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Acute and Chronic Effects of Sulpiride on Serum Prolactin and Gonadotropin Levels in Castrated Male Rats (38581)

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(Introduced by A. L. Gimeno)

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It is a well known fact that some tranquilizing drugs can alter gonadotropin and prolactin levels in the serum of patients (1, 2), and also in other mammalian species (3). *N*-(ethyl-1-pyrrolidinil-2)methyl-methoxy-sulfamoyl-5-benzamide (Sulpiride) was demonstrated to induce galactorrhea and amenorrhea in humans (4) and to cause considerable hypertrophy of mammary glands in the rat (5). It was then considered of interest to study the effects of acute and chronic administration of sulpiride on serum prolactin and gonadotropin levels in rats.

Materials and Methods. Male adult rats of the Wistar strain (mean wt: 300 *g*) were used. The animals were housed in quarters equipped with controlled temperature (21 C) and illumination (14 hr light–10 hr darkness), and were fed Purina laboratory chow, fresh vegetables and tap water *ad lib*. The rats were castrated through a scrotal incision and were given 30,000 IU of penicillin to prevent infections. The rats were used 3 mo later. Initially, the effect of an acute intravenous injection of sulpiride was studied. Fifteen rats were anesthetized with 25% urethane (0.6 ml/100 *g* body wt) and divided into three groups of five rats each. In all the rats the jugular veins were then exposed and the animals of the first group received an intravenous injection of saline, whereas the animals of the second and third group were injected with a solution of sulpiride sulphate at a dose of 1 mg or 7 mg per rat, respectively. Blood was collected from the jugular vein before and 30, 60 and 120 min after the injections of saline or sulpiride. Sera were separated by centrifugation and kept frozen until assayed for gonadotropins and prolactin. In the second part of the experiment, the effect of a chronic administration of sulpiride was studied. Twenty four castrated rats were separated into three groups of eight rats each, the first being treated with saline, the second and the third group were treated with sulpiride sulphate at a daily dosage of 1 mg or 7 mg per rat, respectively. The solutions were administered subcutaneously and the treatment lasted 13 days. The day after the last injection the rats were decapitated and blood was collected from the trunk. Sera were separated and kept frozen until assayed for gonadotropins and prolactin. Prolactin was measured in duplicates in each serum sample using the double-antibody radioimmunoassay described by Niswender *et al.* (6) with kits distributed by the N.I.A.M.D.D., Bethesda. FSH was determined using the double-antibody radioimmunoassay described by Daane and Parlow (7), with kits also distributed by the N.I.A.M.D.D. LH was determined with the double-antibody radioimmunoassay described by Niswender *et al.* (8), using purified ovine LH (gift of Dr. L. E. Reichert) which was iodinated with ^{125}I, an anti-ovine LH serum (obtained from Dr. G. D. Niswender) and NIAMDD-Rat-RPl as a standard preparation. The significance of the differences among groups was determined using the analysis of variance and Student's *t* test in the first part of the experiment, whereas in the second part Student's test was applied, using as variance estimate the combination of the variance estimates of the three groups. The *t* test was applied to the log of data in the case of prolactin values (9).

Results and Discussion. In the first experiment (Fig. 1), the injection of 1 mg or 7 mg of sulpiride sulphate induced a significant increase in serum prolactin levels ($P < 0.01$)

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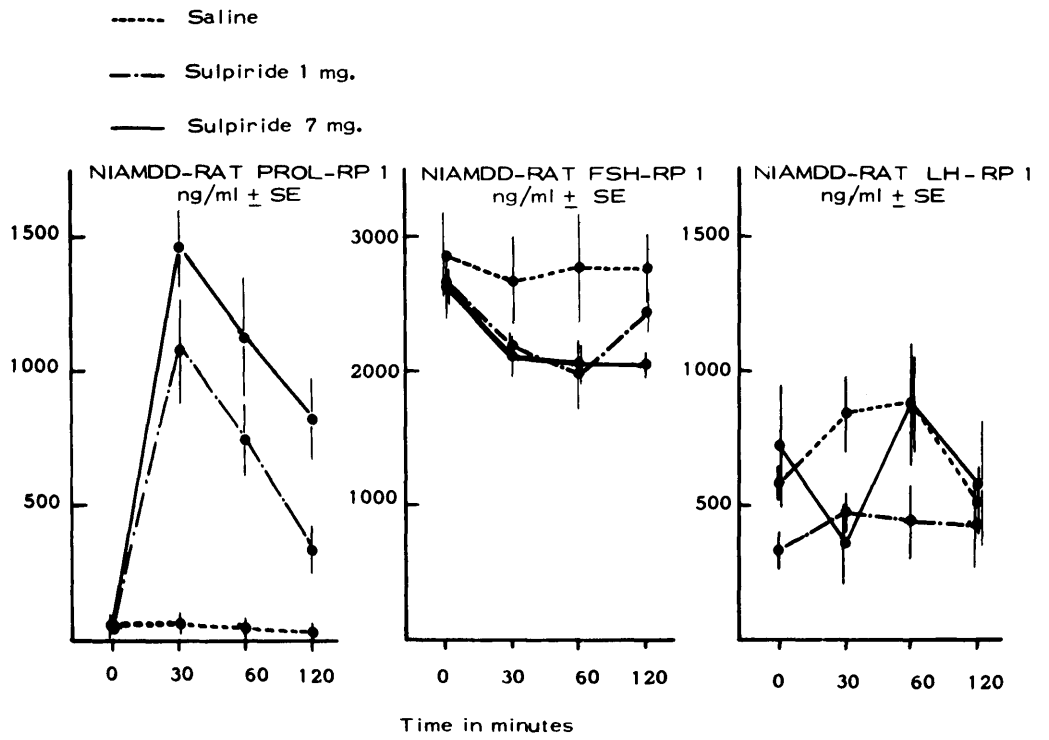

FIG. 1. Effect of an acute intravenous injection of sulpiride on prolactin, FSH, and LH level in serum of castrated male rats.

TABLE I. EFFECT OF SULPIRIDE ON PROLACTIN, FSH AND LH LEVELS IN SERUM OF CASTRATED MALE RATS

Treatment	Dose/Rat	Prolactin[a]	FSH[b]	LH[c]
Saline (8)[e]	—	61.0 ± 16.3	3209 ± 466.0	899.0 ± 134.2
Sulpiride (8)	1 mg	247.7 ± 73.6[d]	4016 ± 408.3	740.0 ± 137.6
Sulpiride (8)	7 mg	186.5 ± 31.5[d]	3478 ± 335.5	594.1 ± 127.7

Duration of the treatment: 13 days.
[a] Expressed as NIAMDD-Rat Prolactin-RP1, ng/ml ± SE.
[b] Expressed as NIAMDD-Rat FSH-RP1, ng/ml ± SE.
[c] Expressed as NIAMDD-Rat LH-RP1, ng/ml ± SE.
[d] $P < 0.01$.
[e] Number of rats in parentheses.

30 min later. Sixty minutes after the injection, serum prolactin levels dropped, but were still significantly higher than before the administration of sulpiride ($P < 0.001$). One hundred and twenty minutes after the injection of sulpiride, serum prolactin levels decreased further as compared with the values at 30 and 60 min, but were still higher than the preinjection values ($P < 0.001$). The injection of saline did not modify significantly the serum prolactin levels. Serum FSH showed no significant modifications after the injection of saline, and showed no significant changes after the injection of sulpiride at both dose levels (Fig. 1). Preinjection values of serum LH in the groups injected with 1 mg of sulpiride were significantly higher than the preinjection values in the group given saline ($P < 0.05$). Sulpiride did not significantly modify serum LH levels.

In the second experiment, chronic treatment with both doses of sulpiride induced a

significant increase in serum prolactin levels as compared with those of saline-injected rats (Table 1) ($P < 0.01$). Serum FSH levels were not significantly different in any of the three groups and serum LH levels in saline and sulpiride-treated rats were not significantly different either.

From the result presented it seems evident that sulpiride was able to induce a fast and considerable increase in serum prolactin levels. Although it is not clear whether this action is exerted on the hypothalamus or directly on the anterior pituitary, our recent in vitro studies indicate that sulpiride can act directly on the pituitary gland (10). It remains to be demonstrated however, whether sulpiride can also act at the hypothalamic level, perhaps through a modification of the biogenic amine content in the hypothalamus. The principal effect of sulpiride on the pituitary secretions seems to be on prolactin, since the effect on gonadotropins was not marked enough to show significant differences in the present investigation. In postmenopausal women, however, sulpiride was shown to decrease significantly serum gonadotropin levels (11). The decrease in serum gonadotropin levels might explain the appearance of amenorrhea in women treated with sulpiride. In case that the effect of sulpiride on prolactin levels described in the present investigation would be confirmed in humans as well, this fact may explain the presence of galactorrhea in women under treatment with sulpiride.

Summary. The effect of the acute or chronic administration of sulpiride on serum prolactin and gonadotropin levels was studied in castrated male rats. Sulpiride administered in acute intravenous injections induced a quick increase in serum prolactin levels and no significant changes in serum gonadotropins. The peak in serum prolactin was observed 30 min after the injection of sulpiride, with a decrease of serum prolactin levels at 60 and 120 min. The subcutaneous administration of sulpiride for 13 days induced a significant increase in serum prolactin levels at the end of the treatment, and no significant changes in serum FSH and LH levels.

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1. Franz, A., *in* "Drug effects on neuroendocrine regulation" (E. Zimmermann, W. H. Gispen, B. H. Marks, and D. de Wied, eds.), p. 311, Elsiever Scientific Publishing Co., Amsterdam (1973).
2. Fournier, P. J. R., Desjardins, P. D., and Friesen, H. G., Amer. J. Obstet. Gynecol. **118,** 337 (1974).
3. Bryant, G. D., Connan, R. M., and Greenwood, F. C., Endocrinology **41,** 613 (1968).
4. Chimenes, H., Presse Med. **78,** 1944 (1970).
5. Marchandise, B., Ann. Endocrinol. (Paris) **32,** 716 (1971).
6. Niswender, G. D., Chen, C. L., Midgley, A. R., Meites, J., and Ellis, S. E., Proc. Soc. Exp. Biol. Med. **130,** 793 (1969).
7. Daane, T. A., and Parlow, A. F., Endocrinology **88,** 653 (1971).
8. Niswender, G. D., Midgley, A. R., Monroe, S. E., and Reichert, L. E., Proc. Soc. Exp. Biol. Med. **128,** 807 (1968).
9. Steel, R. G. D. and Torrie, J. H., "Principles and Procedures of Statistics" McGraw-Hill, New York (1960).
10. Debeljuk, L., Daskal, H., Rozados, R. and Guitelman, A., Experientia **30,** 1355 (1974).
11. Mancini, A. M., Guitelman, A., Debeljuk, L. and Vargas, C. A., J. Endocrinol. (submitted for publication).

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