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BRIEF COMMUNICATION

Influence of Chorionic Gonadotrophin on Brain Amine Levels in Male Rats

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MONTGOMERY, R. L. AND E. L. CHRISTIAN. *The influence of chorionic gonadotrophin on brain amine levels in male rats.* PHARMAC. BIOCHEM. BEHAV. 1(6) 735–737, 1973. Chorionic gonadotrophin did not influence brain amine levels significantly in control animals. Hypophysectomized rats treated with chorionic gonadotrophin showed nonsignificantly elevated levels of brain amines. Castration of male rats resulted in significant decreases in brain amine levels ($p<0.005$). Castrated rats treated with chorionic gonadotrophin showed no significant changes in brain amine levels.

Castration Hypophysectomy Brain amines Chorionic gonadotrophin Hypophysectomized-castrated

EXPERIMENTAL evidence suggests that biogenic amines, norepinephrine (NE), dopamine (DA) and serotonin (5-HT) are involved in regulating the release of pituitary luteinizing hormone (LH). Stefano and Donoso [17] reported that hypothalamic norepinephrine in rats is highest at proestrus when compared with the levels at other states of the estrous cycle. Several investigators have also reported that drugs which deplete brain norepinephrine block ovulation [4]. Kordon and Glowinski [12] devised two experiments, both of which strongly suggested that dopamine and not norepinephrine was involved in regulation of LH release. This information appeared to be in agreement with the histochemical data obtained by Fuxe and Hokfelt [8]. In addition to the possibilities that norepinephrine (NE) and dopamine (DA) have regulating influences on the release of LH, the indolamine serotonin (5-HT) has been reported to inhibit ovulation [11,15]. Moreover, the concentrations of these brain amines are influenced by the level of gonad function. The castration of male or female rats has been reported [6,17] to cause increased levels of hypothalamic norepinephrine.

The negative-feedback concept proposes that the release of LH from the pituitary is inversely related to gonadal steroid levels. Current concepts of neuroendocrine functions include the assumption that sex steroids influence both the anterior pituitary and hypothalamus [5,7]. In addition to the more classical external feedback mechanism, current data indicate that LH may exert some control over its own production by way of a short of internal feedback mechanism [14,16].

Since the location of steroid sensitive areas of the brain [13,18] apparently correlates well with the monoaminergic synaptic regions, it is therefore the purpose of this experiment to determine if aminergic systems are involved in steroid feedback mechanisms.

METHOD

One-hundred-sixty male, adult rats of the Sprague-Dawley strain weighing 250–350 g were used in this experiment. They were housed in a room having a constant temperature of 24° to 25°C. Food and water were available for consumption ad lib.

Forty rats were hypophysectomized and forty were castrated. Forty rats received both operations. The endocrine glands of the forty remaining rats remained intact and these animals were grouped as controls.

Two weeks following the manipulation of endocrine glands, rats of each of the four groups (controls, hypophysectomized, castrated and castrated-hypophysectomized) were equally divided. Twenty rats in each of the groups were injected subcutaneously with 1.5 ml of isotonic salt solution daily for a period of three days. The remaining twenty rats in each of the four groups received 1.5 ml (300 international units total) of chorinoic gonadotrophin (HCG) daily for a period of three days. The chorionic gonadotrophin (HCG) was obtained from the Nutritional Biochemicals Corporation in Cleveland, Ohio with the understanding that it contained primarily LH with a small amount of FSH.

All rats were sacrificed with a guillotine on the day following the third injection. The brains were removed, weighed, and levels of brain amines determined by spectrophotofluorometric techniques, using a modified version (we

did not extract 5-HIAA) of the Welch and Welch method [19].

RESULTS

Our data clearly indicated that the manipulation of certain endocrine glands of the rat influenced brain amine levels. As shown in Table 1, hypophysectomy alone, resulted in no significant alterations in NE, DA or 5-HT levels. Castration, alone, however, resulted in significant decreases ($p<0.005$) in all three brain amines studied. A similar decrease was observed with the rats which were both hypophysectomized and castrated. When compared with control rats injected with isotonic salt solution, the controls injected subcutaneously with 300 international units of HCG daily for three days showed no significant differences in NE, DA, and 5-HT concentrations. As compared with the level of these amines found in HCG injected control rats, the following alterations in levels of NE, DA, and 5-HT were found in the endocrine manipulated, HCG injected rats: (1) Significant increases ($p<0.005$) in brain amines in the hypophysectomized groups. (2) Significant decreases ($p<0.005$) of brain amines in the castrated rats. (3) Significant decreases ($p<0.005$) of brain amines in the castrated-hypophysectomized rats.

When the levels of amines of each of the hypophysectomized, castrated and castrated-hypophysectomized non-HCG-injected groups were compared with those of the HCG injected groups of each similarly endocrine manipulated type, nonsignficant increases of amine levels were found in each case.

DISCUSSION

Several investigators [17] have reported increased norepinephrine and decreased dopamine levels in the anterior hypothalamus of castrated animals. Anton-Tay and Wurtman [1] reported that the total norepinephrine content of the brain was increased about 15 percent twenty days after gonadectomy; however, these increases were not always statistically significant. Graber and Nalbandov [9] reported significant increases in norepinephrine content of the ventral hypothalamus of castrated chickens. We observed statistically significant decreases in norepinephrine, dopamine, and serotonin in whole brains of castrated rats ($p<0.005$). Bernard and Paolino [1] reported norepinephrine levels in castrated mice to be significantly lower than the sham-castrated controls. They also reported the turnover rate of norepinephrine in castrated animals to be significantly decreased. A plausible explanation for decreased levels of brain amines versus increased levels following castration may be associated with specific areas of the brain (hypothalamus) versus whole brain studies.

The failure of hypophysectomy to alter brain amine levels may be related to the presence of adrenal glands which may produce androgenic compounds. The significant decrease of brain amines ($p<0.005$) in castrated-hypophysectomized rats is probably related to the effects

TABLE 1

THE INFLUENCE OF CHORIONIC GONADOTROPHIN ON BRAIN AMINE LEVELS IN MALE RATS

	Nontreated Rats ngs/g of Brain	Chorionic Gonadotrophin (HCG) Treated Rats, ngs/g of Brain
NOREPINEPHRINE NE		
Controls	396 ± 82	425 ± 80
Hypophysectomized	475 ± 107	*595 ± 86
Castrated	*187 ± 58	*208 ± 69
Castrated-Hypophysectomized	*204 ± 53	*210 ± 34
DOPAMINE DA		
Controls	1213 ± 308	1238 ± 204
Hypophysectomized	1368 ± 259	*1878 ± 351
Castrated	*365 ± 55	*389 ± 100
Castrated-Hypophysectomized	*422 ± 99	*495 ± 136
SEROTONIN 5-HT		
Controls	677 ± 114	756 ± 122
Hypophysectomized	994 ± 350	*1241 ± 296
Castrated	*235 ± 31	*247 ± 34
Castrated-Hypophysectomized	*249 ± 43	*263 ± 29

n = 160 *$p<0.005$

of castration and not hypophysectomy. Bliss *et al.* [3] reported decreased testicular functions in rats following the depletion of brain amines which may be indirectly related to our data which shows decreased brain amine levels following the removal of the testes.

Our data concerning chorionic gonadotrophin and its influence on brain amines add to the hypothesis that testosterone levels influence brain amine levels. The failure of HCG to alter brain amine levels in control rats may be due to one of several reasons. (1) HCG does not influence the level of brain amines in animals with normal functioning testes. (2) Larger injections of HCG for longer durations are required. (3) A pure pituitary luteinizing hormone (LH) is required. Hypophysectomized rats injected with HCG showed statistically significant increases ($p<0.005$) in brain amine levels when compared with HCG injected control rats. The HCG probably stimulated the testis and resulted in increased levels of testosterone which may have increased levels of NE, DA and 5-HT. If this hypothesis is correct, then reasons 2 and 3 may be eliminated. Castrated rats injected with HCG showed statistically significant decreases in brain amine levels when compared with HCG injected control rats. ($p<0.005$). This decrease in brain amines was related to the influence of castration and not HCG. Similar observations were noted with the castrated hypophysectomized rats ($p<0.005$). The same hypothesis applies here, whereby castration and not hypophysectomy caused decreased brain amine levels.

We therefore, conclude from this data the following: (1) castration causes a decrease in brain amines (NE, DA and 5-HT); (2) HCG has no significant influence on the level of brain amines in rats with normal testicular function; (3) HCG has a significant influence on the level of brain amines in hypophysectomized animals with intact testes; and (4) testosterone levels influence brain amines.

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