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Effects of prostaglandin E_2 on the ultrastructure of the golden hamster parathyroid gland

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Summary. The effects of different ages on large vacuolar bodies in the parathyroid glands of golden hamsters after administration of prostaglandin E_2 (PGE_2) were investigated. In the parathyroid glands of the young and senile animals 15 min and the senile animals 60 min after administration of PGE_2, the mean serum calcium concentration was significantly higher when compared to that of the young and senile control animals, respectively. In the experimental adult animals 60 min after administration of PGE_2, the serum calcium concentration was seen to increase. In the parathyroid glands of the young animals 15 min and the adult and senile animals 60 min after administration of PGE_2, the percent area occupied by large vacuolar bodies was significantly increased when compared to that of the young, adult and senile control animals, respectively. These findings suggest that the percent area occupied by large vacuolar bodies is increased in response to hypercalcemia induced by PGE_2. It is thought that in the parathyroid glands suppressed by hypercalcemia there is a relationship between the percent area occupied by large vacuolar bodies and aging.

Key words: Parathyroid gland, Large vacuolar body, Prostaglandin E_2, Different ages, Golden hamster

Introduction

PGE_2 has been shown by many investigators to stimulate bone resorption (Tashjian et al., 1972, 1977; Powles et al., 1973; Dietrich et al., 1975; Seyberth et al., 1975; Santoro et al., 1977; Rifkin et al., 1980; Yamasaki et al., 1980). The effects of PGE_2 are similar to those of parathyroid hormone (Klein and Raisz, 1970). Franklin and Tashjian (1975) have reported that constant intravenous infusion of PGE_2 produced an elevation of the plasma calcium concentration in unanaesthetized rats in a period of 4 h.

The parathyroid gland of the golden hamster contains a small number of large vacuolar bodies, besides many secretory granules and a few large secretory granules (Emura et al., 1988, 1989; Shoumura et al., 1988a,b, 1989a,b, 1990, 1991; Chen et al., 1990, 1991; Isono et al., 1990). Recently, we have reported that the percent area occupied by large vacuolar bodies increased in response to acute hypercalcemia, and we thought that in the parathyroid glands suppressed by hypercalcemia there was a relationship between the percent area occupied by large vacuolar bodies and aging (Emura et al., 1992). However, there is no study on the effects of PGE_2 on the ultrastructure of the parathyroid gland.

This investigation was undertaken to study the effects of different ages on the large vacuolar bodies in the parathyroid glands of golden hamsters after administration of PGE_2.

Materials and methods

Twenty-two 1-month-old (young), fifteen 3-month-old (adult) and seventeen 15-month-old (senile) golden hamsters of both sexes were divided into three groups. Seven young, 5 adult and 6 senile golden hamsters served as controls (control groups). The remaining two groups of 15 young, 10 adult and 11 senile animals were intraperitoneally given PGE_2 solution at a dose of 3 mg/Kg body weight and sacrificed at 15 or 60 min after injection. The parathyroid glands of the control and PGE_2-treated groups were removed under sodium pentobarbital anaesthesia. The glands were immersed in a mixture of 2.5% glutaraldehyde and 2% OsO_4 in Millonig's buffer at pH 7.4 for 1 h, dehydrated through increasing concentrations of acetone and embedded in Epon 812. Thin sections were cut on a Porter-Blum MT-1 ultramicrotome, stained with uranyl acetate and lead salts, and examined with a Hitachi H-700 H electron microscope.

In all animals from each group, 20 micrographs (final magnification x 12,000) were taken from different

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regions of the parathyroid glands. The area of cytoplasm and large vacuolar bodies was estimated with the aid of an image analyser (Digigrammer Model-G, Mutoh).

In the control adult group and the experimental groups 15 and 60 min after administration of PGE_2, 20 micrographs (final magnification x 12,000) were taken from different regions of the parathyroid glands. The area of cytoplasm, Golgi complexes, lysosomes and lypid droplets, and number of secretory granules were estimated with the aid of the same image analyser.

The serum calcium levels of all animals were measured using a Corning calcium analyser 940.

All values are presented as means±SEM. In the serum calcium concentration and the percent area occupied by large vacuolar bodies, Golgi complexes, lysosomes and lipid droplets, and the number of secretory granules in each group, mean values were compared by Anova (one-way analysis of variance) followed by Scheffe's method.

Results

Serum calcium level

The mean serum calcium concentrations (mg/100 ml) of the control and PGE_2-treated groups are shown in Table 1. In the young golden hamster 15 min after administration of PGE_2, the mean serum calcium concentration was significantly high ($p<0.05$) when compared to that of the young control group, and the young group 60 min after administration, respectively. In the senile gorups 15 and 60 min after administration, it was significantly high ($p<0.05$) when compared to that of the control senile group.

Table 1. Percent area occupied by the large vacuolar bodies (VB) and serum calcium level (mg/100ml).

	ANIMALS, n	VB	SERUM CALCIUM LEVEL
Young animals			
Control group	7	0.26±0.03	10.98±0.20
15-min-treated group	8	0.41±0.05[a]	12.37±0.12[a]
60-min-treated group	7	0.31±0.04	11.51±0.25[b]
Adult animals			
Control group	5	0.24±0.04	11.77±0.21
15-min-treated group	5	0.27±0.05	12.41±0.33
60-min-treated group	5	0.41±0.04[a,b]	12.56±0.37
Senile animals			
Control group	6	0.16±0.02	10.44±0.09
15-min-treated group	5	0.27±0.04	13.17±0.10[a]
60-min-treated animals	6	0.53±0.04[a,b]	13.47±0.33[a]

The percent area is presented as percentage of cytoplasmic area. Values are means±SEM. [a]: $p<0.05$ versus control group; [b]: $p<0.05$ versus 15-min-treated group (Anova and Scheffe's multiple-comparison test).

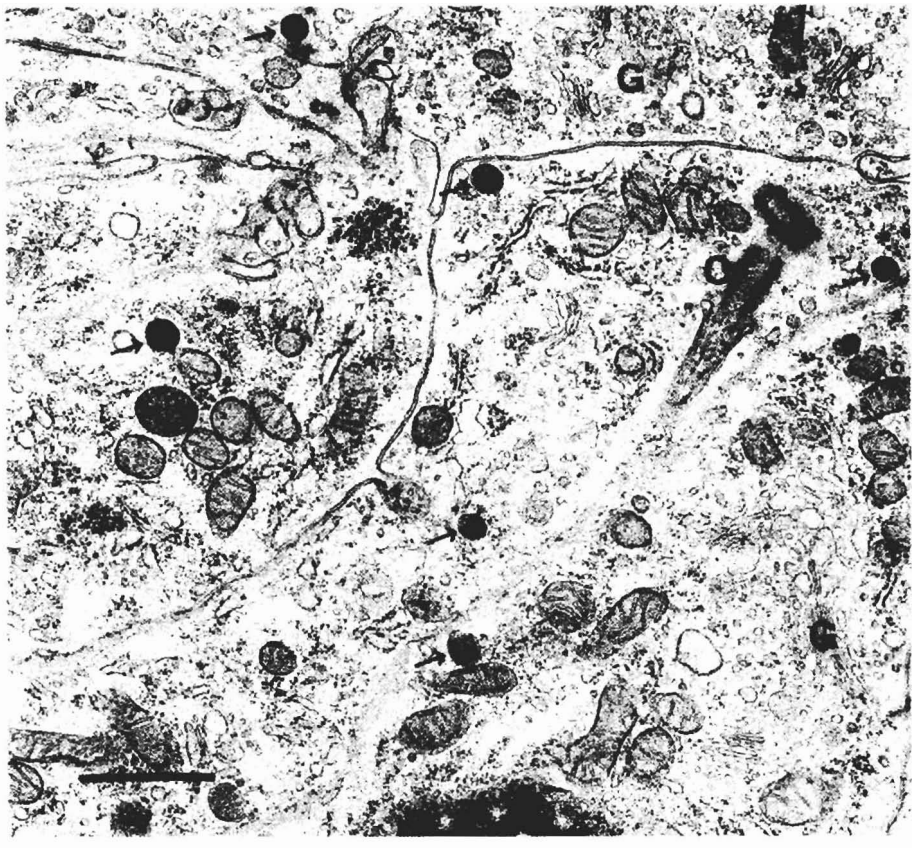

Fig. 1. Parathyroid chief cells from a control adult golden hamster. Golgi complexes (G) are relatively well developed and contain some prosecretory granules. Many secretory granules (arrows) are located in the peripheral cytoplasm. C: Cilium and its basal body. Bar= 1 μm.

Ultrastructure of the parathyroid gland, percent area occupied by cell organelles and number of secretory granules in the parathyroid gland

In the parathyroid glands of the control adult golden hamsters, the chief cells were oval or polygonal in shape. The intercellular spaces were generally narrow, and occasional enlargements had floccular or finely particulate material (Fig. 1). The cytoplasm was scattered diffusely with free ribosomes and randomly with abundant mitochondria (Fig. 1). Cisternae of the granular endoplasmic reticulum were randomly distributed or sometimes arranged in parallel arrays (Fig. 1). Most Golgi complexes were relatively well developed and associated with several prosecretory granules containing floccular material (Fig. 1). Secretory granules of 150-300 nm in diameter were frequently observed in the cytoplasm and sometimes located in a peripheral position adjacent to the plasma membrane (Fig. 1). Large vacuolar bodies of 350-750 nm in diameter, lysosomes and lipid droplets were sometimes observed in the cytoplasm. Large vacuolar bodies contained floccular material and/or vesicles.

In the adult animals 60 min after administration of PGE_2, the chief cells contained abundant free ribosomes, and poorly developed Golgi complexes associated with a few prosecretory granules and cisternae of the granular endoplasmic reticulum (Fig. 2). Many secretory granules were scattered in the cytoplasm and frequently located in the peripheral cytoplasm (Fig. 2). Large vacuolar bodies were fequently observed in the cytoplasm (Fig. 2).

The results obtained from the control adult group and the adult group 15 and 60 min after administration of PGE_2 are shown in Table 2. In the experimental adult group 15 min after administration of PGE_2, the percent area occupied by lipid droplets showed a significant increase ($p<0.05$) compared with that of the control adult group. There were no significant differences between the control and experimental adult groups with regard to lysosomes and secretory granules. The Golgi complexes in the experimental adult group 60 min after administration of PGE_2 appeared to decrease, but the difference was no significant.

Table 2. Percent area occupied by the Golgi complex (G), lysosome (LY) and lipid droplet (L).

ADULT ANIMALS	G	LY	L	SG
Control group	7.77±0.38	0.94±0.04	0.04±0.02	7.23±0.64
15-min-treated group	6.98±0.51	1.07±0.08	0.20±0.04[a]	8.19±1.07
60-min-treated group	6.60±0.36	0.98±0.10	0.09±0.03[b]	7.16±0.91

The percent area is presented as percentage of cytoplasmic area. SG: number of secretory granules per 100 μm^2 in the cytoplasm. Values are means±SEM. [a]: $p<0.05$ versus control group; [b]: $p<0.05$ versus 15-min-treated group. (Anova and Scheffe's multiple-comparison test).

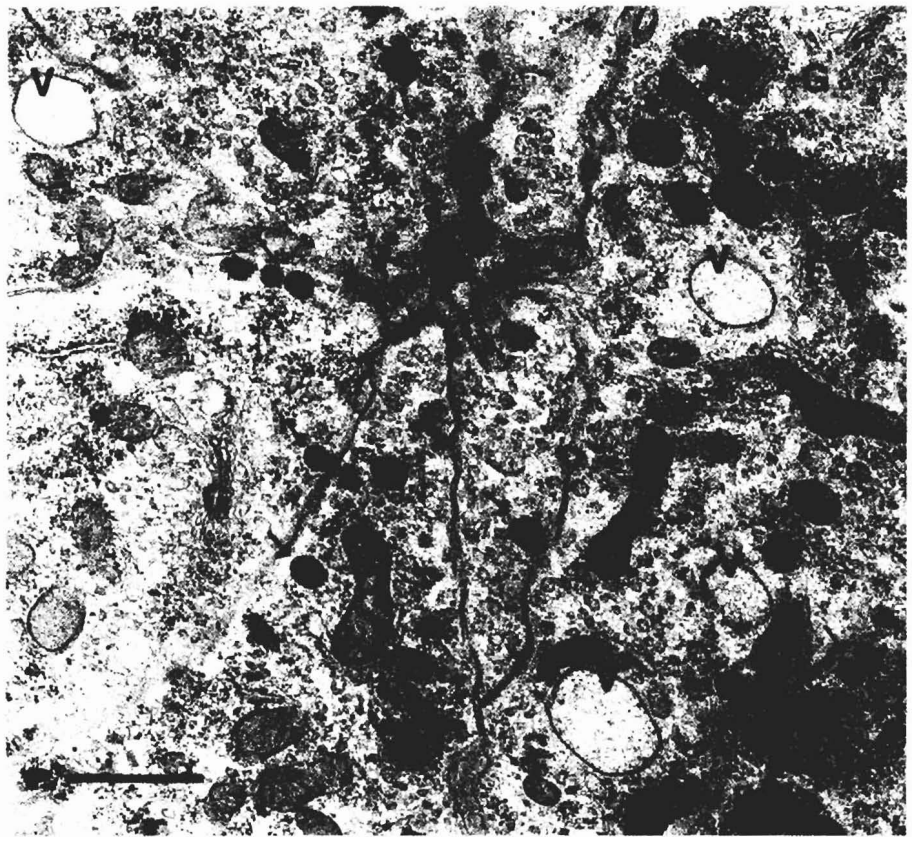

Fig. 2. Parathyroid chief cells from an adult golden hamster 60 min after PGE_2 administration. Many large vacuolar bodies (V), many secretory granules (arrows) and poorly-developed Golgi complexes (G) are observed. Bar= 1 µm.

Percent area occupied by large vacuolar bodies in the parathyroid gland

The results obtained from the control and PGE_2-treated groups are shown in Table 1. In the parathyroid gland of the young group 15 min after administration of PGE_2, the percent area occupied by large vacuolar bodies was significantly increased ($p<0.05$) when compared to that of the control young group. In the adult and senile groups 60 min after administration, the percent area occupied by large vacuolar bodies was significantly increased ($p<0.05$) when compared to that of the control and 15-min treated groups (Figs. 1, 2).

Discussion

PGE_2 is known to have marked effects on calcium metabolism. It has been reported that the serum calcium level in rats after administration of PGE_2 was increased (Franklin and Tashjian, 1975). However, Klein and Raisz (1970), Beliel et al. (1973) and Robertson and Baylink (1977) have reported that serum calcium levels were not affected by the prostaglandin infusion. In the present study, in the young and senile groups 15 min and in the senile group 60 min after administration of PGE_2, the serum calcium level was significantly increased when compared to that of the young and senile control groups, respectively. The reasons for this discrepancy between the findings of the present study and the observations of other investigators are not clear.

In the present study, the lipid droplets of the experimental adult group 15 min after administration showed a sinificant increase compared with that of the control adult group. Some authors have reported that the hypoactive chief cells of the parathyroid glands of adult mice show an increase in lipid droplets (Isono et al., 1980, 1981, 1983, 1985; Hayashi et al., 1981). It has been described that in the parathyroid glands of bats during early hibernation the active chief cells are characterized by many lipid droplets (Nunez et al., 1972). Furthermore, the chief cells of the parathyroid glands of the adult group 60 min after administration contained poorly developed Golgi complexes associated with a few prosecretory granules and cisternae of the granular endoplasmic reticulum when compared to those of the control adult group. These findings are fairly consistent with the observations of a decrease in functional activity of the parathyroid gland (Roth and Schiller, 1976; Isono et al., 1977, 1980, 1981, 1982, 1983, 1985; Hayashi et al., 1981; Emura et al., 1984; Iwasaki et al., 1987; Ishizaki et al., 1989).

Recently, we investigated the effects of different ages on large vacuolar bodies in the parathyroid glands of golden hamsters after short-term treatment with calcium (Emura et al., 1992). The results suggested that large vacuolar bodies in the parathyroid glands of the golden hamsters are increased in response to acute hypercalcemia (Emura et al., 1992). Setoguti et al. (1985) reported that type-I storage granules may be transformed into vacuolar bodies via type-II granules as a result of hydrolysis, and that these processes may be accelerated during hypercalcemia in rats. The present study showed that in the young group 15 min and in the adult and senile groups 60 min after administration of PGE_2, the large vacuolar body was significantly increased when compared to that of the young, adult and senile control groups, respectively. Accordingly, it is thought that the secretory activity of the parathyroid gland may be suppressed by treatment of PGE_2 and there is a relationship between the percent area occupied by large vacuolar bodies and aging.

Prostaglandins, especially the E-series, are potent stimulators of bone resorption in several in vitro systems (Klein and Raisz, 1970; Tashjian et al., 1972; Powles et al., 1973; Dietrich et al., 1975). On the other hand, Gardner et al. (1978) demonstrated that the prostaglandins directly stimulate parathyroid hormone secretion. Shemerdiak et al. (1981) reported that exogenous prostaglandins may affect parathyroid secretion, and endogenous prostaglandins do not appear to play a role in its secretion. These reports are not consistent with our observations of the parathyroid glands suppressed by PGE_2-induced hypercalcemia. Further work is required to clarify the effects of PGE_2 on the parathyroid gland or bone.

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