Plasma LH response to LH-RH injection in immature intact, castrated and cyproterone-treated male rats

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Treatment of intact males of many species with cyproterone mimics the effects of surgical castration, but whereas castration suppresses gonadal androgen secretion, cyproterone acts centrally and peripherally by inhibiting the action of testosterone at androgen receptor sites (Davidson & Bloch, 1969; Neumann *et al.*, 1969). Cyproterone is considered to be a 'pure' antiandrogen since it has no progestagenic effect, and can be used to stimulate the hypothalamo-pituitary axis (Bloch & Davidson, 1967; Viguiet-Martinez & Pelletier, 1972). The present study was undertaken to examine if the pituitary response of male rats to LH-RH was modified by cyproterone, and the response was compared to that observed in intact and castrated animals.

Three groups, each of 20 male Wistar rats, were used: Group I, intact rats; Group II, intact rats injected s.c. with a daily dose of 10 mg cyproterone (Schering AG, Berlin, West Germany) dissolved in 0.2 ml arachis oil/benzyl alcohol (9:1 v/v) from 20 to 35 days of age; Group III, animals castrated at 20 days of age.

At 35 days of age, all the animals were anaesthetized with an i.p. injection of 50 mg sodium pentobarbital (Nembutal)/kg. One carotid artery was cannulated with a heparinized polyethylene tube for blood sampling. In each group, 5 animals received 0.5 ml saline, 10 animals received an i.v. injection of 0.5 µg synthetic LH-RH (Cyclochemical, Los Angeles, California) prepared in 0.5 ml saline, and 5 animals received 1 µg LH-RH. Two blood samples were collected from each rat immediately before the injection of LH-RH, and 8 samples were collected during the next 4 hr. All the animals were killed 4 hr after the injection and the testes, seminal vesicles and ventral prostate were immediately dissected out and weighed to assess the efficiency of the antiandrogen treatment (Table 1).

Plasma samples were stored at -20°C and plasma LH concentration was later measured by radioimmunoassay. The purified hormone for iodination was provided by the NIAMDD (Rat-LH-14); the antiserum to purified rat LH was raised in the rabbit. The results are expressed in terms of the standard LH-SX 1-1, one unit of which is equivalent to 1.58 units of NIH-LH-S11.

Results and discussion

The results are shown in Table 1. The total LH response to LH-RH was defined as the area under the curve of LH concentration plotted against time: the response was measured by weighing the paper graphically representing this area and expressed in $\text{ng} \cdot \text{ml}^{-1} \cdot \text{hr}^{-1}$, as if all the release occurred during 1 hr at a constant rate.

Cyproterone caused a highly significant weight decrease ($P < 0.001$) of the accessory sex organs, as found by Neumann *et al.* (1969), but testicular weight, compared to body weight by covariance analysis, was not affected. In all groups, anaesthesia, catheterization and frequent blood sampling of the control rats did not affect the levels of plasma LH, and there was little or no fluctuation during the period of the experiment, as previously shown by Marek, Jaquet, Gogan, Lombardi & Vague (1974). This basal level is increased after chronic treatment with cyproterone. As shown by Viguiet-Martinez & Pelletier (1972) and Walsh, Swerdloff & Odell (1972), cyproterone treatment resulted in an increase in the LH plasma level which was intermediate between those for intact and castrated rats. Within the 3 groups, the discharge of plasma LH after injection of LH-RH increased with the dose injected, but the increase was significant only for the castrated rats ($P < 0.01$). In the intact rats,

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Table 1. Response (mean $\pm$ S.E.M.) of rats to treatment with cyproterone: Group I, intact animals; Group II, rats receiving 10 mg cyproterone/day for 15 days; Group III, castrated animals

	Group I				Group II				Group III			
	Saline	0.5 μ g LH-RH	1.0 μ g LH-RH	Saline	Saline	0.5 μ g LH-RH	1.0 μ g LH-RH	Saline	0.5 μ g LH-RH	1.0 μ g LH-RH	0.5 μ g LH-RH	1.0 μ g LH-RH
No. of rats	5	10	5	5	5	10	5	5	10	5	10	5
Body weight (g)	137.8 $\pm$ 3.15	125.9 $\pm$ 5.20	136.2 $\pm$ 6.39	120.0 $\pm$ 1.37	120.0 $\pm$ 1.37	110.5 $\pm$ 1.92	118.0 $\pm$ 4.51	130.0 $\pm$ 1.78	127.9 $\pm$ 2.10	120.2 $\pm$ 4.88	127.9 $\pm$ 2.10	120.2 $\pm$ 4.88
Weight of testes (g)	1.21 $\pm$ 0.03	1.10 $\pm$ 0.06	1.23 $\pm$ 0.12	1.04 $\pm$ 0.06	1.04 $\pm$ 0.06	0.85 $\pm$ 0.03	1.01 $\pm$ 0.11	—	—	—	—	—
Weight of ventral prostate (mg)	68.2 $\pm$ 3.48	58.4 $\pm$ 3.43	60.2 $\pm$ 2.13	28.1 $\pm$ 5.34	28.1 $\pm$ 5.34	25.8 $\pm$ 3.13	21.4 $\pm$ 2.00	11.0 $\pm$ 0.65	10.5 $\pm$ 0.35	9.6 $\pm$ 0.36	10.5 $\pm$ 0.35	9.6 $\pm$ 0.36
Weight of seminal vesicles (mg)	43.4 $\pm$ 3.10	38.7 $\pm$ 2.67	43.7 $\pm$ 3.49	15.9 $\pm$ 1.57	15.9 $\pm$ 1.57	15.3 $\pm$ 0.89	14.6 $\pm$ 0.73	10.6 $\pm$ 0.62	11.3 $\pm$ 0.30	10.5 $\pm$ 0.80	11.3 $\pm$ 0.30	10.5 $\pm$ 0.80
Plasma LH (ng/ml)												
0 ₁ min	14.8 $\pm$ 1.11	15.3 $\pm$ 0.95	16.4 $\pm$ 1.34	19.0 $\pm$ 0.96	19.0 $\pm$ 0.96	20.5 $\pm$ 2.67	19.2 $\pm$ 1.83	21.8 $\pm$ 2.78	21.6 $\pm$ 1.14	23.7 $\pm$ 2.27	21.6 $\pm$ 1.14	23.7 $\pm$ 2.27
0 ₂ min	15.4 $\pm$ 1.50	15.0 $\pm$ 0.82	16.8 $\pm$ 2.08	17.6 $\pm$ 2.78	17.6 $\pm$ 2.78	19.9 $\pm$ 2.66	19.7 $\pm$ 1.59	20.8 $\pm$ 2.70	21.6 $\pm$ 0.91	24.2 $\pm$ 1.27	21.6 $\pm$ 0.91	24.2 $\pm$ 1.27
5 min	12.9 $\pm$ 1.30	22.4 $\pm$ 1.67	23.7 $\pm$ 1.63	20.0 $\pm$ 3.05	20.0 $\pm$ 3.05	35.7 $\pm$ 4.25	26.7 $\pm$ 1.25	21.9 $\pm$ 2.17	77.1 $\pm$ 2.30	89.9 $\pm$ 3.41	77.1 $\pm$ 2.30	89.9 $\pm$ 3.41
10 min	13.8 $\pm$ 1.26	24.9 $\pm$ 0.87	27.2 $\pm$ 1.30	20.7 $\pm$ 4.13	20.7 $\pm$ 4.13	36.6 $\pm$ 4.27	30.4 $\pm$ 1.62	22.9 $\pm$ 2.07	81.9 $\pm$ 4.95	104.0 $\pm$ 3.89	81.9 $\pm$ 4.95	104.0 $\pm$ 3.89
20 min	12.7 $\pm$ 1.27	30.8 $\pm$ 1.27	31.8 $\pm$ 1.74	17.6 $\pm$ 3.27	17.6 $\pm$ 3.27	40.7 $\pm$ 4.60	44.7 $\pm$ 4.11	23.2 $\pm$ 2.51	100.4 $\pm$ 5.14	138.7 $\pm$ 7.92	100.4 $\pm$ 5.14	138.7 $\pm$ 7.92
30 min	13.1 $\pm$ 1.26	28.9 $\pm$ 1.74	30.9 $\pm$ 1.56	18.2 $\pm$ 2.71	18.2 $\pm$ 2.71	49.3 $\pm$ 4.86	51.7 $\pm$ 7.00	23.0 $\pm$ 2.16	101.0 $\pm$ 2.68	118.0 $\pm$ 11.25	101.0 $\pm$ 2.68	118.0 $\pm$ 11.25
50 min	12.7 $\pm$ 0.80	26.6 $\pm$ 2.45	35.8 $\pm$ 3.41	20.5 $\pm$ 3.34	20.5 $\pm$ 3.34	53.9 $\pm$ 5.12	60.0 $\pm$ 11.53	21.5 $\pm$ 2.07	94.8 $\pm$ 6.71	116.6 $\pm$ 11.85	94.8 $\pm$ 6.71	116.6 $\pm$ 11.85
90 min	12.9 $\pm$ 1.06	22.5 $\pm$ 2.70	32.7 $\pm$ 5.74	17.3 $\pm$ 3.46	17.3 $\pm$ 3.46	55.0 $\pm$ 6.12	63.3 $\pm$ 6.85	23.2 $\pm$ 1.13	73.6 $\pm$ 7.19	84.3 $\pm$ 12.57	73.6 $\pm$ 7.19	84.3 $\pm$ 12.57
120 min	13.7 $\pm$ 0.73	16.7 $\pm$ 2.13	22.4 $\pm$ 2.84	17.8 $\pm$ 1.55	17.8 $\pm$ 1.55	46.9 $\pm$ 5.16	47.6 $\pm$ 7.31	24.2 $\pm$ 1.35	40.0 $\pm$ 4.40	58.9 $\pm$ 18.38	40.0 $\pm$ 4.40	58.9 $\pm$ 18.38
180 min	15.2 $\pm$ 1.57	14.4 $\pm$ 1.35	16.3 $\pm$ 2.25	20.8 $\pm$ 1.41	20.8 $\pm$ 1.41	37.6 $\pm$ 11.66	40.5 $\pm$ 9.37	22.2 $\pm$ 1.87	27.5 $\pm$ 2.92	47.0 $\pm$ 10.0	27.5 $\pm$ 2.92	47.0 $\pm$ 10.0
240 min												
Total LH release (ng·ml ⁻¹ ·hr ⁻¹)	53.6 $\pm$ 3.50	83.8 $\pm$ 5.43	105.9 $\pm$ 9.35	74.5 $\pm$ 10.90	74.5 $\pm$ 10.90	166.1 $\pm$ 13.90	185.8 $\pm$ 22.15	91.9 $\pm$ 6.82	284.9 $\pm$ 15.4	353.4 $\pm$ 46.15	284.9 $\pm$ 15.4	353.4 $\pm$ 46.15

the time interval between the injection of 1.0 µg LH-RH and the LH peak was 90 min and the total LH release was the smallest of all the groups. The total LH release in cyproterone-treated rats was twice that of intact animals; the LH peak was reached 2 hr after injection of LH-RF, but the total release had not been achieved by the end of the experiment. In castrated rats, the total LH release after LH-RH injection was four times greater than that observed in intact rats and the interval between injection and the LH peak was very short (20 min).

These results show that the LH release which follows injection of LH-RH in the intact male rat (Debeljuk, Arimura & Schally, 1972) or in the castrated rat (Debeljuk, Vilchez-Martinez, Arimura & Schally, 1974) can be modified by treatment with an antiandrogen. This can be partly explained by the increase of pituitary LH content observed after cyproterone administration (Von Berswordt-Wallrabe & Neumann, 1971; Viguier-Martinez & Pelletier, 1972). The magnitude and duration of the LH release and the delay between the injection of LH-RH and the LH peak seem to denote activation of LH synthesis in cyproterone-treated rats. Alternatively, pituitary sensitivity to LH-RF may increase after cyproterone treatment; by inhibiting the action of testosterone this antiandrogen may decrease the intensity of the negative feedback action of testosterone, even though plasma testosterone levels remain unchanged after 15 days of treatment (M. C. Viguier-Martinez, unpublished data) or even increase, as shown by Hasan, Neumann & Schenck (1973).

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