Pharmacology Biochemistry and Behavior, Vol. 43, pp. 1241–1246, 1992
Printed in the U.S.A.

0091-3057/92 $5.00 + .00

2-Deoxy-D-Glucose Antinociception and Serotonin Receptor Subtype Antagonists: Test-Specific Effects in Rats

MICHELLE C. FISHER AND RICHARD J. BODNAR[1]

Department of Psychology and Neuropsychology Doctoral Sub-Program, Queens College, CUNY, Flushing, NY 11367

Received 29 June 1992

FISHER, M. C. AND R. J. BODNAR. *2-Deoxy-D-glucose antinociception and serotonin receptor subtype antagonists: Test-specific effects in rats.* PHARMACOL BIOCHEM BEHAV 43(4) 1241–1246, 1992.—The antinociceptive actions of 2-deoxy-D-glucose (2-DG) are mediated in part by endogenous opioid, dopaminergic, cholinergic, histaminergic, and neurohormonal influences. Although 2-DG antinociception was not affected by tryptophan hydroxylase inhibition, a possible serotonergic role in 2-DG antinociception was investigated because of the existence of serotonin [5-hydroxytryptamine (5-HT)] receptor subtypes. The present study examined the effects of general (methysergide: 5 and 10 mg/kg), 5-HT_2 (ritanserin: 2.5 mg/kg), and 5-HT_3 (ICS-205,930: 0.25–5 mg/kg) receptor subtype antagonists upon 2-DG antinociception on the tail-flick and jump tests in rats. On the tail-flick test, 2-DG (450 mg/kg) antinociception was significantly reduced by all ICS-205,930 doses (48–58%) but unaffected by either methysergide (22–29% reduction) or ritanserin (6% reduction). In contrast, 2-DG antinociception on the jump test was significantly potentiated across the 120-min time course and across the 2-DG dose-response curve (100–650 mg/kg) by methysergide, ritanserin, and ICS-205,930 pretreatment. Each of the three antagonists produced significant leftward shifts in the peak and total 2-DG dose-response curve for the jump test. These data suggest different sites of action for 2-DG antinociception as a function of the pain test employed and a differential modulation by serotonin receptor subtypes at those sites.

2-Deoxy-D-glucose antinociception Serotonin Methysergide Ritanserin ICS-205,930 Rats

THE antimetabolic glucose analog, 2-deoxy-D-glucose (2-DG), activates such physiological responses as glucoprivation, peripheral sympathomedullary discharge, hyperglycemia, and hyperphagia (14,19,34,39). 2-DG elicits an antinociceptive response following peripheral (8) and central (12) administration that displays both antinociceptive tolerance (6) and cross-tolerance with morphine antinociception (36). The opioid mediation of 2-DG antinociception is partial because it displays synergy with morphine antinociception but is unaffected by the opiate receptor antagonist naloxone (9). The neurochemical substrates of 2-DG antinociception have been evaluated using other pharmacological and hormonal manipulations. Whereas dopaminergic receptor agonists (amphetamine and apomorphine) decrease 2-DG antinociception (7), dopaminergic receptor agonists (chlorpromazine and haloperidol) potentiate 2-DG antinociception (13). Further, 2-DG antinociception is potentiated by muscarinic cholinergic receptor antagonists [scopolamine and methylscopolamine (35)], histaminergic (H_2) receptor antagonists [zolantidine (24)], hypophysectomy (10), and medial-basal hypothalamic damage (1). In contrast, central pretreatment with alloxan reduces 2-DG antinociception, presumably through actions upon central glucoreceptors (25).

Serotonin [5-hydroxytryptamine (5-HT)] has been postulated as a major transmitter implicated in antinociceptive processes [for reviews, see (2,4)]. A role for serotonin in the mediation of 2-DG antinociception was evaluated using the tryptophan hydroxylase inhibitor parachlorophenylalanine, which failed to alter this antinociceptive response at doses that reduced morphine antinociception (11). However, multiple 5-HT receptors [for reviews, see (32,33)] have since been classified into $5HT_{1A}$, $5HT_{1B}$, $5HT_{1C}$, $5HT_{1D}$, $5HT_2$, and $5HT_3$ subtypes. Both opioid and nonopioid forms of antinociception have been affected following administration of selective $5HT_{1A}$ (27–29), $5HT_2$ (23,30,31), and $5HT_3$ (17,18,20) receptor agonists and antagonists. Indeed, nonopioid antinociception following continuous cold-water swims was unaffected by parachlorophenylalanine pretreatment (11), but significantly reduced by $5HT_2$ antagonists (23). To characterize further the role of serotonin receptor subtypes in the mediation of 2-DG antinociception, the present study examined whether either general 5-HT (methysergide), 5-HT_2 (ritanserin), or 5-HT_3

[1] To whom requests for reprints should be addressed.

(ICS-205,930) receptor subtype antagonists would alter this form of antinociception in the tail-flick (15) and jump (16) tests.

METHOD

Male, albino Sprague-Dawley rats (300–550 g) were housed individually on a 12 L : 12 D cycle with ad lib access to rat chow and water.

Nociceptive Tests

Tail-flick latencies were ascertained with a radiant heat source (IITC Analgesia Meter, Woodland Hills, CA) in which heat was applied to the dorsum of the rat's tail 3–8 cm proximal to the tip. Each session consisted of three latency determinations made at 10-s intertrial intervals. To avoid tissue damage, the determination was terminated if no response occurred after 12 s. Immediately thereafter, jump thresholds were ascertained in a chamber (30 × 24 × 26.5 cm) with 14 grid bars spaced 1.9 cm apart. Electric shocks (0.3 s) were delivered through the grids by a shock generator (BRS/LVE, Beltsville, MD) and shock scrambler (Campden Instruments, Chicago, IL). An ascending method of limits procedure was employed for each of six trials with shock initially delivered at 0.1 mA and increased in 0.05-mA increments at 5-s intervals. The jump threshold was defined as the lowest of two consecutive intensities at which the rat simultaneously removed both rear paws from the grids or if a cutoff of 1.0 mA was reached.

Protocol

Following 4 days of baseline latency and threshold determinations to ensure stability, a first group of rats (n = 10) received the following injection conditions at 2–6 h into the light cycle according to an incompletely counterbalanced design at weekly intervals: a) vehicle/vehicle; b) vehicle/2-DG (450 mg/kg, Sigma Chemical Co., St. Louis, MO); c) 5 and d) 10 mg/kg methysergide (Sandoz Laboratories, Basle, Switzerland) paired with 2-DG; e) 2.5 mg/kg ritanserin (Janssen Laboratories, Beerse, Belgium) paired with 2-DG; and f) 0.25, g) 1, and h) 5 mg/kg ICS-205,930 (Sandoz) paired with 2-DG. A 15-min interval elapsed between injection conditions. Tail-flick latencies and jump thresholds were assessed 30, 60, 90, and 120 min following the last injection in each condition. 2-DG was administered IP in a distilled water vehicle. Methysergide was dissolved in 0.9% normal saline and administered IP. Ritanserin was administered SC and initially prepared in 100% methanol at a concentration of 10 mg/ml and then titrated with 0.9% normal saline to its desired concentration 0.5 h prior to treatment. ICS-205,930 was administered SC and initially prepared in 100% dimethyl sulfoxide at a concentration of 10 mg/ml and then titrated with 0.9% normal saline to its desired concentration 0.5 h prior to treatment.

Subsequent groups of rats were pretreated at weekly intervals with vehicle/vehicle and either vehicle, methysergide (5 mg/kg), ritanserin (2.5 mg/kg), or ICS-205,930 (5 mg/kg) paired with 2-DG doses of 100 mg/kg (n = 6), 225 mg/kg (n = 10), and 650 mg/kg (n = 5). Jump thresholds were assessed 30, 60, 90, and 120 min following the last injection in each condition. Finally, a last group (n = 10) of rats were tested with either vehicle, methysergide (5 mg/kg), ritanserin (2.5 mg/kg), or ICS-205,930 (5 mg/kg) at weekly intervals. Tail-flick latencies and jump thresholds were assessed after 30, 60, 90, and 120 min to determine basal nociceptive effects.

Statistical Analyses

Split-plot analyses of variance (ANOVAs) assessed significant differences among conditions and across the time course for each of the 2-DG doses and each test. Significant 2-DG effects relative to vehicle and significant 5-HT antagonist effects relative to 2-DG were assessed with Dunnett and Dunn comparisons, respectively. To determine alterations in 2-DG dose–response functions under vehicle and 5-HT antagonist pretreatment, linear regression analyses evaluated peak (30 min) and total antinociceptive effects relative to the 2-DG doses to determine differences between slopes, intercepts, and the ED_{50}. Calculation of the total antinociceptive effect for each condition relative to vehicle was accomplished by summing the differences between each experimental and vehicle score across the time course.

RESULTS

2-DG (450 mg/kg) Antinociception (Tail-Flick Test)

In comparing ICS-205,930 effects upon 2-DG antinociception, significant differences were observed among groups, $F(4, 44) = 5.35$, $p < 0.001$, across the time course, $F(3, 132) = 18.91$, $p < 0.0001$, and for the interaction between groups and time, $F(12, 132) = 2.74$, $p < 0.002$. 2-DG significantly increased tail-flick latencies 30 and 60 min following injection. 2-DG antinociception was significantly reduced after 30 min by pretreatment with ICS-205,930 doses of 0.25 (58% reduction), 1 (50% reduction), and 5 (48% reduction) mg/kg (Fig. 1A). The small antinociceptive effect induced by 2-DG after 60 min was eliminated by ICS-205,930 pretreatment.

In comparing methysergide and ritanserin effects upon 2-DG antinociception, significant differences were observed among groups, $F(4, 42) = 3.99$, $p < 0.008$, across the time course, $F(3, 126) = 27.78$, $p < 0.0001$, and for the interaction between groups and time, $F(12, 126) = 2.18$, $p < 0.017$. The significant increases in tail-flick latencies 30 min following 2-DG failed to be significantly altered by pretreatment with either methysergide at doses of 5 (29% reduction) and 10 (22% reduction) mg/kg or ritanserin (2.5 mg/kg: 6% reduction) (Fig. 1B). Because lower doses of 2-DG failed to produce appreciable antinociceptive effects on the tail-flick test, and higher 2-DG doses produced similar magnitudes of antinociception to that elicited by the 450-mg/kg dose, no other doses of 2-DG were analyzed in the tail-flick test for 5-HT antagonist effects.

2-DG (450 mg/kg) Antinociception (Jump Test)

In comparing ICS-205,930 effects upon 2-DG antinociception, significant differences were observed among groups, $F(4, 44) = 39.60$, $p < 0.0001$, across the time course, $F(3, 132) = 53.44$, $p < 0.0001$, and for the interaction between groups and time, $F(12, 132) = 6.13$, $p < 0.0001$. 2-DG significantly increased jump thresholds across the time course. 2-DG antinociception in the jump test was significantly potentiated after 60- to 120-min pretreatment with ICS-205,930 doses of 0.25 (58% increase), 1 (29% increase), and 5 (27% increase) mg/kg (Fig. 2A). In comparing methysergide and ritanserin effects upon 2-DG antinociception, significant differences were observed among groups, $F(4, 42) = 39.40$, $p < 0.0001$, across the time course, $F(3, 126) = 62.04$, $p < 0.0001$, and for the interaction between groups and time, $F(12, 126) = 7.25$, $p < 0.0001$. 2-DG antinociception in the jump test was significantly potentiated by 5 (22% increase) and 10 (32% increase)

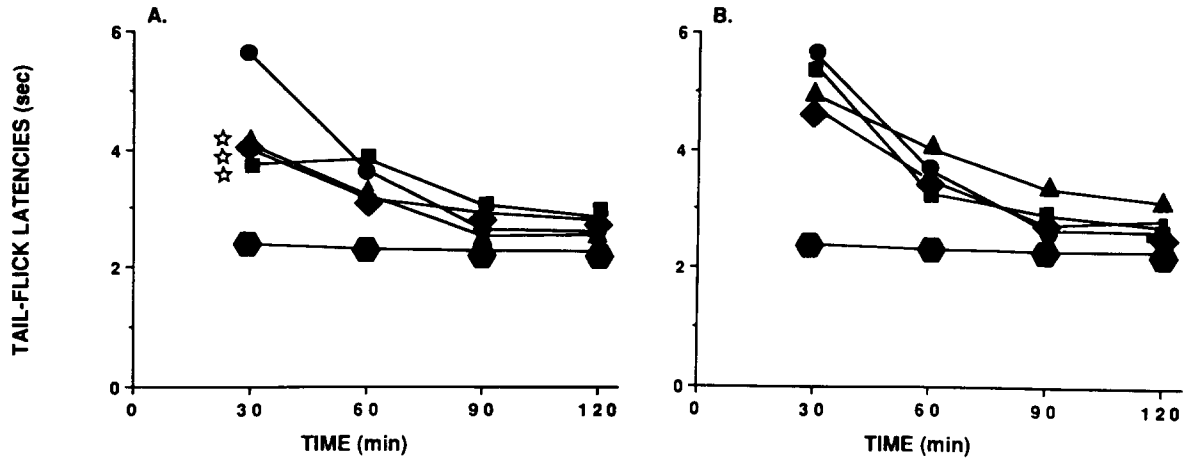

FIG. 1. (A). Alterations in tail-flick latencies (seconds) in rats pretreated with either vehicle (●), 2-deoxy-D-glucose (2-DG, 450 mg/kg; ●), or 2-DG paired with ICS-205,930 doses of 0.25(■), 1 (♦) or 5 (▲) mg/kg. (B). Alterations in tail-flick latencies in rats pretreated with either vehicle (●), 2-DG (450 mg/kg; ●), or 2-DG paired with either ritanserin at a dose of 2.5 mg/kg (■) or methysergide at doses of 5 (♦) or 10 (▲) mg/kg. The open stars indicate significant reductions in the magnitude of 2-DG antinociception in the tail-flick test (Dunnett comparisons, $p < 0.05$).

mg/kg methysergide and by 2.5 mg/kg (69% increase) ritanserin (Fig. 2B).

2-DG Dose-Response Curves (Jump Test)

Evaluation of the effects of 5-HT receptor subtype antagonists upon a dose of 100 mg/kg 2-DG revealed a failure to discern significant differences among groups, across the time course, or for the interaction between groups and time (Fig. 3). Evaluation of the effects of 5-HT receptor subtype antagonists upon a dose of 225 mg/kg 2-DG revealed significant differences among groups, $F(4, 45) = 5.70$, $p < 0.0008$, across the time course, $F(3, 135) = 23.49$, $p < 0.0001$, and for the interaction between groups and time, $F(12, 135) = 2.40$, $p < 0.016$. The significant increases observed in the jump test following this dose of 2-DG were significantly potentiated by ICS-205,930 (90–120 min: 32% increase), methysergide (30–120 min: 56% increase), and ritanserin (30–120 min: 76% increase). Evaluation of the effects of 5-HT receptor subtype antagonists upon a dose of 650 mg/kg 2-DG revealed significant differences among groups, $F(4, 20) = 7.65$, $p < 0.0007$, across the time course, $F(3, 60) = 22.57$, $p < 0.0001$, and for the interaction between groups and time, $F(12, 60) = 2.64$, $p < 0.007$. The significant increases observed in the jump test following this dose of 2-DG were significantly potentiated by ICS-205,930 (30–60 min: 43% increase) and ritanserin (30–90 min: 53% increase) but not by methysergide (30% increase).

Regression analyses of the 2-DG dose-response curves following pretreatment with vehicle, ICS-205,930, methysergide,

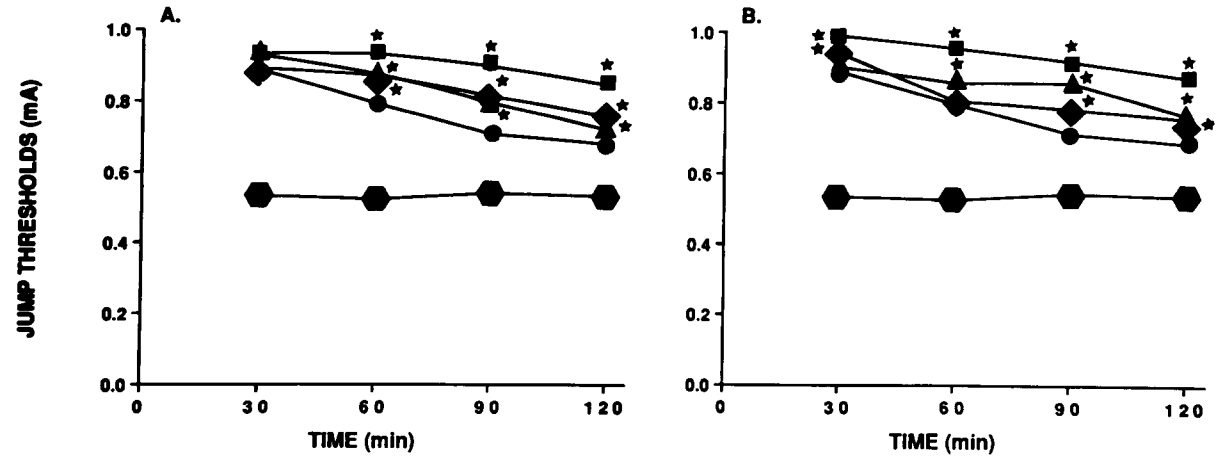

FIG. 2. (A). Alterations in jump thresholds (mA) in rats pretreated with either vehicle (●), 2-deoxy-D-glucose (2-DG, 450 mg/kg; ●), or 2-DG paired with ICS-205,930 doses of 0.25 (■), 1 (♦), or 5 (▲) mg/kg. (B). Alterations in jump thresholds in rats pretreated with either vehicle ( ), 2-DG (450 mg/kg; ●), or 2-DG paired with either ritanserin at a dose of 2.5 mg/kg (■) or methysergide at doses of 5 (♦) or 10 (▲) mg/kg. The solid stars indicate significant increases in the magnitude of 2-DG antinociception in the jump test (Dunnett comparisons, $p < .05$).

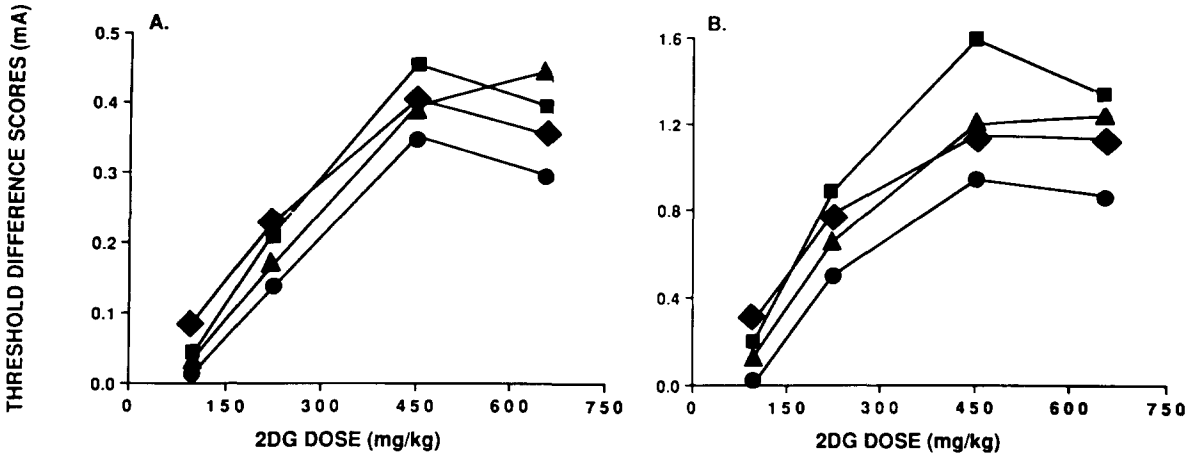

FIG. 3. Alterations in the magnitude of peak (30 min, A) and total (B) 2-deoxy-D-glucose (2-DG) antinociception on the jump test in rats pretreated with either vehicle (●), ICS-205,930 (▲), methysergide (♦), or ritanserin (■). Calculation of the total antinociceptive effect for each condition relative to vehicle was accomplished by summing the differences between each experimental and vehicle score across the time course. Regression analyses indicated significant leftward shifts in the dose-response curves for 2-DG antinociception following methysergide, ritanserin, and ICS-205,930 pretreatment.

and ritanserin revealed significant differences among groups for peak, $F(6, 114) = 2.34$, $p < 0.036$, and total, $F(6, 114) = 4.37$, $p < 0.0005$, antinociceptive effects on the jump test (Table 1). Analyses of ED_{50} values indicated significant reductions in the 2-DG doses necessary to increase peak antinociceptive effects following pretreatment with methysergide (25% reduction), ritanserin (27% reduction), and ICS-205,930 (21% reduction) and total antinociceptive effects following pretreatment with methysergide (22% reduction), ritanserin (43% reduction), and ICS-205,930 (27% reduction).

5-HT Antagonists and Basal Nociception

Neither methysergide, ritanserin, nor ICS-205,930 altered basal tail-flick latencies or jump thresholds across the 2-h time course.

DISCUSSION

The magnitude of 2-DG antinociception was significantly and differentially altered by pretreatment with 5-HT receptor subtype antagonists. The differential effects were due to test-specific effects and the effectiveness of the antagonists employed. First, 2-DG antinociception was significantly *reduced* in the tail-flick test by the selective 5-HT_3 antagonist ICS-205,930 but not by either the general 5-HT antagonist, methysergide or the selective 5-HT_2 antagonist ritanserin. The *reductions* in 2-DG antinociception in the tail-flick test occurred across a wide range of ICS-205,930 doses but failed to occur at peak effective doses for either methysergide or ritanserin. This reduction in antinociceptive effectiveness could not be attributed to any basal effect of ICS-205,930 on tail-flick latencies. A complete analysis of the reductions in 2-DG antinociception in the tail-flick test by 5-HT_3 antagonists was somewhat hampered by the narrow and asymptotic dose-response function for 2-DG antinociception in the tail-flick test. Whereas 2-DG doses lower than 450 mg/kg produced a small and inconsistent antinociception in the tail-flick test, 2-DG doses higher than 450 mg/kg produced a similar magnitude of antinociception in this measure.

Second, 2-DG antinociception was significantly *increased*

TABLE 1

REGRESSION ANALYSES OF THE DOSE-RESPONSE FUNCTIONS OF 2-DG ANTINOCICEPTION ON THE JUMP TEST FOLLOWING 5-HT RECEPTOR SUBTYPE ANTAGONISTS

Condition	Vehicle	Methysergide	Ritanserin	ICS-205,930
Peak effects				
Slope	0.000604	0.000543	0.000731	0.000817
Intercept	0.0035	0.0935	0.0358	−0.0151
ED_{50} (mg/kg)	457.8	343.5	334.1	361.2
Ratio vehicle	–	0.75	0.73	0.79
Total effects				
Slope	0.00162	0.00150	0.00224	0.00202
Intercept	0.0543	0.3535	0.2855	0.1561
ED_{50} (mg/kg)	657.8	511.0	372.5	477.2
Ratio vehicle	–	0.78	0.57	0.73

on the jump test by the general 5-HT antagonist methysergide, the 5-HT_2 antagonist ritanserin, and the 5-HT_3 antagonist ICS-205,930. The ability of these antagonists to each potentiate 2-DG antinociception occurred across all 2-DG doses (225, 450, and 650 mg/kg) capable of eliciting an antinociceptive response itself. This potentiation in antinociceptive effectiveness could not be attributed to any basal effect of either methysergide, ritanserin, or ICS-205,930 on jump thresholds. In contrast, the 2-DG dose of 100 mg/kg failed to elicit antinociception in the jump test and also failed to produce antinociception when paired with any of the 5-HT receptor subtype antagonists. Thus, it appears that these 5-HT antagonists require the presence of antinociception to exert potentiating actions. The peak and total dose–response functions for 2-DG antinociception were significantly shifted to the left following pretreatment with methysergide, ritanserin, and ICS-205,930. This potentiating action of 5-HT antagonists upon 2-DG antinociception in the jump test appears to be due to changes in nociceptive reactivity rather than nonspecific actions for the following reasons: First, tail-flick latencies and jump thresholds were evaluated simultaneously in the analysis of 5-HT effects upon antinociception induced by the 450-mg/kg 2-DG dose. The clearly divergent, selective, and differential actions of 5-HT antagonists upon 2-DG antinociception in the tail-flick and jump tests argues against a nonspecific action. Second, the hyperphagic actions of 2-DG are selectively altered by 5-HT antagonists such that ritanserin only transiently reduced 2-DG hyperphagia after 2, but not after 4 or 6, h (3). Neither methysergide nor ICS-205,930 altered 2-DG hyperphagia. If the potentiations in 2-DG antinociception in the jump test were due to a nonspecific decrement in motor responses, then reductions in 2-DG hyperphagia would be expected as well; this did not occur.

The dissociative effects of 5-HT antagonists upon 2-DG antinociception as a function of the nociceptive test is in keeping with other recent instances of test specificity in nociceptive responding [for review, see (5)]. Nonopioid antinociception induced by continuous cold-water swims is reduced far more effectively in the tail-flick test than in the jump test by the 5-HT_2 antagonist pirenpirone (23) and is decreased in the tail-flick test and increased in the jump test in a diabetic model using alloxan (26). Further, opioid antinociception induced by intermittent cold-water swims is significantly decreased in the tail-flick test and significantly increased in the jump test by pirenpirone (23).

In a schema (38) indicating that different forms of stress-induced antinociception can be characterized as combinations of either opioid or nonopioid and either neural or neurohormonal, 2-DG antinociception appears to possess some opioid characteristics. Whereas it displays both synergy and two-way cross-tolerance with morphine antinociception, it is unaltered following naloxone pretreatment (9,36). Whereas parachlorophenylalanine significantly reduced morphine antinociception, 2-DG antinociception was unaffected by this manipulation (11,37). The 5-HT_3 antagonist ICS-205,930 significantly and similarly reduces antinociception in the tail-flick test induced by 2-DG, morphine, and the κ-agonist U-50,488H (20, 22). However, ICS-205,930 reduced morphine antinociception in the jump test (22), but potentiated 2-DG antinociception in the jump test in the present study. Further, whereas both peripheral and central forms of morphine antinociception are reduced by general and 5-HT_2 antagonists (21,22,30,31,40) these antagonists failed to alter 2-DG antinociception in the tail-flick test, and actually potentiated this effect in the jump test. Thus, whereas 2-DG may activate endogenous opioid pain-inhibitory pathways to produce part of its antinociceptive actions, it appears that other systems are involved in the mediation of the antinociceptive response following this form of glucoprivation.

ACKNOWLEDGEMENTS

This research was supported by PSC/CUNY Grants 662225 and 663216. The authors thank Sandoz Laboratories for their generous gifts of methysergide and ICS-205,930 and Janssen Laboratories for their generous gift of ritanserin.

REFERENCES

1. Badillo-Martinez, D.; Nicotera, N.; Butler, P. D.; Kirchgessner, A. L.; Bodnar, R. J. Impairments in analgesic, hypothermic and glucoprivic stress responses following neonatal monosodium glutamate. Neuroendocrinology 38:438–446; 1984.
2. Basbaum, A. I.; Fields, H. L. Endogenous pain control systems: Brainstem spinal pathways and endorphin circuitry. Annu. Rev. Neurosci. 7:309–338; 1984.
3. Beczkowska, I. W.; Koch, J. E.; Bodnar, R. J. Naltrexone, serotonin receptor subtype antagonists, and glucoprivic intake: I. 2-Deoxy-D-glucose. Pharmacol. Biochem. Behav. 42:661–670; 1992.
4. Besson, J.-M.; Chaouch, A. Peripheral and spinal mechanisms of nociception. Physiol. Rev. 67:67–186; 1987.
5. Bodnar, R. J. Measurement of stress-induced analgesia. In: Conn, P. M., ed. Methods in neuroscience. vol. 14. Paradigms for the study of behavior. New York: Academic; in press.
6. Bodnar, R. J.; Kelly, D. D.; Brutus, M.; Glusman, M. Chronic 2-deoxy-D-glucose treatment: Adaptation of its analgesic, but not hyperphagic properties. Pharmacol. Biochem. Behav. 9:763–768; 1978.
7. Bodnar, R. J.; Kelly, D. D.; Brutus, M.; Greenman, C. B.; Glusman, M. Reversal of stress-induced analgesia by apomorphine, but not amphetamine. Pharmacol. Biochem. Behav. 13:171–175; 1980.
8. Bodnar, R. J.; Kelly, D. D.; Brutus, M.; Mansour, A.; Glusman, M. 2-Deoxy-D-glucose-induced decrements in operant and reflex pain thresholds. Pharmacol. Biochem. Behav. 9:543–549; 1978.
9. Bodnar, R. J.; Kelly, D. D.; Glusman, M. 2-Deoxy-D-glucose analgesia: Influences of opiate and non-opiate factors. Pharmacol. Biochem. Behav. 11:297–301; 1979.
10. Bodnar, R. J.; Kelly, D. D.; Mansour, A.; Glusman, M. Differential effects of hypophysectomy upon analgesia induced by two glucoprivic stressors and morphine. Pharmacol. Biochem. Behav. 11:303–308; 1979.
11. Bodnar, R. J.; Kordower, J. H.; Wallace, M. M.; Tamir, H. Stress and morphine analgesia: Alterations following *p*-chlorophenylalanine. Pharmacol. Biochem. Behav. 14:645–651; 1981.
12. Bodnar, R. J.; Merrigan, K. P.; Wallace, M. M. Analgesia following intraventricular administration of 2-deoxy-D-glucose. Pharmacol. Biochem. Behav. 14:579–581; 1981.
13. Bodnar, R. J.; Nicotera, N. Neuroleptic and analgesic interactions upon pain activity measures. Pharmacol. Biochem. Behav. 16:411–416; 1982.
14. Brown, J. Effects of 2-deoxy-D-glucose on carbohydrate metabolism: A review of the literature and studies in the rat. Metabolism 11:1098–1112; 1962.
15. D'Amour, F. E.; Smith, D. L. A method for determining loss of pain sensation. J. Pharmacol. Exp. Ther. 72:74–79; 1941.
16. Evans, W. O. A new technique for the investigation of some

analgesic drugs on a reflexive behavior in the rat. Psychopharmacology 2:318–325; 1961.
17. Glaum, S. R.; Proudfit, H. K.; Anderson, E. C. Reversal of the antinociceptive effects of intrathecally administered serotonin by a selective $5HT_3$ antagonist. Neurosci. Lett. 95:313–317; 1988.
18. Hasegawa, Y.; Kurachi, M.; Okuyama, S.; Araki, H.; Otomo, S. $5HT_3$ receptor antagonists inhibit the response of K opioid receptors in the morphine-reduced Straub tail. Eur. J. Pharmacol. 190:399–401; 1990.
19. Himsworth, R. L. Hypothalamic control of adrenaline secretion in response to insufficient glucose. J. Physiol. 198:451–465; 1970.
20. Ho, B. Y.; Takemori, A. E.; Attenuation of the antinociceptive action of the selective K-opioid receptor agonist U50,488H by ICS205930. Eur. J. Pharmacol. 178:371–373; 1990.
21. Kiefel, J. M.; Cooper, M. L.; Bodnar, R. J. Inhibition of mesencephalic morphine analgesia by methysergide in the medial ventral medulla of rats. Physiol. Behav. 51:201–205; 1992.
22. Kiefel, J. M.; Cooper, M. L.; Bodnar, R. J. Serotonergic and opioid antagonists in the ventral medulla inhibit mesencephalic morphine analgesia in rats. Soc. Neurosci. Abstr. 18:834; 1992.
23. Kiefel, J. M.; Paul, D.; Bodnar, R. J. Reduction in opioid and non-opioid forms of swim analgesia by $5\text{-}HT_2$ receptor antagonists. Brain Res. 500:231–240; 1989.
24. Koch, J. E.; Hough, L. B.; Bodnar, R. J. Potentiation of 2-deoxy-D-glucose antinociception, but not hyperphagia by zolantidine, a histamine (H_2) receptor antagonist. Pharmacol. Biochem. Behav. 41:371–376; 1992.
25. Lubin, E.; Bodnar, R. J. Intracerebroventricular alloxan reduces 2-deoxy-D-glucose analgesia. Physiol. Behav. 42:465–470; 1988.
26. Lubin, E.; Bodnar, R. J. Differential actions of central alloxan upon opioid and nonopioid antinociception in rats. Pharmacol. Biochem. Behav. 34:511–516; 1989.
27. Millan, M. J.; Bervoets, K.; Colpaert, F. C. 5-Hydroxytryptamine $(5\text{-}HT)_{1A}$ receptors and the tail-flick response. I. 8-Hydroxy-2-(di-*n*-propylamino)tetralin HBr-induced spontaneous tail-flicks in the rat as an in vivo model of $5HT_{1A}$ receptor-mediated activity. J. Pharmacol. Exp. Ther. 256:973–982; 1991.
28. Millan, M. J.; Colpaert, F. C. 5-Hydroxytryptamine $(5\text{-}HT)_{1A}$ receptors and the tail-flick response. II. High efficacy $5HT_{1A}$ agonists attenuate morphine-induced antinociception in mice in a competitive-like manner. J. Pharmacol. Exp. Ther. 256:983–992; 1991.
29. Millan, M. J.; Colpaert, F. C. 5-Hydroxytryptamine $(5\text{-}HT)_{1A}$ receptors and the tail-flick response. III. Structurally diverse $5HT_{1A}$ partial agonists attenuate mu- but not kappa-opioid antinociception in mice and rats. J. Pharmacol. Exp. Ther. 256:993–1001; 1991.
30. Paul, D.; Mana, M. J.; Pfaus, J. G.; Pinel, J. P. J. Attenuation of morphine analgesia by the serotonin type-2 receptor blockers, pirenpirone and ketanserin. Pharmacol. Biochem. Behav. 31: 641–647; 1989.
31. Paul, D.; Phillips, A. G. Selective effects of pirenpirone on analgesia produced by morphine or electrical stimulation at sites in the nucleus raphe magnus and periaqueductal gray. Psychopharmacology (Berl.) 88:172–176; 1986.
32. Peroutka, S. J. 5-Hydroxytryptamine receptor subtypes. Annu. Rev. Neurosci. 11:45–60; 1988.
33. Peroutka, S. J.; Schmidt, A. W.; Sleight, A. J.; Harrington, M. A. Serotonin receptor "families" in the central nervous system: An overview. Ann. NY Acad. Sci. 600:104–113; 1990.
34. Smith, G. P.; Epstein, A. N. Increased feeding response to decreased glucose utilization in the rat and monkey. Am. J. Physiol. 217:1083–1087; 1969.
35. Sperber, E. S.; Kramer, E.; Bodnar, R. J. Effects of muscarinic receptor antagonism upon two forms of stress-induced analgesia. Pharmacol. Biochem. Behav. 25:171–179; 1986.
36. Spiaggia, A.; Bodnar, R. J.; Kelly, D. D.; Glusman, M. Opiate and non-opiate mechanisms of stress-induced analgesia: Cross-tolerance between stressors. Pharmacol. Biochem. Behav. 10: 761–765; 1979.
37. Tenen, S. S. Antagonism of the analgesic effect of morphine and other drugs by *p*-chlorophenylalanine, a serotonin depletor. Psychopharmacologia 12:278–285; 1968.
38. Watkins, L. R.; Mayer, D. J. The neural organization of endogenous opiate and nonopiate pain control systems. Science 216: 1185–1192; 1982.
39. Wick, A. N.; Drury, D. R.; Nakada, H. I.; Wolfe, J. B. Localization of the primary metabolic block produced by 2-deoxy-D-glucose. J. Biol. Chem. 224:963–979; 1957.
40. Yaksh, T. L. Direct evidence that spinal serotonin and noradrenaline terminals mediate the spinal antinociceptive effects of morphine in PAG. Brain Res. 160:180–185; 1979.