INVESTIGATION OF SOME EFFECTS OF LEVAMISOLE ON DOG BLOOD PRESSURE

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Summary : The effects of levamisole were investigated on the blood pressure of anaesthetized dog. Levamisole (0.5 to 4.0 *mg/kg*) elicited a biphasic effect, an initial brief depressor response followed by a pressor response. The pressor response was dose-related and was blocked by phenoxybenzamine. The residual depressor response was blocked by propranolol. Repeated administration of a high dose of levamisole produced tachyphylaxis. The pressor response to levamisole was not modified by either reserpinization, acute bilateral adrenalectomy or pretreatment with cocaine, whereas pretreatment with dexamethasone, nialamide or pyroaallol shifted the dose-response curve to the right. Levamisole potentiated the pressor responses to noradrenaline, angiotensin and acetylcholine. The effects of levamisole are ascribed to inhibition of monoamine oxidase, catechol-0-methyl transferase, catecholamine uptake$_2$ mechanism and cholinesterase.

Key words : levamisole dog blood pressure biphasic effect tachyphylaxis

INTRODUCTION

Levamisole is a recently introduced broad-spectrum anthelmintic. Levamisole improves mood and psychotonicity in depressed patients (1, 2). The therapeutic dose produces depression of central nervous system and occasional muscular twitching (5, 11). Doses greater than this produce salivation and frequent muscular tremors.

The present study was undertaken to investigate the mechanism of action of some autonomic effects of levamisole.

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MATERIAL AND METHODS

Healthy mongrel dogs of either sex (10 to 16 *kg*), were anaesthetized with pentobarbitone sodium (30 *mg/kg*, iv) with supplementary doses as needed. Blood pressure was recorded from left common carotid artery through mercury manometer on smoked kymograph. Drugs were given iv as solutions in normal saline through right femoral vein.

Effect of levamisole on the blood pressure : The dose-response curve of levamisole was elicited by spacing the doses (0.5, 1.0, 2.0 and 4.0 *mg/kg*) at 15 min intervals. The effect of levamisole (4.0 *mg/kg*), administered repeatedly at 5 min intervals was also studied.

Modification of the effects of levamisole by various procedures : Pressor responses to levamisole (4.0 *mg/kg* and 8.0 *mg/kg*), were elicited before and after one hr of injection of phenoxybenzamine (2.0 *mg/kg*) or propranolol (1.0 *mg/kg*).

Dose-response curves of levamisole were obtained before and 15 min after treatment with 2.0 *mg/kg* of cocaine or 30.0 *mg/kg* dexamethasone or 200 *mg/kg* of pyrogallol and 1 hr after 50 *mg/kg* of nialamide : to study the effect of combination of pyrogallol and nialamide, the animals were first treated with nialamide and 45 min later with pyrogallol.

Reseprine (5.0 *mg/kg*, ip) was injected 36 hr before the experiments. Reserpinization was confirmed by absence of responses to tyramine and dose response curve of levamisole was elicited in reserpinised dogs. Control dose-response curves of levamisole and responses to dimethylphenyl-piperazinium (DMPP; 2 $\mu g/kg$) were elicited before and after acute bilateral adrenalectomy performed by dorsal approach.

Modification of reponses to various agonists by levamisole : The control responses to noradrenaline (NA; 1.0 $\mu g/kg$), angiotensin (ANG; 50 $\mu g/kg$) and isoprenaline (ISO; 1.0 $\mu g/kg$), histamine (HIS ; 1.0 $\mu g/kg$) and acetylcholine (ACH; 1.0 $\mu g/kg$) were elicited before and after levamisole (4.0 *mg/kg*).

All results were expressed as the mean $\triangle$ blood pressure (the change from resting blood pressure).

Student's 't' test was employed to determine the level of significance.

The following drugs were used : levamisole hydrochloride (Khandelwal, Bombay), noradrenaline bitartarate (Sigma, St. Louis), angiotensin amide (Ciba Summit, New Jersey), isoprenaline sulphate (Ward, Blenkinsop & Co., London), histamine acid phosphate (BDH, England), tyramine mono-hydrochloride (Hoffman-la Roche, Basle), phenoxybenzamine hydrochloride (SKF, Philadelphia) ; reserpine (Serpasil, Ciba Geigy, Bombay), 1-dimethyl-4-phenyl-piperazinium (Parke Davis, Detroit), nialamide (Pfizer, Bombay), Pyrogallol (BDH, England), pentobarbitone sodium (Abbot Laboratories Bombay) and dexamethasone (Pharmaceutical Company of India, Bombay).

RESULTS

Effects of levamisole on blood pressure of dogs : Levamisole elicited biphasic effects on the blood pressure. It produced an initial brief depressor response (10 to 15 mm Hg; duration about 5 sec) which was not dose-related and was well marked at low doses (0.5 *mg/kg*). This was followed by a pressor response, The rise in pressure was immediate and dose dependent, lasted for 30 to 150 seconds. It was dose-related in the dose range of 0.5 to 4.0 *mg/kg*. On repeated administration of a high dose (4,0 *mg/kg*) of levamisole, there was a progressive decrease of the pressor response (Table I).

TABLE I : Effect of levamisole on the blood pressure (mm Hg) of anaesthetized dogs.

	Levamisole mg/kg, iv (n = 6)	*Mean rise in blood pressure (mm Hg) ± S.E.M.*
A	0.5 *mg*	18.0 ± 3.75
	1.0 *mg*	28.88 ± 4.15
	2.0 *mg*	53.66 ± 5.32
	4.0 *mg*	74.1 ± 4.9
B	4.0 *mg* (I)	70.0 ± 7.5
	(II)	52.0 ± 4.5
	(III)	36.0 ± 6.5
	(IV)	25.0 ± 6.0

Levamisole was given at 15 min intervals (A) or the dose was repeated at 5 min intervals (B)

Effects of ∝ and β-adrenoceptor antagonists : After phenoxybenzamine (2.0 *mg/kg*) the pressor response (74.1 ± 4.9 mm Hg, n = 6) was totally blocked ; depressor response was not altered. At this stage 4.0 *mg/kg* and 8.0 *mg/kg* levamisole produced only depressor responses of 20.0 ± 5.0 mm Hg and 36.0 ± 4.0 mm Hg, respectively (n=6). Propranolol (1.0 *mg/kg*) totally blocked these depressor responses.

Effects of "uptake blockers" : Responses to levamisole (0.5 to 4.0 *mg/kg*) were not modified by prior administration of the uptake$_1$ blocker cocaine (2.0 *mg/kg*) whereas uptake$_2$ blocker dexamethasone (30 *mg/kg*) shifted the dose-response curve of levamisole to the right with significant ($P < 0.05$) depression of maxima (Fig. 1).

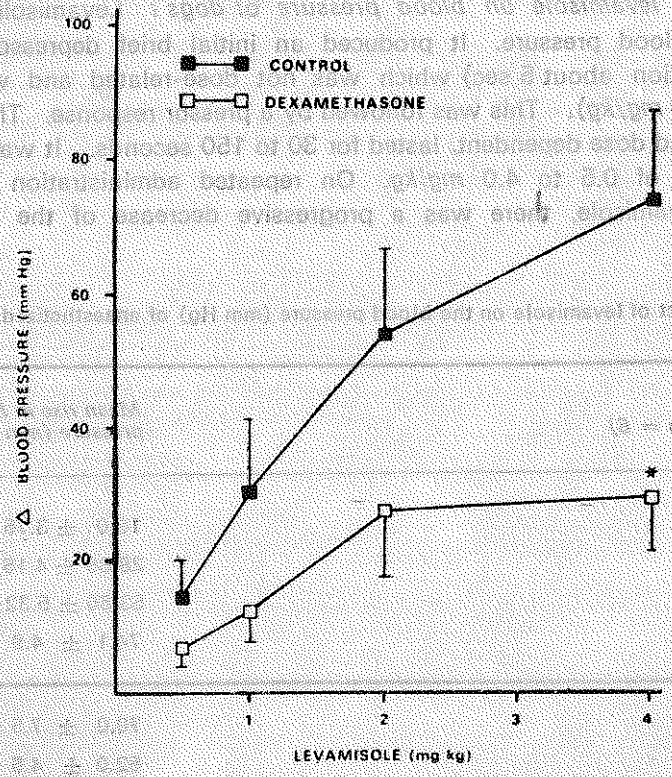

Fig. 1 : Dose response curve of levamisole before (■ — ■) and after pretreatment with dexamethasone (30 *mg/kg*, iv, □ — □) in anaesthetized dogs. Values are means from 6 experiments (vertical bars ± S.E.M.).

* Differs significantly from the control ($P < 0.05$).

Effects of reserpinization and acute bilateral adrenalectomy : Pressor responses to levamisole (0.5 to 4.0 *mg/kg*) were not modified by reserpinization (n=5) or by acute bilateral adrenalectomy (n=5).

Effects of nialamide and pyrogallol pretreatment alone and in combination : Pretreatment with nialamide or pyrogallol produced 30 to 50% inhibition of the pressor responses to levamisole. Prior administration of both nialamide and pyrogallol produced 82 to 90% inhibition of the pressor responses to levamisole (Fig. 2).

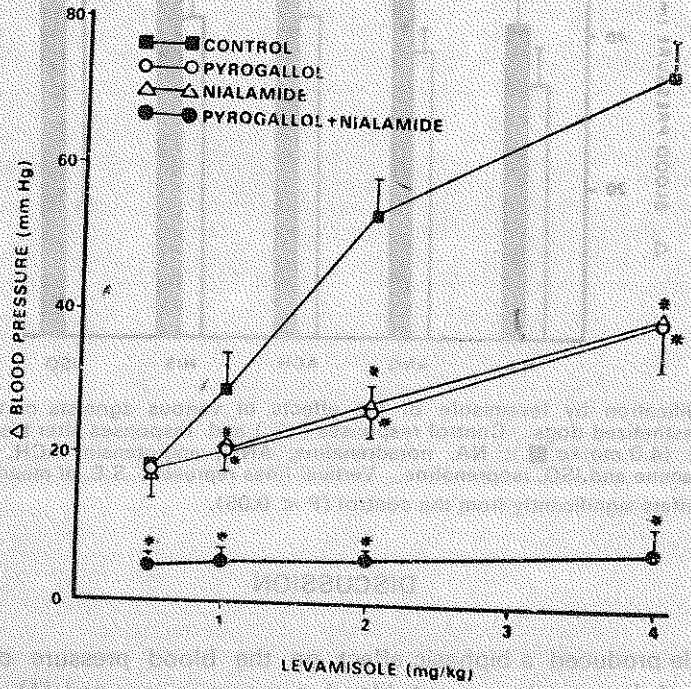

Fig. 2 : Modification of the pressor responses to levamisole by pyrogallol (200 *mg/kg*) and/or nialamide (50 *mg/kg*) in anaesthetized dogs. Control dose-response curve (■ — ■). Dose-response curves after treatment with pyrogallol (○ — ○), nialamide (△ — △) and pyrogallol + Nialamide (● — ●). Vertical lines represent S.E. of means (n = 5). *Differ significantly from the control ($P < 0.05$).

Modification of the effects of various agonists by levamisole : Levamisole (4.0 *mg/kg*) potentiated the pressor responses to NA, ANG and the depressor responses to ACH significantly ($P < .05$) but had no effect on those to HIS and ISO (Fig. 3). Levamisole did not further potentiate the pressor response to NA (4 *μg/kg*) after cocaine pretreatment (control response 80 ± 3.2 mm Hg : after cocaine, 130 ± 3.5 mm Hg : after levamisole 132 ± 4 mm Hg ; n=5).

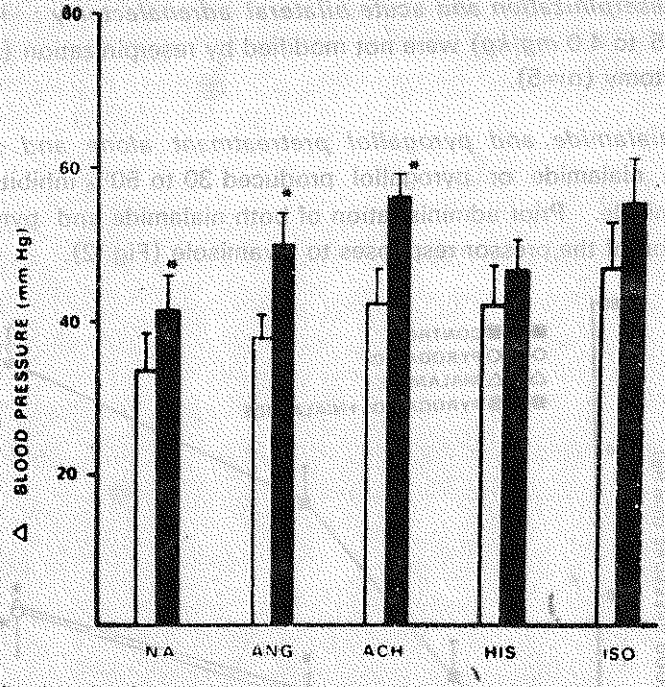

Fig. 3 : Modification by levamisole of the effects of various agonists on the blood pressure of anaesthetized dogs. Control responses (□) and responses after treatment with levamisole (4.0 *mg/kg* ■); NA, noradrenaline; ANG, angiotensin; ACH, acetylcholine; HIS, histamine and ISO, isoprenaline. Vertical lines represent S.E. of means (n = 6).

* Differs significantly from the control ($P < 0.05$).

DISCUSSION

Levamisole produced a biphasic effect on the blood pressure of dog. Biphasic pressure effect of levamisole on cat blood pressure is reported (4). The pressor and depressor effects of levamisole were blocked by phenoxybenzamine and propranolol respectively, suggesting that the pressure effects of levamisole were adrenergically mediated.

The immediate occurence of pressor response suggested a direct action while its persistance suggested an indirect action. Tachyphylaxis to pressor response observed supports the latter action. Adrenalectomy and reserpinization did not alter the response to levamisole which excludes adrenal medulla and reserpine-sensitive catecholamine stores, respectively, as the sites of action of levamisole. Cocaine, a prototype neuronal uptake blocker did not alter the response to levamisole. In contradiction to the present

findings, it was demonstrated (10) that levamisole blocked the uptake of 3H-NA in dog saphenous vein and in the guinea pig heart levamisole had half the potency of cocaine in blocking the neuronal uptake (8). The reason for this discrepancy is not clear.

Responses to levamisole were blocked significantly by prior administration of MAO or COMT inhibitors alone or in combination, suggesting that the action of levamisole may be partly through inhibition of MAO and COMT. In this respect our observations are at least partly in agreement with those of Vanhoutte *et al.* (10) who demonstrated a MAO inhibitory action of levamisole. Thus the conclusion that levamisole may produce its pressor effect by blocking the enzymes MAO and COMT fits in with the conclusion about the NA-releasing action of levamisole (10).

The potentiating effect of levamisole on the pressor responses to NA observed in the present study could also be explained on basis of its MAO and COMT inhibitory effect.

Responses to levamisole were blocked by prior administration of dexamethasone, an uptake$_2$ blocker (9) suggesting that levamisole might also have atleast in part, the same mode of action as dexamethasone i.e. by occupation of uptake$_2$ sites and making NA free for action at the receptor site.

While diffusing to the site of extraneuronal uptake a considerable proportion of NA is O-methylated. A block of COMT by levamisole as suggested above would result in increase in the concentration of the amine at the site of extraneuronal uptake as well as an increase in extraneuronal accumulation. Thus a close relationship between extraneuronal uptake of NA and COMT inhibition envisaged from the present data is in line with the literature evidence (3). It is postulated that uptake$_2$ and metabolism might be the predominent mechanism for NA inactivation in tissues such as vascular smooth muscle in certain parts of which the density of sympathetic innervation is low (6).

The anti-depressive action of levamisole (1, 2) might be explained on the basis of its inhibitory action of MAO as proposed above.

Potentiation of responses to ACH by levamisole may be explained by its inhibitory effect on cholinesterase (7). Muscular twitchings, tremors and salivation observed during clinical use of levamisole (5, 11) may be ascribed to potentiation of the cholinergic influences

In conclusion the effect of levamisole on dog blood pressure may be based on actions like MAO and COMT inhibition and blockade of catecholamine uptake$_2$ process.

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