Possible Reversion of Pituitary “Thyroidectomy Cells” into their Original Acidophils in Rats

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Synopsis

Fifteen days after thyroidectomy, the pituitary acidophils of rats were definitely deprived of their large granules, about 350 m\(\mu\) in diameter each, and in due succession, they displayed the changes characterized by the features of “thyroidectomy cells” (TX-cells). When these rats were given a single or three injections of thyroxine, 50 mg/rat, most of TX-cells might revert to their original acidophils in a short time of 6 hr. The cytomorphological course of possible reversion of the TX-cells into acidophils was clearly demonstrated on our electron micrographs. It was learned that thyroxine exhibited a strong ability to produce large granules, 350 m\(\mu\) in diameter each, in TX-cells which might return to their original acidophils through the reduction of cell size probably due to contraction of ER and loss of intracisternal granules and through the re-production of the large granules in the ground matrix. If the rats, 15 days after thyroidectomy, were given dexamethasone (DM), 0.2 mg/100 g/day, for 14 days, most of TX-cells tended to slightly recover from their modifications due to thyroidectomy. DM had ability to produce both the small (130-150 m\(\mu\) in diameter) and the large (350 m\(\mu\)) granules in some TX-cells, but the rest of them remained unchanged and preserved a number of dilated cisternae and intracisternal granules. DM was greatly inferior to thyroxine in the ability to produce the acidophils of the large granule type (somatotrophs), while the former was superior to the latter in the ability to produce the acidophils of the small granule type. Our previous views that some TX-cells might originate from the degranulated acidophils was confirmed validly by the present observation. In conclusion, all TX-cells have not always been considered as hyperactive thyrotrophs. Mechanism of ER morphodynamics associated with vesiculation in TX-cells has been discussed with special reference to the development and reversion of TX-cells.

It was claimed by Yoshimura et al. (1973) that most of the “thyroidectomy cells” (TX-cells) might genetically originate from the acidophils. Hyperfunctional thyrotrophs were not, however, regarded by them as being substantially identical with TX-cells. The purpose of this electron microscopic study was to present evidence for the possible reversion of TX-cells into their original or genetic acidophils in rats under the present experimental conditions. Such evidences as showing the cell conversion toward the reverse direction would afford a strong support to our theory that most of TX-cells may originate from degranulated acidophils.

Materials and Methods

The conditions of the animals used in this study accord with those used in our preceding work (Yoshimura et al., 1973). The technique with which the electron microscopy was carried out in this experiment was the same as that used in that work.

1. The animals given thyroxine

The animals were divided into two groups; one
consisted of male Wistar-Imamichi rats, 45 days of age, and they were surgically thyroparathyroidectomized. Five animals were picked up from among the animals whose thyroidectomy had been successfully done. A single dose of D. L. thyroxine, 50 mg/rat, was given to each one of them on the 15th postoperative day, and the animals were sacrificed 6 hr later. Another group of successfully thyroidectomized rats were given thyroxine, 15 days after operation, 50 mg/rat day, for 3 days. These animals were sacrificed 6 hr after the final injection. Five animals were picked up as the control from among them that lived 15 days after thyroidectomy without injection.

2. Another group given dexamethasone

Five thyroparathyroidectomized rats were injected subcutaneously with dexamethasone (DM), 0.2 mg/100 g/day, for 14 days from the 15th postoperative day. The non-injected rats were sacrificed on the 30th day after thyroidectomy as the control.

Results

Reversion of the “thyroidectomy cell” into their original acidophils by thyroxine administration

Both single and three injections of thyroxine had a strong ability to dismiss the TX-cells accumulating after thyroidectomy. The dosage of thyroxine, 50 mg/rat, seemed appropriate for their recovery from marked changes which were incurred during their deficiency in thyroxine. As a matter of fact, it was proved electron microscopically that large granules (about 350 μ in diameter), which had been depleted on the 15th day after thyroidectomy, reappeared in clusters in somatotrophs 6 hr

Fig. 1. “Thyroidectomy cells” (TX-cells) from the pituitary of a thyroidectomized rat given a single injection of thyroxine, 50 mg/rat. The TX-cells which were numerosely emergent before injection have decreased in number rapidly 6 hr after injection. Most of the enlarged TX-cells are shrunken and the size of their cisternal ER is reduced as is observed in the TX-1 cell. In the left TX-2, shrinkage of cisternae is pronounced and their intracisternal granules are lost from the cells. Note the encircled stepwise distribution of Golgi vacuoles (Golgi-rings) in both cells. × 10000
after a single injection of thyroxine.

In this study, the course of reversion following thyroxine administration of the TX-cells into the acidophils was observed electron microscopically. The first response seen in the TX-cells was a reduction in luminal size of the endoplasmic reticula (ER) which had been excessively dilated. The reduction of the size varied considerably from cell to cell. In proportion to the advancement of the reduction, the ER became gradually elongated as a narrow canalliculi (TX-1 in Fig. 1 and TX-1 in Fig. 2). The size of cisternal lumen was gradually reduced and their opposite limiting membranes came into contact. The narrowest canalliculi were arranged in parallel (TX-2 in Fig. 1 and TX-3 in Fig. 2). Probably due to sinking of inner pressure of cells which might be associated with the reduced luminal size of the ER, the enlarged polygonal TX-cells soon became dimensionally reduced and changed their shape to ellipsoid. The second response was that the intracisternal granules disappeared completely from TX-cells (Figs. 1 and 2). This finding clearly indicates that these granules occur specifically during their deficiency in thyroxine. The third response was that the small granules ranging from 130 to 150 nm in diameter were kept for a long term without being discharged from their ground matrix.

Fig. 2. The TX-cells from a thyroidectomized rat given three injections of thyroxine. The reversion of the TX-cells is promoted to the same degree as that caused by a single injection of thyroxine. The TX-1 cell indicates the first step of reverted TX-cells. The TX-3 cell has been provided with the fine structural features common to an immature acidophil. This TX-3 cell is ellipsoidal in shape and possesses narrow canalicular ER with a rough surface. At the same time, the occurrence of large mitochondria and lysosomes may help to indicate that their cells belong to the acidophils. Go, Golgi-rings; Ls, Lysosomes. × 10000
Fig. 3. An immature acidophil (rA) which may have just reverted from a TX-cell of a thyroidectomized rat given a single injection of thyroxine. This cell (iA) is characterized by dense arrangement of lamellar ER and the prominent Golgi-ring (Go). The cytoplasm is scattered with large granules 200 to 300 mμ in diameter. The large granules are intermingled with the small granules about 130 mμ in diameter which have been present in the former TX-cells in the pretreatment stage. × 8000

even though the TX-cells were deformed or reduced in size. When an ellipsoid cell, in which lamellar ER are distributed as shown in Figure 3, is compared in detail with a cell (TX-3) shown at the right side of Figure 2, it is conceivable that the former may belong to a cell involved in the next stage of the latter. There was evidence to show that the small granules grew to the large ones, but some of them remain unchanged in size (Fig. 3). The large granules tended to increase in number along with the growth in population of the cells which were recovering from the TX-cells. The concentric arrangement of the lamellar ER was one of the characteristic features of the cells which developed immediately after the completion of reversion from TX-cells. In these cells enlarged Golgi vacuoles continued to form encircled stepping-stones (Golgi-ring) (right cell in Fig. 2). Eventually large granules, 250-350 mμ in diameter, accumulated in the cytoplasm of a certain cells recovered from TX-cells similar in cytological feature to somatotrophs (Figs. 4 and 5). A series of photomicrographs may account for the reversion of TX-cells into their original or genetic acidophils packed with the large granules which are regarded as GH granules (Fig. 5).

Reversion of **“thyroidectomy cells” into their original acidophils by administration of dexamethasone (DM)**

In thyroidectomized control rats, 15 days
after thyroidectomy, most of the acidophils were conspicuously degranulated. Some of the acidophils which were completely deprived of their large granules were quite different in granularity and in shape from the typical acidophils. But some acidophils kept the small granules for a long time instead of the large ones. Thirty days after thyroidectomy, all the acidophils of the large granule type were lost, while the acidophils of the small granule type discharged their remaining small granules and resulted in the transformation into agranular acidophils as observed for the first time by Harumiya (1972), one of the present authors.

In the thyroidectomized rats given DM for 14 days from the 15th postoperative day, there was a slight tendency to reduction in the size of enlarged TX-cells. In ability to decrease the number of the TX-cells DM was inferior to thyroxine. The population of TX-cells was as large as that in the control thyroidectomized rats without injection. Detailed observation showed that the signet-ring cells having large intracellular cavities were still remaining (Fig. 6). However, the TX-cells had a general tendency to contain the atrophic ER. Despite the presence of the atrophic ER, the intracisternal granules did not disappear completely by DM injection (Figs. 6 and 7). It was confirmed by our present electron microscopy that the cisternal ER in the TX-cells were greatly contracted by DM. When contraction of the ER was significant, the cells had no internal fine structure which was characteristic
Fig. 7. The TX-cells in a rat treated with DM under the same condition as that indicated in Figure 6. Some of the TX-cells remain unchanged, but they undergo a considerable reduction in size. Irrespective of the reduced size of the cisternae this cell still keeps intracisternal granules in its cell-body Go, Golgi area. × 10000

to the TX-cells (Fig. 8). The reverted acidophils (rA) shown in Figures 9 and 10 were provided with the appearance of immature acidophils developed immediately after the complete reversion from the TX-cells. In these immature acidophils the elongated slender ER changed into the narrow canaliculi which were distributed in parallel or irregularly. However, generally speaking, the contraction of the ER by thyroxine was more prominent than the induced by DM. The formation of lamellar ER seems to follow an excessive contraction of cisternae. This is indicative of one step of reversion into the acidophils. The prominent Golgi-ring which seemed to play an important role in the subsequent granula-

Fig. 5. A mass of immature and mature acidophils, both of which have reappeared in the pituitary after three injections of thyroxine to the thyroidectomized rat. The ability of thyroxine to make the TX-cells come back to the acidophils is quite remarkable. The parallel distribution of lamellar ER is obviously observed in immature acidophils (rA) which have just reverted from the TX-cells. S, somatotroph; G, gonadotroph. × 6000
Fig. 6. A signet-ring cell in a thyroidectomized rat given an injection of DM, 0.2 mg/100 g/day, for 14 days. The particular TX-cells which, in their structure, resemble the signet-ring-typed castration cells are often detectable. This does not mean a full recovery from the response to thyroidectomy. G, gonadotroph. × 12000
Fig. 8. Profoundly shrunken TX-cells in a rat treated with DM. Although cisternae are conspicuously atrophic and their lumens are narrow, the intracisternal granules are as they were. × 9000

ination was often observed in the cells which were recovered from the TX-cells as illustrated in Figure 9 (Go). Thus, the TX-cells might return to their original acidophils by DM administration. These kinds of acidophils can be classified into three types. First (A-1), a cell having only the small granules of about 130 μ in diameter (Fig. 11); second of (A-2), a cell granulated with both the small and large granules of about 350 μ in diameter (Fig. 12); third (A-3), a cell containing singly the numerous large ones of 300–350 μ in diameter (Fig. 13). Acidophils of the third type are generally immature in cytological characteristics revealing infrequent maturation into somatotrophs in which lamellar ER are abundantly distributed (Fig. 13). In the initial phase of granulation, the immature acidophils which had reverted from the TX-cells occasionally produced cored vesicles in the Golgi area (the left cell in Fig. 10). When these cells were polygonal in shape, they resembled the fine structures of “corticotrophs” denominated by Kurosumi and Kobayashi (1966). In Participation with Golgi-ring, both the small and large granules began to accumu-

Fig. 9 and Fig. 10. Possible immature acidophils (rA) which may appear immediately after the complete reversion from the TX-cells in a rat treated with DM. These cells keep their shape polygonal and contain a small number of slender or atrophic canicular ER (arrow in Fig. 9). A number of cored vesicles (arrow in Fig. 10) which may be associated with the initial granulation are located near the Golgi area (Go), when no evident accumulation of the granules takes place. × 8000, 10000
late gradually after the reversion of the TX-cells into acidophils of immature type. The cells which had reverted from TX-cells were developed again into the acidophils of small and large granule types. But in the ability to produce the large granules DM was inferior to thyroxine. Following the injections of DM, TX-cells were apt to return to their original state, i.e. acidophils of the small granules type, with some exception of returning to another original state, i.e., acidophils of the large granule type (somatotrophs).

Discussion

It was generally accepted that some of acidophils which had been deprived of the α-granules following thyroidectomy were regranulated by the injection of corticosteroids with a slight recovery of pituitary GH content from its remarkable reduction (Meyer and Evans, 1964). Yokoyama (1972), one of the present authors, calculated light microscopically the number of various types of cells following thyroidectomy, and pointed out a significant decrease in the number of acidophils and their almost complete disappearance in the chronic phase. Carrying out the bio- and radioimmunoassay, he reported that pituitary GH content diminished whereas ACTH concentration remained at its constant level. When thyroidectomized rats were injected with DM for 14 days from the 15th postoperative day, Yokoyama (1972) observed that acidophils were moderately recovered from their remarkable lowering along with a rise of pituitary GH content to a normal level. His observation that pituitary ACTH content increases beyond normal following DM administration may give orientation to our present electron microscopical findings. On the other hand, when thyroidectomized rats were given thyroxine, acidophils which had disappeared completely following thyroidectomy emerged again numerously along with the recovery of pituitary GH content from its remarkable reduction (Koneff et al., 1949: Contoupoulos et al., 1958: Suzuki and Shibasaki, 1970: Ieiri, 1971).

Ishikawa et al. (1972) separated, by the ultracentrifuging method, large (350 μm in diameter) and small granules (130–150 μm in diameter) from the pellets of the isolated acidophils from the fresh rat pituitaries. Through the examination of hormonal activities of the small and large granules, these authors found GH activity predominant in the large granules while ACTH activity only in the small ones, although a great part of ACTH activity was easily lost during isolation. Accepting the statement made by Ishikawa et al. that the large granules are storage machineries of GH and the small ones are those of ACTH, it would be reasonable to claim that our present electron microscopic findings on the formation of either size of granules in the TX-cells which have returned to their original cells following thyroxine and DM administration are compatible with the changes of pituitary GH and ACTH contents determined by Yokoyama under the conditions similar to those of the present experiment. Since the entire course of reversion of TX-cells into their original somatotrophs was corroborated by a series of our electron micrographs, our theory that many TX-cells may originate from the degranulated somatotrophs seems to be fully justified.

Figs. 11, 12 and 13. Three types of mature acidophils which may have reverted from the TX-cells in a rat treated with DM. Granulation is advanced in these acidophils. A-1 shows the first type of the reverted acidophil granulated only with the small granules about 130 μm in diameter; A-2, the second type of acidophil granulated with both small and large ones 200 to 300 μm in diameter; A-3, the third type of acidophil granulated only with the large ones (somatotroph). × 8000, 8000, 10000
thyroidectomized rats, the acidophils of the large granule type (somatotrophs) re-appeared numerously, but the acidophils of the small granule type did not. On the other hand, DM displayed its ability to revert TX-cells into the two types of acidophils, one with large granules and the other with small granules. However, the acidophils of the large granule type (somatotrophs) occurred sporadically. Our electron micrographs showed that the ratio of the number of acidophils of the large granule type to that of the small granule type was roughly estimated to be 1:3. This ratio leads us to consider that the frequent occurrence of the acidophils of the small granule type is closely related to the high content of pituitary ACTH. It was impossible to determine clearly whether the “corticotrophs,” denominated by Kurosumi and Kobayashi (1966), which were hypergranulated in response to DM administration was really responsible for ACTH production, or merely belonged to a phase of the immature acidophils which had reverted from the TX-cells, so far as the present observation was concerned, but we are rather inclined to favor the latter.

Some of TX-cells were stained weakly with PAS whereas their intracellular T-granules were intensively stainable with adehyde thionine (Yoshimura et al., 1973). These authors speculated the existence of two groups of original cells of TX-cells, i.e., one consisted of a large number of cells arising from acidophils and the other consisted of a small number of cells from basophilic intermediates. Nevertheless, it is still not determined whether the TX-cells are really basophilic or acidophilic by nature. If we admit the assumption that all the TX-cells are fundamentally basophilic in character, we cannot help accepting the interpretation that acidophils substantially are converted into basophils following thyroidectomy. However, we do not believe that acidophils may be functionally converted into basophils. Even though the TX-cells are provided with the morphological characteristics as seen in the basophils, the acidophils essentially may never, in our opinion, be switched into basophils in their function by modifying their activity in production of simple protein hormone to that of glucoprotein hormone. It seems justifiable, in reference to the foregoing bioassay data of Yokoyama (1972), that most of the TX-cells may produce the same kinds of hormones as produced in the acidophils which are free of the large granules.

One of the main objectives of this investigation was to elucidate the significance of vesiculation which is a most fundamental phenomenon of the TX-cells. It was already assumed in the preceding paper that deficiency in thyroxine gave rise to vesiculation at random in all the adenohypophyseal cells. Some information was obtained in this study concerning the relationship between morphokinetics of ER and the cell function. It was reasonable to assume that a certain level of circulating thyroxine might regulate the contractility of ER within the range of normal value. When deficiency in thyroxine persisted, ER in TX-cells were dilated extraordinarily, and in the event of high thyroxine level they were contracted and their lumen became narrow or closed, and eventually organized to the parallel arrangement of lamellar ER. Vesiculation, therefore, may be a sort of regressive biological response observed upon the majority of the hypophyseal cells, and is rather indicative of dysfunction of the cells. Also corticosteroides given exogenously to thyroxine-deficient rats inhibited the TX-cells from vesiculation of their ER. However, whether this inhibition was mediated by the endogenous overshoot secretion of ACTH or by direct corticosteroid action on the hypophyseal cells was not fully clarified. Baker and Yu (1971) demonstrated immunologically the absence of hormonal activity of the TX-cells. Since the TX-cells were all negatively reacted to the anti-TSH, -GH, -prolactin, -LH and -ACTH, Baker and Yu regarded them as inactive for the hormone production. This served as a guide to our original opinion
as to the functional significance of vesiculation of the TX-cells.

References


