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EFFECTS OF AMITRAZ ON THE ARTERIAL BLOOD PRESSURE AND BODY RECTAL TEMPERATURE OF CONSCIOUS RATS*

EFEITOS DO AMITRAZ SOBRE A PRESSÃO ARTERIAL E A TEMPERATURA RETAL DE RATOS

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SUMMARY

The present study examined the effects of acute amitraz (100.0 mg/kg) administration on arterial blood pressure and body rectal temperature of male Wistar rats. Amitraz decreased arterial blood pressure and also produced hypothermia. Tyramine (100.0 mg/kg) administration induced a significant ($p < 0.05$) increase in the pressure of the rats dosed with the pesticide, but did not change the arterial blood pressure of the rats in the control group. Half of the rats given amitraz plus tyramine, but not the control solution plus tyramine died 3 to 10 hr after the drug administration. The indicative signs of intoxication included sedation, motor incoordination and coma. Yohimbine (10.0 mg/kg) did not change amitraz-induced hypotermia ($p < 0.05$). Although possible action of amitraz on brain alpha$_2$ noradrenergic receptors could not be excluded, the results were interpreted on a possible monoaminoxidase inhibitor (MAOI) - like action for the pesticide.

UNITERMS: Amitraz; Pesticides; Monoamine oxidase inhibitors

INTRODUCTION

Amitraz is a pesticide widely used in veterinary clinical practice for the treatment of demodicosis (MÜLLER[19], 1983; LARSSON and GONÇALVES[16], 1986). Acute intoxications with this pesticide are uncommon. Nevertheless, sedation, loss of the righting reflex, motor incoordinations and other overt symptoms of Central Nervous System (CNS) depression were described after acute amitraz poisonings (FOLZ et al.[8], 1984; LARSSON; GONÇALVES[16], 1986; HSU; SCHAFFER[13], 1988). Other symptoms reported after amitraz intoxications included intestinal impactation and colics (ROBERTS; ARGENZIO[23], 1986). A dose of 800.0 mg/kg was reported as the oral acute amitraz LD_{50} for rats (HOLLINGWORTH[12], 1976).

Amitraz was recently described to decrease the motor activity of adult rats in an open-field, probably through a MAOI-like interference with CNS function (FLÓRIO et al.[7], 1993). In the experiment, the pesticide increased the brain levels of noradrenaline, dopamine and serotonin, at the same time decreasing the levels of homovanilic and 5-hydroxyindoleacetic acids. An effect of amitraz on alfa$_2$-adrenoceptors was described and related to its toxiciy (GILBERT; DYER[10], 1988; FLÓRIO et al.[7], 1993). The present experiment investigates the effects of amitraz on the arterial blood pressure and body rectal temperature of rats in an attempt to better characterize the mechanisms of the pesticide toxicity.

MATERIAL AND METHOD

Animals. Fifty-six genetically similar male Wistar rats weighting 220-250 g were used. Seven days before the experiments, the rats were housed in groups of 3 in wire mesh cages (16 x 30 x 19 cm), in a temperature controlled room (22ºC ± 1) with a 12 hr-light-dark cycle (lights on 7:00 AM). Food and water were provided at *ad libitum* consumption, except during experimental periods when they were withdrawn.

Drugs and dosing. Amitraz (Coopers Co, São Paulo, Brasil); tyramine hydrochloride and yohimbine hydrochloride (Sigma Chemical Co, St Louis, Mo) were used. The drugs were prepared in distilled water immediately before use, except amitraz and yohimbine which were suspended in distilled water with Tween-80. This vehicle alone was used as amitraz and yohimbine control solutions. Amitraz, control solutions and tyramine were administered *per os* to rats; the i.p. route was used for yohimbine. Doses were given in volumes not greater than 2.0 ml/kg body weight.

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Blood Pressure Studies. Sixteen rats were randomly and equally divided in to control and experimental groups. The arterial blood pressure (Basal pressure = Bp) was measured in all rats and 24 hr after the experimental rats received 100.0 mg/kg of amitraz, while those in the control group received the same volume (2.0 ml/kg) of control solution; 90 min later, the arterial blood pressure (Po) were measured in both groups for the 2nd time. Immediately after that, all the rats received 100.0 mg/kg of tyramine, being studied 60 min later for the 3rd arterial blood pressure (P_{60}) determination.

Arterial blood pressure was measured by tail plesthymography as described by CORDELINI et al.[4] (1990) in unanesthetized rats using an E & M Instrument Co. programmed electrosphygmomanometer (Narco BioSystem, Texas, USA). The rats were placed for 10 min in a warming chamber maintained at 40ºC in order to allow pulse rates to be recorded; the cuff pressure then was monitored automatically, since systolic beats were detected.

Body rectal temperature studies. Forty rats were randomly and equally divided in 2 experimental groups (E_1 and E_2) and 2 control groups (C_1 and C_2). The animals basal retal temperature (Bt) was measured sixty min after, and the experimental animals received 100.0 mg/kg of amitraz as well as the controls received the same volume (2.0 ml/kg) of control solution; ninety min later, groups C_1 and E_1 received distilled water (2.0 ml/kg) and those of groups C_2 and E_2 received 10.0 mg/kg of yohimbine; sixty min later, the body rectal temperatures were determined in all animals.

Body temperature was measured in ambient temperature (22ºC ± 1) in unanesthetized animals using a digital thermometer (Digitrom, São Paulo, Brasil) coupled to a transducer; the thermocouple was inserted 2.5 cm into the rectum. During the measurements, the rats were housed individually.

Statistical analysis. Since homocedasticity is necessary for the analysis of variance, Bartlet's test (JOHNSON; LEONE[14], 1974) was performed. It was concluded that the present results were parametric. Thus, Student's "t" test and ANOVA, followed by Duncan's test (DUNCAN[6], 1955) were used to analyse the obtained data, and were considered significant when $p < 0.05$.

RESULTS

Blood pressure studies. Amitraz decreased ($p < 0.05$) the arterial blood pressure of rats (Fig. 1). When compared to the blood pressures recorded before amitraz administration (Bp), the pesticide induced a 21.2% decrease in these values (P_0). Fig. 1 also shows that the effects of tyramine administration were completely different in the control and experimental groups, whereas tyramine did not change the arterial blood pressure of the control rats (P_{60}), it induced a significant increase ($p < 0.05$) in the P_{60} values of the rats dosed with amitraz. The arterial blood pressure of the rats dosed with 100.0 mg/kg of amitraz plus 100.0 mg/kg of tyramine was 21.2% higher than that recorded before dosing (Bp). Half of the rats dosed with amitraz plus tyramine died 3 to 10 hr after the treatments, but none of the rats dosed with control solution plus tyramine. The indicative signs of intoxication included sedation, motor incoordination and coma.

Body rectal temperature studies. Tab. 1 shows that the 100.0 mg/kg amitraz and 10.0 mg/kg yohimbine dosings did not change ($F = 27.85$; $df = 3/36$; $P < 0.05$) the rectal temperature (Ft). Compared to the data from group C_1 (control solution) both amitraz (group E_1) and yohimbine (group C_2) decreased ($p < 0.05$) the body temperature (Tf) of the rats. There were no differences between the data from groups C_2 and group E_2, i.e., yohimbine did not modify amitraz effects on the rectal temperature of the rats. Similar results were also observed ($F = 15.79$; $df = 3/36$; $p < 0.05$) on the differences (Bt - Ft) in the core temperatures of the different groups.

DISCUSSION

Acute amitraz administration had a hypotensive action in normotensive unanaesthetized rats. This is similar to the observation of TABEI et al.[27] (1970) with pargyline. These authors were the first to demonstrate the antihypertensive action of a MAOI inhibitor. This observation agress with the described MAOI effects on pressor responses (SCHULS et al.[25], 1989). MAOI agents are thought to lower blood pressure, either by a peripheral (SCHÖEPKE; SWEET[24], 1967) or central mechanism (FUENTES et al.[9], 1979). There is evidence suggesting a sustained increase in central noradrenergic transmission after MAOI administration, with a resultant decrease in peripheral sympathetic activity (MURPHY et al.[20,21], 1979, 1982).

The pressor response to tyramine, which has been used clinically to diagnose MAOI inhibition (BIECK; ANTONIN[2], 1982), is augmented by amitraz. This result is supported by other work with MAOI agents (BENEZET et al.[1], 1978; BONSALL; TURBULL[3], 1983; MOSER; MACPHAIL[18], 1985) and by the present data related to the effects of the pesticide on rats arterial blood pressure. They indicate that amitraz inhibits MAOI activity, as well as suggest a pesticide effect on MAOI type A. Inhibition of MAOI type A (by clorgyline) enhanced tyramine pressor responsiveness more strongly than inhibition of MAOI type B (by l-deprenyl) (KERECSEN; BUNAG[15], 1989).

Amitraz also induced a decline in the rectal body temperature of the rats. This is similar to observations after dosing with different alpha-noradrenergic agonists, such as norepinephrine, clonidine and phenylephrine, when they are micro-injected directly into the anterior hypothalamic/preoptic area of the rats (MYERS et al.[22], 1987). Contrary to that observed for clonidine, yohimbine did not antagonize the hypothermic effects of amitraz. Yohimbine, a selective alpha$_2$-noradrenergic antagonist (STARKE; ALTMANN[26], 1973) was able to inhibit the thermolytic response produced by clonidine (MYERS et al.[22], 1987). Although differences in yohimbine doses, routes of administration and animals studied could account for the presently observed discrepancies, as suggested elsewhere (MYERS et al.[22], 1987; MAJ et al.[17], 1988), it should not be forgotten that amitraz elevates brain levels of serotonin and decreases the levels of 5-hydroxyindoleacetic acid, by supressing MAOI activity (FLÓRIO et al.[7], 1993). Serotoninergic receptors activated directly (MAJ et al.[17], 1988) or indirectly through cholinergic (GLICK; MARSANICO[11], 1974) or dopaminergic (COX; LEE[5], 1975; YAMADA et al.[29], 1988) neurons are associated with the production of hypothermia. The presently observed hypothermia, after amitraz dosing could also indicate a MAOI-like activity for the pesticide.

The observation that yohimbine produced hypothermia "per se" might seem paradoxical in light of the above discussion. However, the net action on body temperature after administering yohimbine may reflect direct blockage of central noradrenergic postsynaptic receptors (WEINER[28], 1987). The physiologic (eg thermic) response induced by yohimbine could be identical to that induced directly (via activation of presynaptic alpha$_2$-autoreceptors), or indirectly (via MAOI inhibition) by amitraz.

Taken together, the possibility is raised that amitraz may have a MAOI-like effect on the CNS. However, an effect of the pesticide on alpha$_2$-adrenoceptors can not totally be excluded by the present data. As future experiments may bring about a better understanding of our present results, they may also allow better characterization of the neural pathways related to the observed effects of amitraz.

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TABLE 1
Effects of yohimbine (10.0 mg/kg) on amitraz (100.0 mg/kg) induced hypothermia in rats. São Paulo, 1991.

Groups[b]	Body rectal temperature (°C)[a]		
	Basal (Bt)	Final[c] (Ft)	Differences (Bt - Ft)
C_1 (CS + CS)	37.2 ± 0.6	37.5 ± 0.5	0.2 ± 0.1
E_1 (A + CS)	37.5 ± 0.5	35.0 ± 1.0*	2.3 ± 1.3*
C_2 (CS + Y)	37.4 ± 0.4	35.31 ± 1.0*	2.3 ± 1.3*
E_2 (A + Y)	37.3 ± 0.4	34.8 ± 0.4*	2.9 ± 0.6*

(a) Means ± SD.

(b) Treated with control solution (CS), amitraz (A) or yohimbine (Y).

(c) Observed 150 min after A or CS and 60 min after Y or CS treatments.

* $p < 0.05$ in relation to group C_1 (ANOVA + Duncan's test)

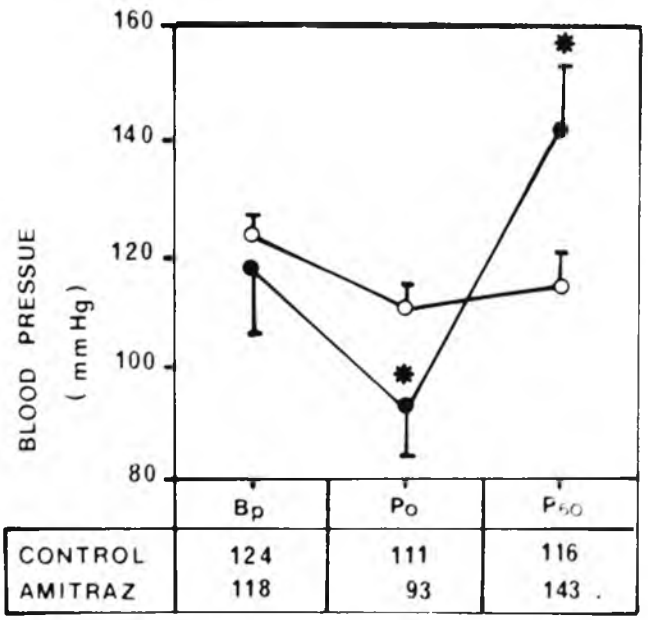

FIGURE 1
Mean (± SD) blood pressure (mm/Hg) in rats treated with 100.0 mg/kg of amitraz + 100.0 mg/kg of tyramine. Bp = basal pressure observed before the dosing; P_0 = pressure recorded 90 min after administering amitraz (black circles) or control solution (white circles); P_{60} = pressure registered 60 min after tyramine administration. * = $p < 0.05$ (Student's "t" test).

RESUMO

O presente trabalho estuda os efeitos da administração única de amitraz (100 mg/kg) sobre a pressão sangüínea arterial e a temperatura corporal de ratos Wistar. A administração de amitraz diminuiu a pressão sangüínea arterial e também produziu hipotermia. A administração de tiramina (100,0 mg/kg) induziu um aumento significativo ($p < 0.05$) na pressão sangüínea arterial de ratos que receberam previamente amitraz, não alterando a dos ratos controle. Metade dos ratos que receberam amitraz e tiramina morreram entre 3 a 10 horas após a administração, fato este não ocorrido com os animais que receberam solução fisiológica e posteriormente tiramina. Os sinais de intoxicação dos animais experimentais incluíram sedação, falta de coordenação motora e coma. A posterior administração de ioimbina (10,0 mg/kg) não alterou a hipotermia produzida pela administração de amitraz ($p < 0.05$). Apesar de não poder ser excluída uma possível ação do amitraz sobre alfa$_2$-noradrenoceptores cerebrais, os resultados indicam uma provável ação deste praguicida inibindo a enzima monoaminoxidase.

UNITERMOS: Amitraz; Pesticidas; Inibidores da monoaminoxidase

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