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Blockade of 5-Hydroxytryptamine$_3$ Receptors Prevents Cisplatin-Induced but not Motion- or Xylazine-Induced Emesis in the Cat

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LUCOT, J. B. *Blockade of 5-hydroxytryptamine$_3$ receptors prevents cisplatin-induced but not motion- or xylazine-induced emesis in the cat.* PHARMACOL BIOCHEM BEHAV 32(1) 207–210, 1989.—5-Hydroxytryptamine$_3$ antagonists have been reported to prevent emesis elicited by cisplatin and radiation. This study investigated the possibility that drugs with this mechanism of action may be useful in preventing emesis elicited by other stimuli. The drugs ICS 205-930 (0.1 and 1.0 mg/kg) and MDL 72222 (0.1 and 1.0 mg/kg) were administered SC to cats before challenging them with either provocative motion or an emetic dose of xylazine. In no instance was a significant reduction in emesis evident. Zacopride was also administered before motion testing (0.01 to 10.0 mg/kg) and found to not have efficacy. To test the possibility that species or route of administration were factors in the negative results, 1.0 mg/kg of ICS 205-930 was administered SC before IV infusion of 7.5 mg/kg of cisplatin. There was a total suppression of emesis for the duration of the six-hour observation periods. This result verifies other work which found 5-hydroxytryptamine$_3$ antagonists to be effective in preventing emesis elicited by cancer chemotherapeutic treatments. However, there is no evidence that they are effective in other syndromes, such as motion sickness and xylazine-induced emesis.

5-HT$_3$ antagonists Motion sickness Xylazine Cisplatin Emesis Cats

CONSIDERABLE interest has been generated by the observation that antagonism of the 5-hydroxytryptamine$_3$ (5-HT$_3$) subtype of receptor by drugs such as MDL 72222, ICS 205-930 and zacopride (6, 10, 15) can prevent emesis elicited by cancer chemotherapeutic agents in ferret and dog (1, 3, 11, 30). However, zacopride does not prevent apomorphine-induced emesis in the dog (32), nor does the 5-HT$_3$ antagonist BDL 24924 prevent apomorphine-induced emesis in the ferret (2). Thus, these drugs might not be effective against a wide spectrum of emetic stimuli. If so, then the antiemetic actions of 5-HT$_3$ antagonists would be fundamentally different from those of 5-HT$_{1A}$ agonists, which prevent emesis elicited by motion, xylazine and cisplatin in the cat (25–28).

These stimuli elicit emesis through different pathways. Xylazine elicits emesis by stimulating alpha-2 noradrenergic receptors (19,24), but does not elicit emesis in cats subjected to area postrectomy (8). However, motion sickness does develop in cats with lesions of the area postrema (5). It is not clear if cancer chemotherapeutic agents use a separate neural pathway. Lesioning the area postrema prevents cisplatin-induced emesis in the cat (29). However, it has been argued that such lesions may interrupt afferents from the periphery which carry the relevant information [(30), see the Discussion section].

In view of the failure of 5-HT$_3$ antagonists to prevent apomorphine-induced emesis, it is important to determine the range of emetic stimuli that these drugs are effective against. In this study, 5-HT$_3$ antagonists were tested for their antiemetic efficacy against motion sickness and xylazine-induced emesis in the cat. The efficacy of a 5-HT$_3$ antagonist against cisplatin-induced emesis in the cat was also verified.

METHOD

Subjects

Subjects were healthy cats weighing 2.3 to 4.9 kg. They were housed in the Laboratory Animal Resources facility and allowed free access to food and water at all times except during testing and immediately postoperatively.

Motion Sickness

In tests of MDL 72222 and ICS 205-930, fifteen female cats with normal free-fall righting and vestibulo-ocular reflexes were selected on the basis of their susceptibility to motion sickness as determined by five screening motion challenges. Of these, five retched on five, three retched on four, five retched on three and two retched on two of five screening tests. In the test of zacopride, twenty cats with normal reflexes were used. The dose-response curve was determined using ten cats, of which two retched on all five screen tests, one retched on three, three retched on two,

three retched on one and one retched on none but had retched previously. An additional ten cats were tested at the dose of 1.0 mg/kg of zacopride, which the dose-response curve indicated as the most promising of having antimotion sickness effects. In this group, six retched on all five screen tests, two retched on four and two retched on three.

The motion stimulus device was motor-driven and modeled after an amusement park Ferris wheel. Cats rode in two Plexiglas boxes suspended from 0.445-m arms that rotated about a central horizontal axle at 0.28 Hz (17 rpm) (12). Motion challenges lasted for 30 min and were followed by one min of observation at rest. The latency to retching was recorded, though in all cases the retching was followed by vomiting. All motion tests were separated by two weeks to prevent the development of habituation to the motion stimulus.

Xylazine

Ten female cats with normal free-fall righting and vestibulo-ocular reflexes but no susceptibility to the motion stimulus were used. The dose of 0.66 mg/kg of xylazine was administered SC and the cats were observed for 30 min or for 15 min after the last emetic episode, whichever occurred later. This procedure has been established as adequate to observe the emesis elicited by xylazine (24). The efficacy of xylazine was verified between all drug studies. Saline alone was administered to evaluate conditioned responses. Xylazine tests were conducted at one-week intervals.

Cisplatin

Sixteen cats of either sex were used. Ten were historical controls reported in other studies [(26,28), submitted]. An additional cat was used as a control to verify the emetic efficacy of the batch of cisplatin used. Of the controls, five were males and six were females. Five cats were used to test the antiemetic effects of ICS 205-930; four males and one female. In four of the controls and all of the cats receiving ICS 205-930, the ability of the cat to vomit was verified by testing for an emetic response to 0.66 mg/kg of xylazine at least one week before catheter implantation.

Jugular catheters were implanted under ketamine (4.5 mg/kg, IM) and pentobarbital (32 mg, IV) anesthesia and sterile conditions. Supplemental pentobarbital and mechanical ventilation were used as necessary. Prophylactic antibiotics were administered after the surgery. Catheters were implanted in the jugular vein, threaded under the skin and externalized at the nape of the neck. Catheter patency was maintained by withdrawing blood, flushing with 50 U/ml heparin and filling with 1000 U/ml heparin. This was done after surgery and on alternate days thereafter. Experiments were done 72–96 hr after surgery.

The dose of 7.5 mg/kg cisplatin was infused through the catheter over a period of 4–5 min. The number and latency of emetic events during the subsequent six-hr observation period were recorded. At the end of the observation period, the cats were euthanized by IV administration of 0.3 ml/kg of T-61 solution (Hoechst, Somerville, NJ).

Drugs

Xylazine HCl (Bayvet, Shawnee, KS) was dissolved in sterile saline to an injection volume of 0.066 ml/kg. Cisplatin (Sigma Chemical Co., St. Louis, MO) was dissolved in sterile water to a concentration of 2 mg/ml by gentle warming and sonicating. ICS 205-930 (Sandoz, East Hanover, NJ) was dissolved in sterile saline to an injection volume of 0.1 ml/kg and administered SC 20 min before motion and xylazine challenges and immediately before cisplatin infusion. MDL 72222 (Merrell Dow Pharmaceuticals, Inc., Cincinnati, OH) was suspended in saline with a few drops of Tween 80 by warming and sonicating to an injection volume of 0.1 ml/kg. MDL 72222 was administered SC 20 min before motion and xylazine challenges. Zacopride (AHR 11190B; Richmond, VA) was dissolved in sterile saline to an injection volume of 0.1 ml/kg and administered SC 60 min before motion testing. All drugs were prepared immediately before each experiment. The doses of all but ICS 205-930 and MDL 72222 are expressed as the base.

TABLE 1

EFFECTS OF MDL 72222 AND ICS 205-930 ON RETCHING ELICITED BY MOTION OR BY 0.66 mg/kg XYLAZINE

Treatment	Motion Number of Cats Retched/Tested	Xylazine Number of Cats Retched/Tested
Saline	10/15	10/10
ICS 205-930		
0.1 mg/kg	10/15	7/10
1.0 mg/kg	9/15	7/9
Saline	—	10/10
MDL 72222		
0.1 mg/kg	8/15	8/10
1.0 mg/kg	8/15	9/10
Vehicle	11/15	8/10
Vehicle + saline	—	0/10

Vehicle for MDL 72222 was saline with a few drops of Tween 80. Vehicle + saline was the vehicle for MDL 72222 followed by saline rather than xylazine.

Statistics

The data from motion and xylazine tests were analyzed using Cochran's Q-test (7). Data from the cisplatin experiment were analyzed using the Fisher Yates test on the number of cats vomiting (14).

RESULTS

Motion Sickness

Neither ICS 205-930 nor MDL 72222 prevented motion sickness ($p>0.3$; Table 1). Zacopride also did not produce a significant decrease in motion sickness at any dose tested ($p>0.2$; Table 2). Because this group was comparatively nonsusceptible, statistical significance could only be achieved by suppression of motion sickness in all cats. Therefore, the dose of 1.0 mg/kg was tested in another ten cats with greater susceptibility. In this group, eight of ten retched when saline administration preceded motion testing and seven of ten retched when 1.0 mg/kg of zacopride preceded motion testing.

Xylazine

Neither ICS 205-930 nor MDL 72222 prevented xylazine-

TABLE 2

EFFECTS OF ZACOPRIDE ON MOTION SICKNESS IN TEN CATS

Treatment	Number Retching
Saline	5
0.01 mg/kg	4
0.1 mg/kg	3
0.316 mg/kg	3
1.0 mg/kg	2
10.0 mg/kg	6
Saline	6

Zacopride was administered 60 min before 30 min of motion testing. Data expressed as the number of cats retching.

TABLE 3

EFFECTS OF 1.0 mg/kg OF ICS 205-930 ON CISPLATIN-INDUCED EMESIS IN THE CAT

Treatment	Number of Cats Vomited/Tested	Latency to First Emesis*	Number of Vomits†
Control	9/11	93.6(13.8)	4.4(0.8)
1.0 mg/kg	0/5	>360	0

*Mean (SEM) latency in min expressed in decimals of cats that vomited. >360; no emesis during the 360-min observation period.
†Mean (SEM) number of emetic events of cats that vomited.

induced emesis ($p>0.1$; Table 1). There was no evidence of conditioned reactions to the experimental situation as evidenced by the absence of retching following injections of saline and vehicle.

Cisplatin

The dose of 1.0 mg/kg of ICS 205-930 prevented emesis in all cats tested (Table 3). The decrease in the number of cats vomiting was significant ($p=0.03$). Because there were no emetic events, further statistical analysis of the latency to the first emetic event and the number of emetic episodes were not performed.

DISCUSSION

The results show that antagonists of 5-HT$_3$ receptors do not prevent emesis elicited by motion or xylazine in the cat. These findings are consistent with reports that drugs with this mechanism of action do not prevent apomorphine-induced emesis in the ferret (2) or the dog (32). However, they do prevent irradiation-induced emesis in the ferret (2) and cancer chemotherapy-induced emesis in dog (3), ferret (10, 11, 30), man (13,23) and cat (present results).

It is unlikely that the negative results are due to dose, route of administration or species. Zacopride was tested over three log units of dose and ICS 205-930 and MDL 72222 were tested at IV doses reported to prevent cisplatin-induced emesis in the ferret (10,30). The efficacy in cat of the higher dose of ICS 205-930 following SC administration against cisplatin-induced emesis was verified in this study (Table 3). However, this treatment was ineffective against motion sickness and xylazine-induced emesis (Table 1).

The anatomical location at which 5-HT$_3$ antagonists prevent emesis is not clear. 5-HT$_3$ receptors were reported to exist only in the peripheral nervous system (16). This led to the suggestion that cisplatin stimulates 5-HT$_3$ receptors in the periphery, which activate afferents that project to or through the area postrema (30). The authors concluded that lesions of the area postrema which prevent cisplatin-induced emesis in the cat (29) interfere with these afferents.

The antiemetic effects of 5-HT$_3$ antagonists in radiation-induced emesis were argued to be extra-abdominal. Vagotomy in ferrets prevents only the early phase of radiation-induced emesis, while the 5-HT$_3$ antagonist BRL 24924 prevents both early and late phases (2). However, authors did not address nonvagal peripheral afferents, which have been implicated in radiation-induced emesis in the cat (4).

More recently, 5-HT$_3$ binding sites have been identified in the central nervous system. The 5-HT$_3$ antagonist ICS 205-930 displaces radiolabeled 5-HT from its binding sites and antagonizes 5-HT-induced stimulation of adenylate cyclase in the spinal cord (17). The 5-HT$_3$ antagonist GR38032 has anxiolytic activity in some animal models (21). It also prevents the effects of increased dopaminergic activity in the nucleus accumbens (9,18). The 5-HT$_3$ antagonist GR65630 binds to sites in the central nervous system, though they have as yet no corresponding behavioral action. These sites are unevenly distributed in the central nervous system (22) and could account for the ability of zacopride to prevent emesis elicited by intracerebroventricular (ICV) administration of cisplatin in cats (31). However, the relevance of ICV administration of cisplatin to emesis elicited by cisplatin clinically is not clear as vagotomy prevents cisplatin-induced emesis in dog (20).

In conclusion, administration of 5-HT$_3$ antagonists to cats prevents cisplatin-induced emesis but not emesis induced by motion or xylazine. Thus, they do not appear to have use for some emetic syndromes. The anatomical location of the antiemetic action remains to be established.

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