

## Lack of Direct Effect of Pineal Extracts on the Secretory Function of the Testis

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A single intravenous injection of bovine pineal extracts in adult male dogs did not significantly alter the secretion of testicular 17-oxosteroids for up to 4 hours after the administration. The testicular secretory response to human chorionic gonadotrophin (HCG) was also not affected by treatment with pineal extracts. It is therefore suggested that pineal extracts have no direct effect on the biosynthesis and/or release of androgens by testicular tissue.

### INTRODUCTION

There are reports in the literature which indicate that the administration of pineal extracts exerts an inhibitory influence on several reproductive processes<sup>1,2)</sup>. This effect is thought to be due to a centrally mediated inhibitory action of melatonin<sup>3-5)</sup>, an endogenously found pineal substance. However, in the present study, attempts were made to see whether pineal extracts exert a selective effect on the secretion of steroids by testicular tissue for up to several hours after the administration.

### MATERIALS AND METHODS

Pineal extracts were prepared from bovine pineal glands collected from freshly slaughtered animals. After removal of bits of brain, blood and connective tissue adherent to the gland, the fresh weight was approximately 150-200 mg. The pineal glands were ground

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with sand with a pestle and motor, and extracted with 2 ml isotonic saline solution per gland. After centrifugation at 2,000 rpm for 30 min, the opalescent supernatant was used for the present experiment. Experiments were performed on adult mongrel dogs weighing 9.5–19.5 kg. The left spermatic vein was exposed by the lumbar route under pentobarbital anesthesia (25 mg/kg, injected iv) and cannulated with a T-shaped glass cannula fitted with a rubber tube by the method described previously.<sup>6)</sup>

To see whether pineal extracts would affect the secretion of testicular steroids first, 2 ml pineal extracts containing the soluble materials from 150 mg of the fresh tissues were injected into the left saphenous vein for 15 sec. Samples of spermatic venous blood were collected 30 and 10 min before, and 15, 30, 60, 120, 180 and 240 min after the injection of pineal extracts.

In other experiments, the effect of pineal extracts on the testicular secretory response to human chorionic gonadotrophin (HCG) was examined. In animals in which the left spermatic vein had been cannulated, a single injection of 1 ml isotonic saline solution containing 20 iu HCG (Sigma Co.)/kg was made into the left saphenous vein. Seventy-five min after the injection of HCG, 2 ml pineal extracts containing 150 mg of wet tissue were injected into the left saphenous vein in a single injection for 15 sec. Samples of spermatic venous blood were collected 30 and 10 min before, and 15, 30, 60, 90, 120, 180 and 240 min after the HCG injection. The volume of each blood sample was approximately 10 ml and the duration of blood collection was 3–6 min. Three ml of plasma from each sample were analysed for 17-oxosteroids by a slight modification<sup>6)</sup> of the method of GARDNER<sup>7)</sup>.  $\Delta^4$ -Androstene-3, 17-dione was used as a reference standard.

## RESULTS AND REMARKS

Intravenous injection of crude saline pineal extracts produced no or little decrease in the secretion of 17-oxosteroids by the testis for up to 4 hours after the injection (Table 1). In animals which received HCG, an injection of HCG caused a marked increase in the secretion of 17-oxosteroids by the testis. In most cases, the highest increase occurred

**Table 1.** Effect of intravenous injection of bovine pineal extracts (BPE) on testicular 17-oxosteroid (17-OS) secretion in dogs.

Body weight (kg)	Dose of pineal tissue (mg)	Rate of testicular 17-OS secretion (ng/kg/min)								
		Before injection of BPE (min)		After injection of BPE (min)						
		-30	-10	15	30	60	90	120	180	240
19.5	150	4.6	6.5	*	2.5	*	*	1.0	6.9	*
9.6	150	*	*	*	5.6	*	*	*	*	*
12.1	150	*	0.4	*	0.4	*	*	*	5.1	4.1
9.5	150	1.2	0.6	*	*	*	2.9	*	2.1	*
13.4	150	3.4	5.2	6.5	7.1	5.7	4.4	4.6	2.3	2.7

\* Below the measurable limit.

within approximately 60 min after the injection. Seventy-five min after the injection of HCG, pineal extracts were given iv but the secretory response to HCG was not affected by this treatment and was sustained during a 4 hour period of observation (Table 2).

The present results indicate that the pineal extract has no direct effect nor does it significantly alter the secretion of testicular steroids and is, in addition, not capable of affecting the increased testicular steroid secretion caused by HCG.

**Table 2.** Effect of bovine pineal extracts (BPE) on testicular 17-oxosteroid (17-OS) secretion caused by human chorionic gonadotrophin (HCG).

Body weight (kg)	Dose of :		Rate of testicular 17-OS secretion (ng/kg/min)										
	HCG (iu/kg)	Pineal tissue (mg)	Before injection of HCG (min)		After injection of HCG (min)								
			-30	-10	15		30		60		After BPE injection (min)		
12.2	20	150	*	6.2	10.5	64.9	152.1	—	136.0	146.4	193.0	125.5	
10.2	20	150	*	*	73.6	113.0	119.3	—	123.3	126.8	76.6	58.8	
10.4	20	150	6.9	5.3	25.1	72.1	67.9	—	78.6	85.3	98.9	92.9	
11.8	20	150	2.3	*	*	65.3	60.2	—	66.9	116.4	102.1	99.8	

\* Below the measurable limit.

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