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Stimulation of Prolactin Release in Rats by GABA (39139)

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Hypothalamic biogenic amines have been reported to be involved in regulating secretion of hypophysiotropic releasing factors and pituitary hormones. In general, catecholamines have been found to be inhibitory and serotonin to be stimulatory to prolactin release (1). More recently, acetylcholine has been observed to inhibit prolactin release (2). There is some evidence that GABA may function as a neurotransmitter in the mammalian central nervous system and it may influence pituitary hormone secretion (3). A significant proportion of the total brain GABA content has been reported to be in the diencephalon (4). Markara *et al.* (5) reported that ACTH release in response to surgical trauma can be blocked by intraventricular infusion of GABA and potentiated by GABA antagonists. Ondo (6) recently observed that intraventricular infusion of GABA into male rats induced LH but not FSH release. It was the purpose of this investigation to determine whether GABA had any effect on PRL release in rats.

Materials and methods. Mature virgin female Sprague–Dawley rats weighing 200–225 g each were obtained from Spartan Research Animals (Haslett, Mich.). The rats were housed in individual cages in a temperature controlled (26 ± 1°) and artificially illuminated room (lights on from 5:00 AM until 7:00 PM daily) and received feed and water *ad libitum*.

In experiments I and II, a cannula made of No. 10 PE tubing was introduced into the right lateral ventricle of each animal under ether anesthesia according to the coordinates and method described by de Balbian-Verster *et al.* (7). In experiment I, after recovery from ventricular cannulation, the rats were infused with solutions of 8 μM GABA (pH 7.4) or 0.9% NaCl (pH 7.4) in a volume of 10 μl on the morning of proestrus. Blood samples were taken by cardiac puncture under light ether anesthesia 5 min prior to and 15, 30, and 60 min following infusion of test solutions. In experiment II, rats were ovariectomized 10 days prior to cannulation, and were infused with 8 or 16 μM GABA (pH 7.4) or 0.9% NaCl in a volume of 10 μl. Blood samples were taken by cardiac puncture under light ether anesthesia 10 min prior to and 15 and 45 min following infusion of test solutions.

To determine if the GABA induced PRL response was mediated through a direct action on the anterior pituitary, mature hypophysectomized male Sprague–Dawley rats weighing 200–225 g were given one anterior pituitary transplant under the left renal capsule and injected with estradiol benzoate (1 μg/100 g) sc once daily for a period of 3 days. On the fourth day, they were injected intraperitoneally with 5, 25, or 50 mg/100 g of GABA (pH 7.4) or 0.9% NaCl (pH 7.4). Blood samples were collected by cardiac puncture under light ether anesthesia at 15 min prior to and 15 and 45 min following injection of test solutions.

Serum PRL was measured by the method of Niswender *et al.* (8). Values were expressed in terms of NIAMD-rat prolactin-RP-1. Student's *t* test was used to test significance of difference between control and experimental blood PRL levels.

Results. In Expt. I (Table I), it can be seen that infusion of 0.9% NaCl into the lateral ventricle of proestrous rats had no effect on serum prolactin levels. A dose of 8 μM GABA infused into the lateral ventricle produced a twofold rise in serum PRL levels by 15 min after infusion ($P < .01$), but by 30 and 60 min serum PRL levels were

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not significantly different from pretreatment values.

In Expt. II (Table I), infusion of saline had no effect on serum prolactin values in ovariectomized rats. A dose of 8 or 16 μM GABA produced approximately a threefold rise in serum PRL levels at 15 min after infusion ($P < .01$), but at 45 min, serum PRL levels had returned to pretreatment values.

In Expt. III (Table II), none of the doses of GABA injected ip into estrogen-primed hypophysectomized male rats with a single pituitary transplant was able to significantly alter serum PRL levels at 15 or 45 min after injection. Injection of saline also did not alter serum PRL values.

Discussion. These results indicate that GABA can stimulate PRL release by 15 min after infusion into the lateral ventricle of proestrus or ovariectomized female rats. This PRL rise showed a return to pretreatment levels by 30 or 45 min after infusion. In a personal communication, Ondo (1975) also observed PRL release after GABA infusion into the third ventricle of rats. Our results suggest that this effect of GABA on serum PRL is not mediated by a direct action on the pituitary, since there was no response when GABA was injected ip into estrogen-primed hypophysectomized male rats with a pituitary transplant. Furthermore, we also have observed that when rat anterior pituitary halves were incubated with several doses of GABA and medium samples were removed during a 4-hr period, there was no significant effect on PRL release (unpublished). This suggests that GABA may act through the CNS.

There have been conflicting reports regarding the role of GABA as an inhibitory or excitatory neurotransmitter in the mammalian CNS. Makara *et al.* (5) stated that GABA acts as an inhibitory neurotransmitter in the neural circuits regulating CRF production and ACTH secretion. Sangiah *et al.* (9) were able to stimulate catecholamine release from isolated bovine adrenal glands perfused with GABA. Dreifuss *et al.* (10) reported that electrophoretic infusions of GABA into the rat hypothalamus were able to reduce the firing rate of cells in the hypothalamus. If the GABA action on PRL release is mediated through biogenic amines, it may be either inhibitory or excitatory. It may stimulate PRL release by in-

TABLE I. EFFECT OF INTRAVENTRICULAR GABA INFUSION ON SERUM PRL LEVELS IN FEMALE RATS.

Treatment and number of rats	Serum PRL (ng/ml)					
	−10 min	− 5 min	+15 min	+30 min	+45 min	+60 min
			Expt I, Proestrous rats			
Saline controls (5)		60.4 ± 12.6	74.0 ± 12.0	85.3 ± 18.5		80.8 ± 23.6
8 μM GABA (10)		43.1 ± 8.9	94.6 ± 13.6*	60.1 ± 12.7		70.1 ± 12.0
			Expt II, Ovariectomized rats			
Saline controls (7)	17.7 ± 3.2		17.3 ± 1.6		18.6 ± 2.4	
8 μM GABA (8)	9.9 ± 1.4		29.5 ± 5.8*		8.1 ± 1.4	
16 μM GABA (8)	14.4 ± 2.0		41.7 ± 6.6*		12.9 ± 1.6	

* $P < .01$.

TABLE II. EFFECT OF SYSTEMIC GABA INJECTION ON SERUM PRL LEVELS IN HYPOPHYSECTOMIZED MALE RATS WITH A PITUITARY TRANSPLANT.

Treatment and number of rats	Serum PRL (ng/ml)		
	−15 min	+15 min	+45 min
Saline controls (8)	49.0 ± 10.4	54.8 ± 11.2	40.7 ± 7.3
GABA, 5 mg/100 g (8)	42.9 ± 10.0	51.2 ± 8.0	43.7 ± 9.3
GABA, 25 mg/100 g (8)	41.8 ± 8.0	40.0 ± 8.0	39.6 ± 10.1
GABA, 50 mg/100 g (11)	51.0 ± 9.1	56.9 ± 10.7	55.2 ± 9.9

hibiting catecholamine activity or by promoting serotonin activity. It is interesting that GABA induces release of both LH (6) and prolactin, since simultaneous release of the two hormones usually does not occur under most conditions. The mechanism of GABA action on the neuroendocrine system as well as any physiological role it may have in regulating prolactin secretion remains to be investigated.

Summary. Infusion of GABA into the lateral ventricle of intact female rats on the morning of proestrus and in ovariectomized rats significantly stimulated PRL release. This response apparently is not mediated through a direct action on the pituitary since injection of GABA into hypophysectomized rats with a pituitary transplant under the kidney capsule did not alter serum prolactin levels. These observations suggest that GABA may have a role in regulating prolactin secretion.

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