but was found to be of small intensity and short duration compared to that of chlorpromazine. Individually these drugs were found to potentiate the hypothermic effect of chlorpromazine to an extent far greater than could be predicted. Indeed, whereas dibenamine or pyribenzamine caused a drop in rectal temperature of less than 1°C, these drugs injected individually with chlorpromazine potentiated the effect of chlorpromazine on body temperature by 3 to 4°C. However, dibenamine and pyribenzamine antagonize one another when given simultaneously to animals treated with chlorpromazine; this antagonism further illustrates the complexity of action of these drugs.

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Synergism of Amines and Antagonism of Reserpine to Morphine Analgesia. (23656)

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It has been demonstrated(1) that reserpine antagonizes the analgetic effect of morphine in mice. Since reserpine is known to deplete tissues of 5-hydroxytryptamine(2) and catecholamines(3), experiments were performed to determine whether morphine analgesia was influenced by administration of various indolalkylamines and catecholamines and some of their derivatives.

Methods. Seven hundred sixty (760) white mice (Webster strain) with an average weight of 20 g were used. The tail of each mouse was subjected to a painful heat stimulus according to the technic of Gross(4) and the reaction time for the occurrence of the tail flick measured. The method has been previously described(1). The effects of morphine sulfate were studied alone and combined with reserpine phosphate, isoreserpine, 5-hydroxytryptamine, 5-hydroxytryptophane, 5-hydroxyindoleacetic acid, tryptophane, tryptamine, epinephrine, amphetamine, mescaline, iproniazid and choline-p-tolyl-ether. In addition, the influence of reserpine was observed on codeine and meperidine analgesia. All drugs were injected subcutaneously and dissolved in distilled water with the exception of isoreserpine which was dissolved in N, N-dimethylacetamide. The significance of the results was evaluated according to the ranking method described by Moroney(5)*.

Results. Table I illustrates and confirms the previous finding that reserpine antagonizes the morphine-induced prolongation of the reaction time in the tail flick test when

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TABLE I. Antagonism of Reserpine to Analgesics (10 Mice/Group).

	Dose, mg/kg	Avg reaction time in sec. Control	30 min.	60 min.	120 min.
Morphine sulfate	5	3.9	7.4	6.4	4.9
Morphine sulfate	5				
0 hr after reserpine	2.5	4.1	7.6 (n.s.)	7.4 (n.s.)	5.0 (n.s.)
2 hr " "	2.5	4.3	5.3 (<5%)	5.1 (")	4.5 (")
4 hr " "	2.5	4.0	4.8 (")	4.8 (")	4.4 (")
16 hr " "	2.5	3.9	4.4 (<1%)	4.5 (<5%)	4.0 (")
Morphine sulfate	10	4.3	8.6	9.9	7.5
Morphine sulfate	10				
2 hr after isoreserpine	10	3.0	8.3 (n.s.)	8.8 (n.s.)	7.7 (n.s.)
Codeine	50	4.2	9.7	5.4	5.1
Codeine	50				
2 hr after reserpine	10	3.8	4.7 (<1%)	4.5 (n.s.)	3.5 (n.s.)
Meperidine	50	3.9	8.9	7.8	5.6
Meperidine	50				
2 hr after reserpine	10	3.8	5.3 (<1%)	4.9 (<1%)	4.2 (n.s.)

n.s. = not significant.

injected 2 to 16 hours prior to the experiment. Similarly reserpine antagonized the analgetic effect of codeine and meperidine, whereas isoreserpine which lacks reserpine-like pharmacodynamic activity(6) failed to have this effect.

Five-hydroxytryptamine (5-10 mg/kg), tryptamine (100 mg/kg) and 5-hydroxytryptophane (30-400 mg/kg) enhanced and prolonged the analgetic effect of morphine markedly (Fig. 1). Very high doses of 5-hydroxytryptamine without morphine (20 mg/kg) were well tolerated and produced also a prolongation of the reaction time. Five-hydroxytryptophane by itself however did not alter the reaction time even in very large amounts (400 mg/kg). Tryptophane and 5-hydroxyindoleacetic acid (a degradation product of 5-hydroxytryptamine) did not affect the analgetic effect of morphine at a dose of 100 mg/kg.

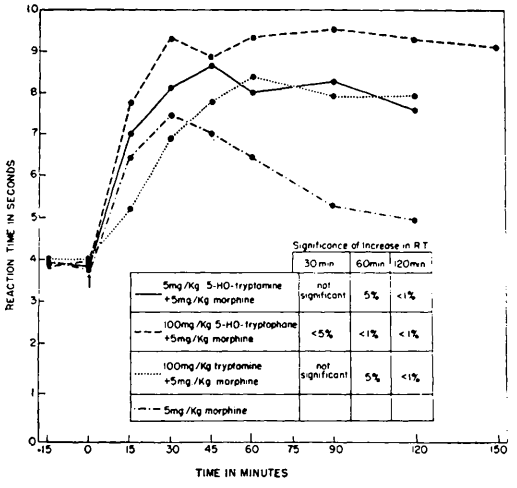

FIG. 1. Influence of 5-hydroxytryptamine and derivatives on morphine analgesia in mice.

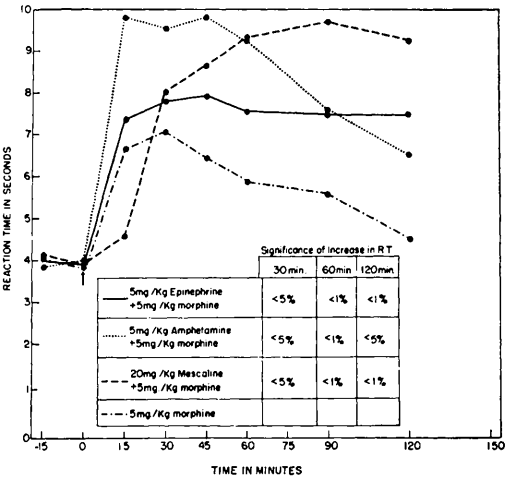

FIG. 2. Effect of some amines on morphine analgesia in mice.

Amphetamine (3-5 mg/kg), mescaline (10-50 mg/kg) and to a lesser extent high doses of epinephrine (2-5 mg/kg) potentiated and prolonged morphine analgesia (Fig. 2). Epinephrine and norepinephrine in doses of

TABLE II. Prevention of Reserpine-Induced Morphine Antagonism by Iproniazid (10 Mice per Group).

	Dose, mg/kg	Avg reaction time in sec. Control	30 min.	60 min.	120 min.
Morphine sulfate	5	4.1	8.3	7.5	5.4
Morphine sulfate 2 hr after reserpine	5 2.5	3.8 (n.s.)	7.0 (5%)	6.0 (5%)	4.9 (n.s.)
Morphine sulfate 6 hr after iproniazid + 2 hr after reserpine	5 100.0 2.5	4.0 (")	9.0 (n.s.)	6.9 (n.s.)	5.3 (")
Morphine sulfate 18 hr after iproniazid + 2 hr after reserpine	5 100.0 2.5	4.0 (")	8.0 (")	7.0 (")	5.4 (")

n.s. = not significant.

less than 2 mg/kg did not alter morphine analgesia. Surprisingly, neither iproniazid (100 mg/kg) nor choline-p-tolyl-ether (100 mg/kg) enhanced the analgesic effect of morphine significantly, no matter whether they were injected simultaneously or several hours before morphine administration. However, reserpine no longer antagonized morphine in mice which were pretreated with iproniazid (Table II).

Discussion. The previous finding(1) that reaction time to the mouse tail flick prolonged by morphine is shortened by reserpine has been confirmed. In addition, reserpine also was shown to antagonize the analgetic effect of codeine and meperidine. Tripod and Gross (7) recently reported a synergistic effect of reserpine on morphine analgesia in the same test. The discrepancy in these results cannot be explained at present. As can be seen from Table I, simultaneous administration of reserpine and morphine did not alter morphine analgesia significantly whereas morphine administration 2-16 hours following reserpine showed a progressive diminution of the analgetic effect.

This latter observation raises the question whether the reserpine antagonism of morphine analgesia is linked with the concentration of indolalkylamines or catecholamines in the central nervous system.

From the investigations of Pletscher *et al.* (2) and Holzbauer and Vogt(3) it is known that reserpine depletes body tissues of their 5-hydroxytryptamine and catecholamine content. If this process plays a role in the mechanism of morphine antagonism of reserpine, an increased level of these substances might be expected to potentiate the analgetic effect of morphine. This is indeed the case for 5-hydroxytryptamine and its precursor, 5-hydroxytryptophane. Five-hydroxytryptamine itself increases the reaction time to a heat stimulus when given in very high doses.

Epinephrine in high doses (2 mg/kg and more) has a distinct potentiating effect, a finding which confirms the report of Hurst and Davies(8). Gross and Kaufmann(9) reported that epinephrine in the dose range of 0.05 to 0.25 mg/kg did not influence morphine analgesia. In this connection it is interesting to note that morphine has been found to release epinephrine(10) and norepinephrine (11) but not 5-hydroxytryptamine.

Choline-p-tolyl-ether and iproniazid, both inhibitors of amino oxydase(12,13) failed to potentiate morphine analgesia. However, if the destruction of 5-hydroxytryptamine and catecholamines is prevented by administration of iproniazid, reserpine no longer antagonizes the analgetic effect of morphine. This indicates that the reserpine-induced morphine antagonism is likely to be due to the depletion of 5-hydroxytryptamine and/or catecholamines.

The result that several other amines such as amphetamine(14), tryptamine and mescaline enhance and prolong the analgetic effect of morphine suggests that the morphine potentiation produced by 5-hydroxytryptamine is not specific and that another mechanism of morphine synergism must be of importance.

Cholinergic mechanisms have attracted attention since morphine action is synergistically influenced by neostigmin(15) and ibogaine(16), both reported to be cholinesterase inhibitors. According to several authors(17, 18) morphine itself inhibits cholinesterase activity markedly. However, the point of view that the mechanism underlying morphine synergism is linked to cholinesterase inhibition cannot be maintained since pilocarpine and carbamylcholine both of which do not affect cholinesterase are active synergists, whereas tetraethyl pyrophosphate, a strong inhibitor, is inactive(19). In addition, no parallelism exists between analgesic action and the degree of cholinesterase inhibition(20). The weak cholinesterase inhibitory activity of 5-hydroxytryptamine and tryptamine(21) can therefore hardly be used to explain the morphine synergism of these agents.

It seems more appropriate to look for a more common factor involved in morphine potentiation. It has been demonstrated that diethylaminoethyl - diphenyl - propyl - acetate which enhances morphine analgesia(22) and prolongs hexobarbital sleeping time(23) inhibits rate of metabolism of hexobarbital and retards demethylation of meperidine thereby prolonging their action(24,25,26). A similar mechanism might play a role in the case of morphine synergism, induced by a variety of drugs.

Summary. (1) Reserpine antagonizes the analgesia in mice induced by morphine, meperidine and codeine. (2) Five-hydroxytryptophane, 5-hydroxytryptamine, tryptamine, amphetamine, mescaline and epinephrine prolong and enhance morphine analgesia whereas iproniazid and choline-p-tolyl-ether fail to do so. (3) Possible mechanisms involved in the synergistic action of amines and the antagonistic action of reserpine on morphine analgesia are discussed.

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