tinemia again shows a difference in the mode of action of the 2 neuroleptics, without offering a meaningful interpretation. Enhancement by MCE of the PRL-releasing effect of another antidopaminergic drug i.e., sulpiride also has been reported in rats[30]. In behavioral studies, MCE, and also ATR, were able to potentiate the effect of LR511 but not that of HAL on the inhibition of conditioned avoidance response[31], results which are in part reminiscent of those obtained in our study.

In contrast to the observed failure of MCE to affect the PRL-releasing effect of HAL is the reported ability of alleged 5-HT antagonists, i.e., SQ 10631 and methysergide, to inhibit the elevation in PRL levels induced by another anti-dopaminergic drug, pimozide[32]. It has to be noted, however, that the neuronal specificy of methysergide as a selective 5-HT antagonist has been questioned[2], and a direct stimulation of pituitary DA receptors by this drug has been demonstrated[33,34].

In conclusion our results demonstrate that: a) HAL-induced PRL release is antagonized by DA agonists and, in addition by drugs capable of enhancing (pituitary?) GABAergic function, thus suggesting an important role for GABA in the control of PRL secretion; b) the novel neuroleptic drug, LR511, behaves differently from HAL as far as PRL secretion is concerned, so that its PRL-releasing effect cannot be accounted for by a simple blockade of DA receptors.

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Immunization of female rabbits against testosterone stimulates testosterone accumulation by isolated ovarian follicles[1]

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Summary. Ovarian follicles isolated from female rabbits after active immunization against testosterone-3-oxime bovine serum albumin produced more testosterone than similar follicles from controls.

Immunization of animals in order to study the role of hormones is widely accepted as a useful tool in reproductive endocrinology. Although testosterone is predominantly a male sex hormone it may have an important role in ovarian physiology[3-7]. In a previous study[6] it was found that immunization of female rabbits against testosterone-3-oxime bovine serum albumin (T-3-BSA) resulted in increased levels of testosterone in circulation, but this could have been due to increased binding to antibodies, as noted, or to decreased metabolic clearance rate as observed in male rabbits[8]. In view of the marked hypertrophy of the interstitial cells observed in T-3-BSA immunized rabbits[7] our working hypothesis was that this cell layer could be another source of testosterone.

Testosterone accumulation in the medium, and follicular testosterone content after incubation of stroma or single follicles isolated from rabbits actively immunized against testosterone

Follicles	BSA immunized	Testosterone immunized	t	df	
Preincubation					
MEM	0.87±0.18	2.68±0.54	3.39	42	$p<0.005$
Incubation with					
LH	3.73±1.61	3.41±0.60	0.17	9	NS
FSH	2.63±0.54	10.82±4.18	2.16	9	NS
LH+FSH	4.45±0.96	7.82±1.65	1.65	9	NS
Stroma preincubation	0.48±0.01	5.41±1.21	2.80	42	$p<0.05$
Follicular homogenate					
MEM	0.58±0.38	2.44±1.33	1.48	9	NS
LH	1.37±0.33	5.28±1.75	2.41	9	NS
FSH	1.28±0.52	3.88±0.69	1.87	9	NS
LH+FSH	1.17±0.30	3.63±1.41	3.03	8	$p<0.025$

Results are expressed as ng/follicle or ng/stroma incubation (mean±SEM) and were analyzed using Student's t-test. MEM, minimal Eagle's medium with 5% normal rabbit serum; LH, luteinizing hormone; FSH, follicle stimulating hormone.

Unless otherwise noted, materials and methods were identical to those previously described[6]. 2 groups of 6 animals were used. After 8–9 weeks, when antibody titre was sufficiently elevated the animals were sacrificed. Blood was collected for hormonal analyses and the uteri were weighed. Follicles and pieces of stroma were isolated and incubated in vitro with luteinizing hormone (LH) and follicle stimulating hormone (FSH) using a 2×2 factorial experimental design. The incubation medium consisted of Eagle's minimal essential medium, supplemented with L-glutamine (292 mg/l) and 5% normal rabbit serum, and buffered to pH 7.4 with 10 ml 1 mM HEPES (N-2-hydroxyethylpiperazine-N[1]-2-ethanesulphonic acid) and 20 ml 7.5% sodium bicarbonate solution per litre (MEM). Incubation conditions were the same as described previously[9], and involved a preincubation period of 2 h in MEM alone, followed by another 2 h incubation period when the medium was replaced with one containing various test substances, as follows:

2 h preincubation	2 h incubation
MEM	MEM or test substances dissolved in MEM

Serum gonadotropins were essentially similar to levels previously found, with LH being significantly elevated in the testosterone immunized group (1.84±0.11 ng/ml vs 1.32±0.07 ng/ml, $p<0.01$) and FSH levels being similar[6]. The uteri of the control animals (4.94±0.53 g) were also significantly heavier than those of the testosterone immunized group (2.85±0.26 g, $p<0.05$).

Testosterone accumulation and follicular content after incubation of follicles and stroma are shown in the table. In the preincubation period the follicles isolated from T-3-BSA animals secreted 3 times more testosterone into the medium than the corresponding follicles from BSA animals. There were no marked differences in the response of follicles of both BSA and T-3-BSA animals to stimulation with gonadotropins, although the follicles from the T-3-BSA animals generally produced more testosterone. There was a 4-fold increase in testosterone accumulation when follicles from BSA animals were incubated with LH, but only a 1.3-fold increase for corresponding follicles from T-3-BSA animals. This discrepancy was probably due to the higher circulating levels of LH found in the T-3-BSA animals, which was responsible for increased testosterone accumulation in the preincubation period and occupancy of LH receptor sites. The response to FSH and a combination of LH and FSH was about 3–5-fold in both BSA and T-3-BSA follicles. The greater production of testosterone by T-3-BSA follicles is also reflected in the higher testosterone content of the homogenized follicles. It is not known whether this represents greater concentrations in the follicular fluid.

Of interest also was the significant accumulation of testosterone in incubations of stromal tissue from T-3-BSA animals. Combined with the marked hypertrophy of this tissue seen in similar animals[7], it could be indicative of interstitial tissue androgen production as suggested previously[10]. However, both studies suffer from uncertainty as to whether all follicular tissue had been completely removed.

Estradiol accumulation was also determined in these studies, but no consistent or different effects were detected when BSA and T-3-BSA immunized follicles were compared.

The relatively higher production of testosterone by isolated follicles from animals immunized against T-3-BSA begs the question of whether local ovarian testosterone is responsible for the greater rate of follicular development observed in similarly treated animals[7]. In similar studies with male rabbits increased testosterone production, Leydig cell hyperplasia and increased responsiveness to human chorionic gonadotropin were observed in testosterone-immunized animals.

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