Goiter Formation Following Prostaglandin Administration in Rats

A. Lupulescu, MD, MS, PhD

Prostaglandins (PGE_1 and PGE_2) induced a hyperplastic microfollicular goiter with a high radioiodine (^{131}I) thyroid uptake, increased endocytosis, a heavy autoradiographic (^{125}I) reaction, and a moderate increase of thyroid hormones (T_4, T_3), thyroxine-binding globulin (TGB), and thyrotropin (TSH) concentrations in adult rats. Ultrastructurally, both prostaglandins (E_1 and E_2) markedly stimulated the thyroid cell activity and increased the number of pseudopodia, the size of colloid and dense granule populations, and the number of polysomes. Conversely, a hypofunction of thyroid glands with low radioiodine (^{131}I) thyroid uptake, a decreased autoradiographic (^{125}I) reaction, and a moderate decrease in T_4, T_3, TGB, and TSH concentrations were observed following prostaglandin $F_{2\alpha}$. Ultrastructurally, a decrease in size of the colloid and dense granule population and the number of degenerative mitochondria occurred in follicular cells. An intense hyperplasia of parafollicular (C) cells, with abundant population of characteristic dense granules, could be seen in $PGF_{2\alpha}$- treated rats. A marked decrease of radioiodine (^{131}I) uptake, endocytosis, and autoradiographic (^{125}I) reaction, and a sharp decline in T_4, T_3, and TBG were observed in hypophysectomized and chronically prostaglandin-treated rats. Light and electron microscopy revealed signs of an advanced thyroid hypofunction with flat cuboidal cells, reduced microvilli, scarce endoplasmic reticulum, and few dense droplets. The present findings demonstrate that the chronic administration of prostaglandins exerts significant effects on thyroid gland and goiter formation (goitrogenesis), radioiodine metabolism, and hormone synthesis, and that these effects are mediated by TSH secretion. (Am J Pathol 85:21–36, 1976)

IT HAS BEEN DEMONSTRATED that the release of radioiodine (^{131}I), endocytosis, adenyl cyclase activity, ^{14}C-glucose oxidation, and ^{32}P incorporation into phospholipids were differently affected by prostaglandins.[1-3] Each prostaglandin increased adenyl cyclase activity and colloid droplet formation (endocytosis) in sheep thyroid slices, but only PGE_1, PGE_2, and PGF_{1B} enhanced glucose oxidation. It was also shown that, in sheep thyroid slices, all prostaglandins tested (E_1, E_2, $F_{1\alpha}$, F_{1B}) increased endocytosis, a result similar to that induced by thyrotropic-stimulating hormone (TSH). Since several effects induced by prostaglandins on thyroid glands are similar to those induced by TSH (such as increased colloid droplet formation or endocytosis, augmented ^{131}I release from the thyroid, increased organification of iodide, augmented glucose oxidation, and increased cyclic AMP in thyroid slices), it was postulated that these effects

From the Department of Dermatology, Wayne State University School of Medicine, Detroit, Michigan.

Accepted for publication May 14, 1976.

Address reprint requests to Dr. A. Lupulescu, 323 Medical Research Building, 550 East Canfield Avenue, Detroit, MI 48201.

could be mediated via the anterior pituitary. However, there is no direct evidence whether or not such effects are mediated by TSH secretion from the adenohypophysis. To ascertain if the cellular and metabolic effects of prostaglandins (PGE_1, PGE_2, $PGF_{2\alpha}$) on thyroid glands are direct effects or are mediated by the pituitary TSH secretion, comparative investigations were performed in intact and hypophysectomized rats. Hence, the results are divergent depending on the species and effectiveness of prostaglandins on various parameters of thyroid glands; the exact mechanism of the action of prostaglandins on thyroid and goiter formation still remains to be determined.

In the present study, we investigate the long-term effects of prostaglandins (PGE_1, PGE_2, and $PGE_{2\alpha}$) on thyroid and goiter induction, radioiodine (^{131}I) uptake, intracellular radioiodine (^{125}I) distribution, and thyroid hormone (T_4, T_3), thyroxine-binding globulin (TBG), and TSH concentrations in intact and hypophysectomized rats.

Materials and Methods

Adult male rats (Sprague-Dawley strain) weighing between 272 to 300 g and hypophysectomized male rats (same strain) weighing between 220 to 250 g were used in these experiments; each group was composed of 20 rats that were treated in one of the following ways for 2 months: a) the first group received, intramuscularly, 150 μg prostaglandin E_1 (PGE_1), every other day; b) the second group received, intramuscularly, 150 μg prostaglandin E_2 (PGE_2) every other day; c) the third group was injected intramuscularly with 150 μg prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$), also every other day; and d) the fourth group served as controls and received, intramuscularly, the same volume of diluent every other day. Hypophysectomized (Hypox) rats, purchased from Charles River Breeding Laboratories, were also of the Sprague-Dawley strain; hypophysectomy was performed by the parapharyngeal approach. Completeness of hypophysectomy was ascertained at autopsy by examining the pituitary region with a 10 × dissecting microscope. These rats were divided as follows: e) the fifth group received, intramuscularly, 150 μg of PGE_1 (Hypox + PGE_1) every other day; f) the sixth group received 150 μg of PGE_2 (Hypox + PGE_2) intramuscularly every other day; g) the seventh group received $PGF_{2\alpha}$ (Hypox + $PGF_{2\alpha}$) also every other day and h) the eighth group (Hypox only) received the same volume of diluent every other day. All prostaglandins were diluted in a mixture of absolute ethanol (1 part) and 0.02% sodium carbonate (9 parts). During the experiments, both intact and hypophysectomized rats were fed Purina laboratory chow with an approximate iodine content of 3 μg ^{127}I g and kept at a constant room temperature. All were studied over the same time period and sacrificed 2 months after treatment with prostaglandins by ether anesthesia.

For the study of radioiodine (^{131}I) in thyroid, 8 rats from intact and prostaglandin-treated groups (I–IV) as well as from each hypophysectomized and prostaglandin-treated groups (V–VIII) were injected intraperitoneally with 10 μCi of ^{131}I, 1 hour prior sacrifice; another 8 rats from each experimental group received the same amount of radioiodine of ^{131}I, 5 hours prior sacrifice and following the last injection of prostaglandins. At that time, thyroid glands were removed for determination of ^{131}I uptake. Scintillation countings were determined by using a liquid scintillation counter (Nuclear Chicago Corporation) type 724 and 725 with a 95% efficiency after the specimens were transferred in a scintillation fluid. Results were expressed as counts per minute and per gram wet thyroid tissue; mean ± standard error was calculated.

Levels of thyroid hormones (T_4, T_3), TBG, and thyrotropin (TSH) in rat sera were estimated as follows: serum thyroxine (T_4) was determined according to the method based on the thyroxine-binding properties for TBG and using radiothyroxine; the results were expressed in micrograms per 100 milliliters serum.[4]

The determination of thyroxine-binding globulin (TBG) was carried out by a procedure employing dextran-coated charcoal.[5] The results were expressed also in micrograms per 100 milliliters. Total triiodothyronine (T_3) and TSH were estimated by radioimmunoassays (RIA).

Total T_3 was determined by a modified radioimmunoassay using T_3 antibodies, sheep antirabbit γ-globulin, and T_3 labeled with ^{125}I (Abbott Company); a gamma-well counter was also used for measurements. Results were expressed in nanograms per 100 milliliters.[6] TSH was assayed using ^{125}I and a Beckman γ-radiation counter. A homologous radioimmunoassay using a highly purified rat TSH (35 IU/mg) (LH contamination = 0.2 and FSH less than 0.1) labeled with ^{125}I and antibody to it prepared in rabbits and provided by the Rat Pituitary Hormone Distribution Program, (NIAMDD) was employed. Radioactivity of the precipitates were determined with a Beckman γ-radiation counter. Results were expressed in microunits per milliliter.[7]

For light and electron microscopic studies, thyroid glands were diced into small specimens and fixed in 2.5% phosphate-buffered glutaraldehyde for 2 hours; some of the specimens were embedded in Paraplast and stained with hematoxylin and eosin; others were postfixed in 1% phosphate-buffered osmium tetroxide,[8] dehydrated in ascending series of ethanol and embedded in a mixture of Epon-Araldite. Thin sections (600 to 900 Å) were cut with a LKB-Ultrotome, mounted on grids, stained with uranyl acetate and lead citrate. They were examined under a Hitachi HS-8 electron microscope.

For light and electron microscopic autoradiography, 4 rats from each intact and prostaglandin-treated group (I–IV) and 4 rats from each hypophysectomized and prostaglandin-treated group (V–VIII) were injected intraperitoneally with 250 μCi radioiodine (^{125}I), 1 and 5 hours before sacrifice and following the hormone injection. Thyroid glands were removed, fixed for 2 hours in 2.5% phosphate-buffered glutaraldehyde, then postfixed in 1% phosphate osmium tetroxide and routinely embedded in the same mixture of Epon-Araldite. Thick plastic sections (1 μ) were covered with Ilford K5 nuclear emulsion and exposed for 4 to 5 days for light microscope autoradiography, then developed in Kodak developer D19, fixed, washed and stained with 1% toluidine blue. For electron microscope autoradiography, thin sections (600 to 900 Å) were covered with Ilford L4 nuclear emulsion diluted 1:2 using a wire loop procedure,[9] exposed for 4 weeks in refrigerated (4 C) boxes, developed in Microdol-X for 5 minutes, fixed, and washed, then examined under an electron microscope.

Cell height was measured with a Zeiss micrometer (× 10) ocular and (× 45) objective. Ten cells in ten fields of each slide and ten slides from thyroids of each experimental group—approximately 1000 cells—were measured. Colloid droplets were counted in paraffin and plastic sections in 20 follicles per each experimental group using the oil immersion objective of a light microscope. All results were expressed as mean ± standard error; *P* values were also calculated.

Results

Radioiodine (^{131}I) Studies

The thyroid uptake of ^{131}I at 1 hour and 5 hours shows a marked increase of ^{131}I in the thyroid glands following PGE_1 and PGE_2 treatment. PGE_1 is more active; a decrease of ^{131}I uptake occurs in the rat thyroid glands following $PGF_{2\alpha}$ administration as compared to the controls (Text-

figure 1A and B). Conversely, a marked decrease of ^{131}I uptake occurs in all hypophysectomized and prostaglandin-treated rats (Text-figure 2A and B), compared to intact and prostaglandin-treated rats ($P < 0.001$).

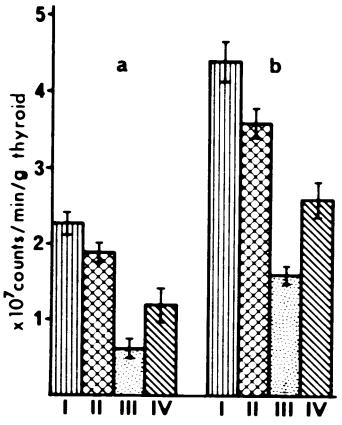
Thyroid Hormone Determinations

Determination of thyroid hormones reveals moderate disturbances in their biosynthesis following prostaglandin administration to rats (Table 1). Thus, serum T_4 values are slightly increased at 1 hour and moderately at 5 hours in PGE_1- and PGE_2-treated rats; they decrease in $PGF_{2\alpha}$-treated rats. These values are significant only at 5 hours ($P < 0.02$). Thyroxine-binding globulin levels moderately are increased by PGE_1 and PGE_2 and lowered after $PGF_{2\alpha}$ as compared to those in control rats also at 5 hours.

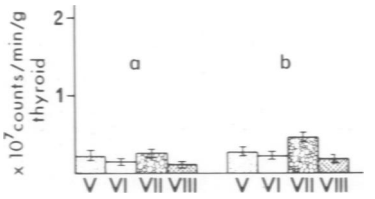
Measurements of total T_3 and TSH in the sera of prostaglandin-treated rats show a moderate increase in their circulating values in PGE_1 and PGE_2-treated rats ($P < 0.02$) and, on the contrary, a slight decrease in $PGF_{2\alpha}$-treated rats at 5 hours as compared to the control groups. Marked decreases of T_4, TBG, and T_3 values occur in sera of hypophysectomized and prostaglandin-treated rats (Table 2). These values are highly significant as compared to intact and prostaglandin-treated rats ($P < 0.001$); but they are not statistically significant between the hypophysectomized groups themselves; no detectable amount of TSH can be found.

Thyroid Weight Measurements

Measurements of thyroid weight in intact and prostaglandin-treated rats show a significant increase following PGE_1 and PGE_2 administration as compared to control rat thyroids; a moderate decrease in thyroid weight occurs after $PGF_{2\alpha}$ administration. Cellular height and colloid



TEXT-FIGURE 1—Radioiodine (^{131}I) thyroid uptake at 1 hour (**a**) and 5 hours (**b**) in intact and prostaglandin-treated rats. (PGE_1-treated rats, *I*; PGE_2-treated rats, *II*; $PGF_{2\alpha}$-treated rats, *III*; Controls, *IV*. Vertical bars represent standard error.)



TEXT-FIGURE 2—Radioiodine (^{131}I) thyroid uptake at 1 hour (**a**) and 5 hours (**b**) in hypophysectomized (Hypox) and prostaglandin-treated rats. (Hypox + PGE_1, *V*; Hypox + PGE_2, *VI*; Hypox + $PGF_{2\alpha}$, *VII*; Hypox only, *VIII*. Vertical bars represent standard error)

droplet formation are markedly increased after PGE_1 and PGE_2 administration and decreased in $PGF_{2\alpha}$-treated rats (Table 3) ($P < 0.001$). Thyroid weight, cellular height, and the number of colloid droplets are sharply decreased in hypophysectomized and prostaglandin-treated rats as compared to intact and prostaglandin-treated rats (Table 4) ($P < 0.001$). Also, these values are significant ($P < 0.02$) when compared to hypophysectomized only.

Light Microscopy

Light microscopic observations reveal marked structural changes in prostaglandin-treated rats. Thus, a hyperplastic parenchymatous goiter with an intense microfollicular hyperplasia and reduced amount of colloid is observed in PGE_1-treated rats (Figure 1). Small follicles (50 to 75 μ in diameter) are predominant with follicular epithelium composed of high columnar cells as compared to cubic flat cells in the controls. Several intracellular colloid droplets are visible at the apical border (Figure 1); follicular colloid is pale, and interstitial hyperplasia is also observed. On the contrary, large follicles filled with dense colloid and low cuboidal epithelium occur in $PGF_{2\alpha}$-treated rats; a marked hyperplasia of parafollicular or C-cells occurs (see Figure 5). An inhibition of thyroid activity with large follicles (250 μ in diameter) composed of cuboidal cells and homogenous colloid is observed in hypophysectomized and prostaglandin-treated rats; sometimes desquamated cells are seen (Figure 2).

Electron Microscopy

Electron microscopic observations of the control thyroid glands reveal a regular follicular pattern with microvilli at the apical border protruding into a dense colloid. Endoplasmic reticulum is well developed, and dense droplets scattered throughout the cytoplasm are visible in control thyroid cells (Figure 3). A marked cellular activity with hyperplastic columnar cells exhibiting a large population of small dense granules concentrated at the apical border is visible in PGE_1-treated rats. In the middle and lower areas of thyroid cells, a second type of numerous large dense granules,

Table 1—Effect of Prostaglandins (PGE_1, PGE_2, and $PGF_{2\alpha}$) on Serum Thyroxine (T_4), Thyroxine-Binding Globulin (TBG), Triiodothyronine (T_3), and TSH in Intact Rats (Mean ± SE)

Experimental groups	Thyroxine (T_4) (μg/100 ml)		TBG (μg/100 ml)		Triiodothyronine (T_3) (ng/100 ml)		TSH (μU/ml)	
	1 hr	5 hrs	1 hr	5 hrs	1 hr	5 hrs	1 hr	5 hrs
I. PGE_1-treated (N = 16)	5.96 ± 0.38	9.94 ± 0.65*	8.56 ± 0.49	14.0 ± 0.49*	44 ± 3.02	69 ± 4.09*	33 ± 3.1	49 ± 3.8*
II. PGE_2-treated (N = 16)	5.23 ± 0.26	9.41 ± 0.81	8.38 ± 0.75	13.3 ± 0.59	41 ± 3.80	64 ± 4.04	31 ± 2.9	46 ± 4.0
III. $PGF_{2\alpha}$-treated (N = 16)	3.01 ± 0.20	4.25 ± 0.40	6.71 ± 0.70	6.9 ± 0.68	29 ± 2.08	34 ± 3.01	10 ± 1.6	9.8 ± 1.4
IV. Controls (N = 16)	4.82 ± 0.29	6.18 ± 0.70	7.38 ± 0.60	9.01 ± 0.70	36 ± 2.45	46 ± 4.01	26 ± 2.2	27.2 ± 2.3

* $P < 0.02$ as compared to controls.

Table 2—Effect of Prostaglandins (PGE_1, PGE_2, and $PGF_{2\alpha}$) on Serum Thyroxine (T_4), Thyroxine-Binding Globulin (TBG), Triiodothyronine (T_3), and TSH in Hypophysectomized Rats (Mean ± SE)

Experimental groups	Thyroxine (T_4) (μg/100 ml)		TBG (μg/100 ml)		Triiodothyronine (T_3) (ng/100 ml)		TSH (μU/ml)	
	1 hr	5 hrs	1 hr	5 hrs	1 hr	5 hrs	1 hr	5 hrs
V. Hypox + PGE_1 (N = 16)	0.88 ± 0.05	0.92 ± 0.04*	2.0 ± 0.02	3.0 ± 0.01*	13.0 ± 0.2	14.0 ± 0.2*	ND	ND
VI. Hypox + PGE_2 (N = 16)	0.85 ± 0.05	0.87 ± 0.04	3.0 ± 0.02	3.0 ± 0.01	13.0 ± 0.01	13.0 ± 0.1	ND	ND
VII. Hypox + $PGF_{2\alpha}$ (N = 16)	0.81 ± 0.04	0.79 ± 0.04	2.0 ± 0.01	3.0 ± 0.01	12.0 ± 0.01	12.0 ± 0.2	ND	ND
VIII. Hypox (N = 16)	0.72 ± 0.04	0.75 ± 0.04	2.0 ± 0.01	2.0 ± 0.01	13.0 ± 0.01	13.0 ± 0.1	ND	ND

ND = not detectable.

* $P < 0.001$ as compared to intact and prostaglandin-treated rats (see Table 1), but not statistically significant when compared to VIII (Hypox only).

Table 3—Effect of Prostaglandins (PGE_1, PGE_2, and $PGF_{2\alpha}$) on Thyroid Weight, Cell Height, and Colloid Droplet Formation in Intact Rats (Mean ± SE)

Experimental groups	Thyroid weight (mg)	Thyroid cell height (μ)	No. of colloid droplets
I. PGE_1-treated (N = 16)	38 ± 2.5	29 ± 2.0*	856 ± 21*
II. PGE_2-treated (N = 16)	37 ± 2.7	27 ± 2.1*	824 ± 22*
III. $PGF_{2\alpha}$-treated (N = 16)	15 ± 1.4	11 ± 0.6	5 ± 0.7
IV. Controls (N = 16)	20 ± 1.8	14 ± 1.5	8 ± 1.0

* $P < 0.001$ as compared to controls.

sometimes agglomerated, occurs following PGE_1 treatment (Figure 4). Large pseudopodia protruding deeply in the follicular colloid and containing cytoplasmic organelles and extensive endoplasmic reticulum are visible in PGE_1-treated rats. Occurrence of dark and light cells is frequently observed following PGE_1 and PGE_2 treatment.

Conversely, ultrastructural findings of hypofunction are visible in thyroids of $PGF_{2\alpha}$-treated rats. Thus, flat cubic cells with reduced endoplasmic reticulum, degenerative mitochondria, and decreased number of small dense granules at the apical border are predominant. An interesting hyperplasia of parafollicular (C) cells occurs following $PGF_{2\alpha}$ administration. Hyperplasia and hypertrophy of parafollicular cells can be seen between follicles; endoplasmic reticulum, a large population of characteristic dense granules, hypertrophic Golgi complex, and polysomes are seen in $PGF_{2\alpha}$-treated rats (Figure 5). In the thyroid glands of hypophysectomized and prostaglandin-treated rats, ultrastructural features of evident hypofunction with flat follicular and few dense droplets are observed. No

Table 4—Effect of Prostaglandins (PGE_1, PGE_2, and $PGF_{2\alpha}$) on Thyroid Weight, Cell Height, and Colloid Droplet Formation in Hypophysectomized Rats (Mean ± SE)

Experimental groups	Thyroid weight (mg)	Thyroid cell height (μ)	No. of colloid droplets
V. Hypox + PGE_1 (N = 16)	10.5 ± 0.18	12 ± 1.5*	20 ± 2*
VI. Hypox + PGE_2 (N = 16)	9.5 ± 0.15	11 ± 1.0	15 ± 2*
VII. Hypox + $PGF_{2\alpha}$ (N = 16)	8.5 ± 0.15	9 ± 0.5	5 ± 0.1
VIII. Hypox (N = 16)	8.0 ± 0.12	8.5 ± 0.5	2 ± 0.1

* $P < 0.001$ as compared to intact and prostaglandin-treated rats (see Table 3); also these values are significant ($P < 0.02$) when they are compared to Hypox (VIII).

pseudopodia or colloid droplets could be seen, but large areas of sclerosis are frequently seen (Figure 6). Electron microscopic observations on pituitary glands from PGE_1 and PGE_2-treated rats reveal a predominance of large thyrotropic cells with several granules and hypertrophic Golgi apparatus, whereas a reduction and degranulation of secretory granules occur in $PGF_{2\alpha}$-treated rats.

Autoradiography

Electron and light microscopic autoradiograms reveal in the control thyroid glands that the great amount of ^{125}I is incorporated over follicular colloid, occurring as filaments (developed grains) located mostly over microvilli and the apical cell border (Figure 7); only a few developed grains are visible within cytoplasmic structures. A heavy autoradiographic reaction with many agglomerated or clumped filaments occurs at 1 hour in PGE_1-treated rats, mainly over follicular colloid and the apical cell border (Figure 8). A characteristic autoradiographic reaction over colloid droplets can also be seen in both PGE_1- and PGE_2-treated rats (Figure 10). A marked decrease of autoradiographic reaction occurs in the thyroid of hypophysectomized and prostaglandin-treated rats; only few developed grains are visible over follicular colloid and apical cell border (Figure 11).

Discussion

The current investigations revealed that a long-term administration of PGE_1 and PGE_2 markedly stimulated the thyroid gland, inducing a hyperplastic microfollicular goiter with intense cellular activity, high thyroid ^{131}I uptake, and moderate increase of serum of T_4, T_3, TBG, and TSH, whereas $PGF_{2\alpha}$ induced a thyroid hypofunction with low ^{131}I uptake, decrease serum T_4, TBG, T_3, and TSH.

PGE_1 and PGE_2 also markedly increased ^{125}I incorporation as several complex filamentous structures mostly over the follicular colloid and the apical cell border, at 1 hour, and increased significantly their intracellular distribution at 5 hours. This confirmed previous investigations that peripheral colloid and the apical zone are the active site of thyroglobulin iodination in control thyroid glands.[10] It is also interesting that PGE_1 and PGE_2 increased the levels of thyroxine-binding globulin (TBG) and $PGF_{2\alpha}$ lowered them, a finding which has not been previously reported. However, it is known that human TBG can fluctuate under the influence of other hormones, such as estrogens, androgens, or anabolic steroids; large doses of glucocorticoids; or thyroid diseases and genetic factors.[11] From the data published already it appears that prostaglandins exert a divergent effect on thyroid structure and function, but there are no reports suggest-

ing their role in goiter formation (goitrogenesis). Comparative investigations have shown that each prostaglandin increases mouse thyroid ^{131}I release (still less than that induced by TSH), but when prostaglandin was added to a submaximal dose of TSH or long-acting thyroid stimulator (LATS), a significant reduction of hormonal effect occurred.[1-3] However, other investigators did not find these interactions in dog thyroid slices.[12] It has been shown that prostaglandins do not directly mediate the action of TRH on TSH secretion,[13] but they can mimic the effects of TSH on thyroid glands and are able to stimulate most of iodine metabolism parameters in a manner similar to TSH.[14]

Our findings reveal that the cellular and metabolic effects induced by prostaglandins (E_1 and E_2) on thyroid gland—such as hyperplastic goiter, stimulation of secretory activity, increased iodine (^{131}I and ^{125}I) incorporation, and moderate stimulation of hormone (T_4 and T_3) synthesis—do not occur in hypophysectomized rats, suggesting a direct effect of prostaglandins on pituitary TSH secretion. Cytologic observations of pituitary glands from prostaglandin-treated rats suggest a direct influence on TSH release from thyrotropic cells. Some investigators have also reported that PGE_2 directly stimulates TSH secretion by the rat pituitary cell culture *in vitro*.[15] It is interesting that thyroid hypofunction induced by $PGF_{2\alpha}$ is due to a decrease in TSH secretion and is abolished in hypophysectomized rats; this also can explain the small increases in radioiodine incorporation in $PGF_{2\alpha}$-treated and hypophysectomized rats. Despite that, the differences between intact and prostaglandin-treated rats as compared to hypophysectomized and treated rats are statistically highly significant ($P < 0.001$) (Tables 1–4); there are only a few statistically significant values ($P < 0.02$) between animals in the hypophysectomized group itself (Table 4).

Inasmuch as we studied the chronic effect of prostaglandins on several parameters of thyroid glands, our findings are somewhat different from other investigators. This chronic effect of prostaglandins on thyroid function is not well studied; most of the reports already published were carried out with small doses and investigated the short-term effect. Combined experiments with PGE_1 and TSH reveal that each stimulates organic binding of iodine and thyroxine synthesis in preincubated canine thyroid slices, but their effects are not additive.[16] In our investigations the chronic stimulation of TSH secretion induced by PGE_1 and PGE_2 can explain the goiter formation, increased radioiodine uptake, and moderate increase of thyroid hormone synthesis. In this respect, the goitrogenic effect of prostaglandins is primarily mediated by TSH secretion, since it does not occur in hypophysectomized rats. However, not all effects of prostaglandins can be

influenced by TSH secretion. Thus, increased values of T_4 and T_3 in the serum of PG-treated rats could be due to disorders occurring in their utilization or conversion in the peripheral tissues.

An interesting finding is the occurrence of a marked hyperplasia of parafollicular or C-cells with an increase in their characteristic granule populations following $PGF_{2\alpha}$. This suggests a relationship between prostaglandin $F_{2\alpha}$ and calcitonin synthesis in rat thyroids. No increase in C-cell population is observed in untreated controls, after PGE administration or in hypophysectomized and treated rats.

Therefore, our data demonstrate that prostaglandins exert significant action on thyroid gland and goiter formation (goitrogenesis), namely by influencing TSH secretion.

References

1. Burke G: Effects of prostaglandins on basal and stimulated thyroid function. Am J Physiol 218:1445–1452, 1970
2. Field J, Dekker A, Zor U, Kaneko T: In vitro effects of prostaglandins on thyroid gland metabolism. Ann NY Acad Sci 180:278–286, 1971
3. Sato S, Szabo M, Kowalski K, Burke G: Role of prostaglandin in thyrotropin action on thyroid. Endocrinology 90:343–356, 1972
4. Murphy BP, Jachan C: The determination of thyroxine by competitive protein-binding analysis employing an anion-exchange resin and radiothyroxine. J Lab Clin Med 66:161–167, 1965
5. Roberts RC, Nikolai TF: Determination of thyroxine-binding globulin: A simplified procedure utilizing dextran-coated charcoal. Clin Chem 15:1132–1140, 1969
6. Nejad I, Bollinger J, Mitnick MA, Sullivan P, Reichlin S: Measurement of plasma and tissue triiodothyronine concentration in the rat by radioimmunoassay. Endocrinology 96:773–780, 1975
7. Kieffer JD, Weintraub BD, Baigelman W, Leeman S, Maloof F: Homologous radioimmunoassay of thyrotrophin in rat plasma. Acta Endocrinol (Kbh) 76:495–505, 1974
8. Millonig G: Advantages of phosphate buffer for OsO_4 solutions in fixations. J Appl Physics 32:1637, 1961 (Abstr)
9. Caro LG, Tubergen RP, Kolb JA: High resolution autoradiography. I. Methods. J Cell Biol 15:173–188, 1962
10. Lupulescu A, Stebner F: Electron microscopic autoradiography of rabbit and rat thyroid glands following calcitonin. Electron Microscopic Concepts of Secretion: Ultrastructure of Endocrinal and Reproductive Organs. Edited by M Hess, New York, John Wiley & Sons, Inc., 1975, pp 357–377
11. Inada M, Sterling K: Thyroxine transport in thyrotoxicosis and hypothyroidism. J Clin Invest 46:1442–1450, 1967
12. Mashiter K, Mashiter GD, Field JB: Effects of prostaglandin E_1, ethanol and TSH on the adenylate cyclase activity of beef thyroid plasma membranes and cyclic AMP content of dog thyroid slices. Endocrinology 94:370–376, 1974
13. Tal E, Szabo M, Burke G: TRH and prostaglandin action on rat anterior pituitary: Dissociation between cyclic AMP levels and TSH release. Prostaglandins 5:175–182, 1974
14. Onaya T, Solomon DH: Stimulation by prostaglandin E_1 of endocytosis and glucose oxidation in canine thyroid slices. Endocrinology 86:423–426, 1970

15. Vale W, Grant G, Amoss M, Blackwell R, Guillemin R: Culture of enzymatically dispersed anterior pituitary cells: Functional validation of a method. Endocrinology 91:562–572, 1972
16. Ahn CS, Rosenberg IN: Iodine metabolism in thyroid slices: Effects of TSH, dibutyryl cyclic 3',5'-AMP, NaF and prostaglandin E_1. Endocrinology 86:396–405, 1970.

Acknowledgments

We kindly thank Dr. J. Pike from Upjohn Company for his generous gift of prostaglandins used in this study. We acknowledge K. Stuart, J. Milkintas, and J. Mansour for their skillful help. The author is also grateful to Dr. A. F. Parlow for preparation of a purified rat TSH and antibody to it, through National Institute of Arthritis, Metabolism and Digestive Diseases (NIAMDD), Rat Pituitary Hormone Distribution Program.

Legends for Figures

Figure 1—PGE_1-treated rat showing hyperplastic parenchymatous goiter. Microfollicles, 50 to 70 μ in diameter, are delineated by a high columnar epithelium containing pale colloid (*C*). Several colloid droplets are seen at the apical border. (H&E, × 500)

Figure 2—Hypophysectomized and PGE_1-treated rat in which no hyperplasia of thyroid follicular cells is observed. The follicles are large (200 to 250 μ in diameter), contain homogenous colloid (*C*), and are delineated by low cuboidal cells. (H&E, × 500)

Figure 3—Electron micrograph of control rat thyroid showing typical thyroid cells with microvilli (*mv*), colloid (*C*), dense granules (*dg*), endoplasmic reticulum (*er*), and basement lamina (*bl*) (Uranyl acetate and lead citrate, × 10,400).

Figure 4—Electron micrograph of PGE_1-treated rat thyroid with hyperplastic follicular cells with microvilli (*mv*), colloid (*C*), several small apical dense granules (*sdg*), large confluent dense granules (*ldg*), and endoplasmic reticulum (*er*). (Uranyl acetate and lead citrate, × 7500)

Figure 5—Electron micrograph of $PGF_{2\alpha}$-treated rat with hyperplasia of parafollicular cells (*Pc*), several small dense granules (*sd*), mitochondria (*m*), hypertrophic Golgi complex (*Gc*), nucleus (*N*), and follicular cells (*Fc*). (Uranyl acetate and lead citrate, × 10,400). **Inset**—Marked hyperplasia and hypertrophy of parafollicular (C) cells (*arrows*) in a 1-μ plastic-embedded section (Toluidine blue, × 500).

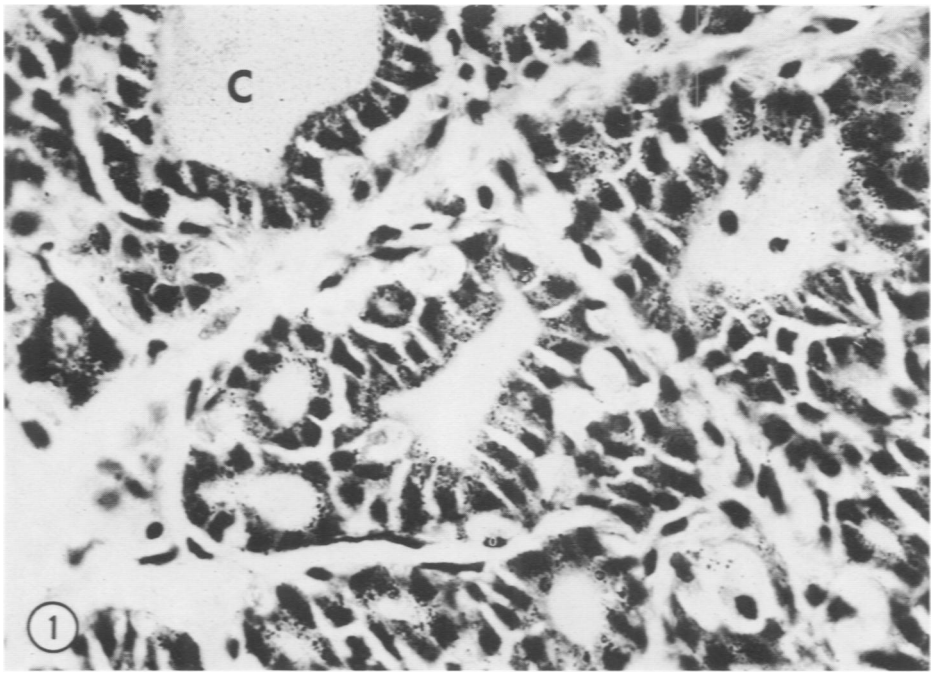
Figure 6—Electron micrograph of a hypophysectomized and PGE_1-treated rat (Hypox + PGE_1) shows flat cuboidal cells with dense colloid (*C*), few microvilli (*mv*) and dense granules (*dg*), scarce endoplasmic reticulum (*er*), sclerosis (*S*), and nucleus (*N*) (Uranyl acetate and lead citrate, × 10,400).

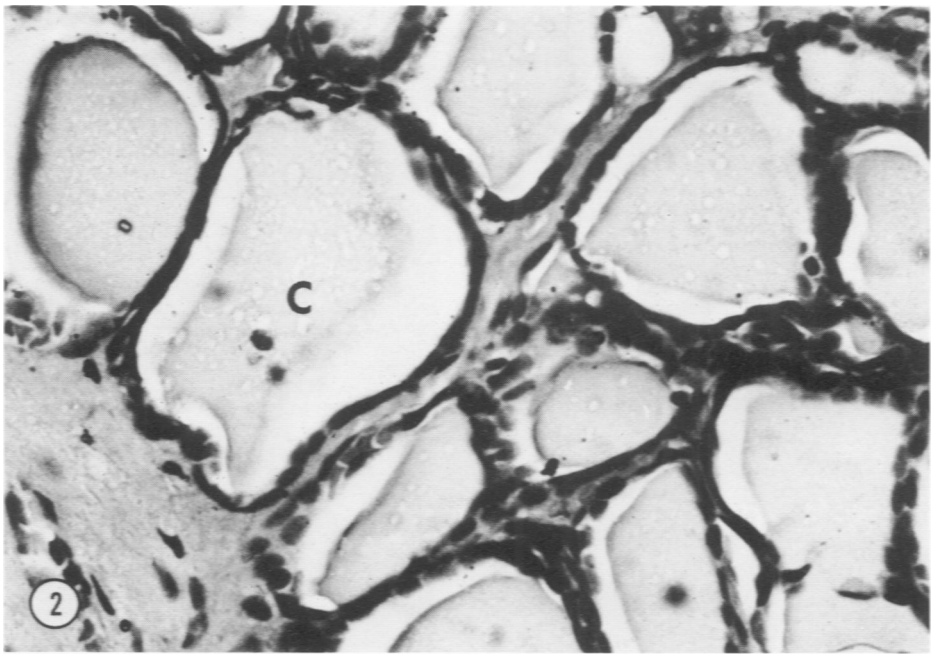
Figure 7—Electron microscopic autoradiogram (^{125}I) of a control rat thyroid at 1 hour. Characteristic intracellular distribution of ^{125}I is seen as developed grains over follicular colloid (*C*) and microvilli (*mv*); only a few grains are scattered in the endoplasmic reticulum (*er*) and dense granules (*dg*). (Uranyl acetate and lead citrate, × 10,400) **Inset**—Light microscopic autoradiogram in a 1-μ plastic-embedded section from the same block that shows a geographic distribution of radioiodine (Toluidine blue, × 500).

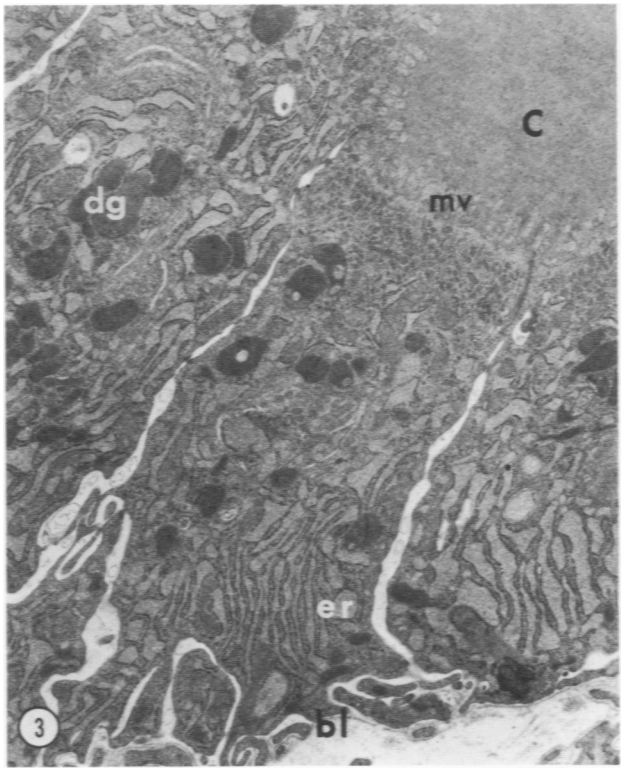
Figure 8—Electron microscopic autoradiogram (^{125}I) of a PGE_1-treated thyroid gland at 1 hour revealed a marked intracellular autoradiographic reaction; several developed grains are agglomerated and clumped together over follicular colloid (*C*) and an apical border with microvilli (*mv*); endoplasmic reticulum (*er*) and dense granules (*dg*) are also seen. (Uranyl acetate and lead citrate, × 10,400) **Inset**—Light microscopic autoradiogram in a 1-μ plastic-embedded section from the same block shows an intense cellular autoradiographic reaction (Toluidine blue, × 500).

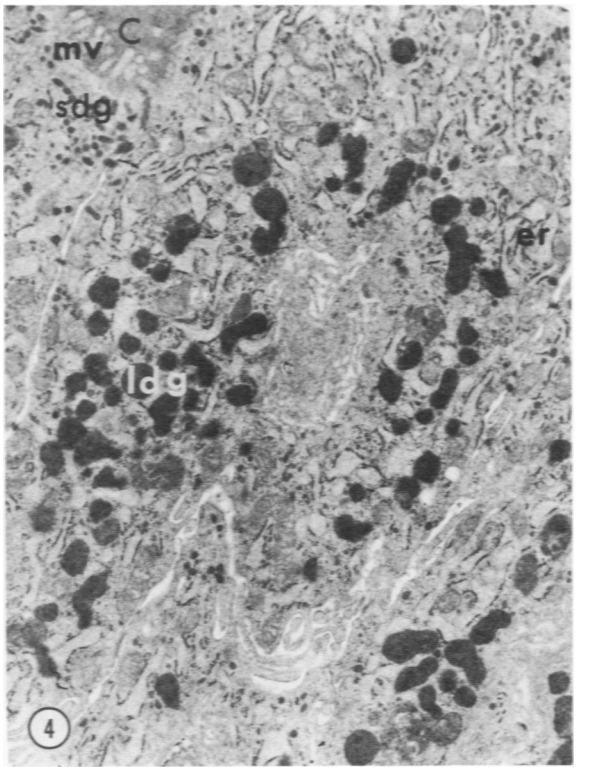
Figure 9—Electron microscopic autoradiogram (^{125}I) of a PGE_2-treated rat thyroid at 1 hour following administration. Characteristic reaction is seen over colloid droplets (*CD*). Colloid (*C*), microvilli (*mv*), dense granules (*dg*), and endoplasmic reticulum (*er*) are also seen. (× 18,400)

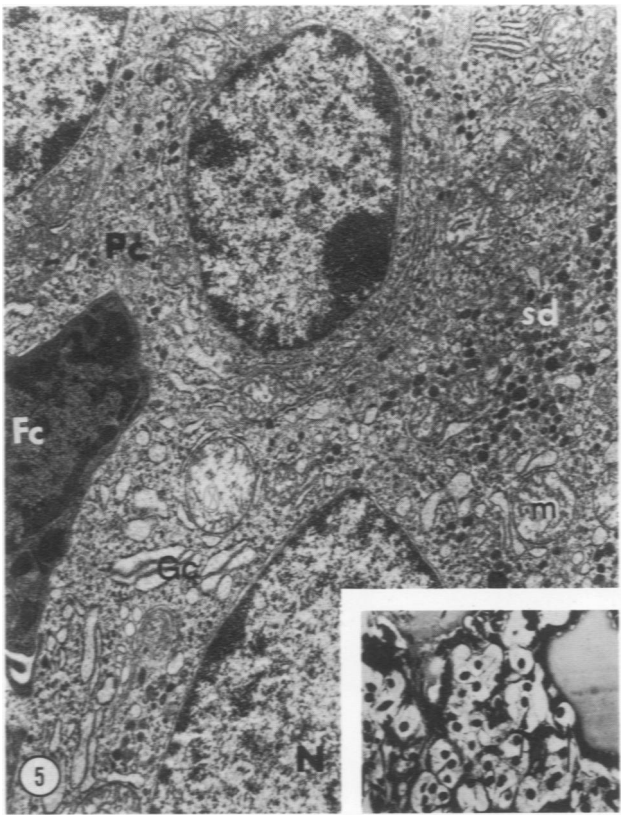
Figure 10—Electron microscopic autoradiogram (^{125}I) of a hypophysectomized and PGE_1-treated rat (Hypox + PGE_1). A marked reduction of autoradiographic reaction (1 hour) is seen. Only a few developed grains were seen over follicular colloid (*C*) and microvilli (*mv*); dense granules (*dg*) are seen. (Uranyl acetate and lead citrate, × 10,400) **Inset**—Light microscopic autoradiogram in a 1-μ plastic-embedded section from the same block reveals a reduced cellular autoradiographic reaction (Toluidine blue, × 500).

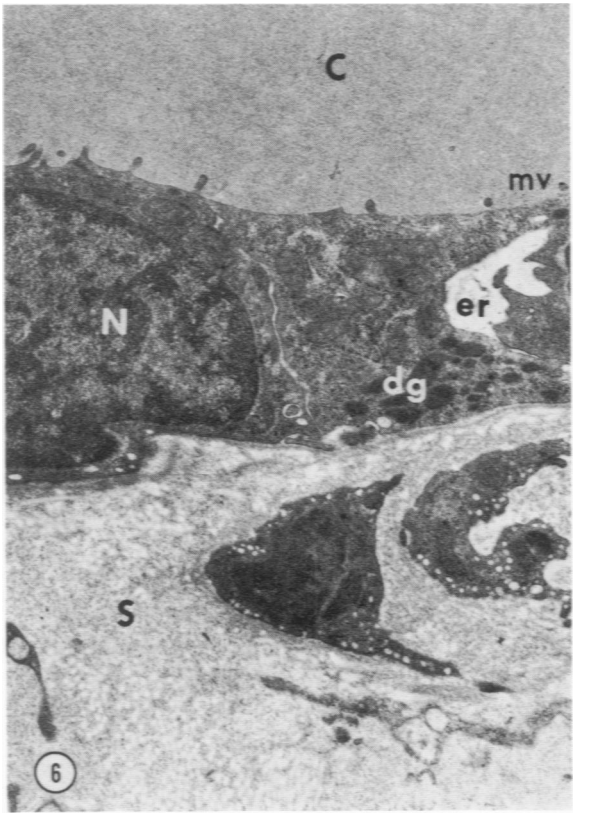

C
1

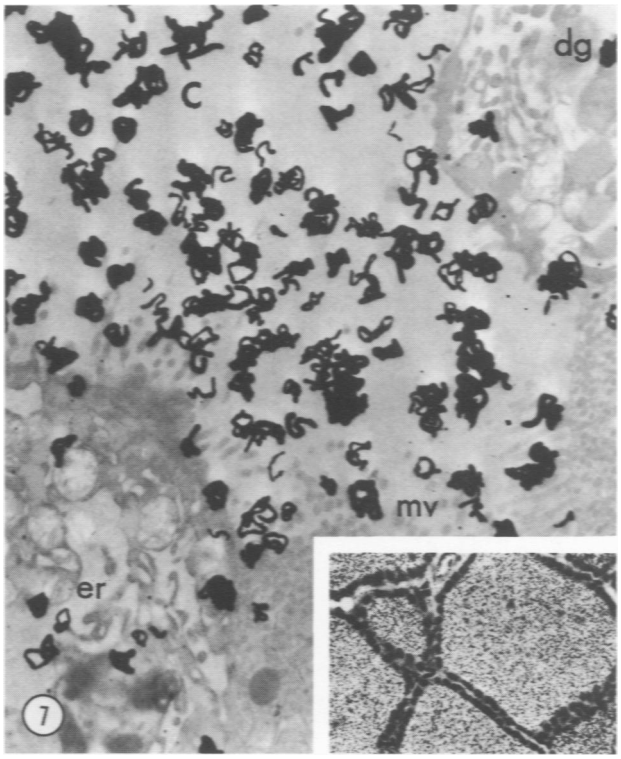

C
2

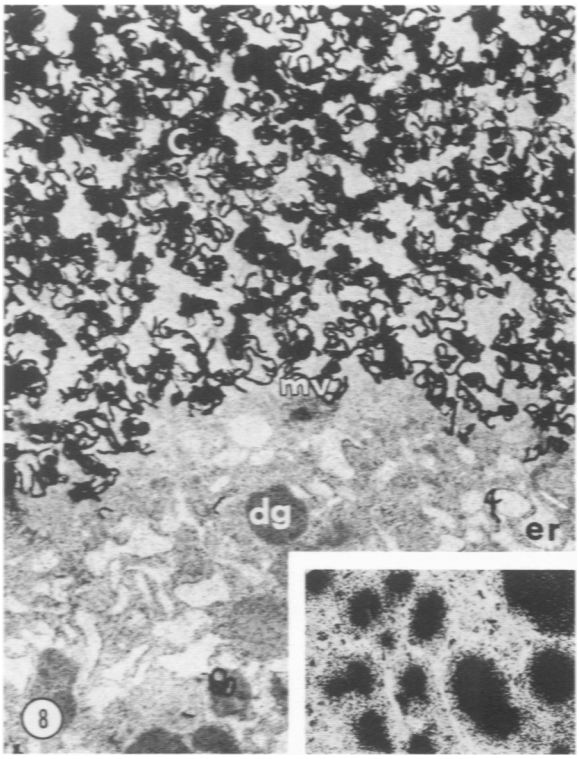

C
dg
mv
er
bl
3

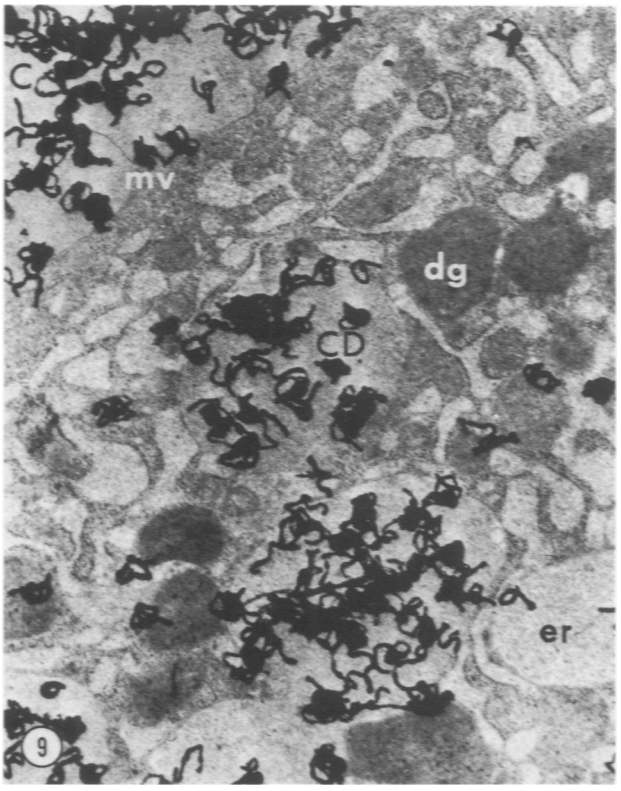

mv
C
sdg
er
ldg
4

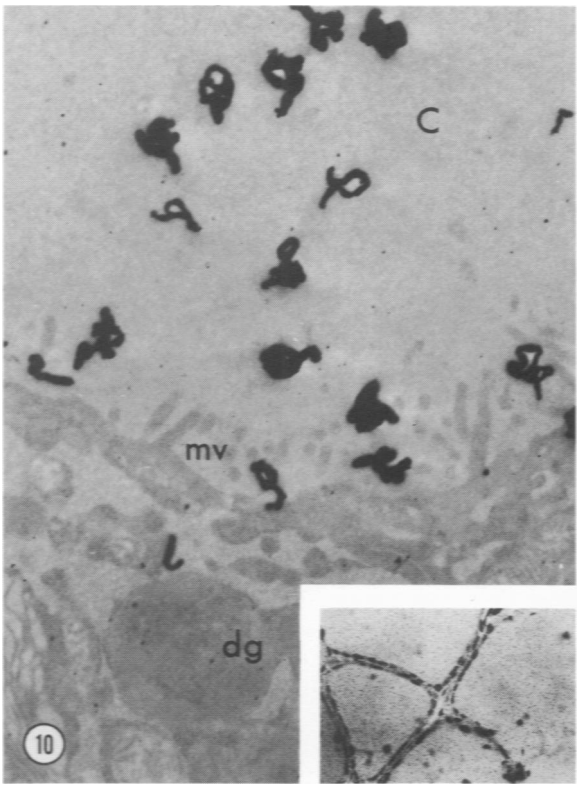

Pc
sd
Fc
m
Ec
N
5


C
mv
N
er
dg
S
6


dg
C
mv
er
7


C
mv
dg
er
8


C
mv
dg
CD
er
9


C
mv
dg
10

[*End of Article*]