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Pharmacologic Characterization of Reserpine Responses in Rats Pre-Treated with Iproniazid.* (23878)

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Reserpine is known primarily for its hypotensive and sedative activities(1). It causes also liberation of pressor amines, notably epinephrine, norepinephrine and serotonin(2). Pressor activity consequent on release of these substances has been unmasked in anesthetized dogs and cats(3-5) by pre-treatment with iproniazid, a mono-amine oxidase inhibitor and attributed to impaired degradation of norepinephrine(4). In conscious mice, rats and rabbits, pre-treatment with iproniazid altered the response to reserpine from sedation to excitation(3,6); this change was attributed primarily to release of serotonin and its delayed inactivation(7,8). The present report concerns effects of reserpine on arterial pressure and behavior of anesthetized and conscious rats pre-treated with iproniazid and attempts to characterize the factors involved in these responses.

Methods. Experiments were done in male Sprague-Dawley rats weighing 175-255 g each. Except when indicated, anesthesia was induced with sodium amytal (9 mg/100 g body weight intraperitoneally) and vascular reflexes were inhibited with pentolinium bitartrate (5 mg solution in 20% polyvinylpyrrolidone subcutaneously); blood pressure was recorded following carotid cannulation (9). When given, iproniazid phosphate (25 mg intraperitoneally) was injected 18-24 hours before the test. Free reserpine dissolved in glycol vehicle was administered intraperitoneally in a dose of 5 mg/kg after arterial pressure had stabilized. Drugs used in evaluating the nature of responses were chlorpromazine hydrochloride (10 mg/kg intraperitoneally), lysergic diethylamide (to 250 μg/kg intravenously) and phenoxybenzamine hydrochloride (to 5 mg/kg intravenously). These were given 30-90 minutes before injection of reserpine. Parallel studies of symptomatic effects were done in conscious animals.

Results. 1. *Pressor effects of reserpine.* In rats treated with pentolinium but not with iproniazid, reserpine produced a small, transient initial decrease in arterial pressure followed by gradual rise to maximum of about 20 mm Hg above control levels; this pressor effect persisted for 90 minutes (Fig. 1A). In rats given both iproniazid and pentolinium, the response to reserpine consisted, as above, of initial small fall in pressure, followed by sharp rise which reached a maximum between 50 and 100 mm Hg within a half hour after injection (Fig. 1B) and remained at this level to 90 minutes. In rats given iproniazid but not pentolinium, reserpine was substantially as pressor as in rats given iproniazid and pentolinium; since post-iproniazid responses were presumably maximal, we could not demonstrate pressor sensitizing effect of the ganglion blocking agent.

2. *Effects of blocking agents on responses to reserpine.* While LSD had no definite effect, chlorpromazine inhibited but did not wholly prevent pressor activity of reserpine in iproniazid treated animals (Fig. 2A): under the same conditions, the same dose of chlorpromazine inhibited almost completely pressor effects of serotonin (1 μg). Phenoxybenzamine, an adrenergic blocking agent, reversed the effect of reserpine in iproniazid treated

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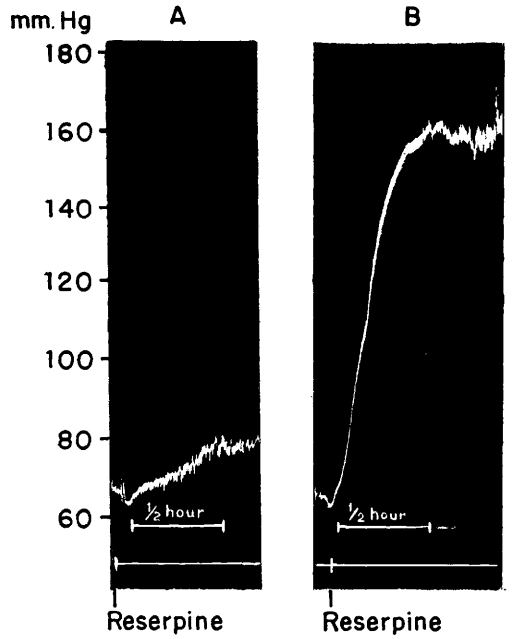

FIG. 1. Anesthetized, pentolinium treated rats. Pressor effects of reserpine in control (A) and in iproniazid treated (B) rats.

rats, so that it caused a prolonged depressor response (Fig. 2B); the maximum fall in pressure varied from 10 to 40 mm Hg. Animals given phenoxybenzamine but not iproniazid, also showed a depressor response, but this was small and corresponded to the small pressor response provoked by reserpine in rats not given phenoxybenzamine. In other experiments it was determined that this dose of phenoxybenzamine was sufficient to reverse responses to .2 μg of epinephrine and norepinephrine from pressor to depressor. As demonstrated in the dog(4,5) the main pressor agent released by reserpine in the rat seems to be norepinephrine; removal of adrenals, principal source of epinephrine, did not decrease pressor response to reserpine.

3. *Effects of "blocking agents" in conscious animals.* Blocking effects of phenoxybenzamine and chlorpromazine on behavioral responses to reserpine after treatment with iproniazid were studied in 3 groups of conscious rats. All animals were given iproniazid and reserpine in dosages noted above. Thirty minutes before administration of reserpine, one experimental group received phenoxybenzamine intravenously and the other, chlorpromazine intraperitoneally. As described by Chessin *et al.*(3) control animals responded to reserpine with an adrenergic reaction, demonstrating piloerection, exophthalmos, salivation, increased motor activity and excitability. This syndrome did not appear in the 2 groups given blocking agents; whatever symptoms appeared were attributable to specific actions of the blocking drugs, namely ptosis with phenoxybenzamine and depression with chlorpromazine.

Discussion. Our experiments extend previous observations in anesthetized cats and dogs (3-5) to the anesthetized rat. They indicate that the pressor effect caused by reserpine depends primarily on release of adrenergic amines, presumably norepinephrine and not serotonin, since LSD did not affect the response while chlorpromazine, a weak adrenergic blocker, and phenoxybenzamine a strong one, did. Further, during these experiments phenoxybenzamine, unlike chlorpromazine, did not affect the pressor response to serotonin in anesthetized rats. Although it has been shown in the rabbit that epinephrine is liberated from the adrenal medulla in response to reserpine (10) the present, like previous(5) experiments, in adrenalectomized animals, indicate that its role is negligible. Similarly the adrenergic response to reserpine in iproniazid-treated rats is, as the descriptive term implies, largely attributable to release of pressor catecholamines. This view is supported by blocking effect of phenoxybenzamine on pressor and central nervous effects of reserpine and

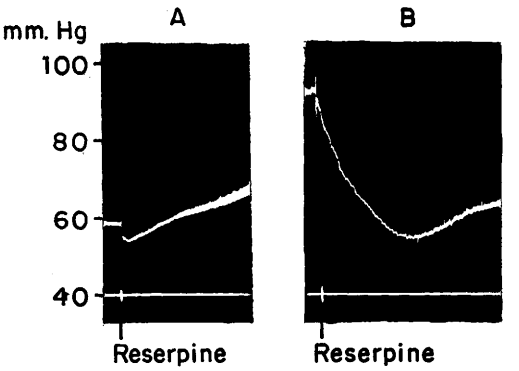

FIG. 2. Anesthetized rats pre-treated with iproniazid and pentolinium. Effect of chlorpromazine (A) and of phenoxybenzamine (B) on the response to reserpine.

by lack of effect of LSD, since in dosage used, LSD has been found to inhibit the effect of injected(11) and of infused(12) serotonin on arterial pressure of anesthetized rats. Further confirmation is found in the fact that in rats pre-treated with iproniazid but not given pentolinium, reserpine was consistently pressor while serotonin was depressor. Clinical corroboration is found in the response to phentolamine of hypertensive patients under treatment with rauwolfia drugs whose response is commonly depressor(13); it seems likely that this reflects a state of underlying adrenergic excitation which in a degree mimics the status of patients with functioning pheochromocytoma.

Summary. In anesthetized rats pretreated with iproniazid, injected reserpine elicits a marked pressor response which from observations on effects of phenoxybenzamine, chlorpromazine and LSD seems primarily attributable to release of adrenergic amines, presumably norepinephrine. Experiments in conscious rats indicate that their response to reserpine is adrenergic. The data support conclusions reached by others in other species but do not support the view that serotonin plays a large role.

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Biliary and Fecal Vit. B_{12} Excretion in Man. An Isotope Study.* (23879)

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In spite of considerable progress in Vit. B_{12} research, it is still not known what amount normal humans have to absorb in order to maintain health; likewise, the optimum maintenance dose for pernicious anemia patients is unknown. One way to elucidate these questions is to study quantitatively the excretion of Vit. B_{12}. As judged from the minimum maintenance dose for pernicious anemia patients, normal humans seem to absorb at least one microgram *pro die* under physiological circumstances(1). When pure Vit. B_{12} is given *per os,* the upper limit of absorption lies well above this figure(2,3). On the other hand,

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