

A Randomized, Cross-Over Trial of Once-Daily Versus Twice-Daily Parathyroid Hormone 1–34 in Treatment of Hypoparathyroidism

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ABSTRACT

Once-daily sc injection of PTH 1–34 can normalize mean serum and urine calcium levels in patients with hypoparathyroidism; however, once-daily PTH has diminishing effects on serum calcium after 12 h, such that serum calcium levels fall below the normal range in some patients. Once-daily PTH also causes a marked increase in bone turnover, with persistent increases in markers of bone formation and resorption. To test the hypothesis that a twice-daily PTH regimen can produce more physiological control than a once-daily regimen, we performed a randomized cross-over trial, lasting 28 weeks, in 17 adult subjects with hypoparathyroidism. Each 14-week study arm was divided into a 2-week inpatient dose-adjustment phase and a 12-week outpatient phase. The PTH dose (given sc once daily at 0900 h or twice daily with one dose at 0900 h and the other at 2100 h) was adjusted to maintain both serum and urine calcium within, or close to, the normal range.

During the second half of the day (12–24 h), twice-daily PTH in-

creased serum calcium and magnesium levels more effectively than once-daily PTH. In patients with calcium receptor mutations (CaR), once-daily PTH normalized urine calcium, provided that serum calcium was maintained at levels below normal range. However, twice-daily PTH treatment produced higher mean serum calcium in patients with CaR with no significant rise in urine calcium excretion, and with no significant differences in either serum or urine calcium levels between CaR and patients with acquired or idiopathic hypoparathyroidism. Thus, treatment with twice-daily PTH is the better regimen for patients with CaR to overcome their tendency to hypercalciuria while producing near-normal levels of serum calcium. The total daily PTH dose was markedly reduced with the twice-daily regimen (twice daily 46 ± 52 vs. once daily 97 ± 60 $\mu\text{g}/\text{day}$, $P < 0.001$). We conclude that a twice-daily PTH regimen provides effective treatment of hypoparathyroidism and reduces the variation in serum calcium levels at a lower total daily PTH dose. (*J Clin Endocrinol Metab* 83: 3480–3486, 1998)

HYPOPARATHYROIDISM is one of the hormonal insufficiency states that is usually not treated by replacing the missing hormone. Conventional therapy for hypoparathyroidism, with calcitriol or other vitamin D analogs, normalizes serum calcium but does not have the renal calcium-retaining effect needed to normalize urine calcium. Thus, patients with hypoparathyroidism who are treated with vitamin D analogs have a tendency toward hypercalciuria. Eventually, this may lead to nephrocalcinosis, nephrolithiasis, or renal insufficiency (1–10).

Synthetic human PTH 1–34 was initially administered to humans as a treatment for osteoporosis (11–13). We have recently found that once-daily sc injections of PTH may be superior to calcitriol in the treatment of hypoparathyroidism (14). Once-daily PTH therapy significantly reduced the level of urine calcium excretion compared with calcitriol therapy and maintained serum calcium in the normal range throughout most of the day. Some patients, however, had periods of hypocalcemia near the end of the 24-h period.

We hypothesized that twice-daily administration of lower doses of PTH would improve the 24-h serum calcium profile

and would continue to maintain urine calcium excretion within the normal range. To test this hypothesis, we performed a randomized cross-over trial, comparing once-daily and twice-daily PTH regimens in 17 adult patients with hypoparathyroidism. We found that twice-daily PTH allowed for a marked reduction in the total daily PTH dose, with less fluctuation in serum calcium, normalization of urine calcium, and significantly improved metabolic control in patients with calcium receptor mutations (CaR).

Subjects and Methods

Subjects

Seventeen adult subjects, ages 19–64 yr, with hypoparathyroidism were studied (Table 1). These study subjects did not participate in a previous pilot study comparing PTH with calcitriol in the treatment of hypoparathyroidism (14). The diagnosis of hypoparathyroidism was determined in each subject by low levels of intact PTH during hypocalcemia (data not shown). Four patients (E–H) were from a single family of autosomal dominant hypoparathyroidism in which we identified a calcium receptor mutation (15). Patient J had a sporadic mutation in the calcium receptor causing severe hypocalcemia and seizures during infancy (16). All patients were receiving calcitriol and calcium supplementation at study entry. Patients who were managed with other vitamin D analogs before the study were switched to calcitriol at least 1 month before study entry. All study patients had received vitamin D and calcium supplementation for at least 3 yr. There was no drug-free interval before starting the protocol, and the last vitamin D dose was given approximately 12 h before the initiation of PTH. Only 5 subjects were receiving magnesium replacement (130–450 mg/day) at study entry. Two other patients had low-normal magnesium levels at baseline, and later had low serum magnesium levels while on PTH. By the end of the

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TABLE 1. Baseline clinical and laboratory features of 17 patients with hypoparathyroidism at study entry

Patients ^a	Age (yr)	Sex	Diagnosis	Duration of Hypoparathyroidism (yr)	Serum calcium (mmol/L; 2.05–2.5) ^b	Serum phosphorus (mmol/L; 0.7–1.4)	Serum magnesium (mmol/L; 0.65–1.05)	Serum intact PTH (pg/mL; 10–65) ^c	Urine calcium (mmol/24 h; 1.25–6.25)	Creatinine clearance (mL/min/m ² ; 90–125)	Nephrocalcinosis by CT ^d
A	35	F	Postsurgical	3	2.3	1.2	0.74	<8.3	12.5	66	–
B	19	F	APS-1 ^e	15	2.45	1.4	0.73	<2.5	9.2	99.1	+
C	64	F	Postsurgical	9	1.8	1.5	0.7	29.7	1.4	58.8	–
D	57	F	Postsurgical	27	1.9	1.6	0.75	<3.3	6.8	86.4	–
E	28	F	Familial	19	2.46	1.2	0.65	<3.3	12.59	44.7	+
F	27	F	Familial	19	2.2	1.5	0.92	<3.3	7.6	21.6	+
G	55	F	Familial	19	2	1.1	0.62	<3.3	9.7	29.1	+
H	25	F	Familial	19	2.42	1.3	0.71	<3.3	9.28	34.6	+
I	50	F	Postsurgical	30	2.4	1.2	0.54	<7.5	12.6	78.7	+
J	20	F	Sporadic	20	1.96	1.4	0.65	<7.5	6.09	97.6	+
K	41	F	Postsurgical	7	2.2	0.9	0.81	24	7.5	84.1	–
L	52	M	Idiopathic	34	2	1.8	0.78	<9.6	10.7	76.5	–
M	38	F	Postsurgical	4	2.02	1.3	0.75	22	4.2	77.3	–
N	42	M	Familial	40	2	1.7	0.75	<12.7	14.3	84.8	–
O	55	M	Postsurgical	4	2.2	1.6	0.83	<12.7	19.2	95	+
P	46	M	Postsurgical	21	2.13	1.4	0.79	<1	23.45	58	–
Q	51	F	Postsurgical	33	2.98	1.3	0.67	<1	5.56	59.5	–

^a Patients were receiving calcitriol and supplemental calcium at time of studies, thus not all patients had calcium levels below normal range.

^b Normal ranges follow units of measure in parentheses.

^c To convert values for PTH to pmol/L multiply by 0.2440.

^d CT, Computerized tomography.

^e APS-1, autoimmune polyglandular syndrome type 1.

initial 14-week period, 11 patients were supplemented with magnesium (100–600 mg/day). All 5 patients with CaR required magnesium supplementation. Eight patients had evidence of nephrocalcinosis by renal computerized tomography scan, and 14 patients (80%) had renal insufficiency (Table 1).

Protocol

The study was approved by the Institutional Review Board of the National Institute of Child Health and Human Development (NICHD). Informed consent was obtained from all subjects. A randomized, cross-over design was used to compare once-daily PTH with twice-daily PTH therapy. The two arms, each lasting 14 weeks, were divided into a 2-week inpatient dose-adjustment phase and a 12-week outpatient phase during which continued dose adjustment was permitted as indicated.

PTH was administered sc (in the extremities with an insulin syringe) once daily at 0900 h or twice daily at 0900 h and 2100 h. The initial dose was 0.7 µg/kg per day for both treatment arms. The dose of PTH was adjusted in increments or decrements of approximately 5–10% to maintain urine and serum calcium within the normal range. For several patients it was necessary to maintain the serum calcium below the normal range to obtain normal urine calcium excretion. Both serum calcium and 24-h urine calcium were measured daily during the initial 2 weeks and weekly thereafter.

Dietary intake of calcium ranged from 1–2 g of elemental calcium during both the inpatient phase (based on daily dietary calcium intake counts) and outpatient phase (based on dietary history). One subject received oral calcium supplementation when her dietary calcium remained below 800 mg/day because of lactose intolerance. This subject was given sufficient calcium supplementation to raise her total elemental calcium intake to 1000 mg/day. No study participant received phosphate binders, diuretics, or other study medications that affected serum calcium, magnesium, or phosphorus levels.

The primary outcome measures were the levels of calcium, phosphorus, and magnesium in serum and urine. These were assessed in two ways. First, 0800 h serum calcium, phosphorus, and magnesium levels (before the morning dose of PTH), along with the corresponding 24-h urine calcium, phosphorus, and magnesium levels, were measured six times between weeks 12 and 14. The mean of these six measurements is referred to as the 14-week level. Second, at 14 weeks, near the conclusion of each treatment arm, patients underwent blood sampling over a 24-h period to assess the time course of PTH effects on mineral metabolism.

Serum was collected at 0900 h (before the dose of PTH) and then every 2 h until 0900 h the next morning. On the same day, urine was collected at 4-h intervals from 0900 h (before PTH) to 0900 h the next morning. Subjects consumed a diet containing at least 1000 mg of elemental calcium during the 24-h test.

The secondary outcome measures were the dose of PTH administered, the serum alkaline phosphatase and osteocalcin (which reflect bone formation) measured before the morning dose of PTH, and corresponding 24-h urine pyridinoline and deoxypyridinoline (which reflect bone resorption). Serum 25-hydroxyvitamin D₃ and 1,25-dihydroxyvitamin D₃ were measured at the beginning of each study arm along with the other serum measures before the morning PTH dose.

Preparation of PTH

Lyophilized human PTH 1–34 was purchased from Bachem California, Inc. (Torrance, CA) and prepared for human administration as previously described (14).

Biochemical assays

All blood and urine samples for calcium, phosphorus, magnesium, creatinine, and alkaline phosphatase were measured at the Clinical Center, NIH. Blood samples were measured using the Hitachi Scientific Instruments, Inc. 917 analyzer (Indianapolis, IN). Urine samples were measured using the Cobas-Mira analyzer (Montclair, NJ). RIAs for intact PTH (17), cAMP, and vitamin D (18), were measured at Corning Hazleton (Vienna, VA), and total urine pyridinoline and deoxypyridinoline and serum osteocalcin (19), were measured at Corning-Nichols Institute Diagnostics (San Juan Capistrano, CA). Total urine pyridinoline and deoxypyridinoline were measured by flurometry after reversed-phase high performance liquid chromatography of hydrolyzed urine (20).

Statistical analysis

Data are presented as mean ± SD unless otherwise stated. Data from the two arms of the study were compared by repeated measures of ANOVA, with Bonferroni's adjustment applied to correct for the number of ANOVAs performed. The sequence of the dose regimens and the etiology of the hypoparathyroidism were the two between-group factors in all ANOVAs. Patients were divided into those with calcium receptor defects and those with any other etiology for hypoparathyroidism (id-

idiopathic and postsurgical hypoparathyroidism). *Post hoc* comparisons at individual time points were performed with paired and unpaired Fisher's protected least significant difference tests. Contingency table analysis was applied to categorical data. Logarithmic transformation was performed, where appropriate, to achieve uniformity of variance. There were no sequence effects on urine or serum calcium and phosphorus levels (data not shown). Because the baseline results in Table 2 were obtained while most patients were receiving the calcitriol regimen prescribed by their referring physician, no statistical comparisons were made to these data because the calcitriol treatment regimens had not been optimized.

Results

Response to treatment at 14 weeks

Serum calcium, phosphorus, magnesium, vitamin D, and alkaline phosphatase levels obtained before the morning dose of PTH were repeatedly measured during the final 2 weeks of each treatment arm, and the means of these values are given in Table 2. Patients with CaR had significantly lower mean 0800 h serum calcium levels during once-daily PTH compared with patients with other etiologies of hypoparathyroidism (1.67 ± 0.12 vs. 2.02 ± 0.18 mmol/L, $P < 0.001$). The mean 0800 h serum calcium was significantly higher in CaR patients receiving twice-daily compared with once-daily PTH (1.91 ± 0.15 vs. 1.67 ± 0.12 mmol/L, $P < 0.02$). By contrast, the 0800 h calcium values were equally well maintained during both dose schedules of PTH for patients with idiopathic or postsurgical hypoparathyroidism. Serum magnesium was significantly greater for all study patients on twice-daily PTH compared with once-daily PTH

(0.68 ± 0.08 vs. 0.64 ± 0.07 mmol/L, $P < 0.008$). The serum phosphorus values were equally well maintained during both dose schedules of PTH.

Patients with CaR defects had similar levels of alkaline phosphatase compared with patients with other etiologies of hypoparathyroidism on both dosage regimens. Although mean serum alkaline phosphatase was lower during twice-daily compared with once-daily PTH therapy (146 ± 51 vs. 243 ± 272 U/L, $P < 0.05$) for all subjects, this difference appeared to be related to an order effect. Patients receiving daily PTH during the second treatment arm, after receiving twice daily PTH, had significantly greater elevations in alkaline phosphatase levels compared with patients receiving daily PTH in the initial 3 months of the study. This order effect was not apparent for twice-daily PTH. Mean serum osteocalcin, 25-hydroxyvitamin D₃, and 1,25 dihydroxyvitamin D₃ levels were similar during both study arms for all etiologies of hypoparathyroidism.

As we have previously reported (14), PTH reduced the level of mean urine calcium excretion for all subjects compared with baseline levels on calcitriol therapy (Table 2). The PTH dose schedule did not significantly affect mean 24-h urinary calcium and phosphorus excretion levels. For both study arms, mean urine calcium levels were within the normal range for the 13 female subjects. However, mean urine calcium excretion was above normal in the 4 male subjects during both study arms. Although patients with CaR and other etiologies of hypoparathyroidism had similar urine

TABLE 2. Effect of PTH regimen on PTH and magnesium doses and on serum and urine markers of calcium and bone metabolism

	Calcitriol and calcium (at Baseline)	PTH regimen		Normal range
		Once daily	Twice daily	
Dose administered				
PTH (μ g/day)				
CaR		141 ± 80	52 ± 28^e	
Acquired and idiopathic		80 ± 41	45 ± 36^e	
PTH (μ g/kg per day)				
CaR		2.62 ± 1.58^b	0.96 ± 0.56^e	
Acquired and idiopathic		1.00 ± 0.81	0.47 ± 0.33^e	
Elemental mg (mg/day)	154 ± 35	381 ± 185	423 ± 533	
Calcitriol (μ g/day)	0.60 ± 0.31			
Serum (0700 h)				
25-Hydroxyvitamin D ₃ (ng/mL)	40 ± 29	29 ± 22	22 ± 14	9–52
1,25-Dihydroxyvitamin D ₃ (pg/mL)	26 ± 9	26 ± 12	23 ± 9	15–60
Calcium (mmol/L)				2.05–2.5
CaR	2.19 ± 0.26	1.67 ± 0.12^a	1.91 ± 0.15^c	
Acquired and idiopathic	2.21 ± 0.22	2.02 ± 0.18	2.03 ± 0.15	
Phosphorus (mmol/L)	1.38 ± 0.24	1.55 ± 0.24	1.58 ± 0.26	0.74–1.4
Magnesium (mmol/L)	0.72 ± 0.10	0.64 ± 0.07	0.68 ± 0.08^d	0.65–1.05
Alkaline phosphatase (U/L)	66 ± 21	243 ± 272	146 ± 51^c	37–116
Osteocalcin (ng/mL)	3.0 ± 2.7	18.5 ± 21.1	13.6 ± 9.9	1.6–9.2
Urine (24-hr)				
Calcium (mmol/day)				
Total	10.5 ± 5.3	7.2 ± 3.6	6.4 ± 3.2	
Female	8.5 ± 3.4	6.1 ± 3.3	5.7 ± 2.4	1.25–6.25
Male	17.0 ± 5.6	10.7 ± 1.8	8.6 ± 4.8	1.25–7.5
Phosphorus (mmol/day)	25.7 ± 8.0	26.6 ± 11.5	26.7 ± 9.4	13–42
Magnesium (mmol/day)	4.9 ± 1.6	5.3 ± 2.0	4.8 ± 1.9^d	3–4.25
Pyridinoline (nmol/mmol creatinine)				
Female	48 ± 20	178 ± 154	143 ± 80	22–89
Male	25 ± 10	99 ± 16	84 ± 5	20–61
Deoxypyridinoline (nmol/mmol creatinine)	11 ± 6	50 ± 30	42 ± 18	4–20

^a $P < 0.001$; ^b $P < 0.05$, CaR vs. other etiologies of hypoparathyroidism; ^c $P < 0.05$; ^d $P < 0.01$; ^e $P < 0.005$, once- vs. twice-daily regimen. Calcitriol treatment was not compared with PTH treatments because calcitriol regimens were not optimized in this study.

magnesium excretion, mean urine magnesium excretion for all subjects was significantly lower during twice-daily PTH (4.8 ± 1.90 vs. 5.3 ± 2.0 mmol/24h, $P < 0.01$), although still above the normal range. Urine pyridinoline and deoxypyridinoline, markers of bone turnover, were similar during both dose schedules.

Twenty four-hour profile of serum calcium, phosphorus, and magnesium

The 24-h profiles of serum calcium, phosphorus, and magnesium were measured at the conclusion of each 14-week treatment phase (Fig. 1). ANOVA demonstrated significant differences in the serum calcium response to PTH related to the etiology of hypoparathyroidism ($P < 0.002$) and related to the number of doses of PTH administered ($P < 0.0001$). The 24-h mean serum calcium level in patients with CaR compared with patients with other etiologies of hypoparathyroidism showed a significantly lower mean serum calcium level during the once-daily arm (1.8 ± 0.16 mmol/L for CaR and 2.13 ± 0.11 mmol/L for others; Fig. 1, $P < 0.0002$). The 24-h mean serum calcium level in patients with CaR compared with others was not significantly different during the twice-daily arm (1.96 ± 0.15 mmol/L for CaR and 2.10 ± 0.17 mmol/L for others; Fig. 1, $P = 0.16$).

Particularly during the second half of the day (12–24 h), twice-daily PTH normalized serum calcium levels more effectively than once-daily PTH. The benefits of twice-daily PTH, assessed by the ability of PTH to normalize serum

calcium concentrations, were distinct for patients with CaR. For these patients, the frequency of hypocalcemia during the 24-h test was significantly less during twice-daily PTH than during once-daily PTH (63 vs. 91%, $P < 0.001$, Cochran test), but was not significantly different in patients with acquired or idiopathic hypoparathyroidism (32 vs. 43%, $P = 0.4$). Patients with CaR did not have hypercalcemia during the 24-h test during either treatment regimen. By contrast, patients with other etiologies for hypoparathyroidism had significantly more hypercalcemia during once-daily than during twice-daily PTH (4% vs. 0%, $P < 0.02$, Cochran test).

ANOVA demonstrated significant differences in the serum magnesium response to PTH related to the number of doses of PTH administered ($P < 0.003$). For patients with CaR, magnesium levels remained subnormal during both treatment arms. For patients with acquired and idiopathic hypoparathyroidism, twice-daily PTH produced normal serum magnesium levels throughout the day. During the twice-daily arm, mean serum magnesium levels were significantly higher during the final 8 h of the day (16–24 h) for patients with acquired and idiopathic hypoparathyroidism (Fig. 1). Once-daily PTH, however, appeared to have diminishing effects toward the end of the day, thus producing subnormal magnesium levels.

Mean 24-h serum phosphorus levels remained supranormal during both the twice- and once-daily regimens (1.42 ± 0.20 mmol/L vs. 1.45 ± 0.15 mmol/L, $P = 0.34$). Mean 24-h phosphorus levels did not differ for patients with CaR compared with other causes of hypoparathyroidism ($P = 0.26$). However, there were significant differences in the serum phosphorus response to PTH related to the number of doses of PTH administered ($P < 0.02$). In contrast to serum calcium, for which the greatest difference in patients with CaR between the two regimens was observed during the latter portion of the day, there were no differences in nighttime phosphorus levels between the two regimens (Fig. 1).

Twenty four-hour profile of urine calcium, phosphorus, magnesium, and cAMP

Twice-daily PTH produced a bimodal 24-h urine profile for urine calcium, magnesium, phosphorus, and cAMP, whereas once-daily PTH produced a unimodal profile ($P < 0.001$ by ANOVA; Fig. 2). For patients with CaR, urine calcium levels were significantly higher during twice-daily PTH than the once-daily arm during most of the latter portion of the day ($P < 0.01$, 12–16 h and 16–20 h). For patients with idiopathic and postsurgical hypoparathyroidism, twice-daily PTH produced urine calcium levels significantly lower than levels on once-daily PTH during the middle portion of the day ($P < 0.05$, 8–12 h, 12–16 h), but significantly higher than once-daily PTH from 16–20 h ($P < 0.05$).

For patients with CaR, urine magnesium levels were significantly lower during twice-daily PTH during most of the initial portion of the day ($P < 0.04$, 0–4 h, 4–8 h, 8–12 h). For the acquired and idiopathic hypoparathyroid patients, on twice-daily PTH, urine magnesium was significantly lower during the middle portion of the day ($P < 0.0001$, 8–12 and 12–16 h).

The tubular reabsorption of phosphorus decreased in re-

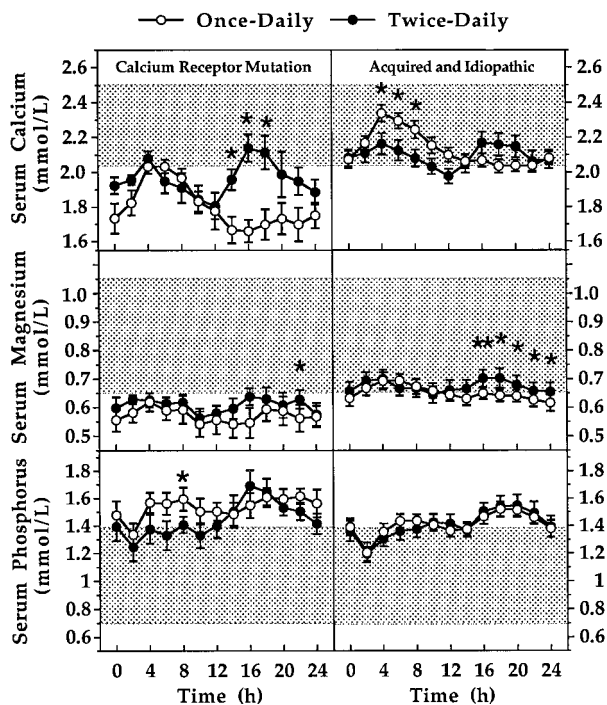


FIG. 1. Twenty four-hour profile of serum calcium, phosphorus, and magnesium in patients with CaR (left) and acquired and idiopathic hypoparathyroidism (right) treated with twice-daily (●) or once-daily PTH (○). Shaded region indicates normal range. PTH was administered at 0 h (0900 h) or 0 and 12 h, during once- and twice-daily arms, respectively. *, $P < 0.05$; **, $P < 0.01$ twice- vs. once-daily PTH treatment.

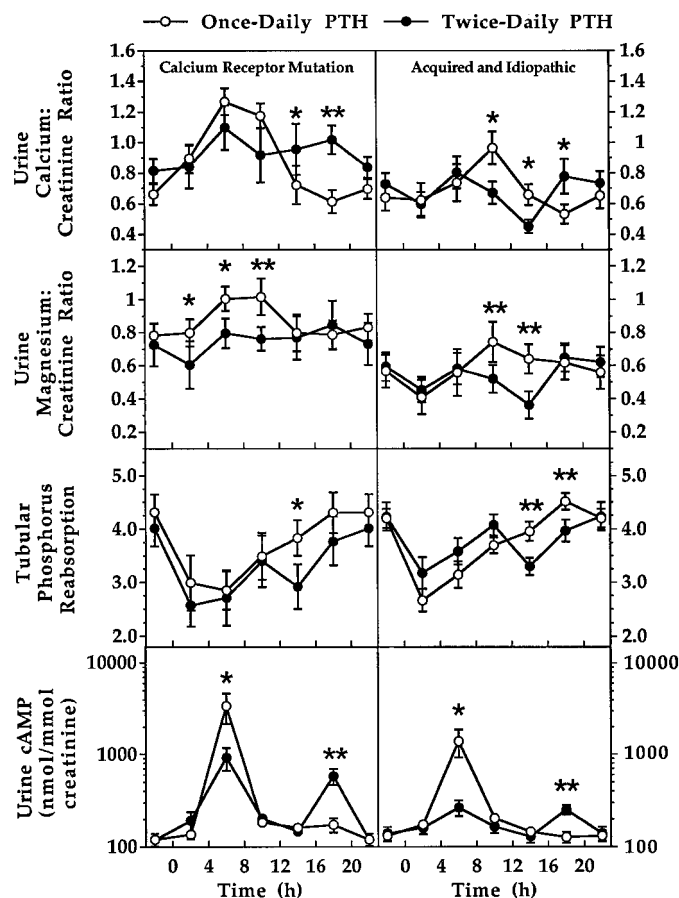


FIG. 2. Twenty-four-hour profile of urine excretion of calcium, phosphorus, magnesium, and cAMP in patients with CaR (left) and acquired and idiopathic hypoparathyroidism (right) treated with twice-daily (●) or once-daily PTH (○). *, $P < 0.05$; **, $P < 0.01$ twice- vs. once-daily PTH treatment.

sponse to PTH within the first 4 h of PTH administration. When patients received twice-daily PTH, tubular phosphorus reabsorption was significantly lower during most of the latter portion of the day ($P < 0.04$, 12–16 h for CaR; $P < 0.01$, 12–16 h, 16–20 h for acquired and idiopathic). Urine cAMP levels rose in response to PTH administration on both dose schedules and peaked at 4–8 h after PTH administration.

PTH dose

The total daily PTH dose required to maintain serum calcium in the normal or near normal range was significantly lower for all subjects receiving twice-daily PTH ($46 \pm 32 \mu\text{g}/\text{day}$) than once daily PTH ($97 \pm 60 \mu\text{g}/\text{day}$, $P < 0.001$, Table 2). After 14 weeks of treatment, the PTH dose administered to individual subjects ranged from 0.07–1.84 $\mu\text{g}/\text{kg}$ per day (mean $0.62 \pm 0.45 \mu\text{g}/\text{kg}$ per day) during the twice-daily arm and 0.14–4.20 $\mu\text{g}/\text{kg}$ per day (mean $1.48 \pm 1.29 \mu\text{g}/\text{kg}$ per day) during the once-daily arm. During the once-daily PTH arm, individuals with CaR required significantly more PTH per kilogram body weight ($2.62 \pm 1.58 \mu\text{g}/\text{kg}$ per day) compared with patients with other etiologies of hypoparathyroidism ($1.00 \pm 0.81 \mu\text{g}/\text{kg}$ per day, $P < 0.05$). For patients who received magnesium supplementation, the

dose of elemental magnesium was similar during both treatment arms.

Adverse events

In this open, unblinded, pilot study, bone pain (tibial discomfort) was reported in four patients during the once-daily arm and in one patient during the twice-daily arm ($P = \text{not significant}$). On three occasions, one patient developed nausea within 4 h of his single-daily PTH injection, whereas no nausea was reported during twice-daily PTH. Two patients had nocturia during twice-daily PTH therapy, which was not observed during the once-daily arm. Two patients complained of fatigue during the once-daily arm, whereas there were no complaints of fatigue during twice-daily therapy. Twelve out of the 17 patients preferred twice-daily over daily PTH despite the inconvenience of the second injection ($P < 0.05$).

Discussion

This study confirms our previous observation that PTH is able to maintain normal urine calcium levels in hypoparathyroid patients who were hypercalciuric on calcitriol therapy. In addition, for patients with acquired or idiopathic hypoparathyroidism, twice-daily PTH corrected the tendency toward nighttime hypocalcemia and maintained serum calcium levels within a more narrow range, thus producing an improved and more physiological profile compared with once-daily PTH. The mean 24-h serum calcium profile during the once-daily arm for subjects with acquired and idiopathic hypoparathyroidism is comparable with the profile obtained on a different group of subjects in our previous study (14). The differences in serum calcium levels over a 24-h period between once- and twice-daily PTH are most notable for the patients with CaR. Twice-daily PTH produced calcium levels just below the normal range throughout the day while maintaining normal levels of urine calcium. The higher mean serum calcium during twice-daily PTH in patients with CaR occurred with no significant rise in urine calcium excretion. During once-daily PTH, patients with CaR experienced profound nighttime hypocalcemia as the effects of PTH appeared to diminish. We conclude that treatment with twice-daily PTH is the better regimen for patients with CaR to overcome their tendency to hypercalciuria while producing near-normal levels of serum calcium.

Serum phosphorus levels normally follow a diurnal variation, with higher levels at night. This rhythm persisted during both dose schedules and was not affected by the second dose of PTH in the evening. We encouraged the patients to eat dairy products, which are high in both phosphorus and calcium. We hypothesize that it might be possible to achieve lower phosphorus levels by eliminating dairy products and supplementing the diet with calcium.

All protocol patients with CaR had magnesium deficiency and required supplementation. Individuals with a gain of function mutation in the calcium receptor may have a greater tendency to hypomagnesemia, because this receptor, in part, controls magnesium reabsorption in the kidney (21). This might reflect reduced magnesium tubular reabsorption, which may increase the tendency toward hypomagnesemia

in this form of hypoparathyroidism. In addition, the overall increased need for magnesium supplementation in the group with other etiologies for hypoparathyroidism may be because of a referral bias or due to the closer monitoring and higher attentiveness to this issue on the part of the investigators.

Previously reported data on the pharmacokinetics of PTH 1–34 have demonstrated maximal serum levels 30 min after sc injection in osteoporotic subjects (22). N-terminal PTH levels measured in hypoparathyroid patients from our previous study (our unpublished data) showed mean peak levels at 40 min after sc PTH injection (14). We also reported that circulating levels of 1,25 dihydroxyvitamin D peaked 8 h after PTH administration. Although these measures were not repeated in the present study group, one can assume a similar response. It is interesting to note that serum calcium levels rise approximately 3 h after the expected time of peak serum PTH level and 4 h before the maximal serum vitamin D level. The rise in serum calcium after PTH administration results from a 2-fold response. First, a rapid direct response at the level of the kidney to decrease calcium clearance and second, a more prolonged effect of endogenous calcitriol on the gastrointestinal tract to increase calcium absorption. We do not know whether mobilization of calcium from the bone contributes to the rise in serum calcium in response to PTH, because markers of bone turnover were not included in the 24-h serum profile.

The experimental design was not conducive to effectively measuring changes in bone in response to these two dose regimens. The relatively brief 14-week time period does not provide sufficient time to observe trends that might reflect the potential for long-term effects of these two dose regimens on the bone. Based on our experience, the decreased incidence of bone pain, the decrease in the levels of bone turnover markers during the twice-daily regimen, and the decreased incidence of hypercalcemia during twice-daily PTH, we hypothesize that twice-daily PTH is more physiological for the bone. There have been no clinical studies exploring the effect of multiple daily doses of PTH 1–34 on bone in humans with hypoparathyroidism. There are, however, animal studies that support our hypothesis that a shorter interval between doses of PTH 1–34 enhances the anabolic response in bone despite the administration of smaller doses (23). Numerous studies have shown that intermittent administration of PTH is more anabolic for the bone compared with continuous infusion of PTH (24, 25).

Nocturia, a side effect that appeared to differ according to the dose schedule, may have occurred more frequently during twice-daily PTH because of the relatively higher nighttime urine calcium levels during that treatment arm. Additionally, PTH is known to have natriuretic effects, which may explain nocturia after the second PTH dose. Although this adverse effect might be reduced by a different dose fractionation, such as giving 40% of the daily dose at night and 60% in the morning, this possibility has not yet been examined.

The total daily PTH dose required to maintain serum calcium in the normal or near-normal range was reduced by nearly 50% with twice-daily PTH. Patients with mild renal insufficiency might be expected to need more PTH than those with normal renal function (26). This does not appear to be

the case in our study. The subject (F) with the lowest glomerular filtration rate (GFR) required similar doses during once-daily PTH (3.4 $\mu\text{g}/\text{kg}$ per day) compared with subject B with normal GFR and malabsorption. Subject F, however, required higher doses during twice-daily PTH (0.95 $\mu\text{g}/\text{kg}$ per day) compared with subject B (0.66 $\mu\text{g}/\text{kg}$ per day). Another example, subject (E) with a GFR of 45 mL/min, required PTH doses of 4.2 $\mu\text{g}/\text{kg}$ per day during once-daily PTH and 1.8 $\mu\text{g}/\text{kg}$ per day during twice daily PTH. Her mother, (subject G) whose GFR was significantly worse (29 mL/min), required much lower doses: 0.7 $\mu\text{g}/\text{kg}$ per day during once-daily PTH and 0.4 $\mu\text{g}/\text{kg}$ per day during the twice daily PTH arm. Although renal function may be a contributing factor, gastrointestinal absorption of calcium may also be an important factor determining individual PTH requirements.

In conclusion, twice-daily PTH provides effective short-term treatment for hypoparathyroidism, with a markedly reduced total PTH dose, an apparent reduction in bone turnover, and a decreased incidence of bone pain compared with a once-daily regimen. Unresolved issues include the long-term effects of PTH therapy on bone density, renal complications, and the possible emergence of resistance to PTH. To address these longer term issues, we are currently conducting a randomized parallel study to compare the long-term outcome of PTH and calcitriol (both given twice daily) in the treatment of chronic hypoparathyroidism.

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