphate pesticides such as diazinon can be toxicity to non-target organisms. If these toxins affect laboratory animals, may be having same effects on mammals too. Therefore, protective methods are necessary against OPI toxicity.

REFERENCES

Ali, F.A. and M.H. Abdalla, 1992. Pathological changes in tests and liver of male albino rats after dermal exposure to DDVP insecticide. J. Egypt. Public Health Assoc., 67: 565-578.

Ansari, B.A. and K. Kumar, 1988. Diazinon toxicity: Effect on protein and nucleic acid metabolism in the liver of zebrafish, *Brachydanio rerio* (Cyprinidae). Sci. Total Environ., 76: 63-68.

Bhatnagar, 1980. Effect of Pesticide Stress Among Pesticide Factory Workers in Delhi. Pub. HIM., 74: 380.

Bomser, J.A., G.B. Quistad and J.E. Casida, 2002. Chlorpyrifos oxon potentiates diacylglycerol-induced extracellular signal-regulated kinase (ERK 44/42) activation, possibly by diacylglycerol lipase inhibition. Toxicol. Applied Pharmacol., 178: 29-36.

Bustos-Obregón, E. and P. Gonzalez-Hormazábal, 2003. Mice testicular damage elicited by Malathion. Int. J. Morphol., 21 (2): 155-159.

Butler, A.M. and M. Murray, 1993. Inhibition and inactivation of constitutive cytochromes P450 in rat liver by Parathion. Mol. Pharmacol., 43: 902-908.

Contreras, H.R., V. Paredes, B. Urquieta, L. Del Valle and E. Bustos-Obregon, 2006. Testosterone production and spermatogenic damage induced by organophosphate pesticides. J. Biocell., 30 (3): 423-429.

Cooke, B.A., 1996. Transduction of Luteinizing Hormone Signal Within the Leydig Cell. In: The Leydig Cell, Payne, A., M. Hardy and D. Russel (Eds.). Casche River Press, Vienna II USA., pp: 351-364.

Cooke, B.A., 1998. *In vitro* Models for the Investigations of Reproductive Toxicology in the Testes. Reproductive Toxicology Del Mazo (Ed.). Plenum Press, New York, pp: 95-103.

Dikshith, T.S., J.R. Behari, K.K. Datta and A.K. Mathur, 1975. Effect of diazinon in male rats. Histopathological and biochemical studies. Environ. Physiol. Biochem., 5: 239-299.

Fatemeh, T., A. Nasim, E. Hadi, M. Azadeh, A. Atousa and A. Mohammad, 2006. Alteration of hepatic cells glucose metabolism as a non-cholinergic detoxication mechanism in counteracting diazinon-induced oxidative stress. Human Exp. Toxicol., 25: 697-703.

Gordon, C.J. and C.M. Mack, 2003. Influence of gender on thermoregulation and holinesterase inhibition in the long-evans rat exposed to diazinon. J. Toxicol. Environ. Health, A 66: 291-304.

Jyotsana, A.P., J.P. Arun and P.G. Sanjay, 2003. Biochemical effects of various on sprayers of garape gardens. Ind. J. Clin. Biochem., 18 (2): 16-22.

Matin, M.A., K. Husain and S.N. Khan, 1990. Modification of diazinon-induced changes in carbohydrate metabolism by adrenalectomy in rats. Biochem. Pharmacol., 39: 1781-1786.

McGill, H.C. Jr., C.A. McMahan, A.W. Kruski and G.E. Mott, 1981. Relationship of lipoprotein cholesterol concentrations to experimental atherosclerosis in baboons. Arteriosclerosis, 1: 3-12.

Morris, I.D., 1996. Leydig Cell Toxicology. In: The Leydig Cell, Payne, A.H., M.P. Hardy and L.D. Russell (Eds.). Cache River Press, Vienna, Ill. USA., pp: 573-596.

Naqvi, S.M. and C. Vaishnavi, 1993. Bioaccumulative potential and toxicity of endosufan insecticide to non-target animals. Comp. Biochem. Physiol. C, 105 (3): 347-361.

Quistad, G.B., S.E. Sparks and J.E. Casida, 2001. Fatty acid amide hydrolase inhibition by neurotoxic organophosphorus pesticides. Toxicol. Applied Pharmacol., 173: 48-55.

Quistad, G.B. and J.E. Casida, 2002. Sensitivity of bloodclotting factors and digestive enzymes to inhibition by organophosphorus pesticides. J. Biochem. Mol. Toxicol., 14: 51-56.

Quistad, G.B., S.E. Sparks, Y. Segall, D.K. Nomura and J.E. Casida, 2002. Selective inhibitors of fatty acid amide hydrolase relative to neuropathy target esterase and acetylcholinesterase: Toxicological implications. Toxicol. Applied Pharmacol., 179: 57-63.

Ray, 1992. Pollution and Health, Wiley Eastern Ltd., New Delhi 1992.

Riebeiro, B.M., R.N.C. Guedes, E.E. Oliveira and J.P. Santos 2003. Insecticide resistance and synergism in Brasilian populations of *Sitophilus zeamais* (Coleoptera: Curculionidae). J. Stored Prod. Res., 39: 21-31.

White, N.D.G., 1995. Insects, Mites and Insecticides in Stored Grain Ecosystems. In: Stored Grain Ecosystem, Jayas, D.S., N.D. White and W.E. Muir (Eds.). Marcel Dekker, New York, USA., pp: 123-168.

Zettler, J.L. and G.W. Cuperus, 1990. Pesticide resistance in *Tribolium castaneum* (Coleopteran: Tenebrionidae) and *Rhyzopertha dominica* (Coleoptera: Bostrichidae) in wheat. J. Econ. Entomol., 83: 1677-1681.