International Journal of Sciences: Basic and Applied Research (IJSBAR)

ISSN 2307-4531
(Print & Online)

http://gssrr.org/index.php?journal=JournalOfBasicAndApplied

Study of Propanil Induced Toxicity on Heamatological Parameter and Amelioration by Taurine in Mice

Arti Chauhan[a]*, Usha Gaur[b], Nidhi Shrivastva[c]

[a,b]*Department of zoology, Holkar Science College, Devi Ahilya University, Indore- 452001, Madhya Pradesh, India*

[c]*Department of Veterinary Pathology, College of Veterinary Science & Animal Husbandry, 453441, Madhya Pradesh, India*

[a]*Email: artichauhan1014@gmail.com*

[b]*Email: drushapankajgaur@gmail.com*

[c]*Email: nidhivinmra@yahoo.com*

Abstract

This study was carried out to evaluate in vivo protective role of taurine on toxic effect caused by propanil in mice on the hematological values. Methods: In an experimental study 24 albino mice were distributed in six equal groups of six animals in each as follows: Control group, 100mg propanil/kg, 100mg taurine/kg, 200mg taurine/kg, propanil (100 mg/kg) + taurine (100 mg/kg), propanil (100mg/kg) + taurine (200mg/kg). Treatment was via oral route and was fed once daily for 90 days. The mice exposed to propanil showed significant decrease in RBC's and Hb level. There is marked decrease in WBC, PCV, and DLC values. Number of MCV values increases which is reciprocal to RBC, PCV and Hb in propanil intoxicated mice. Co-administration of taurine with propanil helped to prevent toxicity by reversing and scavenging propanil induced hematological alterations in mice.

Key words: hematology; propanil; taurine; mice.

---

* Corresponding author.

1. Introduction

Taurine (2-aminoethanesulphonic acid) is an important intra cellular amino acid found in minute concentration in many animal tissues like brain, liver, muscles, heart etc [1]. It is an essential sulfonated beta amino acid derived from methionine and cysteine metabolism. Taurine is present in high amount too, particularly in polymorphonuclear phagocytes and in the retina. Metabolic action of taurine includes bile acid conjugation, detoxification, membrane stabilization, osmoregulation and modulation of cellular calcium level [2, 3,4,5]. The beneficial effect of taurine as an antioxidant in biological system have been imparted to its ability to stabilize biomembranes [1] and scavenging reactive oxygen species [6]. Taurine may also have protective effect on the tissue damage that results due to ROS formed by propanil toxicity. With this point of view, the present study has been aimed to find out the ability of taurine to normalize the blood profiles of intoxicated mice by co administration with taurine. Modern cultivation is dependent on various agrochemicals viz. pesticides, insecticides mainly herbicides. Herbicides are quite an important class of agricultural pesticides. Propanil (3 ,4 dichloropropionanilide) is an acetanilide which is used to control broadleaf weeds and most extensively used as post emergent herbicides for rice wheat and potato production worldwide. Pesticide poisoning is a big issue in parts of the developing world. Occupational and accidental exposure can never be avoided, but majority of death are due to deliberate self-poisoning. Propanil and its major metabolite, 3,4-dichloroanilide salt, induce the conversion of Fe^{2}+ in haemoglobin to Fe3+, forming methaemoglobin (metHb) which leads reduction in oxygen carrying capacity of blood. Propanil is among top twenty pesticides used in agriculture [7]. Propanil is used on those crops which we take on daily basis which suggests that we are at high risk regularly. World Health Organization (*WHO*) has considered propanil slightly hazardous in terms of human risk [8]. Taurine (2-aminoethanesulphonic acid) acts as an antioxidant which is present in many animal tissues like kidney, liver and brain majorly in minute quantity [1, 9]. To the best of our knowledge, there are no studies concerning the effect of taurine on hematological alteration due to propanil intoxication. Therefore, the present study was carried out to investigate (a) the adverse effect of sub chronic propanil intoxication on mice blood parameters (b) the probable ameliorating effect of taurine against propanil intoxication in mice.

2. Methodology

2.1 Chemicals

Herbicide propanil PESTANAL#, of analytical standard was purchased from Sigma-Aldrich Co.Ltd.St. Louis, USA and taurine was purchased from LOBA chemie. All other chemicals were of technical grade and purchased from Loba Chemie, Mumbai India.

2.2 Animals

Colony bred Swiss albino mice weighing 18-20gm obtained from Institute of Animal health and Veterinary and Biological Products, Rasalpura, Mhow, Madhya Pradesh were used for this study. The animals were maintained at 22±3^{o}C with 50-70% relative humidity and 12:12 hrs of light and dark cycles and were kept in well ventilated cages. The animals were fed with calculated amount of laboratory pellet diet procured from government

agricultural college, Indore, India, and water *ad libitum*. Animals were maintained as per the guidelines laid down by departmental ethical committee for handling and maintenance for experimental animals and the committee for the purpose of control and supervision on experiments in animals (CPCSEA) Ref.No-1063/DDS/2014-15.

2.3 Experimental Protocol

Mice were divided into six groups of six animals in each and were allowed free access to feed and water for 20 days before the commencement of the experiment. As both the drugs were given in pellet diet, so mice were closely studied for a period of 20 days to evaluate the consumption of food according to already studied equation.

- Group 1- Control animals (no treatment)
- Group 2- Propanil treatment (100 mg/kg BW)
- Group 3 - Taurine treatment (100mg/kg BW)
- Group 4-Taurine treatment (200mg/kg BW, double dose treatment)
- Group 5- Propanil (100mg/kg BW) + Taurine (100mg/kg BW)
- Group 6-propanil (100mg/kg BW) + Taurine (200mg/kg BW)

The dose of potential ameliorating drug was increased as it was observed that the protection was improved by increasing the dose to twice. All the above groups except group 1 were treated daily for the period of 90 days. After the administration of the last dose animals were given rest overnight and were sacrificed next day by exposing them to mild ether anesthesia. Blood from each animal was collected and preceded further for hematological parameters.

2.4 Blood Sampling

Blood from each animal was collected and serum was isolated for the estimation of different biochemical and hormonal parameters. The various blood profile viz., RBC, WBC, PCV, DLC and Hb. PCV was determined by Wintrobe's method. TEC and Hb were calculated by [10]. WBCs and DLC were determined by improved Neubauer's counting chamber [10].

3. Results

The hematological properties of mice exposed to propanil are shown in table 1. Group intoxicated with propanil leads to significant decrease ($P < 0.05$) in pac cell volume (PCV20.24±0.23 >14.18±0.32), white blood cells (WBC 5.16±0.66 > 3.12±0.15), red blood cells (RBC 6.08±1.12 > 3.43±1.32), and (Hb 10.90±0.43 > 6.12±0.23) as compared to respective control values. There were significant rise in mean cell volume (MCV 56.13±1.12 > 50.10±0.22) and decrease in mean cell hemoglobin (MCH 26.45±0.12 > 23.46±0.14) in propanil treated mice as compared to control group. The level of Hb and PCV significantly increased ($P < 0.05$) when taurine is fed along with propanil. There were moderate increase in WBC with taurine extract alone but when taurine was administered along with propanil, it was found that taurine is able to increase the values to normal level of WBC

(6.17±1.11 and 6.18±0.7), RBC (6.34±0.89 and 6.56±0.78), Hb (14.45±0.38 and 14.24±0.34), PCV (27.83±0.21 and 27.28±0.15), by significant amount ($P < 0.05$) of all blood profile. There was significant decrease in MCV (58.26±0.78 and 58.43±0.37) with both concentration of taurine. But we did not find any significant difference by giving double dose of taurine. Co-administration of taurine and propanil lead to significant increase ($P < 0.05$) MCH level (31.21±1.12 and 31.56±1.16) it was worth to note that the level of MCH reached above the normal level present in control on co- administration of taurine with propanil. Table 2 signify ($P < 0.05$) decrease in values of lymphocytes (49.16±0.16 > 40.27±0.14) monocyte (11.32±0.23 > 6.16±0.53) and neutrophils (28.32±0.28 > 24.14±0.76) count in propanil treated mice as compared to normal control group. Administration of taurine along with propanil resulted in significant increase in ($P < 0.05$) level of all leukocyte count, while giving double dose of taurine there were moderate increase in monocyte and lymphocyte values as compared to normal animals. Interestingly co administration of taurine and propanil increased the neutrophil count and monocyte than untreated animals which suggests that there is significant amelioration by taurine.

Table1: Protective effects of taurine on differential leukocyte count in mice intoxicated with propanil.

Groups	Monocyte(%)	Lymphocyte(%)	Neutrophils(%)
Control	11.32±0.23*	49.16±0.16*	28.32±0.28*
Propanil	6.16±0.53$^{\#}$	40.27±0.14$^{\#}$	24.14±0.76$^{\#}$
Tau 1	14.37±0.52$^{*\#}$	52.13±0.62$^{\#*}$	32.43±0.26$^{*\#}$
Tau 2	14.74±0.54$^{*\#}$	53.02±0.15$^{*\#}$	33.56±1.04$^{\#*}$
Propanil+Tau1	12.25±0.70*	38.89±0.21*	27.20±0.13*
Propanil+Tau2	12.28±0.49*	39.09±0.24$^{\#}$	28.09±0.24*

Values are mean ±SD of six mice in each group. Significance at P< 0.05

* compared against propanil; # compared against control

Table 2: Protective effect of taurine on some hematological profiles in propanil exposed mice.

Groups	Hb(g/dl)	PCV(%)	RBC(millions/µl)	WBC(10^3/mm^3)	MCV(%)	MCH (%)
Control	10.90±0.43*	20.24±0.23*	6.08±1.12*	5.16±0.66*	50.10±0.22*	26.45±0.12*
Propanil	6.12±0.23$^{\#}$	14.18±0.32$^{\#}$	3.43±1.32$^{\#}$	3.12±0.15$^{\#}$	56.13±1.12$^{\#}$	23.46±0.14$^{\#}$
Tau 1	14.45±0.38$^{\#*}$	27.83±0.21$^{*\#}$	6.34±0.89*	6.17±1.11*	58.26±0.78$^{\#}$	31.21±1.12$^{\#*}$
Tau 2	14.24±0.34$^{\#*}$	27.28±0.15$^{\#*}$	6.56±0.78*	6.18±0.76*	58.43±0.37$^{\#}$	31.56±1.16$^{\#*}$
Propanil+Tau1	9.34±0.42*	19.17±0.45*	5.30±1.13*	5.23±0.64*	48.12±0.34*	27.78±1.13*
Propanil+Tau2	9.78±0.87*	19.79±0.36*	5.14±1.13*	5.43±0.47*	48.31±0.42*	27.86±1.15*

Values are mean ±SD of six mice in each group. Significance at P< 0.05

* compared against propanil

compared against control

4. Conclusion

The result of present study depicts that propanil affects hematological system by inhibiting the heme and hemoglobin synthesis and leads to hemolytic anemia. It decreases the amount on RBC, PCV, WBC count and Hb concentration in mice (Table 1). The decrease in PCV values in blood of propanil treated mice indicated the increased destruction of erythrocytes which results to leucopenia, erythrocytopenia and methaemoglobinemia are direct effect of propanil on the hematological system. RBCs are without biosynthetic pathways they are actually terminally differentiated cells where many biosynthetic pathways are absent. Whatever be the root of exposure, inhalation, dermal, peritoneal or deliberate ingestion, RBCs are prior most cell to be exposed to xenobiotics. RBC acts as oxygen carrier, so they are attacked continuously by external agents and free radicals. The amount of polyunsaturated lipids is high in erythrocytes which makes them prone to oxidative damage. So, RBCs should have strong antioxidant system to shield themselves from cytotoxic effect of reactive oxygen species. DLC count decreased in propanil treated mice. Neutropenia and lymphocytopenia resulted in propanil treated group is an indication of immune suppression. Co-adminstration of taurine with propanil produced moderate to significant effect in almost all blood profile, thus indicating the protective role of taurine in propanil toxicity. It is further noted that taurine significantly prevented the influence of propanil on differential leukocyte count. Earlier studies with known antioxidant such as vitamin C and vitamin E showed that they aid in reducing the propanil toxicity from blood. Similarly in our findings suggests that taurine is capable of scavenging propanil induced hematological alterations to significant extent. Subsequent N-oxidation of 3,4-DCA produces 3,4-DCPHA that is responsible for the production of the hemolytic anemia and methemoglobinemia. The propanil metabolite 3,4-DCA can also induce nephrotoxicity in vivo [11] and in vitro in rat renal cortical slices [11, 12]. The result of present study goes with the findings [9,13,14] where it was suggested that propanil metabolite shows hematotoxic effects due to hydrolysis of propanil to yield 3,4-DCA. So it can be concluded that co administration taurine with propanil protects the blood from the oxidative damage. Taurine affects the non-enzymatic and enzymatic AO system of the RBCs and also blocks oxidation of Hb to MetHb. The protective effects of taurine reported in this study are summarized in table 1 and table 2. [15, 16, 17]. The protection by taurine against propanil can be due to its intrinsic biochemical and natural AO properties. These findings clearly depicts the importance of taurine on the functions of blood, especially red blood cells, and show us a mechanism by which propanil induced toxicity can be stopped. However, more elaborated research with varying doses of taurine and propanil need to be done to delineate the exact molecular steps of the protective effects of this amino acid.

References:

[1]. C.E. Wright, H.H. Tallan, Y.Y.Lin, G.E. Gaull. “Taurine: biological update”. Annu. Rev. Biochem. vol.

55, pp.427–453, 1983

[2]. R.W.Chesney, X. Han, A.B. Patters . “Taurine and the renal system”. J. Biomed. Sci.vol. 17, pp. 1-10, 1985.

[3]. R.J. Huxtable. “Physiological actions of taurine”. Physiol. Rev. vol.72, pp.101–163, 1992

[4]. H.P. Redmond, P.N. Stapleton and H.D. BouchierImmunonutrition. “The role of taurine”. Nutrition, vol.14, pp. 599- 604, 1998.

[5]. T.C. Birdsall. “Therapeutic applications of taurine”. Alt. Med. Rev.,vol. 3pp.128–36, 1998.

[6]. C.E.Wright, T.T. Lin, J.A. Syurman, G.E. Gaull. “Taurine scavenges oxidized chlorine in biological systems”. Taurine: biological actions and clinical perspectives vol.3, pp.137–147,1985.

[7]. M.T. Moore, J.L. Farris. “Acute and Chronic toxicity of the herbicide Stam M-4 in field and laboratory exposures”. Arch. Environ. Contam. Toxicol, vol. 33,pp.199-202, 1997.

[8]. D.C. McMillan, J.P. Freeman and J.A. Hinson. “Metabolism of the arylamide herbicide propanil. I. Microsomal metabolism and in vitro methemoglobinemia.” Toxicol. Appl. Pharmacol, vol.103,pp. 90-101, (1990 (a)).

[9]. S.S. Alam, N.A. Hafiz, A.H. Abd El-Rahim (2011). “Protective role of taurine against genotoxic damage in mice treated with metho- trexate and tamoxifen.” Environ. Toxicol. Pharmacol. vol. 31,pp.143–152, 2011.

[10]. N.C. Jain. “Schalms veterinary Haematology.” 4th edn Lea and Febiger, Philadelphia. pp.121-123, 1986.

[11]. H.H. Lo, P.I. Brown, G.O. Rankin. “Acute nephrotoxicity induced by isomeric dichloroanilines in Fischer 344 rats.” Toxicology vol.63,pp. 215–231, 1980.

[12]. M. Valentovic , J.G. Ball, S. Stoll, G.O. Rankin.“3,4-Dichlorophenylhydroxylamine cytotoxicity in renal cortical slices from Fischer 344 rats.” Toxicology,vol. 162,pp.149–156, 2001.

[13]. A.Y.K. Chow, and S.D. Murphy. “Propanil (3,4-dichloropropionanilide)-induced methemoglobin formation in relation to its metabolism in vitro.” Toxicol. Appl. Pharmacol. Vol. 33, pp. 14–20, 1975. [PubMed: 1162701]

[14]. D.C. McMillan, T.P. Bradshaw, J.M. McMillan, J.A. Hinson and D.J. Jollow. “Contribution of 3,4-dichlorophenylhydroxylamine in propanil-induced hemolytic anemia.” Adv. Exp. Med. Biol.vol. 283,pp. 343-345, (1991 (b))..

[15]. D.C. McMillan, T.A. McRae, J.A. Hinson. "Propanil-induced methemoglobinemia and hemoglobin binding in the rat." Toxicol. Appl. Pharmacol. Vol. 105, pp.503–507, (1990(b)).

[16]. M. Bidri and P. Choay. "Taurine: A particular amino acid with multiple functions." Ann. Pharm. Fr. Vol. 61, pp. 385–391, 2003.

[17]. J. Das, J. Ghosh, P. Manna, P.C. Sil. "Arsenic-induced oxidative cerebral disorders: Protection by taurine." Drug Chem. Toxicol. Vol. 32, pp. 93–102, 2003.