FURTHER OBSERVATIONS OF THE HYPOTHALAMIC CONTROL OF THE THYROID GLAND

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On the hypothalamic control of the thyroid gland, many intensive studies have been carried out since Gräfe and Grünthal (1927), and many workers seem to recognize in the hypothalamus the regulatory center for the thyroid. As reported previously, we found a thyroid stimulating neurohumor in the hypothalamus (Shibusawa *et al.*, 1956 a, b), and further confirmed the presence of this substance in the urine, and of its active principle in -7.8 fraction in the zone electrophoresis of the urinary polypeptides. It was provisionally designated by us as TRF (Thyrotropin Releasing Factor). Greer (1951 a, b and c) advanced a view that the thyrotrophic agent in the hypophysis consists of two factors—metabolic and growth factors. Against Greer, we considered that there is only a single factor, TRF, which regulates the pituitary-thyroid system.

MATERIALS AND METHODS

(a) Experimental animals: Wistar strain male rats weighing about 150 g were used. Female animals were rejected because of the possible influence of menstrual cycle on the thyroidal function. The animals were fed pressed barley, vegetables and fish powder to secure sufficient calories and vitamins, and given tap water for drinking. To avert the influence of temperature, it was kept at 10–15°C, and also with respect to noise, light and other environmental conditions, care was taken to maintain the constancy.

(b) Exp. I: The animals were divided into 4 groups, each group consisted of 5 rats, according to the dosage of 0.5% TRF—minute dose (0.2 cc), small dose (0.5 cc), medium dose (1.0 cc), and large dose (2.0 cc). Each dose was daily injected for one week.

Exp. II: Experiments on rats with lesion in the hypothalamus. Lesion was placed using the Krieg's stereotaxic machine and that devised by the Brain Research Institute of Tokyo University. For anesthesia, a 0.0125 g×0.5 cc solution of pentobarbital (Tanabe Co.) was used intraperitoneally. A monopolar needle attached to the stereotaxic machine was inserted into the skull through holes (0.2 mm in dia.) made symmetrically on the right and the left.

The site of lesion was determined using the coordinate by Krieg (1946) and hole on the skull was made just over the nucleus to be injured. An electric

Fig. 1. The current of electric source

current was passed with the arrangement as illustrated in Figure 1. As the electric source, a 22.5 V National Cell was used. (The operation was carried out under the direction of Prof. Koikegami, Department of Anatomy, Niigata University). The current strength was 2 mA $\times$ 5 mm, which was reported by Krieg to be most suited for placing lesion in the hypothalamus. The animals were divided into 3 groups according to the site of lesion—anterior, middle and posterior areas of the hypothalamus. Each group was injected with 50 mg/kg of methylthiouracil (MTU) and 2.0 cc/kg of TRF for 3 and 5 consecutive weeks, as tabulated below:

Group I. Lesions on the anterior hypothalamus.

Control.
 { MTU given for 3 weeks. MTU given for 5 weeks.
 { TRF given for 3 weeks. TRF given for 5 weeks.

Group II. Lesions on the middle hypothalamus.

Control.
 { MTU given for 3 weeks. MTU given for 5 weeks.
 { TRF given for 3 weeks. TRF given for 5 weeks.

Group III. Lesions on the posterior hypothalamus.

Control.
 { MTU given for 3 weeks. MTU given for 5 weeks.
 { TRF given for 3 weeks. TRF given for 5 weeks.

Exp. III: After the effective destruction of the anterior hypothalamus, 0.5% TRF was injected for 2 consecutive weeks in 4 different doses—minute dose (0.2 cc), small dose (0.5 cc), medium dose (1.0 cc) and large dose (2.0 cc)—similarly as in Exp. I.

On the day of the completion of the injection, all the animals including controls were intraperitoneally injected with 2 μ c of I^{131} , which was supplied by the Japanese Association of Radioactive Isotope.

Two μ c of I^{131} was contained in 0.5 cc of solution. At the 24th hr. after the injection of I^{131} , blood was collected by heart puncture, serum was separated, and radioactivity by the count by minute (c.p.m.) was determined with a Geiger-Müller counter.

The thyroid was removed after sacrifice by exanguination, I^{131} uptake was measured with a Geiger-Müller counter for 3 mins. And T/S was calculated by the following formula: T/S = c.p.m. per 100 g thyroid/c.p.m. per 0.1 cc serum.

RESULTS

Exp. I: After the administration of the minute (0.2 cc) and the small dose (0.5 cc) of TRF, the weight of the thyroid was 4.8 mg and 5.6 mg, respectively, showing scarcely any increase as compared with the control (4.4 mg). By contrast, the medium (1.0 cc) and the large dose (2.0 cc) increased the thyroid weight remarkably to 10.2 mg and 9.6 mg, respectively (Fig. 2).

The thyroid uptake of I^{131} was 42 c.p.m./mg after the injection of TRF in the minute dose, 48 c.p.m./mg in the small dose, 65 c.p.m./mg in the medium dose and 66 c.p.m./mg in the large dose. Thus in the case of the minute and small dose, no significant increase was seen, while in the case of the medium and large dose increase was prominent as compared with the control (40 c.p.m.) (Fig. 2). T/S ratio was 36.8% in the case of the minute dose, 54.4% in the case of the small dose, 59.3% in the case of the medium dose, and 60.5% in the case of the large dose. Thus increase was remarkable as compared with the control in the case of the minute dose, and became still more conspicuous as the dose was increased

Fig. 2. Effect of TRF (0.2cc, 0.5cc, 1.0cc and 2.0cc) repeated injection

Fig. 3. Anterior hypothalamus lesion

(Fig. 2). Namely, when the hypophysis and the thyroid were intact, the weight and uptake of I^{131} of thyroid showed no remarkable response to the minute and the small dose of TRF, whereas T/S ratio responded remarkably even to the minute dose. That is to say, T/S ratio is more sensitive to TRF than the thyroid weight if the hypophysis and the thyroid are intact.

Exp. II: (I) Effects of administrations of MTU and TRF after destroying the anterior hypothalamus.

(a) In the case of lesions alone (without MTU and TRF): As shown in Figure 3, lesions were placed rostrally to the median eminence reaching the preoptic area, supraoptic, and suprachiasmatic nuclei. After 3 weeks, the thyroid weight was 5.2mg, and after 5 weeks 4.9mg, showing 25.5% and 30.0% decrease, respectively as compared with the normal (Fig. 4).

The thyroid uptake of I^{131} was 34 c.p.m./mg 3 weeks after placing lesions, and 32 c.p.m./mg 5 weeks after, respectively, showing 24.5% and 28.9% decrease as compared with the normal (Fig. 4). The T/S ratio of I^{131} concentration was 17.6% after 3 weeks, and 17.1% after 5 weeks, showing evident decrease in each case (Fig. 4).

The pituitary weight was 3.4 mg (5.5% decrease) after 3 weeks, and 3.2 mg (11.1% decrease) after 5 weeks (Fig. 4).

The average height of the follicular epithelium of the thyroid was 4.7μ 3 weeks after placing lesions, and 4.6μ 5 weeks after, respectively showing 18.8% and 20.6% decrease as compared with the normal (Fig. 4).

Histological findings, which will be reported elsewhere, were summarized as follows:

Follicular epithelium of the thyroid was low, follicles were large in size, nuclei were flattened, and colloid stained well with acid dye was plentiful. The absorption of colloid was not seen. In the hypophysis, beta cells were decreased in number, their nuclei were round or polygonal, rather small in size, and found in eccentric positions, and cytoplasm contained large granules, presenting a picture of hypofunction in thyrotrophin releasing.

(b) Administration of MTU after the placement of lesions: The thyroid weight was 6.0 mg after the 3 weeks administration of MTU following the placement of lesions and 6.5 mg after 5 weeks, which are 14.2% and 7.1% decrease from the control level, respectively. Thus the goitrogenic effect of MTU as demonstrated in the normal animal could not be found in the hypothalamectomized animal (Fig. 4).

The pituitary weight was 2.9 mg after 3 weeks, and 2.7 mg after 5 weeks, showing 19.4% and 16.6% decrease, respectively, as compared with the normal. Also in this organ, MTU did not produce increase in weight (Fig. 4).

The uptake of I^{131} by the thyroid was 40 c.p.m./mg (11.2% decrease) after 3 weeks, and 37 c.p.m./mg (17.8% decrease) after 5 weeks, showing slight decrease in both cases (Fig. 4).

T/S was 20.3% 3 weeks after, and 20.5% 5 weeks after, showing nearly the same values as the normal (Fig. 4).

The average height of the follicular epithelium of the thyroid was 5.4μ 3 weeks after, and 5.5μ 5 weeks after, respectively, showing decrease from the normal value (Fig. 4).

Fig. 4. Effects of lesion on the anterior hypothalamus, and effects of administration of MTU and TRF.

Summary of the histological findings: Follicular epithelium of the thyroid was rather low, nuclei were flattened, follicles were large in size, plentiful of colloid well stainable, and figures of colloid absorption were not seen. In the hypophysis, beta cells were decreased in number as compared with alpha cells, circular or polygonal in shape, and their granules were coarse.

(c) Administration of TRF after the placement of lesions.

The thyroid weight was 11.6 mg after the 3 weeks administration of TRF following the placement of lesions and 17.3 mg 5 weeks after, that is, it showed remarkable increase of 65.5% and 143% from the normal value (Fig. 4).

The pituitary weight was 3.8 mg after 3 weeks, and 4.0 mg after 5 weeks. The value after 3 weeks was showed 5.6% increased, while the value after 5 weeks showed 11.1% increase (Fig. 4).

The thyroid uptake of I^{131} was 69 c.p.m./mg after 3 weeks, and 84 c.p.m./mg after 5 weeks, showing 34.8% and 46.5% increase, respectively (Fig. 4).

The T/S ratio of I^{131} concentration was 42% after 3 weeks, and 42.7% after 5 weeks, showing increase in both cases as compared with the normal (Fig. 2).

The average height of the follicular epithelium of the thyroid was 6.4μ after 3 weeks, and 6.8μ after 5 weeks, showing 12.1% and 19.2% increase, respectively (Fig. 4).

Summary of the histological findings: Follicular epithelium of the thyroid was thickened and proliferated in various places into follicles in a papillary form. Nuclei were circular or elliptic and large in size. Follicles were small, scanty of colloid, displaying a vacuolar picture of absorption.

In the hypophysis, enlargement and proliferation of beta cells were prominent, displaying a picture of releasing beta granules.

(II) Effects of administrations of MTU and TRF after the lesions destroying the middle hypothalamus.

(a) In the case of lesions alone (without MTU and TRF): As illustrated in Figure 5, lesions were placed dorsally to the median eminence along the medio-basal region, keeping the former uninjured as far as possible.

Fig. 5. Middle hypothalamus lesion

The thyroid weight was 7.0 mg 3 weeks after the placement of lesion, and 6.3 mg after 5 weeks, showing slight decrease of 1.4% and 11.1%, respectively (Fig. 6).

I^{131} uptake by the thyroid was 34 c.p.m./mg after 3 weeks, and 20.4 c.p.m./mg after 5 weeks, scarcely different from the normal (Fig. 6).

Fig. 6. Effects of lesion on the middle hypothalamus, and effects of administration of MTU and TRF

The pituitary weight was 3.6 mg after 3 weeks, and 3.6 mg after 5 weeks, were same with the normal (Fig. 6).

The average height of the follicular epithelium of the thyroid was 5.5μ after 3 weeks, and 5.4μ after 5 weeks, showing slight decrease of 5.2% and 5.3%, respectively (Fig. 6).

Summarized histological findings: In the thyroid, there was no remarkable difference from the control after 3 weeks, but after 5 weeks, follicular epithelium showed no enlargement, nuclei were normal, and colloid was plentiful, showing a picture of depressed function. In the hypophysis, beta cells were slightly smaller in number than alpha cells, irregular in form, and granules were coarse, indicating the depressed thyrotrophin secretion.

(b) Administration of MTU after the placement of lesions. The thyroid weight was 7.5 mg after the 3 weeks administration of MTU, nearly the same as normal, and 8.9 mg after 5 weeks, showing slight increase (Fig. 6).

The thyroid uptake of I^{131} was 36 c.p.m./mg after 3 weeks, and 37 c.p.m./mg after 5 weeks, showing 20% and 17.8% decrease, respectively (Fig. 6).

The average height of the follicular epithelium of the thyroid was 5.6μ after 3 weeks, nearly the same with the normal, and 6.3μ after 5 weeks, showing a slight increase (Fig. 6).

Summarized histological findings: There were no remarkable changes in 3 weeks. After 5 weeks, however, follicular epithelium of the thyroid was rather thickened, nuclei assumed circular form, follicles were relatively small, and colloid was scanty but without showing a picture of absorption.

(c) Administration of TRF after the placement of lesions.

The thyroid weight was 11.9 mg after the 3 weeks administration of TRF, and 13.5 mg after 5 weeks, showing remarkable increase of 70.0% and 92.8%, respectively (Fig. 6). The pituitary weight was 4.1 mg after 3 weeks, and 4.2 mg after 5 weeks, showing 5.1% and 7.6% increase, respectively (Fig. 6).

The thyroid uptake of I^{131} was 67 c.p.m./mg after 3 weeks, and 69 c.p.m./mg after 5 weeks, showing 48.8% and 53.3% increase, respectively (Fig. 6).

The T/S ratio of I^{131} concentration was 40.6% after 3 weeks, and 48% after 5 weeks, showing remarkable increase in both cases (Fig. 6).

The average height of the follicular epithelium of the thyroid was 6.3μ after

Fig. 7. Posterior hypothalamus lesion

3 weeks, and 6.7μ after 5 weeks, showing 8.6% and 15.5% increase, respectively (Fig. 6).

Summarized histological findings: The follicular epithelium of the thyroid was thickened, assuming columnar form in various places, nuclei were circular, follicles were small, and scanty of colloid. Also vacuoles were seen there. In the hypophysis, beta cells were increased, showing a picture of releasing granules, that is, accelerated secretion of the thyrotrophin.

(III) The effects of administrations of MTU and TRF after the placement of lesions in the posterior hypothalamus.

(a) In the case of lesions alone (without MTU and TRF). As illustrated in Figure 7, lesions were given dorsally to the infundibular nuclei reaching the mamillary nuclei. The thyroid weight was 7.0 mg 3 weeks after the placement of lesions, and 6.5 mg 5 weeks after, scarcely different from the control in both cases (Fig. 8).

The pituitary weight was 3.6 mg after 3 weeks, and 3.7 mg after 5 weeks, nearly the same as the normal (Fig. 8).

The thyroid uptake of I^{131} was 22 c.p.m./mg after 3 weeks, and 24 c.p.m./mg after 5 weeks, scarcely showing significant difference from the control (Fig. 8).

The T/S ratio of I^{131} concentration was 16.6% after 3 weeks, and 17.8% after 5 weeks, showing some decrease in both cases (Fig. 8).

The height of the follicular epithelium of the thyroid was 5.6μ after 3 weeks, and 5.5μ after 5 weeks, nearly the same with the normal values (Fig. 8).

Summarized histological findings: The follicular epithelium was rather low, some nuclei were flattened, and follicles, though not so large, were relatively plentiful of colloid, displaying, in general, a picture of the depressed function.

In the hypophysis, beta cells were relatively small in number, and no prominent changes were seen in their form, granules and nuclei.

(b) Administration of MTU after the placement of lesions.

The thyroid weight was 19.5 mg after the 3 weeks administration of MTU, and 20.9 mg after 5 weeks, showing remarkable increase of 178.5% and 198.2%, respectively (Fig. 8).

The pituitary weight was 3.8 mg after 3 weeks, and 3.7 mg after 5 weeks (Fig. 8).

The thyroid uptake of I^{131} was 40 c.p.m./mg after 3 weeks, and 41 c.p.m./mg after 5 weeks, slightly lower than the control value (Fig. 8).

The T/S ratio of I^{131} concentration was 20.9% after 3 weeks, and 22.7% after 5 weeks, similarly to the normal (Fig. 8).

The height of the follicular epithelium of the thyroid was 7.0μ after 3 weeks, and 7.3μ after 5 weeks, showing prominent increase of 20.7% and 25.8%, respectively (Fig. 8).

Summarized histological findings: The follicular epithelium was small, colloid was scanty, and vacuoles were seen in some parts. In the hypophysis, beta cells were not significantly different from the normal in number and form, or with regard to their nuclei and granules.

(c) Administration of TRF after the placement of lesions.

The thyroid weight was 15.4 mg after the 3 weeks administration of TRF,

Fig. 8. Effects of the lesion on the posterior hypothalamus, and effects of administration of MTU and TRF

and 17.6 mg after 5 weeks, showing extremely prominent increase of 120.0% and 151.4%, respectively (Fig. 8).

The pituitary weight was 4.1 mg after 3 weeks, and 4.4 mg after 5 weeks, showing slightly increase of 13.8% and 22.2%, respectively (Fig. 8).

The height of the follicular epithelium of the thyroid was 6.9μ after 3 weeks, and 7.1μ after 5 weeks, showing remarkable increase of 21% and 24.5%, respectively (Fig. 8).

The thyroid uptake of I^{131} was 69 c.p.m./mg after 3 weeks, and 69.5 c.p.m./mg after 5 weeks, showing remarkable increase of 53.3% and 54.4%, respectively (Fig. 8).

The T/S ratio of I^{131} concentration was 34.1% after 3 weeks, and 38.0% after 5 weeks, showing increase in both cases (Fig. 8).

Summarized histological findings: The follicular epithelium of the thyroid was markedly thickened, showing papillary proliferation in various parts.

Nuclei were circular and large, but follicles were small and scanty of colloid, displaying a picture of absorption at various parts. In the hypophysis, proliferation and hypertrophy of the beta cells were prominent, displaying a picture of releasing beta granules, that is, a picture of accelerated thyrotrophin secretion.

The above mentioned results were put in order as follows: When the anterior hypothalamus was injured effectively, evident decrease in the weight of the thyroid and hypophysis resulted, indicating their depressed functions. MTU (50 mg/kg) given after the injury did not exert any goitrogenic effect as in the normal animal. When, however, TRF was administered, the thyroid weight increased remarkably, and histological findings also presented a picture of hyperfunction. At the same time the pituitary weight increased. When lesions were given in the middle hypothalamus, the thyroid displayed a morphological as well as functional picture of slight depression. The administration of MTU produced some goitrogenic effect in the injured animals though not so conspicuously as in the normal animal. The administration of TRF elicited remarkably activated response both morphologically and functionally. Also in this case the increase of pituitary weight concurrently occurred. When the posterior hypothalamus was injured, there was scarcely any depression in the thyroidal function. The administration of MTU produced remarkable enlargement of the thyroid, but the I^{131} accumulating function remained almost unchanged. The administration of TRF, however, increased both the weight and the function of the thyroid. At the same time pituitary weight increased, and histologically a picture of enhanced thyrotrophin secretion was seen.

Exp. III: By the same procedures as Exp. II, lesions were made in the effective site of the anterior hypothalamus. And the minute (0.2 cc), the small (0.5 cc), the medium (1.0 cc) and the large dose (2.0 cc) of TRF were administered.

After the administration of TRF in the minute and the small dose for 2 consecutive weeks, thyroid weight was 5.2 mg and 5.1 mg, respectively, showing some increase as compared with 3.8 mg of the hypothalamectomized control. But the difference was not so evident. When the medium dose was given, thyroid weight remarkably increased to 7.0 mg. And in the case of the large dose, it was 8.6 mg with still greater increase (Fig. 9). The thyroid uptake of I^{131} was 39 c.p.m./mg in the case of the minute dose, and in the case of the small dose, showing

no significant difference from the control. It was 45 c.p.m./mg in the case of the medium dose, and 57 c.p.m./mg in the case of the large dose, showing remarkable increase in both cases (Fig. 9).

The T/S ratio of I^{131} concentration was 36.1% and 39.1% in the cases of the minute and the small dose, respectively, with significant increase as compared

Fig. 9. Effects of the lesion on the anterior hypothalamus, and effect of administration of TRF (0.2cc, 0.5cc, 1.0cc and 2.0cc)

with the control. It was 47.5% and 50.1% in the cases of the medium and the large dose, respectively, showing prominent increase in both cases (Fig. 9).

These results were in agreement with those of Exp. I. That is to say, the T/S ratio responded more sensitively than thyroid weight to a small amount of TRF even when the anterior hypothalamus was effectively damaged.

DISCUSSION

It is well known that the hypothalamus-pituitary connection must be intact to maintain unmolested release of TSH. As pointed out early by Cahane and Cahane (1938), the regulatory center of TSH secretion is considered to exist in the hypothalamus.

Greer (1951, 1952) made electric lesions to the hypothalamus of the rat in the area from suprachiasmatic to ventromedial nuclei, and then fed them with PTU for 1~3 weeks; he failed to find the thyroid hypertrophy, which is observed in the normal animal. In this case, T/S ratio of I^{131} concentration was elevated similarly as in the normal. This was confirmed by Bogdanove and Halmi (1952, 1953).

Since then two important questions have been raised: One is concerned with the localization of the regulatory center for the thyroid, and the other is whether two factors—growth and metabolic—can be recognized in thyrotrophin.

In the extract of the anterior hypothalamus we confirmed the presence of TRF, a substance which stimulates the release of TSH, and claimed that the control of TSH secretion can be ascribed to TRF (Shibusawa *et al.*, 1956a, b).

As pointed out by Olivecrona (1957), when the median eminence or the mediobasal tissue in front of the latter was injured, the thyrotrophic function was so depressed that it became impossible to analyse the central system of the hypothalamus. Bogdanove and Halmi (1953) recognized the effective focus in the basal region near the midline between the median eminence and the optic chiasma. Greer and Erwin (1954) regarded the lesions involving paraventricular nuclei as effective. Ganong, Frederickson and Hume (1954), and Ganong, Frederickson and Hume (1955) confirmed in dogs that the lesions made on the median eminence and on the immediately cranial tissue were effective. Olivecrona (1957) found that the lesions completely destroying the paraventricular nuclei did not prevent the goitrogenic response to PTU feeding.

According to our results, lesions rostrally to the median eminence extending from supraoptic and suprachiasmatic nuclei to the preoptic area were most effective in preventing the goitrogenic response to MTU feeding. And the lesions extending from the tuber cinereum to the infundibulum in the middle hypothalamus exerted the similar effect to some degree. In this case, the effect of lesions in the median eminence could not be denied. Our results were in this way approximately in agreement with those of Bogdanove and Halmi (1953), and of Bogdanove (1957).

TRF is found in the extract of tissue of this region.

Bogdanove and Halmi (1953) reported that the effective lesions in the anterior hypothalamus could not make follicular cells of the thyroid lower than the

cuboidal form, and consequently they could not render serious curtailment in the thyrotrophin release. According to D'Angelo and Traum (1957), the effective lesions in the anterior hypothalamus could inhibit the production and release of thyrotrophin in the lesser degree than hypophysectomy. Reichlin (1957) obtained the results that despite the presence of the effective lesions in the anterior hypothalamus, compensatory hypertrophy following the 3/4 removal of the thyroid, still developed.

In our experiments, the effective lesions in the above mentioned anterior hypothalamus induced slight atrophy and hypofunction of the thyroid, but to a higher degree than lesions in the middle and posterior hypothalamus.

It may be considered that the release of TSH would autonomously be performed to some degree after destroying the central system of the hypothalamus.

Assumedly owing to this, MTU could elevate the T/S I^{131} concentration ratio, and TRF was able to elicit an evident increase in the thyroid weight and T/S ratio. We claimed that the stimulating action of TSH secretion by MTU would be exerted partly through the mediation of TRF, and consequently that TRF is the TSH stimulating factor.

As to growth and metabolic factors, proposed by Greer (1951), extensive discussions have been carried out between Greer and Bogdanove. According to Bogdanove and Halmi (1953), Bogdanove, Spirtos and Halmi (1955), T/S I^{131} concentration ratio was more sensitive than the thyroid weight, and PTU induced elevation of T/S was dependent on a potentiation of I^{131} concentrating mechanism of TSH, secreted in small amount in animals with the effective hypothalamic lesions.

Recently Bogdanove and Bogdanove (1957) maintained the goitrogen-treated rats at several levels of TSH secretion, and found that the "T/S responded to very slight thyrotrophic stimulation and achieved virtually which failed to affect the thyroid structure greatly, if at all." On account of this finding, they seem to deny the presence of the two separate factors. Previously we (Shibusawa *et al.*, 1956 a, b) reported that TRF could elevate the T/S ratio and the thyroid weight in animals with intact hypothalamo-hypophyseal connection as well as in animals with stalk section, and on account of this finding we postulated that TRF could be the single factor to stimulate the secretion of TSH. In the present series of experiments, it was confirmed that a small dose of TRF could significantly elevate T/S ratio, but not affect the thyroid structure.

And it seemed that, as claimed by Bogdanove and Bogdanove (1957), T/S ratio responds sharply to the slight thyrotrophic stimulation, while the thyroid weight responds only to medium or intensive thyrotrophic stimulation. And the thyrotrophic stimulation is considered to be mediated through a single factor—TRF.

From these results conclusions are drawn that the regulatory center of the thyroid will be found in the anterior hypothalamus, and that TRF extracted from this central tissue will be a single neurohumor which controls the secretion of TSH.

SUMMARY

(1) After placing the lesions in the anterior, middle and posterior hypothalamus of rats with the Krieg's stereotaxic machine, and the apparatus devised by the Brain Research Institute of Tokyo University, MTU or TRF was administered for 3 and 5 consecutive weeks, and the weight of pituitary and thyroid, I^{131} uptake of the thyroid, T/S ratio of I^{131} concentration and the histological changes in thyroid and hypophysis were investigated.

(2) When the lesions extended from the rostrum of the median eminence to the frontal area in the hypothalamus, both the thyroid and the pituitary weight decreased, the thyroidal function was slightly depressed, and the goitrogenic response to MTU was prevented. However, when TRF was given, the weight and function of the thyroid remarkably increased.

(3) When the lesions were placed in the middle and posterior hypothalamus, scarcely any depression was observed either morphologically or functionally. Depression was seen only when the lesions involved the median eminence. The goitrogenic effect of MTU was evidently observed. The administration of TRF produced both morphological and functional activation of the thyroid together with enlargement of the hypophysis.

(4) A minute (0.2 cc), small (0.5 cc), medium (1.0 cc) and large dose (2.0 cc) of TRF were given to both intact rats and those with effective lesions in the anterior hypothalamus. In the case of the minute and small dose a remarkable increase was not seen in the weight of the thyroid and the uptake of the thyroid, but the increase was observed only in the case of the medium and the large dose. By contrast, T/S I^{131} accumulation ratio was evidently increased by the minute dose, and the increase became still more pronounced when larger doses were given.

(5) It was discussed that the regulatory factor of TSH secretion would be TRF, a neurohumor elaborated in the anterior hypothalamus and excreted in urine.

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