Physiology & Behavior, Vol 20, pp 91–93 Pergamon Press and Brain Research Publ , 1978 Printed in the U S.A

BRIEF COMMUNICATION

Effects of Ether Stress on Prolactin and Corticosterone Levels in Prenatally-Stressed Male Rats as Adults

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(Received 25 July 1977)

POLITCH, J A , L R HERRENKOHL AND R R GALA *Effects of ether stress on prolactin and corticosterone levels in prenatally-stressed male rats as adults* PHYSIOL BEHAV 20(1) 91–93, 1978 – The effect of a potent prenatal stressor on adult levels of prolactin and corticosterone was investigated in male rats It was found that prenatal stress had no effect on initial levels of either of these two hormones Ether stress levels of prolactin were significantly lower in prenatally-stressed animals compared to prenatally-nonstressed subjects, whereas corticosterone levels in response to ether stress were only marginally affected in prenatally-stressed animals reared by prepartally-stressed mothers It is suggested that alterations in endocrine function in the adult animal may result from prenatal stress effects on the development of neural mechanisms that regulate the hormonal response to ether stress

Ether stress Prolactin Corticosterone Prenatal stress

IT HAS been shown that during early postnatal life, several factors, including behavioral experience and hormonal milieu, can affect the development and function of various endocrine subsystems and their responsiveness to stress in later life [1, 11, 17, 21] In the case of prenatal factors, it has been shown that alterations in the maternal pituitary-adrenocortical system during pregnancy have long-lasting effects on the pituitary-adrenocortical system of offspring as adults [11,13] However, few studies have investigated the role of prenatal experience on later endocrine function Previous studies have shown that prenatal stress may feminize and demasculinize the sexual behavior of male rats [8, 22, 23] The following experiment was performed to determine the effects of a potent prenatal stressor on adult levels of corticosterone and prolactin in male rats, both of these hormones are known to be responsive to stress [2, 4, 7, 9, 15, 18, 20]

METHOD

Animals and Procedure

Twenty primiparous, pregnant Sprague-Dawley rats, weighing about 250 g were obtained from Zivic Miller (Allison Park, PA) one week before stressing They were housed singly under a standard 12 hr light/dark cycle and maintained on ad lib food and water On Day 14 of gestation, 10 pregnant females were selected at random and subjected to simultaneous stressors of bright light, heat and restraint through Day 21 according to methods modified from Ward [22] Stressed females were placed individually in 18 × 8 cm semicircular Plexiglas restraining cages grouped in rows under four bright incandescent lights which produced a surface illumination of more than 400 ft-candles and surface temperature of 34°C The remaining 10 pregnant females were left unhandled in their home cages On the day of birth, litters were cross-fostered according to a 2 × 2 experimental design in which prepartally-stressed mothers reared nonstressed pups, nonstressed mothers reared prenatally-stressed pups, prepartally-stressed mothers reared stressed foster pups and nonstressed mothers reared nonstressed foster pups At 21 days of age, prenatally-stressed and nonstressed offspring were weaned, segregated by sex and housed 2 per cage At approximately 60 days of age, all animals were placed in single cages

At approximately 160 days of age, 10 prenatally-stressed males reared by stressed or nonstressed mothers (n = 5 in each condition) and 10 nonstressed males (n = 5 per group)

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TABLE 1

EFFECTS OF ETHER STRESS ON CORTICOSTERONE AND PROLACTIN IN PRENATALLY-STRESSED MALES AS ADULTS

	Groups			
	Prenatally Stressed Males reared by		Prenatally Nonstressed Males reared by	
	Stressed Mothers	Nonstressed Mothers	Stressed Mothers	Nonstressed Mothers
Measures	SS (n = 5)	SN (n = 5)	NS (n = 5)	NN (n = 5)
Prolactin ($ng/ml \bar{X} \pm SE$)				
Initial Level	21 08 ± 1 85	21 40 ± 1 62	22 14 ± 1 96	26 74 ± 3 22
Combined	21 24 ± 1 16		24 44 ± 1 94	
Ether Stress	22 74 ± 1 77	31 34 ± 5 53	44 24 ± 10 65	38 10 ± 7 83
Combined	27 04 ± 3 08		* 41 17 ± 6 31	
Corticosterone ($\mu g/100\ ml \bar{X} \pm SE$)				
Initial Level	13 40 ± 6 73	15 00 ± 2 07	12 80 ± 2 56	12 00 ± 3 24
Combined	14 20 ± 3 33		12 40 ± 1 95	
Ether Stress	32 60 ± 5 10	35 80 ± 7 70	32 20 ± 4 96	47 40 ± 6 73
Combined	34 20 ± 4 38		39 80 ± 4 69	

were selected at random and subjected to a modification of a procedure routinely used to evaluate the role of a variety of experimental manipulations on pituitary-adrenocortical activity in the adult rat [11] All animals were subjected to 2 samplings of blood, each approximately 1 5 ml, via cardiac puncture Each animal was placed under ether anesthesia and an initial sample was obtained within 2 min after removal from the home cage This sample was used to determine initial levels of corticosterone and prolactin Ether stress levels of these hormones were obtained from a second sample taken from the same animal following additional ether anesthesia administered 15 min after the initial removal from the animal's home cage In the interim between the first and second samplings, animals were placed in neutral retaining cages The order of blood sampling was counter-balanced over the various stress and rearing conditions and blood collection took place between 9 30 a m and 1 30 p m in order to minimize diurnal fluctuations in hormone secretion Upon obtaining each 1 5 ml sample, 0 5 ml was placed in a heparinized cuvette and subsequently centrifuged for plasma removal The remaining 1 0 ml was placed in a non-heparinized cuvette and was allowed to stand for 3–4 hr This blood was then centrifuged for serum removal The plasma was assayed for corticosterone according to a modification of the microfluorometric procedure of Glick, von Redlich and Levine [5] and the serum was assayed for prolactin by a double antibody radioimmunoassay at two dilutions each in duplicate as previously described [10]

RESULTS

Table 1 summarizes the means ± the standard errors of the initial and ether stress-induced levels of serum prolactin and plasma corticosterone Analysis by two-tailed Mann-Whitney U tests revealed no significant differences among prenatal stress or rearing conditions in the initial levels of serum prolactin or plasma corticosterone Furthermore, levels of ether stress prolactin did not differ between rearing conditions within the prenatal conditions However, when the rearing conditions under each prenatal condition were combined to form two larger groups, a prenatal stress group and a prenatal nonstress group, a Mann-Whitney U test revealed that the prenatal stress group had significantly lower ether stress levels of serum prolactin than the prenatal nonstress group ($U = 23$, $n_1 = n_2 = 10$, $p<0\ 05$) None of the differences in plasma corticosterone levels was significant, although there was some tendency for stressed animals (regardless of rearing conditions) and nonstressed animals reared by stressed mothers to have lower ether stress corticosterone levels than prenatally-nonstressed animals reared by nonstressed mothers

DISCUSSION

The results of the present study show that prenatal stress produces a decrease in the levels of prolactin in response to ether stress in adulthood in male rats The suppression of stress-induced prolactin levels may occur as the result of the effect of prenatal stress on the development of neural mechanisms that regulate the prolactin response to ether stress In support of this possibility is the observation that a number of neurotransmitters, including dopamine, norepinephrine, serotonin, and histamine, have been implicated in the ether-induced release of prolactin [6, 12, 19] Furthermore, Moyer, Herrenkohl and Jacobowitz [14] have found that the same prenatal stressors as those utilized in the present experiment cause widespread changes in steady state levels of dopamine and norepinephrine in the adult brain Specifically, they found significantly lower

steady state concentrations of norepinephrine in the medial preoptic nucleus and median eminence of prenatally-stressed males compared to nonstressed males The norepinephrine levels of the prenatally-stressed males in these two areas were not significantly different from those of normal (nonstressed) females It is interesting to note that electrochemical stimulation of the medial preoptic nucleus has been found to facilitate the release of prolactin as evidenced by the induction of pseudopregnancy in female rats [16] Furthermore, the median eminence is thought to be a site involved in the estrogen-stimulated release of prolactin in female rats [3]

There was a tendency for prenatally-stressed males to have lower ether stress corticosterone levels than prenatally-stressed males reared by non-stressed mothers In the case of corticosterone, although the results are only suggestive, they are consistent with studies involving the offspring of mothers having high levels of pituitary-adrenal activity during pregnancy Offspring of mothers bearing ACTH-secreting tumors show suppressed adrenal response to ether stress as adults [13] In addition, Levine [11] reported that male offspring of adrenalectomized mothers exhibit a similar suppression of adrenocortical activity in response to ether in adulthood Both of these effects are thought to be the result of elevated plasma corticosteroid levels in the fetuses of adrenalectomized mothers and mothers implanted with ACTH-secreting tumors [11] In a similar fashion, it is possible that elevated corticosteroid levels (of fetal and/or maternal origin) in prenatally-stressed males as fetuses may have had a suppressive effect on the corticosteroid response to ether in adulthood

ACKNOWLEDGEMENTS

This research was supported by a Biomedical Science Support Grant awarded to the Interdisciplinary Graduate Program in Psychobiology at Temple University and by a Grant-in-Aid from Temple University The authors thank Dr Alan Leshner for the use of his facilities in the Psychology Department, Bucknell University, where the plasma corticosterone assays were performed Serum prolactin assays, performed in Dr Gala's laboratory, were supported in part by NSF Grant BMS 74–17332

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