from the end face of the guide slide. Cuts are made clear through the mucilage and object to the glass. With successive cuts the razor blade is brought more nearly erect (positions 1 through 4 in Fig. 5). These cuts bring the razor blade more nearly perpendicular and nearer and nearer to the end face of the guide slide. This procedure controls the thickness of the sections. By the time sufficient sections have been cut to bring the razor blade nearly perpendicular the operator has the feel of the particular object-mucilage mount and of the razor blade. The material should be so placed that the critical sections can now be made by continuing the process of slicing and straightening up the blade with each successive slice (positions 4 through 7 in Fig. 5). The best sections, i.e., the thinnest and most perpendicular, are obtained, as a rule, just when the razor blade is brought perpendicular (position 5 in Fig. 5) and past perpendicular (positions 6 and 7 in Fig. 5) by slanting the razor blade to cut under the guide slide edge.

After slicing is completed through the desired area, the guide slide is removed and a drop of water or dilute glycerine is placed over the mucilage-impregnated sections. In a minute or two the sections will float free, or nearly free, as the mucilage softens. The thick, unsectioned portions of the object may be removed with a dissecting needle and the remaining sections spread about. The sections may then be stained or treated in any manner which may be required.

The technique works equally well with fresh, dried, or preserved materials. Moreover, a section may be obtained with a high degree of success, with almost no equipment, and with a minimum of experience when just one pin-point object is available. Sections have been obtained easily and consistently of materials that are somewhat less than 5 μ thick.

Effect of a Folic Acid Antagonist on Hormonally Induced Changes in the Rat Prostate

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A definite relationship has been demonstrated in the chick between estrogen utilization and folic acid requirement (*1*). That this relationship is not based simply on a general impairment of body growth has been shown by the fact that optimal oviduct responses to stilboestrol occur in riboflavin- and pyridoxine-deficient chicks. In a more recent development, Hertz has found that the tissue growth response of the chick oviduct to stilboestrol can be blocked by the administration of a folic acid antagonist, presumably by competitive displacement of essential folic acid (*2*). This explanation is supported by the observation that the inhibitive effect can be completely reversed by the administration of excess folic acid.

In the course of an investigation into growth mechanisms of the rat prostate,[1] the foregoing findings raised several questions: 1. The use of a folic acid antagonist to inhibit the *stimulating* potential of an estrogen had been clearly shown in the chick. Could the usual *depressant* effect of estrogen on the rat prostate be nullified in a similar manner? 2. If such were the case, was there a possibility that the folic acid antagonist possessed androgenic characteristics *sui generis?* 3. If the antifolic principle did not inhibit the usual effect of the estrogen on the prostate, the probability was good that the blocking action was directed only at that function of the biological activity of the estrogen concerned with tissue growth. If so, might it not therefore inhibit androgen-stimulated growth as well?

TABLE 1

Exp.	Type of rat*	No. used	Average rat wt. in g	Mode of treatment	Average prostate wt. in mg†
	Intact adult	7	201	α-estradiol	190
1	" "	7	231	α-estradiol and aminopterin	373
	Intact adult	5	277	aminopterin	469
2A	Castrate adult	5	238	aminopterin‡	89
	Intact immature	5	62	.....	33
	" "	5	61	testosterone	59
2B	" "	7	54	aminopterin	28
	" "	3	59	testosterone and aminopterin	53
	Castrate adult	5	223	testosterone‡	380
3	" "	5	202	testosterone‡ and aminopterin‡	406

* The adult rats used in these experiments were of the Sherman strain, and ranged in age from 90 to 120 days. The immature rats were obtained from Carworth Farms (Wistar) and were approximately 30 days old.

† Combined weight of anterior (ventral) and posterior lobes, except in Exp. 2B, where only anterior lobes were used.

‡ Begun 24 hr postcastrationally.

The results of experiments designed to test the above hypotheses are summarized in Table 1. It will be seen that: 1. The use of the folic acid antagonist[2] partially,

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[2] Aminopterin (4-amino pteroyl glutamic acid), a product of the Lederle Laboratories, was kindly furnished by Dr. E. B. Schoenbach, of the Johns Hopkins School of Medicine, who also supplied much valuable information derived from his experiences with the compound. The daily dose used in these experiments was 0.2 mg/kg body weight in aqueous solution. Subcutaneous injections were given for 6–7 days, and the animals were sacrificed 24 hr after the last injection.

if not completely, interfered with the ordinary depressive influence of α-estradiol[3] on the rat prostate. (Table 1, exp. 1.) This finding in a sense confirms, but also extends Hertz' recent observations in the chick (*2*). 2. Furthermore, the folic acid antagonist possessed no androgenic potential itself, as judged by its inability to stimulate prostatic growth in castrate adult, and intact immature rats. (Table 1, exps. 2A and 2B.) 3. Lastly, no interference with androgen[3] stimulation of tissue growth is occasioned by the folic acid antagonist in castrate adult or intact immature rats. (Table 1, exps. 2B and 3.) From this we may reason that folic acid is not a prerequisite for hormonally induced growth in general, but is only essential for the proper utilization of estrogen, either in a stimulant or depressant capacity.

Further investigation of this problem is in progress, using other folic acid antagonists than the one employed here. These studies will be reported subsequently in a paper which will also include complete histological details.

References

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Metabolism of I^{131} in Severe Anoxic Anoxia

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Clinical reports (*3, 4, 6*) have indicated symptomatic improvement of hyperthyroid patients who sojourn at high altitudes. Rats exposed to severe anoxic conditions demonstrated an oxygen consumption 40–50% of normal (*1, 7*). These results suggest involvement of the thyroid gland in accommodation to anoxia. The present investigation was undertaken to determine whether any significant alterations in metabolism of radioactive iodine could be detected in relatively acute exposures to severe anoxic anoxia.

Eighteen rats were fed a low iodine diet (*5*) for 8 weeks, after which half of the animals served as controls and the other half were exposed to 268 mm Hg air pressure (which stimulated a 27,000-ft altitude) at 15–20° C. The low pressure rats were subjected to this anoxia for 12 hr and were then returned to sea level for injection of I^{131}. Within 3 min they were re-exposed to the low pressure for an additional 24 hr, after which they were sacrificed. The sea level control animals were also kept at 15–20° C and likewise injected with I^{131} 24 hr before they were sacrificed. All injections of radioactive iodine consisted of 10 μc in 0.5 cc of physiological saline (pH = 8.0) administered intraperitoneally. In each rat the total I^{131} was determined in the thyroid gland and in a sample of plasma.

[3] Crystalline α-estradiol and testosterone were obtained through the courtesy of the Schering Corporation. The daily dose of these substances was 0.1 mg in sesame oil; the same injection schedule was used as that for aminopterin.

TABLE 1

FRACTIONATION OF PLASMA RADIOACTIVE IODINE

Experimental conditions	Male rats		Female rats	
	% Plasma I^{131}			
	Filtrate*	Ppt.†	Filtrate*	Ppt.†
Sea level controls	22	78		
	22	78		
	14	86	62	38
	47	53	46	54
	26	75	46	54
avg	26	74	51	49
Anoxic rats	88	12		
	90	10	98	2
	95	5	98	2
	95	5	97	2
avg	91	8	98	2

* Trichloro-acetic acid filtrate.
† Trichrolo-acetic acid precipitate (protein bound fraction).

Plasma I^{131} was fractionated by the addition of 5 volumes of 10% trichloroacetic acid to separate "protein bound" I^{131} (*2*). Urinary excretions from the two groups of male animals were pooled as two samples. The data are summarized in Tables 1 and 2.

The anoxic rats demonstrated a pronounced reduction in protein bound I^{131} in plasma. The average values expressed as percent of plasma I^{131} for male animals were as follows: trichloro-acetic acid filtrate of plasma of control rats 26%, and of the anoxic rats 91%; trichloroacetic acid precipitate of plasma of control rats 74%, and the anoxic rats 8%. In the case of female rats, the anoxic animals demonstrated an even greater depression of the protein bound I^{131} fraction, but the control values were also somewhat different from those of the male animals. It appears inadvisable to account for the ap-

TABLE 2

DISTRIBUTION OF I^{131} IN URINE, PLASMA AND THYROID GLAND

Experimental conditions	Male (10 rats 170–230 g)			Female (6 rats 140–180 g)		
	% of injected dose I^{131}					
	Total urine (pooled*)	Plasma (1 cc)	Thyroid gland	Total urine	Plasma (1 cc)	Thyroid gland
Sea level controls	30	0.78	76	(Not determined)	0.23	50
		0.64	76		0.23	53
		0.78	40		0.38	
		0.78	55			
		0.57				
		0.14				
avg		0.62	62		0.28	52
Anoxic	22	0.57	53	(Not determined)	0.57	31
		0.64	43		0.67	28
		0.16	46		0.57	28
		0.71	66			
avg		0.52	42		0.60	29

* Pooled urine sample, calculated as % of dose per rat.