IJBPAS, September, 2014, 3(9): 2150-2156

ISSN: 2277–4998

International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)

'A Bridge Between Laboratory and Reader'

www.ijbpas.com

STUDY ON THE EFFECTS OF AMITRAZ ON SOME SERUM BIOCHEMICAL PARAMETERS IN RATS

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ABSTRACT

Amitraz is an insecticide of the formamidine group, initially synthesized in England in 1969, that is used as an acaricide and tickicide in veterinary medicine. The mechanism of action of amitraz in arthropods occurs by the activation of octopaminergic receptors.The aim of present study was to determine the effects of amitraz on some serum biochemical parameters in rats.40 Wistar rats, weighing 200-250g, obtained from veterinary faculty of Islamic Azad University. Animals were divided into 2 identical groups by chance. Group 1 considered as normal control which received no treatment with exception of water. Group 2 received amitraz at the dose of 200 mg/kg in drinking water for 30 days. After 30 days, blood samples were obtained from animals and after separation of serum, biochemical parameters such as Bilirubine, Ca, P, Albumin, Protein, cholesterol, Glucose, Urea, BUN, Creatinine, triglyceride, HDL, AST, ALT, ALP were measured using of commercial available kits. The results of present study showed that in rats treated with amitraz the serum value of urea, ALP and bilirubin increased significantly ($P<0.05$) and level of HDL decreased significantly ($P<0.05$). Other parameters showed difference with normal control values but it was not significant.

Keywords: Amitraz, Toxicity, Serum, Biochemical Parameters, Rats

INTRODUCTION

Amitraz is a non-systemic acaricide and insecticide **[1]**. It was first synthesized by the Boots Co. in England in 1969 **[2]**. Amitraz has been found to have an insect repellent effect, works as aninsecticide and also as a pesticide synergist. Its effectiveness is traced back on alpha-adrenergic agonist activity, interaction with octopamine receptors of the central nervous system and inhibition of monoamine oxidases and prostaglandin synthesis **[3]**. Therefore, it leads to overexcitation and consequently paralysis and death in insects. Because amitraz is less harmful to mammals, amitraz is among many other purposes best known as insecticide against mite- or tick-infestation of dogs **[1].**

Amitraz is particularly effective against acarids **[4]**, but it is used as a pesticide in many different fields. Thereforeamitraz is available in many different forms, as in a wettable powder, emulsifiable concentrate, soluble concentrate/liquid, and impregnated collar (for dogs). It is characterized as an insect repellent, insecticide and pesticide synergist. These are the properties which make it especially useful as a pesticide **[3]**. The repellent effect causes insects to turn away from their target as this is treated with amitraz **[3]**. It acts as an insecticide, which means that it can be used to control insects that are directly or indirectly harmful to men **[3]**. As apesticide synergist it also increases the effect of some other pesticides if they are combined with amitraz **[3]**. These can be traced back to the mechanisms of action, which lead to a wide field of effects, including directlethality, excitant-repellant behavioral effects, and chemosterilization for the target species **[5]**. In addition it generally causes low damage to nontarget species, which is one of the advantages of amitraz. Furthermore amitraz is especially effective against insects as spider mites and ticks in their juvenile and resistant forms **[5]**. For agricultural purposes amitraz is primarily used to control the pear psylla (Cacopsyllapyricola) on Oregon pear crops and whiteflies and mites on cotton or pear crops. It's also applied to pome fruit, citrus fruit, cotton, stone fruit, bush fruit, strawberries, hops, cucurbits, aubergines, capsicums, tomatoes and ornamental plants to control all stages of tetranychid and eriophyid mites, pear suckers, scale insects, mealybugs, whiteflies, aphids and eggs and first instar larvae of lepidoptera **[1]**. To apply amitraz, various techniques can be used as for example an airblast and concentrate spray to pears or by ground boom and aircraft to cotton. Territorial differences in amitraz use depend on the species of mites that infest the crops/trees/etc., the local practice and the number and size of the pear trees. An infestation e.g. by Tetranychusspp requires higher rates of amitraz. Taking those factors into consideration the application volumes of amitraz have been standardized in terms of maximum spray concentration and in the rate of amitraz per hectare.

Besides its application as pesticide on plants, amitraz is also used as an animal ectoparasiticide on cattle, goats, sheep, pigs and dogs **[1]**. Thereby it's exclusively applied externally **[6]**. It achieves special efficiency against mites (first of all Demodexcanis), but it also works against lice, flies and all development stages of ticks **[6, 7]**. In combination with additional agents it can be used against flea-infestation as well **[6, 7]**. For the treatment of dogs amitraz is available as a collar or as a spray- or washsolution and has an immediate effect against tick-infestation as well as a preventive effect. In some countries amitraz-emulsions are also applied to treat Demodicosis of cats or dogs, an exceeding infestation of mites of the family Demodicidae **[6, 7]**. For the treatment of cattle, sheep, goats and pigs amitraz is available as spray or washsolution, to treat or prevent infestations by mites, lice, flies and ticks. Thereby pigs and cattle should be sprayed and sheep and goats bathed **[6]**. Other animal species as for example horses or Chihuahuas should not be treated with amitraz, because adverse effects may occur **[6, 7]**. The aim of present study was to determine the effects of amitraz on some serum biochemical parameters in rats.

MATERIALS AND METHODS

The experiment was approved by the Ethical Committee. 40 Wistar rats, weighing 200-250g, obtained from veterinary faculty of Islamic Azad University. The rats were sorted by health condition and only those with physical, hematological and biochemical normal values were used. The day before running the experiment, the rats were socially isolated and held in individual stainless steel cages under 12:12 artificial light-dark cycle, room temperature at about 25°C, and fed ad libitum. Animals were divided into 2 identical groups by chance. Group 1 considered as normal control which received no treatment with exception of water. Group 2 received amitraz at the dose of 200 mg/kg in drinking water for 30 days. After 30 days, blood samples were obtained from animals and after separation of serum, biochemical parameters such as Bilirubine, Ca, P, Albumin, Protein, cholesterol, Glucose, Urea, BUN, Creatinine, triglyceride, HDL, AST, ALT, ALP were measured using of commercial available kits.

Data were analyzed using SPSS software and T-test statistical method and p<0.05 considered as statistical significance.

RESULTS

The results of present study showed that in rats treated with amitraz the serum value of urea, ALP and bilirubin increased significantly (P<0.05) and level of HDL decreased significantly (P<0.05). Other parameters showed difference with normal control values but it was not significant **(Table 1)**.

Table 1: The Effect of Administration of Amitraz on Some Biochemical Factors

Group / Parameter	Control	Treatment	P value
Glucose (mg/dl)	119.8±0.86	111±2.46	0.087
Urea (mg/dl)	37.5±1.6	49±2.46	0.043
BUN (mg/dl)	20.48±0.43	22.87±1.16	0.361
Creatinine (mg/dl)	1.26±0.05	1.38±0.08	0.399
ALT (IU/L)	63.6±1.36	60±0.63	0.097
AST (IU/L)	92.2±3.9	92±0.7	1.00
ALP (IU/L)	48.6±3.12	56±0.63	0.046
Bilirubine (mg/dl)	0.41±0.01	0.52±0.02	0.003
Ca (mg/dl)	8.3±0.12	8.1±0.47	0.981
P (mg/dl)	4.28±0.11	4.2±0.39	0.997
Albumin (g/dl)	2.2±0.1	2.34±0.1	0.607
Protein (g/dl)	5.84±0.18	5.9±0.15	0.995
Cholesterol (mg/dl)	45.4±1.69	41±1.58	0.321
HDL (mg/dl)	43.6±0.24	39.2±0.37	0.000
Triglyceride (mg/dl)	69.6±6.94	86±8.72	0.239

DISCUSSION AND CONCLUSION

Amitraz is an insecticide of the formamidine group, initially synthesized in England in 1969, that is used as an acaricide and tickicide in veterinary medicine **[8]**. The mechanism of action of amitraz in arthropods occurs by the activation of octopaminergic receptors. In mammals, the main mechanism of action consists of activation of α2-adrenoceptors similar to the mechanism of action of xylazine and clonidine, agonist α2-adrenergics, and in the inhibition of the enzyme monoaminoxidase (MAO) **[9]**.

Clinical signs occur due the α2-adrenergic effects nduced by amitraz and include: sedation, loss of reflexes, lethargy and incoordination, bradycardia, bradyarrhythmia, hypotension, hypothermia, polyuria, transitory hyperglycemia, emesis, mydriasis and decrease of intestinal motility **[8, 9]**.

Amitraz is approved in Brazil by the Ministry of Agriculture for use in bovines, swine, sheep and dogs **[8]**. Although its use is not recommended for cats by the manufacturer, it is an effective treatment of feline scabies and demodicosis **[10, 11]** and is a very popular product, cheap and readily available, which could be an excellent alternative for acaricide treatment in this species **[8]**. However, few reports of amitraz intoxication are related to cats **[8, 12]**. Thus, the objectives of the present study were to investigate experimental amitraz intoxication and record its effect on some biochemical factors.

Amitraz toxicity has been studied in animals of different species. So that administration of Amitraz in rats at a dose of 4 mg/kg for 28 days showed that the concentration of toxin is bigger in the tissues of the thyroid, adrenal, liver, kidney, skin and eyes in compared with other tissues.

Amitraz effects on enzymes and some biochemical parameters in mice has been investigated and it was found that oral administration at a dose of 100 mg/kg with corn oil for 2 weeks cause mortality, increased liver microsomal enzymes, increase liver weight and increased oxidative enzyme activity in mice.

Amitraz acute toxicity in mice and rats is identified with increased irritability, ataxia, tremors and paralysis. Rats receive Amitraz at a dose of 200 mg/kg show irritable bladder spasms. Weakening of the central nervous system, decreased rectal temperature, decreased heart rate and respiratory rate is observed in rabbits 48 hours after administration of Amitraz at a dose of 100 mg/kg **[13]**.

The Amitraz toxicity in dogs at a dose of 20-100 mg/kg is determined with weaken the central nervous system imbalance, muscle spasms, loss and decreased rectal temperature and pulse rate. Increased levels of glucose, blood urea nitrogen and serum potassium concentration are other Amitraz toxicity in dogs. It has been shown in studies of acute Amitraz toxicity and Biochemical changes in tissues of Wistar rats. So after 7 days of administration of Amitraz at dose of 200 mg/kg, irritability, weight loss, elevated alkaline phosphatase, elevated bilirubin and increased ALT and AST activity and increased in serum sodium and potassium concentrations was observed **[14]**.

The rise in both AST and ALT levels ($p<0.05$) in mice given Amitraz was one of the most familiar indicators of hepatocellular damage. Also, **[15]** had also reported an increase in serum AST levels in mice given amitraz. However, a significant decrease in ALP level may refer to liver dysfunction. Besides, increasing bilirubin levels indicated diffused harm to the liver.

[16] revealed the amitraz effects on liver biochemical concentrations, whereas propolis led to a significant decrease in these levels in treated group. But, hepatocytes of mice treated with amitraz + propolis demonstrated positive stained nuclei, by using Ki67 immunostaining, less than those of amitraz treated only.

In a study by **[17]** it shown that Increase in urea, phosphorous, aspartate transaminase and alanine amino transferase values were observed only in the group given 45 mg/kg body weight amitraz. A decrease in creatinine and alkaline phosphatase concentrations was observed in all groups. No differences were observed in serum calcium and bilirubin concentrations. No pathological changes were detected in the kidneys and livers.

[15] indicated that features of toxicity were depression, incoordination of movement, paresis of the limbs, hepatonephrotoxicity, muscular hemorrhage at site of injection and peritonitis following i.p. injection. These changes, accompanied by leucopenia, were correlated with alterations in serum AST and concentrations of serum constituents. Amitraz did not inhibit serum ChE activity.

In conclusion it can be driven that the clinical signs observed in Rats intoxicated byamitraz were: sedation, loss of reflexes, hypothermia, bradycardia, bradyarrhythmia, hypotension, bradypnea and mydriasis. Some Rats also presented vomiting, diuresis, sialorrhea, vocalization and increased appetite. Metabolicand hormonal alterations were transitory hyperglycemia and hipoinsulinemia and transitory decrease of plasma levels of cortisol. Amitraz intoxication in Rats increased urea, ALP and bilirubin and decreased HDL concentration.

REFERENCES

[1] Corta E, Bakkali A, Berrueta LA, Gallo B, and Vicente, F, Kinetics and mechanism of amitraz hydrolysis in aqueous media by HPLC and GC-MS. Talanta, 48(1), 1999, 189-199.

[2] Harrison IR, Kozlik A, McCarthy JF, Palmer BH, Wakerley SB, Watkins TI and Weighton DM. 1,3,5-Triazapenta-1, 4-dienes: Chemical aspects of a new group of pesticides, Pesticide Sci., 4 (6), 1973, 901-910.

[3] Bonsall JL and Turnbull GJ, Extrapolation from safety data to management of poisoning with reference to amitraz (a formamidine pesticide) and xylene, Hum. Toxicol. 2(4), 1983, 587-92.

[4] Brown PM, Toxicological problems associated with the manufacture of triazapentadienes, Proceedings of the Royal Society of Med., 70 (1), 1977, 41-43.

[5] Hollingworth RM, Chemistry, biological activity, and uses of formamidine pesticides.

Environmental Health Perspectives, 14, 1976, 57-69.

[6] Peter R, De Bruin C, Odendaal D and Thompson PN, The use of a pour-on and spray dip containing Amitraz to control ticks (Acari: Ixodidae) on cattle, J. S. Afr. Vet. Assoc., 77 (2), 2006, 66-9.

[7] Tarallo VD1, Lia RP, Sasanelli M, Cafarchia C and Otranto D, Efficacy of Amitraz plus Metaflumizone for the treatment of canine demodicosis associated with Malasseziapachydermatis, Parasit Vectors, 2 (1), 2009, 13.

[8] Andrade SF and Sakate M, Intoxicaçãoporamitraz. Vet. Not., 10, 2004, 101-109.

[9] Hsu WH, Antiparasitic Agents, In: AHRENS, F.A. (Ed), Farmacol., Baltimore: Willians and Wilkins, 1996, 243-260.

[10] Cowan LA and Campbell K, Generalized demodicosis in a cat responsive to amitraz, J. Am. Vet. Med. Assoc., 192, 1988, 1442-1444.

[11] Scott DW, Miller WH and Griffin CE, (Eds), Dermatologia de pequenosanimais, Rio de Janeiro: Interlivros, 1996, 1130.

[12] Soli NE, Braseth TA, Ectodex poisoning in a cat reversed with Antisedan, Norsk-Veterinaertidsskrift, 104, 1992, 25-26.

[13] Costa LG, Gastel J and Murphy SD, The formamidine pesticides chlordimeform and amitraz decrease hepatic glutathione content in mice through an interaction with alpha2-adrenoceptors, J. Toxicol. Environ. Health, 33, 1991, 349-358.

[14] Grossman MR, Amitraztoxicosis associated with ingestion of an acaricide collar in a dog, J. Am. Vet. Med. Assoc., 203, 1993, 55-57.

[15] Al-Qarawi AA, al-Damegh MS and Adam SE, Effects of amitraz given by different routes on rats, Vet. Hum. Toxicol., 41 (6), 1999, 355-7.

[16] El-Kott AF and Owayss AA, Protective Effects of PropolisAgainst the Amitraz Hepatotoxicity in Mice, J. Pharmacol. and Toxicol., 3, 2008, 402-408.

[17] Filazi A, Sireli M and Kalkan F, The influence of amitraz on biochemical parameters in mice, Hum. Exp. Toxicol., 22 (2), 2003, 99-101.