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BRIEF COMMUNICATION

Studies of Thyrotropin-Releasing Hormone (TRH)-Induced Defecation in Cats

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BELESLIN, D B , D JOVANOVIĆ-MICIĆ, R SAMARDZIĆ AND B TERZIĆ *Studies of thyrotropin-releasing hormone (TRH)-induced defecation in cats* PHARMACOL BIOCHEM BEHAV **26**(3) 639–641, 1987 —In unanesthetized cats, defecation produced by thyrotropin-releasing hormone (TRH) was investigated after its injection into the cerebral ventricle (ICV) through chronically implanted cannulae TRH injected in doses from 0 1 to 1 0 mg into the cerebral ventricle evoked defecation which was not dose-dependent The antimuscarinic drug, atropine, the ganglionic blocker, mecamylamine, the alpha and beta adrenergic blocking agents, yohimbine and propranolol, the dopamine antagonist, chlorpromazine, the 5-hydroxytryptamine antagonist, methysergide, and the antihistamine, antazoline, all injected into the cerebral ventricle had virtually no effect on the defecation evoked by TRH injected similarly In cats pretreated with ICV reserpine, 5,6-dihydroxytryptamine and hemicholinium-3, the defecation induced by ICV TRH was not significantly changed On the other hand, in cats pretreated with ICV 6-hydroxydopamine, the defecation caused by ICV TRH was potentiated Therefore, it is concluded that TRH-induced defecation could not be related to central catecholaminergic, 5-hydroxytryptaminergic and cholinergic receptors, but rather to central TRH sites in the cat

TRH Defecation Intracerebroventricular injections Cats TRH sites
Catecholaminergic receptors 5-Hydroxytryptaminergic receptors Cholinergic receptors

STIMULATION of the gastrointestinal motor activity after central administration of thyrotropin-releasing hormone (TRH) in rabbits and cats has already been described [6–10, 13, 14] Further investigations have revealed that centrally administered TRH activates colonic transit via a vagally mediated serotonergic mechanism [6,8] On the other hand, few studies have been done on the mechanism by which central administration of TRH in unanesthetized cats induces defecation. The present study was designed, therefore, to characterize more fully the central regulation of defecation produced by TRH injected into the cerebral ventricle of unanesthetized cats.

METHOD

Experiments were conducted with 154 cats of either sex, weighing 2–4 kg. After the cats were anesthetized with pentobarbital sodium a Collison cannula was then screwed into the calvarium with the tip resting in the left lateral ventricle according to procedures described previously [5]. The lower end of the cannula shaft was positioned with the lumen facing the foramen of Monro. Post-mortem dye studies indicated that the injected material passed from the lateral ventricle into the third and fourth ventricle. Postoperatively penicillin was administered intramuscularly. An interval of five days was allowed to elapse before an experiment began.

On the day of the test, the cat was acclimated to the test environment in a wire-mesh cage, for at least one hour before ICV drug injection. The behavior of the animals was under direct observation continuously throughout the experiments for a period of 4 hr and intermittently for the next 24 hours.

The substances injected ICV were dissolved in sterile, pyrogen-free 0 9% sodium chloride. The solutions were then injected manually in a volume of 0.1–0.2 ml over a period of 15–20 sec and washed in with 0 1 ml of saline Sterile precautions were followed throughout the experiment with each cat used only once Student's t-test was used to determine the significance of the difference between control and various experimental groups The results were considered statistically significant when $p<0.05$.

The compounds used were. synthetic thyrotropin-releasing hormone (Relisorn T, Serono, Rome), atropine sul-

fate, mecamylamine hydrochloride, yohimbine chloride, propranolol chloride, chlorpromazine chloride, methysergide bimaleate, antazoline chlorhydrate, reserpine, 6-hydroxydopamine bromide (6-OHDA), 5,6-dihydroxytryptamine (5,6-DHT) creatinine sulfate and hemicholinium-3. 6-Hydroxydopamine and 5,6-dihydroxytryptamine were dissolved in 0 9% saline containing 0.1 mg/ml ascorbic acid. All drug doses refer to the salt except those of thyrotropin-releasing hormone which refer to the tripeptide, while those of reserpine and hemicholinium-3 refer to the drug.

RESULTS

TRH-Induced Defecation

As shown in Fig 1A, 0 1–1 0 mg TRH injected ICV in the cats tested induced defecation with an average latency ranging from 30 to 90 seconds, the response was not dose-dependent Apart from defecation, ICV TRH evoked the autonomic effects of salivation, licking, tachypnea, panting and micturition, emotional behavior, miaowing, occasional restlessness, and motor responses such as ear twitching and seldom myoclonic jerks

The antimuscarinic drug, atropine (0.005–0.05 mg), the ganglionic blocker, mecamylamine (0.01–0.2 mg), the alpha adrenergic blocking agent, yohimbine (0 005–0.05 mg), the beta adrenergic blocking drug, propranolol (0.01–0 1 mg), the dopamine antagonist, chlorpromazine (0.01–0.1 mg), the antihistamine, antazoline (0 005–0.05 mg) and the 5-hydroxytryptamine antagonist, methysergide (0.1–0.2 mg) were injected ICV 15–20 min before 0.4 mg TRH was injected similarly. As shown in Fig. 1C the defecation caused by TRH was not significantly ($p>0$ 05) changed by any of these pharmacological antagonists

The effect of ICV reserpine on TRH-induced defecation was evaluated over 24 hours after its single injection in a dose of 1 0 mg, while the effect of ICV 6-OHDA on the same response to TRH was examined over 10–14 days after two consecutive days of treatment with daily doses of 2 0 mg of 6-OHDA. Hemicholinium-3 in doses of 0.05 mg was injected ICV twice daily for 5 days, whereas the effect of 5,6-DHT on TRH defecation was evaluated over 10–14 days after two consecutive days of treatment in daily doses of 0 2 mg of 5,6-DHT

In cats treated with reserpine, 5,6-DHT and hemicholinium-3, the defecation produced by (0 4 mg) ICV TRH was not significantly ($p>0.05$) altered (Fig 1B). On the other hand, in 6-OHDA-treated animals the defecation induced by this dose of TRH was potentiated (Fig. 1B, $p<0.05$)

Control Experiments

An ICV injection of 0.2 or 0 3 ml of 0 9% NaCl into the cerebral ventricle of unanesthetized cats (n=4) did not induce any visible behavioral, autonomic or motor phenomena

DISCUSSION

As revealed in these experiments, the immediate action of TRH injected ICV in the unanesthetized cat is defecation, which agrees with other experiments in which defecation was produced by TRH injected ICV or into the reticular substance of the cat brainstem [7, 9, 10, 11] However, the

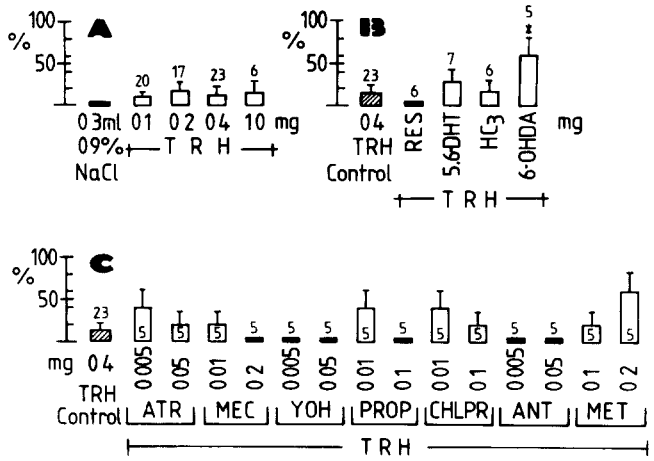

FIG 1 A Mean percent defecation ±S E produced by TRH injected ICV in the cat B Percent defecation ±S E after reserpine (RES), 5,6-DHT, hemicholinium-3 (HC-3) and 6-OHDA injected ICV before ICV TRH C Mean (percent) defecation ±S E after atropine (ATR), mecamylamine (MEC), yohimbine (YOH), propranolol (PROP), chlorpromazine (CHLPR), antazoline (ANT) and methysergide (MET) injected before ICV TRH On abscissae in B and C, the first column (hatched) represents the control value for defecation Defecation was measured over 24 hr after RES, over 10–14 days after 6-OHDA or 5,6-DHT and after five consecutive days of HC-3 The number of experiments at each dose is denoted on each bar Significant differences *$p<0$ 05

defecation response was not dose-dependent and occurred in about 15% of animals. Although the motor activators of the colon produced by ICV TRH in anesthetized rabbits was also not dose-dependent [13], the stimulant effect of ICV TRH on colonic transit in the rabbit was dose-dependent [6] In the rat, ICV administration of TRH enhanced the EMG of the duodenum [15], whereas the intracerebral administration of the tripeptide inhibited gastrointestinal transit in the mouse [3]

TRH injected into the lateral or third cerebral ventricle or cisterna magna may stimulate gastrointestinal motility by an action on central cholinergic mechanisms [8] However, the present experiments do not favor such a mode of action of TRH in the cat, since ICV administration of atropine or hemicholinium-3 did not prevent the induced defecation In addition, yohimbine, propranolol, mecamylamine, chlorpromazine, antazoline, as well as methysergide, injected similarly, had virtually no effect on defecation evoked by ICV TRH. Therefore, defecation induced by ICV TRH does not appear to be associated with central muscarinic, nicotinic, alpha and beta adrenergic, dopaminergic, histaminergic or 5-HT receptors. In this connection, it should be noted that atropine and mecamylamine, but not yohimbine, propranolol, chlorpromazine, antazoline or methysergide also injected ICV in the cat antagonized the salivation produced by ICV TRH [1] Further, the defecation evoked in the cat by ICV nicotine is abolished by ICV atropine, hexamethonium, yohimbine, propranolol, chlorpromazine, antazoline and methysergide [2], whereas atropine, hexamethonium, propranolol and chlorpromazine prevented the defecation produced by the nicotinic ganglionic stimulant dimethylphenylpiperazinium injected ICV [12]

Since specific TRH binding sites have been found in the brain [4,11], and defecation induced by ICV TRH is not blocked by pharmacological antagonists, it is likely that TRH produces defecation in the cat by an action on central TRH binding sites. Although TRH-induced defecation does not seem to be related to catecholaminergic, serotonergic or cholinergic neurons, the salivation induced by TRH does seem to require intact catecholamine pathways in the brain [1].

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