

particles which are possibly of diverse origin. The bulk of the particles probably have not been transported more than a few kilometers, however, so that the analysis represents, for the most part, a mixture of rocks derived from a small area on Mare Tranquillitatis.

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Ribonucleic Acid: Control of Steroid Synthesis in Endocrine Tissue

Abstract. *Ribonucleic acid extracted from either adrenal or testis altered the enzyme activity of adrenals and ovaries maintained in organ culture. The pattern of steroid hormone synthesis of the cultured endocrine tissue reflected the origin of the RNA.*

Preparations of ribonucleic acid from adrenals or gonads altered the pattern of steroid hormones synthesized in endocrine tissues maintained in organ culture. Ribonucleic acid was extracted from adrenals or gonads with phenol and dodecyl sulfate by the method of Hiatt (1). After precipitation from 0.1M NaCl with ethanol, the RNA was treated twice with deoxyribonuclease at room temperature for 5 minutes and then was precipitated with ethanol. The final precipitate was washed once with 95 percent ethanol and twice with a mixture of ethanol (95 percent) and ether (2:1). The RNA was dissolved in water, and its absorption was measured spectrophotometrically over the range 220 to 290 m μ . For each RNA preparation used, the ratios of the absorption at 260/280 and 260/230 m μ were very nearly two.

Adrenal glands can be maintained in organ culture in a histologically and biochemically differentiated state for

short periods of time (2). Explants of ovaries or adrenals weighing no more than 5 mg were cultured sterily on stainless steel rafts in a chemically defined medium (Parkers 1066) containing RNA extracted from either adrenal or testis. The concentration of the RNA in the medium ranged from 25 to 250 μ g/ml. The cultures were maintained at 26°C for 24 hours with 95

Table 1. Radioactive steroid products, expressed as counts per minute per milligram of adrenal, isolated from cultures of rat adrenals previously exposed to RNA from either rat adrenal or rat testis. The substrate was pregnenolone-7 α -³H (0.5 μ c per milligram of adrenal).

Product	Yield (count min ⁻¹ mg ⁻¹)	
	Adrenal + adrenal RNA	Adrenal + testis RNA
Pregesterone	2950	4000
Testosterone	72	114
Deoxycorticosterone	1920	3820
Corticosterone	2080	1250

percent oxygen 5 percent carbon dioxide as gas phase. The explants were then removed from the chamber and assayed for enzyme activity by one of two methods. In one method the explants were placed in fresh chambers containing nutrient medium plus pregnenolone-7 α -³H (0.25 μ c per milligram of tissue) and cultured for an additional 24 hours. The radioactive products in the tissue and medium were isolated and characterized by reverse isotope dilution. In all such experiments only 10 to 20 percent of the radioactive substrate was metabolized. In the second method, the explants after exposure to RNA were homogenized in phosphate buffer and incubated in a Dubnoff shaker for 1 hour at 37°C with an excess of substrate (pregnenolone-7 α -³H), NAD, NADP (nicotinamide adenine dinucleotide and the phosphate), and glucose-6-phosphate. Radioactive products were isolated as in the first method of assay. After separation of the steroid products on thin-layer chromatography, radiochemical homogeneity of radioactive products and carrier steroids was achieved by crystallization of the carrier steroids to constant specific activity followed by the formation of a derivative of the steroid and crystallization of the derivative.

The influence of RNA from rat testis on steroid synthesis in rat adrenals in culture was significant (Table 1). Adrenals previously exposed in organ culture to RNA from rat testis produced more radioactive progesterone, testosterone, and deoxycorticosterone, but less corticosterone when subsequently cultured in the presence of pregnenolone-7 α -³H. These results might be explained by assuming that RNA from rat testis led to a greatly increased activity of 3 β -hydroxysteroid dehydrogenase in the adrenal gland in culture. One must also assume that the RNA led to some inhibition of the 11 β -hydroxylase in the adrenal.

Ovaries previously exposed to RNA from the adrenal formed more radioactive deoxycorticosterone and corticosterone from pregnenolone-7 α -³H than did control ovaries or ovaries previously exposed to RNA from the testis (Table 2). Those ovaries from a pregnant rat synthesized a large amount of testosterone and perhaps for this reason, RNA from rat testis produced no effect on their synthesis of testosterone. However, ovaries from nonpregnant rats were clearly affected by testis RNA, and when subsequently cultured

with pregnenolone-7 α -³H formed larger amounts of testosterone-³H than did control ovaries. Thus the activity of enzymes involved in the synthesis of steroid hormones in the ovary and adrenal of the adult rat was influenced by previous exposure of these tissues in organ culture to homologous RNA extracted from the adrenal or testis.

Similar effects of RNA on the pattern of steroid synthesis could be demonstrated by the second method of enzyme assay. Mouse adrenals previously exposed to RNA from mouse testis, and subsequently incubated in a cell-free system in the presence of excess pregnenolone-7 α -³H, NAD, NADP, and glucose-6-phosphate, formed more testosterone-³H and less corticosterone-³H than control adrenals did (Table 3). In a similar experiment, rat adrenals previously exposed to RNA from rat testis formed more testosterone-³H than adrenals exposed to RNA from rat adrenal did. These results are in complete agreement with those in which the enzymes were assayed in organ culture (Table 1). Rat ovaries previously exposed to RNA from human fetal adrenals formed more progesterone-³H, deoxycorticosterone-³H, and corticosterone-³H than control ovaries (Table 3) did. In this instance, heterologous RNA

showed an effect on the pattern of steroid synthesis in rat ovary in organ culture. These results are similar to the effects of homologous RNA from the adrenal noted in Table 2. Under both assay conditions, rat ovaries previously exposed to adrenal RNA formed more progesterone and corticosterone than control ovaries did.

Because of the extensive purification procedures necessary for isolation and identification of the radioactive steroid products, it was not feasible to do a large number of duplicate experiments and apply the usual criteria of statistical significance. However, many experiments have been completed in which the material added to the organ culture was RNA from the same gland or an inactive fraction of RNA isolated from a methylated-albumin column. In such experiments, in which no effect was expected, the agreement between the steroid products isolated from control and experimental incubations was quite striking (Table 4).

The evidence from these experiments supports the concept that RNA from one steroid-producing gland can alter the enzyme activity of another steroid-producing gland and that the pattern of steroid hormone synthesis reflects the origin of the RNA. There are several

Table 4. Radioactive steroid products, expressed as counts per minute per milligram of adrenal, isolated from cell-free incubations of mouse adrenals, previously exposed to saline or to an inactive fraction of testicular RNA eluted from a methylated-albumin column. The substrate was pregnenolone-7 α -³H (0.5 μ C per milligram of adrenal).

Product	Control	Inactive RNA
Progesterone	5,310	4,910
Androstenedione	755	844
Testosterone	55,900	48,000
Corticosterone	164,400	175,000

reports (3) which indicate that preparations of RNA administered by one or another route to tissues in vivo may alter the morphology or enzyme activity of the recipient cells. The present experiments extend these observations to cells in organ culture. It should be emphasized that the preparation of RNA used is a crude one and a small contamination with protein or DNA has not been ruled out. The type of RNA molecule that may be responsible for the biological effect has not yet been defined.

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Table 2. Radioactive steroid products, expressed as counts per minute per milligram of ovary, isolated from organ cultures of rat ovaries previously exposed to RNA from either rat adrenal or rat testis. The substrate was pregnenolone-7 α -³H (0.25 μ C per milligram of ovary).

Product	Amounts (count min ⁻¹ mg ⁻¹)				
	Ovaries from pregnant rat			Ovaries from nonpregnant rat	
	Control	Adrenal RNA	Testis RNA	Control	Testis RNA
Progesterone	3,800	8,560	8,500	13,300	13,000
Testosterone	3,000	2,820	3,080	333	417
Androstenedione				559	506
Deoxycorticosterone	12	763	240		
Corticosterone	48	210	23		

Table 3. Radioactive steroid products, expressed as counts per minute per milligram of tissue, isolated from cell-free incubations of adrenals or ovaries, previously exposed to RNA from either adrenal or testis. The substrate was pregnenolone-7 α -³H (0.5 μ C per milligram of tissue).

Product	Amounts (count min ⁻¹ mg ⁻¹)					
	Mouse adrenals		Rat adrenals		Rat ovaries	
	Control	Testis RNA	Adrenal RNA	Testis RNA	Control	Human adrenal RNA
Testosterone	8,480	12,500	671	2,170		
Corticosterone	53,000	6,580	37,500	32,900	82	217
Cortisol					70	848
Estrone					11,700	14,200
Progesterone					4,550	8,120

Pediplain in Northern Chile and the Andean Uplift

Abstract. *A pediplain in the Chilean Atacama Desert formed during Oligocene and Miocene time when the aridity of the region started and was later displaced by north-trending faults associated with the Andean uplift. Block basins and some horsts were later concealed by Upper Tertiary and Quaternary orogenic sediments and ignimbrites.*

Late Tertiary block faulting, associated with the Andean uplift in northern Chile, displaced a Tertiary pediplain that had formed in the preceding 30 million years and initiated deposition of orogenic sediments and ignimbrites in large areas of the Atacama Desert. The