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Analgesia Following Intraventricular Administration of 2-Deoxy-D-Gluocse

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BODNAR, R. J , K P MERRIGAN AND M. M WALLACE. *Analgesia following intraventricular administration of 2-deoxy-D-glucose*. PHARMAC. BIOCHEM. BEHAV. **14**(4) 579–581, 1981 —The glucose analogue, 2-deoxy-D-glucose (2-DG) elicits hyperphagic and analgesic responses in rats. The former response appears to be mediated by central processes since overeating is elicited following intraventricular administration of 2-DG at low (3.5 and 5 0 mg/kg) doses The present study examined whether flinch-jump thresholds would increase 30, 90 and 180 min following intraventricular injections of 2-DG at low (3.5, 5.0 and 10.0 mg/kg) doses and compared these effects with systemically-applied 2-DG doses of 350 and 500 mg/kg. Intraventricular 2-DG administration increased jump thresholds for up to 180 min across all test doses Flinch thresholds were also increased, but in a manner dissociated from jump thresholds. Animals with cannulae located near, but not in the lateral ventricle, displayed delayed analgesic effects. The magnitude of intraventricular 2-DG analgesia was not as potent as the 100-fold higher systemic injections It appears that central mechanisms mediated intraventricular 2-DG analgesia at the low dose range since higher, systemic 2-DG doses have previously failed to increase flinch-jump thresholds.

Pain Analgesia 2-Deoxy-D-glucose Rats

THE GLUCOSE analogue, 2-deoxy-D-glucose (2-DG) [5,8] elicits hyperphagic responses in rats [13]. Several studies have postulated that this response is due to activation of central nervous system pathways rather than general cellular glucoprivation. Support for this hypothesis include the observations that systemic 2-DG injections induce peripheral sympatho-medullary and pituitary-adrenal discharge [5,8] and can prevent norepinephrine depletions induced by acute exposure to inescapable foot shock [12]. Furthermore, while acute exposure to either inescapable foot shock or hypothermic stress depletes hypothalamic norepinephrine and eliminates the hyperphagic response to systemic 2-DG, chronic exposure to these two stressors produces the reverse effects [11]. Finally, lesions placed in the lateral hypothalamus and zona incerta eliminate 2-DG hyperphagia [7,15].

That the hyperphagia effects induced by systemic 2-DG are not linked to glucoprivation is demonstrated by the finding that overeating occurs in the absence of glucoprivation if food deprivation occurs 6 hr following the 2-DG injection. Moreover, the time courses of hyperphagic and glucoprivic effects are not correlated since the former is diminished at the 2 hr peak of the latter [10]. Furthermore, while glucoprivation is eliminated by glucose infusion, the hyperphagic response is reduced only when the glucose treatment prevents the onset of glucoprivation [9]. However, one of the most compelling arguments favoring central mediation of 2-DG hyperphagia is the recent observation that 2-DG doses (3.5 and 5.0 mg), incapable of eliciting overeating following systemic injection, elicit hyperphagia following intraventricular (ICV) administration [6].

Systemic administration of 2-DG also induces dose-dependent and time-dependent increases in both operant and reflex pain thresholds, effects which are potentiated by concurrent food deprivation [2]. Moreover, the analgesic, but not the hyperphagic, effects of systemic 2-DG develop tolerance with repeated injections [1] and cross-tolerance with both cold-water swim and morphine analgesia [14]. In addition, morphine and 2-DG analgesia exhibit synergy [3]. The analgesic effects of 2-DG appear to be mediated by specific central nervous system processes since administration of either apomorphine, a dopamine receptor agonist, or capsaicin, a Substance P antagonist, reduces 2-DG analgesia [3,4]. Since all of the above studies involved systemic administration of 2-DG, a further important control to infer central nervous system mediation of 2-DG analgesia would be to determine whether ICV administration of 2-DG would induce analgesia at those doses that induced hyperphagia

METHOD

Male albino Sprague-Dawley rats weighing between 330 and 420 g were anesthetized with Pentothal (50 mg/ml sterile water/kg body weight, IP) and stereotaxically (Kopf) implanted with three stainless steel anchor screws and one stainless steel 22 gauge guide cannula aimed 0.3 mm above the left lateral ventricle With the incisor bar set at +5 mm, lateral ventricle coordinates were 0.5 mm anterior to the bregma suture, 1.3 mm lateral to the sagittal suture, and 3.6 mm from the top of the skull. Five days after surgery, the animals were tested for flinch-jump thresholds in which electric shocks were delivered through a 30 cm by 24 cm floor composed of 16 grids by a 60 Hz constant current shock

TABLE 1

ALTERATIONS IN JUMP AND FLINCH THRESHOLDS FOLLOWING INTRAVENTRICULAR (ICV) AND INTRAPERITONEAL (IP) INJECTIONS OF 2-DEOXY-D-GLUCOSE (2-DG) AS COMPARED TO BASELINE (BL)

Route	2-DG Dose (mg/kg)	n	Time Course (min)		Jump Threshold BL	Jump Threshold 2-DG	Flinch Threshold BL	Flinch Threshold 2-DG	r (Flinch/Jump)
ICV	3 5	11	30	Mean	0 429	0.490	0 152	0 167	+0.76†
				t	3 59†		1 04		
				w^2	0 52		0 01		
			90	Mean	0 436	0 479	0.155	0 177	+0 26
				t	2 15*		1.54		
				w^2	0 25		0.11		
			180	Mean	0 463	0.517	0 169	0 195	+0 48
				t	2 35*		1 61		
				w^2	0.29		0 13		
ICV	5.0	11	30	Mean	0 429	0 489	0.152	0.192	+0.82†
				t	2 41*		2.50*		
				w^2	0 30		0 32		
			90	Mean	0.436	0 503	0 155	0 180	+0 26
				t	2 16*		1 95		
				w^2	0.25		0 20		
			180	Mean	0.463	0.502	0.169	0 203	+0 68*
				t	1.18		1 58		
				w^2	0.03		0 12		
ICV	10 0	7	30	Mean	0 435	0.486	0 157	0 176	+0 11
				t	2 32*		1 34		
				w^2	0 38		0 10		
			90	Mean	0.454	0.476	0.152	0.179	+0 40
				t	1.16		2.01		
				w^2	0 05		0 30		
			180	Mean	0 474	0 506	0.173	0.196	−0.30
				t	2 46*		2 12		
				w^2	0.42		0.33		
IP	350 0	8	30	Mean	0 406	0 715	0 165	0 300	+0 07
				t	7.54†		3.88†		
				w^2	0 87		0 64		
			90	Mean	0 399	0 595	0 156	0.270	+0 12
				t	4 26†		4.67†		
				w^2	0.68		0 72		
			180	Mean	0 421	0.518	0 159	0.206	−0 25
				t	5 11†		2 60*		
				w^2	0 76		0 42		
IP	500.0	8	30	Mean	0 406	0.756	0 165	0.378	+0.73*
				t	4.86†		5 44†		
				w^2	0 74		0 78		
			90	Mean	0 399	0 624	0 156	0.288	+0 67*
				t	4 33†		4 51†		
				w^2	0.69		0 71		
			180	Mean	0 421	0.542	0 159	0 215	+0 81*
				t	3 67†		4 16†		
				w^2	0 61		0.67		

*$p<0$ 05; †$p<0$ 01.

generator and an electromechanical grid scrambler. Using an ascending method of limits of successively more intense shocks, the flinch threshold was defined in mA as the lowest intensity that elicited a withdrawal of a single paw from the grids. The jump threshold was defined as the lowest of two consecutive intensities that elicited simultaneous withdrawal of both hindpaws from the grids. Each trial began with the animal receiving a 300 msec foot shock at a current intensity of 0.1 mA Subsequent shocks occurred at 10 sec intervals and were increased in equal 0.05 mA steps until each nociceptive threshold was determined. After each trial, the current intensity was reset to 0.1 mA for the next trial until 6 trials were completed. Daily flinch and jump thresholds, computed as the mean of these six trials, were determined post-operatively over 4 days.

Rats were tested on a weekly schedule which consisted of

paired placebo and experimental drug sessions with at least 48 hr elapsing between sessions. In one group, animals received intraperitoneal injections in an incomplete counterbalanced fashion: (a) 350 mg/kg of 2-DG (300 mg/ml sterile water/kg body weight); (b) 500 mg/kg of 2-DG; and (c) vehicle (1.67 ml sterile water/kg body weight). In a second group, animals received ICV injections in an incomplete counterbalanced fashion: (a) 3.5 mg of 2-DG dissolved in 7 μl of sterile water; (b) 5.0 mg of 2-DG dissolved in 10 μl of sterile water; and (c) 10 μl of sterile water. In a third group, half of the animals received ICV injections of 10 mg of 2-DG dissolved in 20 μl of sterile water and 20 μl of sterile water alone, while the remainder received the reverse sequence. The ICV injections were infused at a rate of 1 μl every 15 sec through a stainless steel 28 gauge internal cannula which extended 0.5 mm ventral to the guide cannula. Flinch-jump thresholds were determined 30, 90 and 180 min after all injections. The experimenter conducting the flinch-jump tests was uninformed of the specific experimental conditions.

After completion of the experiment, all animals were overdosed with pentothal and perfused through the heart with normal saline followed by 10% Formalin. Serial frozen 40 μm sections were taken around the cannula placement and were stained with luxol fast blue and cresyl violet. The cannula placement of each animal was determined by light microscopic examination of the sections by an observer uninformed as to the behavioral results.

RESULTS

As summarized in Table 1, ICV administration of 2-DG induced significant increases in jump thresholds across the three chosen doses which persisted over the post-injection time course. These significant effects accounted for between 25 and 52% of the variance. As expected, intraperitoneal injections of 2-DG produced significant time-dependent increases in jump thresholds which accounted for between 61 and 87% of the variance. Flinch thresholds produced more inconsistent effects and only correlated consistently with jump thresholds following the 500 mg/kg dose of systemic 2-DG. Finally, the cannulae of five additional animals impinged on structures dorsal or lateral to the ventricle. These animals displayed delayed increases in jump thresholds following central injections: 3.5 mg/kg–93% of baseline at 30 min, 107% at 90 min, 116% at 180 min; 5.0 mg/kg–103% at 30 min, 117% at 90 min, 116% at 180 min.

DISCUSSION

The present study supports the contention that the analgesic effects of 2-DG are mediated by central pain-inhibitory mechanisms since ICV administration of 2-DG at doses of 3.5, 5.0 and 10.0 mg produce significant analgesia that persists for up to 3 hr. By contrast, while systemic 2-DG injections at doses of 350, 500 and 700 mg/kg increase jump thresholds, the lower doses of 100 and 225 mg/kg administered systemically do not [2]. Furthermore, animals with cannulae near, but not within the lateral ventricle displayed delayed analgesic effects. Moreover, the more systematic increases occurred in the jump rather than the flinch thresholds suggesting that such changes occurred in reactivity to the noxious properties of, rather than the detection of electrical stimuli. Although central administration of 2-DG at these low doses produced significant analgesia, the accounted variance of between 25 and 52% was less powerful than the accounted variance of between 61 and 87% for the systemic injections. This result parallels the smaller hyperphagic responses induced by ICV 2-DG as compared to systemic 2-DG at 100-fold concentrations [6]. Taken together, however, while central sites of action do not account for the entire hyperphagic and analgesic effects of 2-DG, they do appear to be necessary for their occurrence [3, 4, 6, 7, 15].

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