Cardiovascular Effects of Thyroxine Treatment in Hypophysectomized Rats

By Margaret Beznak, M.D.

With the technical assistance of A. Marcsan and T. Fournier

Earlier experiments showed that the low blood pressure of hypophysectomized rats was due to a decreased cardiac output, while peripheral resistance was slightly above normal. This was also the case in hypophysectomized man. Acute loading by infusion of polyvinylpyrrolidone, PVP, into the right side of the heart resulted in an increase of cardiac output and work to 2-4 times the basal, pre-infusion values. It was concluded that the hypotension and low cardiac output in hypophysectomized rats was not due to the inability of the heart to raise its output but rather to the fact that some hemodynamic prerequisite of a higher output was absent. The atrophy of the heart which is known to take place after hypophysectomy (the loss of cardiac weight is proportionately greater than the loss of body weight) was considered to be due to an adaptation of the heart to the lower demands. If this is so then any increase in the demand on the heart should prevent the cardiovascular changes occurring after hypophysectomy. Early and Leblond showed that thyroxine treatment of hypophysectomized rats prevented the fall in oxygen consumption of the animals, in the heart rate and in the heart rate/body weight ratio. Scow found that in rats hypophysectomized at weaning and treated with thyroxine the weight of the heart was heavier than in untreated hypophysectomized rats and in normal control rats.

The present experiments were undertaken to ascertain the effect of thyroxine on the cardiovascular system of hypophysectomized rats in which all changes due to hypophysectomy had already fully developed. The problems were to determine: (a) whether thyroxine could restore the weight of the heart to normal or only prevent its atrophy, (b) whether aortic constriction in hypophysectomized and thyroxine treated rats led to cardiac hypertrophy, (c) what the effect of thyroxine treatment was on various cardiovascular parameters and (d) whether thyroxine had any effect on the cardiac reserve of hypophysectomized rats.

Methods

FIRST SERIES

Male rats, Wistar strain, were hypophysectomized and kept in individual cages for two to three months. Four groups of such rats were then injected with thyroxine, subcutaneously, in increasing doses so that the total amount of thyroxine administered was as follows: a total of 0.5 mg thyroxine in three weeks, a total of 1.0 mg thyroxine in four weeks, a total of 1.5 mg thyroxine in four weeks, and a total of 2.5 mg thyroxine in five weeks.

A group of 17 untreated hypophysectomized rats and a group of 21 normal rats were included as controls.

At the end of the injection period the rats were anesthetized with pentobarbital. Blood coagulation was prevented by heparin, 0.3 ml of a 3% solution, intravenously, per rat. Cardiac output was determined on the basis of the direct Fick
The effect of thyroxine treatment on oxygen consumption, weight and rate of the heart, blood pressure and peripheral resistance of hypophysectomized rats.

1st group = 17 untreated rats hypophysectomized 2 to 3 months previously. 2nd group = 6 rats which received a total of 0.5 mg thyroxine in 3 weeks, 2 to 3 months after hypophysectomy. 3rd group = same as above, but receiving a total of 1.0 mg thyroxine in 4 weeks (6 rats). 4th group = same as above but receiving a total of 1.5 mg thyroxine in 4 weeks (6 rats). 5th group = same as above but receiving a total of 2.5 mg thyroxine in 5 weeks (11 rats). 6th group = 17 untreated normal control rats. Gross hatched area = standard deviation of the individual cases. Doubly cross hatched area = standard error of the mean. T.P.R. = calculated total peripheral resistance.

The oxygen content of mixed venous and arterial blood was estimated with the micro-method of Roughton and Scholander. Blood pressure was recorded in the left carotid artery with a Hathaway gauge. The rats breathed pure oxygen from a microspirometer made of plexiglass. The drop of the spirometer was recorded electrically on the same photographic paper as the arterial and venous pressures. Heart rate was calculated from the records. Peripheral resistance and work of the heart were calculated from output and blood pressure. Only the work of the left ventricle was considered and the kinetic factor was omitted. The load on the heart was increased by infusing PVP into the right side of the heart. The cardiovascular parameters described above were determined at regular intervals during infusion. Details of the procedures have been described elsewhere. "Basal" output and work refer to the values before infusion was started and "maximum" output and work to the maximum values measured during infusion. The rats were autopsied at the end of the experiment and the wet and dry weights of the heart and different organs were determined. The statistical significance of the differences was checked by Student's t test.

SECOND SERIES

One hundred and ten male rats, Wistar strain, were hypophysectomised and kept in individual cages for two to three months. Then a group received a total of 0.5 mg thyroxine in three weeks and another group 1.5 mg thyroxine in four weeks. Towards the end of the 3rd week of treatment the aortae of the rats were constricted just below the diaphragm with a silver ring of 0.8 mm diameter. Because of the very large mortality, 5 mg hydrocortisone (Hydrocortone, Merck, Sharp and Dohme) were injected, intraperitoneally, into rats on the day before and at the time of aortic constriction. Five to ten days later the same measurements were performed in the surviving rats as described in the first series. A group of 16 se-
The effect of thyroxine treatment on basal cardiac output and work of hypophysectomized rats. Groups are the same as in Figure 1.

Results

FIRST SERIES

Disregarding operative mortality and deaths occurring during the first two to three postoperative days, 25% of the rats survived hypophysectomy for a period of two to three months. Of the 70 rats available at this time, 17 were used as untreated controls (group 1, body weight 174 ± 14 g). The remaining 53 were injected with thyroxine. By the end of the 3rd week of injection, 12 rats, about 23%, had died. At this time six rats were removed (group 2, body weight 173 ± 16 g) and the measurements performed as described above. Of the 35 remaining rats seven (20%) died during the 4th week of injections and 13 were removed for the experiments (group 3, body weight 179 ± 27 g and group 4, body weight 166 ± 12 g). Of the remaining 15 rats four died during the 5th week of injection and the experiment was performed on the remaining 11 at this time (group 5, body weight 173 ± 27 g). Group 6 (body weight 175 ± 27 g) was made up of 17 normal control rats.

Figure 1 shows that thyroxine treatment of rats hypophysectomized two to three months previously restored the low oxygen consumption, the weight of the heart, the rate of the heart and the mean carotid blood pressure to normal. There was a tendency for the higher peripheral resistance of hypophysectomized rats to be lowered towards normal by the treatment.

Figure 2 demonstrates that the stroke volume was not significantly different in hypophysectomized and in normal rats and that thyroxine treatment had not much effect on it. On the other hand, the minute volume was much lower in hypophysectomized rats than in normal rats, in consequence of the bradycardia which developed after hypophysectomy. The minute volume could be restored to normal by thyroxine treatment, due to the in-
crease in the rate of the heart caused by the treatment. Stroke work, and to a much more marked degree minute work, were lower in hypophysectomized rats than in normal rats and both were restored to normal by thyroxine treatment.

Figure 3 shows the maximum values obtained during PVP infusion. While the basal, pre-infusion values were completely restored to normal by thyroxine treatment (fig. 2), the maximum values measured in these groups did not reach the level found in normal rats during similar PVP infusion. This seemed to indicate that the strength of the heart or its reserve force, was less in hypophysectomized rats treated with thyroxine than in normal rats, although under basal conditions (without loading) none of the cardiovascular parameters measured was different.

SECOND SERIES
Since the experiments indicated that the maximum cardiac output and work (PVP infusion) were less in hypophysectomized rats treated with thyroxine than in normal rats, the performance of the heart during chronic loading was next measured. Chronic load was imposed on the heart by constricting the aorta.

Figure 4 shows that fewer hypophysectomized rats survive aortic constriction if they are treated with thyroxine than if they have no treatment. A comparison of curve 1 and curve 2 indicates that in the group of untreated hypophysectomized rats (curve 1) the first mortality occurred on the 3rd day after aortic constriction, whereas most of the hypophysectomized rats treated with thyroxine had died by this time. Not one of the 50 rats in this latter group survived longer than 10 days, but 50% of the untreated hypophysectomized rats were alive 14 days after aortic constriction.

Since the greatest number of the rats (some 70%) died within one day of narrowing, the possibility arose that the mortality might be due to surgical shock, i.e., the operation (laparotomy) itself. A sham operation was therefore performed in 12 hypophysectomized rats treated with thyroxine (curve 3). It was immediately evident that the large initial mortality was not due to aortic constriction. Such initial mortality did not occur in untreated

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**FIGURE 3** The effect of thyroxine on maximum cardiac output and work of hypophysectomized rats. Groups are the same as in Figure 1.
rats hypophysectomized two to three months previously, even after aortic constriction (curve 1). It seemed likely therefore that by restoring metabolic rate thyroxine made hypophysectomized rats more liable to succumb to surgical shock. Ten rats hypophysectomized two to three months previously were therefore treated with thyroxine for four weeks and then laparotomized. They received 5 mg hydrocortisone on the evening before the operation and 5 mg immediately following the operation (curve 4). Mortality, particularly early mortality, was significantly reduced in this group (compare curves 3 and 4). In the last group of 40 rats hypophysectomized two to three months previously and then treated with thyroxine over three weeks, the aorta was constricted. On the night before and immediately following constriction the rats received 5 mg hydrocortisone (curve 5). Although immediate mortality was reduced by hydrocortisone (curves 2 and 5) the death rate of hypophysectomized rats treated with thyroxine was much greater throughout than was mortality of untreated hypophysectomized rats or of normal rats on aortic constriction.

Figures 5 and 6 indicate the hemodynamic parameters measured 5 to 10 days after aortic constriction. Figure 5 shows the basal values determined before infusion and figure 6 the maximum values reached during PVP infusion. Thyroxine treatment restored the weight of the heart of hypophysectomized rats to normal (fig. 1); aortic constriction caused no further increase in the weight of the heart in these rats. In normal rats, with cardiac weights similar to those of the hypophysectomized thyroxine treated group, similar aortic constriction led to a cardiac hypertrophy of 33%. Cardiac output and especially cardiac work were considerably less in thyroxine treated hypophysectomized rats after aortic constriction than before it, whereas this was not the case in normal rats (compare figures 2 and 3 with figures 5 and 6). This was true of measurements made before, as well as during, infusion of PVP.

Discussion
The experiments indicate that thyroxine treatment not only prevents the appearance of cardiovascular changes following hypophysectomy, but can restore all cardiovascular parameters to normal at a time when all changes had developed maximally. Under basal conditions, the weight and rate of the heart, the blood pressure, cardiac output and work reached the values found in normal rats. It is evident from figure 2 that although the doses of thyroxine used in the present experiments were relatively large (0.5 to 2.5 mg over three to five weeks) compared to those used by other investigators, e.g., Reti and Leblond, Scow, and Evans et al., they caused a graded restoration of some of the hemodynamic values. Thus minute volume...
The effect of aortic constriction in hypophysectomized rats treated with thyroxine. Basal, pre-infusion values.

1 = hypophysectomy (2 to 3 months) + aortic constriction (10 rats, 172 ± 15 g). 2 = hypophysectomy + 0.5 mg thyroxine over 3 weeks + aortic constriction (8 rats, 154 ± 16 g). 3 = hypophysectomy + 1.5 mg thyroxine over 4 weeks + aortic constriction (10 rats, 153 ± 12 g). 4 = normal rats with aortic constriction (12 rats, 175 ± 13 g).

Percentage change in cardiac weight was calculated from the equation $y = 2.14x + 295.10$, where $y$ represents expected weight of the heart in mg and $x$ body weight of the rats in g. The equation was calculated by the method of least squares as described earlier.1

Administration of 2 × 5 mg hydrocortisone decreased the immediate mortality following laparotomy and aortic constriction in hypophysectomized rats treated with thyroxine. The possibility that hydrocortisone might have had other significant effects was discarded for several reasons. (a) When the experiments were started hydrocortisone was not used. It was introduced because of the high mortality. There were no differences in any of the cardiovascular measurements made whether hydrocortisone was given or not. (b) Hydrocortisone was given together with growth hormone to hypophysectomized rats without having any effect.10 (c) It was found earlier that treatment of hypophysectomized rats with various adrenocortical preparations did not restore cardiovascular parameters to normal.11

Although thyroxine treatment raised the...
THYROXINE AND HYPOPHYSECTOMY


Groups are the same as in figure 5. The number of rats was 7 in the first group, 9 in the second group, 10 in the third group and 12 in the fourth group.

low hemodynamic parameters of hypophysectomized rats to normal, and the weight of the heart increased to the normal level, the myocardium still seemed to be qualitatively different from that of normal hearts. This difference was brought to light by measuring cardiac output and work during loading, either acutely (by infusing PVP) or chronically (by constricting the aorta). In these tests, cardiac output and work in hypophysectomized rats treated with thyroxine were less than in normal rats of comparable size, although there was no difference under basal conditions.

These particular results serve to emphasize a more general rule, namely that measurements designed to compare the effects of different treatments in experimental animals should be made not only in the basal state but also during the application of a physiological load. This point was emphasized by A. B. L. Beznak et al. in connection with the comparison of the nutritional value of diets. With various oils in the diet, no difference appeared in the growth rate of the animals under resting conditions. However, very great differences became apparent in the extent of loss of body weight during forced exercise and in the speed of recovery in the post-exercise resting period.

It was concluded that thyroxine treatment restored oxygen consumption of hypophysectomized rats to normal. With it the blood pressure, rate and weight of the heart, cardiac output and work also reached normal values. Under basal conditions, therefore, there were no apparent differences in the cardiovascular system of normal rats and hypophysectomized
rats treated with thyroxine. Significant differences appeared, however, when the heart was forced to do extra work. The heart of the normal rat could raise its output and work considerably under such conditions, whereas the performance of hearts of hypophysectomized rats treated with thyroxine fell below normal. In fact, in some measurements, these hearts did not even do as well as the much smaller hearts of untreated hypophysectomized rats. It is evident that hypophysectomized rats require another factor besides thyroxine to enable the cardiovascular system to parallel the performance of normal rats. Experiments indicate that cardiovascular parameters, in basal conditions and during loading, are practically normal and in some cases even exceed the normal level in hypophysectomized rats treated with a combination of thyroxine and growth hormone.

**Summary**

Oxygen consumption, weight and rate of the heart, blood pressure, cardiac output and work are restored to normal if rats hypophysectomized two to three months previously are treated with thyroxine over a period of three to five weeks. Although under basal conditions the above parameters in these rats are the same as those in normal rats, the maximum output and work during loading by infusion of polyvinylpyrrolidone into the right side of the heart are less in hypophysectomized rats treated with thyroxine than they are in normal rats. Similarly, if a load is imposed on the heart by constriction of the aorta, cardiac output and work are well below the values measured in normal rats with aortic constriction. It is concluded that thyroxine treatment of hypophysectomized rats can restore to normal the performance of the cardiovascular system under basal conditions but that during loading (polyvinylpyrrolidone infusion, aortic constriction) the performance of the cardiovascular system is less than in normal rats.

**References**

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doi: 10.1161/01.RES.12.4.333

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