trically induced tachycardia. Apparently the vasodilating effect of Isuprel does not offset completely the vasoconstriction due to the reduction of CO produced by the tachycardia, and Isuprel raises MABP when administered during tachycardia. It is possible that this increase in MABP might disappear if administration of Isuprel was continued longer than in the present experiments.

Other drugs such as nitroglycerine increase both HR and CO(13,14), apparently exclusively through their vasodilating property since they are not known to have any direct effect on the myocardium. The present experiments concerning Isuprel do not necessarily permit the conclusion that nitroglycerine, like Isuprel, would increase cardiac output even if it were prevented from increasing HR. Arteriovenous fistula is also known to increase both HR and CO(15) and the increase in HR has been proven experimentally not to be a *sine qua non* for the increase in CO(15).

Summary. The effect of Isuprel on MABP, CO and TPR has been investigated in the anesthetized dog in which the effect of the drug on HR is allowed to occur, as well as in the anesthetized dog in which electrically induced atrial or ventricular tachycardia is initiated before and maintained throughout period of action of the drug to prevent the increase in heart rate normally induced by the drug. Under the circumstances of drug dosage, rate of the induced tachycardia as well as control values of HR and CO, the effect of the drug on CO and TPR is essentially the same, whether the effect of the drug on HR is permitted or prevented.

1. Konzett, H., *Arch. f. Exp. Path. Pharm.*, 1940, v197, 27.

2. Ahlquist, R. P., *Am. J. Physiol.*, 1948, v153, 586.

3. Aviado, D. M., Jr., Wnuck, A. L., De Beer, E. J., *J. Pharm. Exp. Ther.*, 1958, v122, 406.

4. Lands, A. M., *Pharm. Rev.*, 1949, v1, 279.

5. Nakano, J., McGiff, J. C., Zekert, H., Guggenheim, F. G., Wégria, R., *Arch. Int. Pharm. et Thér.*, to be published.

6. Wégria, R., Frank, C. W., Misrahy, G. A., Siousat, R. S., McCormack, G. H., Jr., Sommer, L. S., PROC. SOC. EXP. BIOL. AND MED., 1950, v74, 551.

7. Wégria, R., Nakano, J., McGiff, J. C., Rochester, D. F., Blumenthal, M. R., Muraviev, T., *Am. J. Physiol.*, 1958, v193, 147.

8. Wégria, R., Nakano, J., McGiff, J. C., Muraviev, T., Zekert, H., Blumenthal, M. R., *J. Pharm. Exp. Ther.*, 1959, v126, 164.

9. Shipley, R. E., Wilson, C., PROC. SOC. EXP. BIOL. AND MED., 1951, v78, 724.

10. Nakano, J., Zekert, H., Griege, C. W., Wang, K. M., Schaefer, H., Wégria, R., *Am. J. Physiol.*, 1961, v200, 413.

11. Wégria, R., Frank, C. W., Wang, H. H., Lammerant, J., *Circulation Res.*, 1958, v6, 624.

12. Remington, J. W., Hamilton, W. F., Ahlquist, R. P., *Am. J. Physiol.*, 1948, v154, 6.

13. Grollman, A., *The Cardiac Output of Man in Health and Disease.* Charles C Thomas, Springfield, Ill., 1932.

14. Wégria, R., Nickerson, J. L., Case, R. B., Holland, J. F., *Am. J. Med.*, 1951, v10, 414.

15. Frank, C. W., Wang, H. H., Lammerant, J., Miller, R., Wégria, R., *J. Clin. Invest.*, 1955, v34, 722.

Received January 23, 1961. P.S.E.B.M., 1961, v107.

Studies with Mouse Pituitary Thyrotropic Tumors. II. Restored Responsiveness in Semi-Responsive Strain Surgically Reduced in Size.* (26571)

SIDNEY C. WERNER AND RAUL GRINBERG

Department of Medicine, Columbia University College of Physicians and Surgeons

A study has been made of the effect upon responsiveness to thyroxine of surgical reduction in size of an autonomous semi-responsive thyrotropic mouse pituitary tumor which had grown so large as to be no longer suppressible by this hormone. Of the 3 types of strains of thyrotropic mouse pituitary tumor isolated by Furth(1), dependent responsive, autonomous

* Aided by contract from Cancer Chemotherapy Nat. Service Center, Nat. Cancer Inst., U.S.P.H.S.

semi-responsive, and autonomous non-responsive, only the autonomous semi-responsive strain changes its responsiveness to thyroid hormone with increasing size after implantation. The growth of tumors of this latter strain in radiothyroidectomized mice can be arrested by administration of thyroid hormone when the tumor is 1 cm or less in size at start of treatment; but growth continues despite treatment with the hormone if the therapy is begun when the tumors are larger(2).

Development of loss of responsiveness raises several questions. The first is whether the concentration of thyroid hormone provided to the tumor cells is a factor in responsiveness, and if so, whether enough hormone could possibly be made available to cause suppression of the larger tumors without killing the animal. Second is whether a change in the tumor, or in the host because of continued presence of the tumor, or both, accounts for the change in responsiveness with tumor growth. The fact that a large, autonomous semi-responsive tumor which has become non-responsive yields, upon implantation, tumors which behave true to form for the original strain, tends to eliminate the likelihood that development of non-responsiveness by the tumor is associated with an inherent change in the tumor cells.

Methods and results. The tumor strain L24, was made available by Dr. Furth. This strain, implanted into radiothyroidectomized mice, appears as a palpable tumor about 4 months after implantation; in intact mice, no tumors appear up to a year after implantation(2). The cells then escape from suppression of growth by thyroid hormone and develop into palpable tumors at 13 months after implantation(2).

The behavior of all strains of thyrotropic mouse pituitary tumors is extremely reproducible so that valid conclusions may be drawn from groups of 4 mice each. Nonetheless, an initial experiment was made with 2 groups of 10 mice each, then the entire experiment repeated with added control groups, using 5 mice per group. Since tumor size must be estimated by palpation until autopsy, the estimated size is somewhat inaccurate. In the second experiment, tumor weight was estimated from palpation, and at autopsy determined by weighing.

The methods used in our laboratory have been described(1,2). In both present experiments the mice were implanted with L24 tumor, 3 weeks after being placed on a standard iodine diet (Ken-L-Kibble medium ration) and 2 weeks after radiothyroidectomy. Two and one-half months later, when the tumors had reached a size approximately 2 g, sodium-L-thyroxine was placed in the Ken-L-Kibble medium ration, 700 mg/100 g feed. Ten days later, the tumors were larger in all groups (Table I).

The tumors were then reduced in size surgically to produce a palpable nubbon approximately 0.5 cm in diameter. Thyroxine feeding was continued in some groups for 30 days. Control groups were fed the diet without hormone. In the first experiment, operation was delayed 1 month during which thyroxine feeding was maintained and tumor size increased.

At 30 days after surgical reduction of tumor size, no tumor had grown, as judged both from palpation and measurement at autopsy, in thyroxine-treated animals; the tumors in untreated control mice, however, had grown and were readily palpable (Table I). In the second experiment (Table I), final tumor weights were determined and again clearly show the same changes. In the second experiment, the failure of thyroxine to inhibit growth of the larger tumors is shown, as well as the inhibition of growth by thyroxine after the tumors were surgically reduced in size (Period A, Table I). Cessation of treatment with thyroxine in these latter then resulted in rapid increase in tumor size (Periods A and B, Table I). The cut-down tumors responded to thyroxine identically with implanted tumors of the same size in unoperated mice.

Discussion and Summary. Tumor implants grown in radiothyroidectomized mice to 2 g in weight or more of an autonomous semi-responsive strain of mouse pituitary thyrotropic tumor, L24, lose their responsiveness to thyroxine. When these were reduced in size surgically to a diameter of about 0.5 cm, and the hosts fed thyroxine, no tumor became palpable over the space of 30 days, although the expected increase in tumor size occurred in

TABLE I. Responsiveness to Thyroxine of Autonomous Semi-responsive L24 Tumor before and after Surgical Reduction in Size.

	Pre-operative period					Post-operative period								
	Treatment			Avg tumor size or wt* ± S.D.		A. Treatment			Avg tumor size or wt* ± S.D.		B. Treatment			Avg tumor size or wt* ± S.D.	
Exp. No.	Type	Duration (days)	No. mice	Initial	Final	Type	Duration (days)	No. mice	Initial	Final	Type	Duration (days)	No. mice	Initial	Final
				cm	cm				cm	cm					
1	T4 + KB†	40	10	2.0	3.5	KB	30	10	.75	3.0 ± .3*					
	Idem	10	10	2.0	3.0	T4 + KB	30	10	.75	.75 ± .2*					
				g	g				g	g				g	g
2	KB	10	5	2.5	4.8 ± .2*										
	T4 + KB	10	5	2.5	5.0 ± .2*				.5	3.7 ± .2*					
	Idem	10	5	2.5	3.5	KB	30	5							
	,,	10	5	2.5	3.5	T4 + KB	30	3	.5	.4 ± .1*					
						Idem	30	2	.5	.5	KB	30	2	0.5	2 ±.2*
	,, ‡	10	5	.5	.5	,,	30	3	.5	.7 ± .2*					
						,,	30	2	.5	.5	KB	30	2	0.5	3.0 ±.3*

* All measurements estimated by palpation except where measured or weighed at autopsy as indicated by asterisks.
† KB = Ken-L-Kibble medium ration; T4 = thyroxine.
‡ Unoperated controls with implanted tumors at start of experiment of same size as those surgically reduced in size in preceding groups.

similarly operated, untreated controls. The suppressed tumors promptly grew in the thyroxine treated group when thyroxine was stopped.

The results suggest that the large non-responsive tumors had not been fundamentally altered in character and that the host mouse too had not undergone any fundamental change to account for loss of responsiveness. Decreasing concentrations of thyroxine reaching the tumor cell as the tumors grew larger would thus appear to be the most likely factor accounting for lack of responsiveness of this tumor strain. Nonetheless, provision of increased amounts of thyroxine, greater than the increase in tumor size would seem to require, fail to suppress growth of the larger tumors.† Thus more is probably involved than hormonal concentration alone: perhaps limited blood supply to the tumor, perhaps loss of exogenous excess hormone from the body through overflow and excretion into the bile(3). In any event, it would appear that lethal amounts of thyroid hormone may have to be administered systemically if the minimal concentrations required to suppress growth of the larger tumors are to be reached.

† Grindberg, R., Werner, S. C. unpublished data.

1. Furth, J., Gadsden, E. L., Burnett, W. T., PROC. SOC. EXP. BIOL. AND MED., 1952, v80, 4.

2. Werner, S. C., Grinberg, R., *Cancer Research,* April 1961.

3. Albert, A., Keating, F. R., Jr., *J. Clin. Endoc. Metab.,* 1949, v9, 1406.

Received October 10, 1960. P.S.E.B.M., 1961, v107.