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BRIEF COMMUNICATION

A Differential Dopamine Receptor Involvement During Stress Ulcer Formation in Rats

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PURI, S., A. RAY, A. K. CHAKRAVARTI AND P. A. SEN. *A differential dopamine receptor involvement during stress ulcer formation in rats.* PHARMACOL BIOCHEM BEHAV **47**(3) 749–752, 1994.—The involvement of dopaminergic (DA) receptors and their possible interactions were evaluated during stress ulcer formation in rats. The DA_1 antagonist SCH 23390 (0.025, 0.05, or 0.1 mg/kg) produced only marginal aggravations in gastric stress pathology when compared to vehicle controls. The DA_2 antagonist sulpiride (10 or 50 mg/kg) had dose-related effects. The lower dose aggravated whereas the higher dose attenuated stress ulcerogenesis. The DA_2 agonist bromocryptine (2.5 or 5.0 mg/kg), however, attenuated gastric stress ulcers. Pretreatment of rats with the DA depletor α-methyl-para-tyrosine or the DA_1-antagonist SCH23390 clearly neutralized the stress ulcer-attenuating effects of bromocryptine. These results reaffirm a gastric cytoprotective role for DA and further suggest that DA_1–DA_2 receptor interactions are crucial during DAergic regulation of gastric mucosal integrity during stress.

Dopamine Stress ulcers DA_1 receptor DA_2 receptor SCH23390 Sulpiride Bromocryptine

STRESS ulceration of the stomach is associated with clinical conditions like trauma, head injury, burns, shock, sepsis, and neurological disorders, and is now recognized as a multifactorial phenomenon (6). It is reported to result from interactions between mucosal, vascular, and neurohumoral factors, and the autonomic nervous system plays a crucial role. The central nervous system (CNS) and, more importantly, the brain-gut axis are important mediators of stress ulcerogenesis, and complex neural mechanisms are proposed (10,18,26). For example, several disruptive and protective mediators are now recognized, and biogenic amines, amino acids, and peptides are implicated (6,9). A gastric cytoprotective role for dopamine (DA) is widely speculated and both peripheral and central mechanisms are suggested (4–8,13,14,25). Further, activation of gut DA receptors and reduced gastric acid output have been proposed as mechanisms for gastric cytoprotective effects in different experimental models (5). The neurotransmitter/neuromodulator role for DA is known and its physiological significance in various gastrointestinal effects has been reported (7). DA is known to activate DA_1 and/or DA_2 receptors for its pharmacological effects, and the functional significance of both DA receptor subtypes have been amply demonstrated. These receptors have been identified in specific brain/peripheral areas, and the biochemical and pharmacological significance of such receptor stimulation is a subject of considerable contemporary research (12,24). More recent data have shown that interactions between DA_1 and DA_2 receptors are possible during the expression of some DAergic effects (3,28). However, the exact role of these DA receptor subtypes and possible interactions during stress and the resultant gastric ulcerogenesis are not clearly defined. The present study, therefore, evaluated the probable role of DA_1 and DA_2 receptors and their possible interactions during stress ulcer formation in rats.

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METHODS

Male Wistar rats (200–250 g) were used. They were housed in standard laboratory conditions of light (12-h light-dark cycle) and temperature (22 ± 2°C) and had free access to food and water. They were food- (but not water-) deprived for 18 h prior to the experimental procedure. The experimental stressor consisted of cold restraint stress (CRS; 3 h at 4°C), with the rats immobilized in Plexiglas restrainers (INCO, Ambala, India) in refrigerated chambers. Immediately after the CRS procedure, the rats were sacrificed with an overdose of anesthetic ether. The stomachs were dissected out, cut open along the greater curvature, washed in cold water, and examined microscopically (× 10) under a dissecting microscope, under "blind" conditions. The number of erosions and the cumulative ulcer length in millimeters (to the nearest 0.1 mm), per rat, were determined.

The drugs used were SCH 23390 (Schering, Kenilworth, NJ); (−)sulpiride, α-methyl-para-tyrosine methylester hydrocloride (α-MT), and bromocryptine (all from Sigma Chemical Co., St. Louis); and haloperidol (Searle, India). All drugs were dissolved in distilled water except for sulpiride, which was dissolved in 0.1 N HCl and neutralized to a pH of 5–5.5 with 0.1 N NaOH, and volume made up with distilled water. The drugs were injected IP in a volume of 1 ml/kg 30 min prior to CRS procedure except for haloperidol (pretreatment time 2 h) and α-MT (total pretreatment time 4 h). The drug effects were compared to appropriate vehicle-treated CRS controls.

The data were analysed using the Mann-Whitney *U* test (two-tailed). A *p* value less than 0.05 was considered to be the level of significance in all statistical tests.

TABLE 1

EFFECTS OF DOPAMINE RECEPTOR AGONISTS AND ANTAGONISTS ON STRESS ULCER FORMATION IN RATS

Treatment (mg/kg)	*n*	Mean Gastric Pathology	
		Ulcer Number	Ulcer Severity (mm)
Vehicle	12	7.1 ± 0.9	1.3 ± 0.6
α-MT*	7	15.2 ± 2.8†	3.4 ± 0.8†
Haloperidol (0.5)	7	11.0 ± 3.5‡	4.2 ± 2.5§
SCH 23390 (0.025)	8	6.3 ± 2.0	1.2 ± 0.6
SCH 23390 (0.05)	8	8.6 ± 2.4	2.0 ± 0.4‡
SCH 23390 (0.1)	7	7.5 ± 2.6	1.3 ± 0.7
Sulpiride (10.0)	10	10.7 ± 1.2‡	2.2 ± 0.6‡
Sulpiride (50.0)	6	3.6 ± 0.8§	0.5 ± 0.3§
Bromocryptine (2.5)	8	1.6 ± 0.6†	0.3 ± 0.1†
Bromocryptine (5.0)	8	2.6 ± 0.6†	0.5 ± 0.2§
Sulpiride (10) + Bromocryptine (2.5)	6	9.0 ± 2.2	2.0 ± 0.6
SCH 23390 (0.05) + Bromocryptine (2.5)	7	4.7 ± 0.9‡	0.9 ± 0.3
α-MT + Bromocryptine (2.5)	8	6.3 ± 1.6	1.1 ± 0.4

*α-MT = α-methyl-para-tyrosine, initially 300 mg/kg followed 2 h later by 150 mg/kg. †$p < 0.002$, ‡$p < 0.05$, §$p < 0.02$ (compared to vehicle control group).

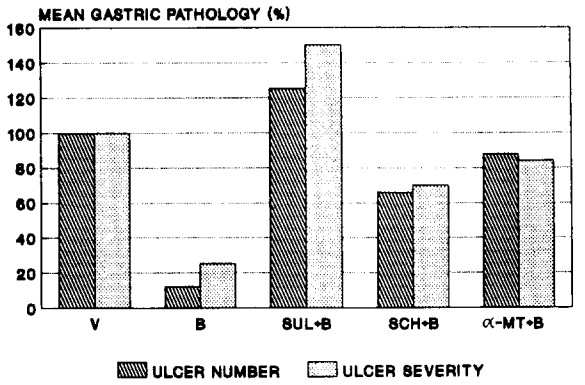

FIG. 1. Dopamine agonist−antagonist/depletor interactions during stress ulcerogenesis. V = vehicle, B = bromocryptine, Sul = sulpiride, SCH = SCH 23390, α-MT = α-methyl-para-tyrosine.

RESULTS

CRS consistently induced gastric mucosal erosions in a manner similar to that seen in some of our earlier studies (18,19,21). The lesions were fairly shallow, did not penetrate the muscularis mucosa, and were mostly seen in the acid-secreting part of the stomach. Pretreatment of rats with the DA blocker haloperidol (0.5 mg/kg) clearly aggravated stress ulcer formation when compared to vehicle controls. As shown in Table 1, both the ulcer number and severity data were significantly greater than the 18-h food-deprived controls. Similar aggravations in stress ulcer pathology were also seen after pretreatment with the DA depletor α-MT (300 + 150 mg/kg), and this group of rats apparently had more number of ulcers per rat than the haloperidol-treated group. SCH 23390 (0.025, 0.05, and 0.1 mg/kg) showed rather variable effects on stress ulcerogenesis. The most marked (i.e., significant) effect was seen with the dose of 0.05 mg/kg, whereas the two dose extremes were less effective in this regard ($p > 0.05$). Sulpiride (10 or 50 mg/kg), on the other hand, showed obvious dose-related effects. The lower dose (10 mg/kg) of the drug clearly augmented the response of the gastric mucosa to CRS, whereas the higher dose (50 mg/kg) showed clear-cut stress ulcer-attenuating effects. The DA agonist bromocryptine (2.5 or 5.0 mg/kg) also showed dose-dependent inhibitory effects on this phenomenon, and both the ulcer number and saverity were significantly lower than that of the control (vehicle) group. In the interaction studies, pretreatment of rats with α-MT clearly reversed the ulceroprotective effects of the DA_2 agonist bromocryptine (2.5 mg/kg). Similarly, prior SCH 23390 administration also blunted significantly the gastric cytoprotective effects of the DA agonist. As shown in Fig. 1, the data of both α-MT + bromocryptine and SCH 23390 + bromocryptine groups were markedly greater than the bromocryptine (alone) group and not significantly different from the vehicle control (+CRS) group.

DISCUSSION

Complex neural mechanisms regulate stress responsiveness, and several lines of data have led to the hypothesis that ergotropic and trophotropic factors maintain the gastric mucosal integrity during stressful experiences (6,9,10). The role of DA in gastrointestinal function is known, and a gastric cytoprotec-

tive role is proposed (4-8,13,14,26). Further, the mesolimbic DA system is probably more important for this protective effect (22). In fact, the amygdaloid complex has been shown to be a crucial neuroanatomical substrate for this effect (6,19,20). Though studies have indicated that the DA_2 receptor may be involved, the effects of specific DA_1 or DA_2 antagonists or agonists are not clearly shown. The initial experiments of this study merely reaffirm a cytoprotective role of DA. The tyrosine hydroxylase inhibitor α-MT, which depletes brain DA, aggravated the ulcerogenic response to CRS. Similar effects were seen with the DA antagonist haloperidol. The effects of the specific DA antagonists were revealing. The DA_1 antagonist SCH 23390 showed only marginal facilitatory effects on CRS ulcers—which would indicate that the DA_1 receptor activation is probably not the only mechanism for DAergic gastric cytoprotection. However, an earlier study has shown that DA_1 receptors in limbic areas are important for this response (19). Our results with the specific DA_2 blocker sulpiride are interesting. Whereas the lower dose (10 mg/kg) aggravated, the higher dose (50 mg/kg) showed inhibitory effects on CRS ulcerogenesis. A previous study also showed similar effects with sulpiride (22). The aggravation is probably due to DA_2 receptor blockade—a mechanism already suggested in some of our earlier reports (19,21). The DA_2 receptor involvement is also highlighted by the clearcut CRS-ulcer attenuating effects of the DA_2 agonist, bromocryptine (15), and is also in agreement with earlier data (22). The atypical properties of the benzamide neuroleptic sulpiride are known, and several of its effects differ from those of the more classical agents (17). In fact, a recent study showed that sulpiride may have anxiolytic effects in several animal models of anxiety (1). Stress responsiveness is a function of the emotionality of the organism, and antianxiety agents attenuate stress responses like ulcerogenesis and elevations in plasma corticosterone (23). Thus, the results with the high dose of sulpiride are in keeping with its atypical nature and/or proposed anxiolytic profile. This is also suggestive of the fact that factors/mechanisms other than DA_2 receptor activation may contribute to the gastric cytoprotective effects of DA. In fact, a recent study showed that DA_1 receptors may be equally important in mediating gastric cytoprotection and reduction in gastric acid output (5). However, while this study suggested a predominant role of DA_1 receptors, we show that the DA_2 receptors are also crucial for stress ulcer development. A central DA_2 receptor involvement has already been suggested for stress ulcerogenesis, and the role of a brain-gut axis (probably DAergic) has been speculated (19-21).

Interactions between DA receptors during the expression/mediation of several biobehavioral responses are known (3,28). For example, the functional integrity of DA_1 receptors is seemingly important for DA_2 receptor-mediated DAergic effects. Though DA_1 and DA_2 receptor agonist or antagonist effects have been shown in some studies (5,7), the probable relationship between these two types of DA receptors during stress reactions has not been studied. Our present data show that such an interrelationship is possible, particularly during stress ulcer formation. Though the DA_1 antagonist SCH 23390 had only marginal effects on CRS ulcers, it clearly blunted the gastric cytoprotection offered by the DA_2 agonist bromocryptine. Similarly, DA depletion (by way of syntheses inhibition) by α-MT also attenuated bromocryptine effects on the stressed gastric mucosa. It can thus be speculated that endogenous DA released during CRS acts on the DA_1 receptors, which in turn increases the sensitivity of the DA_2 receptor to its agonist. Blockade of the DA_1 receptor by SCH 23390 or depletion of endogenous DA by α-MT neutralizes DA_2 agonist effects. An earlier study had suggested the SCH 23390 may even activate/block the DA_2 receptor (11), and this also could be a reason for such SCH 23390-bromocryptine interactions observed during stress ulceration. A similar hypothesis involving DA_1-DA_2 receptor interactions has also been proposed during other experimental situations (16). Taken together, it is possible that DA_1 and DA_2 receptors act in tandem in the DAergic regulation of gastric mucosal integrity during stress and the functional integrity of one regulates that of the other. Nevertheless, the DA_2 receptors still play a dominant role in this phenomenon. These results highlight the recently increasing body of evidence implicating complex neurotransmitter receptor interaction during neurobehavioral states like stress.

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