Calcemic Activity of 19-Nor-1,25(OH)$_2$D$_2$ Decreases with Duration of Treatment

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Abstract. 19-Nor-1,25(OH)$_2$D$_2$ (19-norD$_2$) has been shown to suppress parathyroid hormone effectively, but with lower calcemic activity than 1,25(OH)$_2$D$_3$. The present study investigated potential mechanisms to explain the reduced calcemic response to 19-norD$_2$. Tissue localization of [^{3}H]19-norD$_2$ or[^{3}H]1,25(OH)$_2$D$_3$ after a single injection was not different. Intestinal calcium absorption and bone mobilization, measured in vitamin D–deficient rats 24 h after single injections of 60 or 600 pmol of 19-norD$_2$ or 1,25(OH)$_2$D$_3$, were enhanced to a similar degree by the two compounds. However, when normal rats were treated every other day with 240 pmol of 19-norD$_2$ or 1,25(OH)$_2$D$_3$, increases in serum calcium were identical 24 h after the first injection but diverged thereafter with significantly lower serum calcium in the 19-norD$_2$–treated rats by 5 d. Intestinal calcium absorption and bone calcium mobilization were reassessed in vitamin D–deficient rats after seven daily injections of 600 pmol of 19-norD$_2$ or 1,25(OH)$_2$D$_3$, and both parameters were significantly lower in the 19-norD$_2$–treated rats. Pharmacokinetic analysis after seven daily injections of 600 pmol of 19-norD$_2$ or 1,25(OH)$_2$D$_3$ showed similar localization to the intestine and bone. In addition, intestinal vitamin D receptor levels were not different after 1 wk of treatment with 19-norD$_2$ or 1,25(OH)$_2$D$_3$. In conclusion, the low calcemic activity of 19-norD$_2$ seems to be due to an acquired, postreceptor resistance of the intestine and bone to chronic treatment with the analog.

Secondary hyperparathyroidism is a common occurrence in patients with chronic renal failure. The high levels of parathyroid hormone (PTH) in these patients produce a high rate of bone turnover and lead to increased fractures. The pathogenesis of the hyperparathyroidism has been attributed to retention of phosphate and the reduction in circulating 1,25(OH)$_2$D$_3$ in these patients, both of which produce a tendency toward hypocalcemia. Low calcium initially stimulates PTH synthesis and secretion, but chronic hypocalcemia induces parathyroid hyperplasia (1). In addition to the indirect control of PTH by its effect on serum calcium, 1,25(OH)$_2$D$_3$ can suppress PTH gene transcription (2,3). Therefore, the low levels of 1,25(OH)$_2$D$_3$ also lead to overexpression of the PTH gene, further exacerbating the hyperparathyroidism.

Correction of secondary hyperparathyroidism involves normalizing serum phosphate, usually with calcium-based phosphate binders that retard intestinal absorption of dietary phosphate, and restoration of the 1,25(OH)$_2$D$_3$ levels by replacement therapy. However, the potent calcemic actions of 1,25(OH)$_2$D$_3$ in the intestine and bone often produce hypercalcemia in renal patients, especially in those who receive oral calcium. To overcome this limitation of 1,25(OH)$_2$D$_3$ therapy, less-calcemic vitamin D analogs have been developed to retain the direct action of 1,25(OH)$_2$D$_3$ to suppress PTH gene expression. These include 19-nor-1,25(OH)$_2$D$_2$ (19-norD$_2$, or paricalcitol) (4–7), 22-oxacalcitriol (OCT) (8–10), and 1α(OH)D$_2$ (Hectorol, Bone Care International, Madison, WI) (11,12). 19-norD$_2$ and OCT have been shown to exert a selective action on PTH in animal models of renal failure, *i.e.*, suppression of PTH levels with less hypercalcemia (4,8).

The mechanisms by which these analogs exert this selectivity on the parathyroid glands are under investigation. The low calcemic activity of OCT seems to be due to its altered pharmacokinetics (13–16). Its low serum vitamin D binding protein (DBP) affinity leads to rapid clearance but greater tissue accessibility. The transient appearance of OCT in target tissues after injection elicits only short-lived effects on intestinal calcium absorption and bone mobilization but a prolonged suppression of PTH gene expression (16).

The mechanism for the selectivity of 19-norD$_2$ is not clear. This analog has been shown to be approximately 10 times less calcemic than 1,25(OH)$_2$D$_3$ in the rat (4), commensurate with a 10-fold lower potency in stimulating intestinal calcium transport and bone mobilization (17). In the present study, we investigated further the calcemic activities of 19-norD$_2$ in the intestine and bone. Our data indicate an induced or acquired resistance to 19-norD$_2$ with chronic treatment that cannot be attributed to pharmacokinetics.

Materials and Methods

Intestinal Calcium Transport and Bone Mobilization

Weanling male rats were maintained for 6 wk on a vitamin D–deficient diet containing 0.4% Ca and 0.3% P. Two d before treatment, the rats were placed on a vitamin D–deficient diet containing 0.02%

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Ca and 0.5% P. The rate of intestinal calcium transport was measured 24 h after the final injection of 1,25(OH)$_2$D$_3$ or 19-norD$_2$ by a modification of the isolated duodenal loop method (16). The first 6 cm of the small intestine distal to the pyloric sphincter were flushed twice with 12 ml of saline, tied off, and filled with 0.6 ml of transport buffer (30 mM Tris-HCl [pH 7.4], 10 mM fructose, 125 mM NaCl, 10 mM CaCl$_2$, 5 μCi/ml ^{45}CaCl$_2$). After 10 min, the rats were exsanguinated and the serum was analyzed for $^{45}Ca^{+2}$. In this protocol, the appearance of $^{45}Ca^{+2}$ in the blood increases linearly for at least 20 min; therefore, the 10-min time point represents a true rate of intestinal calcium absorption. The blood was also analyzed for total calcium. Because the rats were on a calcium-deficient diet, increases in serum calcium provide a measure of bone mobilization.

Pharmacokinetics

Normal male rats (250 g) were injected with [^{3}H]-1,25(OH)$_2$D$_3$ or [^{3}H]-19-norD$_2$ (600 pmol, 0.5 mCi) and killed by exsanguination at the specified times. The amount of tritiated parent compound remaining in the blood was determined by normal phase HPLC as described previously (16). The parathyroid glands, one kidney, bone marrow from one femur, and the mucosa of the first 8 cm of the small intestine were dissolved in tissue solubilizer (BTS-450, Beckman Instruments, Fullerton, CA), and the tritium was determined by liquid scintillation. The data are expressed as disintegrations per minute per gram of tissue.

Intestinal VDR Content

The first 6 cm of the duodenum was removed and flushed with saline, trimmed of mesentery, and slit lengthwise. The mucosa was isolated by scraping with a cold microscope slide and washed three times in cold phosphate-buffered saline containing 200 μg/ml soybean trypsin inhibitor. The mucosa was then homogenized in 10 mM Tris-HCl [pH 7.4], 1.5 mM ethylenediaminetetraacetate, 5 mM dithiothreitol, and 200 μg/ml soybean trypsin inhibitor and centrifuged at 100,000 $\times$ *g* for 60 min. Aliquots of the supernatant (100 ml, 100 μg protein) were incubated with [^{3}H]-1,25(OH)$_2$D$_3$ (2 nM final concentration) with or without 500 nM unlabeled 1,25(OH)$_2$D$_3$ for 16 h at 4°C. The samples were then mixed with charcoal/dextran, placed on ice for 15 min, and then centrifuged for 15 min at 2000 $\times$ *g*. The VDR-bound [^{3}H]-1,25(OH)$_2$D$_3$ in the supernatant was measured by liquid scintillation. Specific binding was determined by subtracting the nonspecific binding ([^{3}H]-1,25(OH)$_2$D$_3$ plus unlabeled 1,25(OH)$_2$D$_3$) from the total binding ([^{3}H]-1,25(OH)$_2$D$_3$ alone).

Statistical Analyses

Data are expressed as mean $\pm$ SD or SEM as denoted. Differences between experimental groups were determined by *t* test and by ANOVA as designated in the figure legends.

Results

Pharmacokinetics after Single Injections of 19-norD$_2$ and 1,25(OH)$_2$D$_3$

The low calcemic activity of many vitamin D analogs has been attributed to their altered pharmacokinetics. Therefore, we compared the pharmacokinetics of 19-norD$_2$ and 1,25(OH)$_2$D$_3$. The rate of appearance in the circulation and peak levels achieved were nearly identical for 19-norD$_2$ and 1,25(OH)$_2$D$_3$ (Figure 1). 19-NorD$_2$ seemed to be cleared from the circulation slightly faster than 1,25(OH)$_2$D$_3$. The time courses for appearance of [^{3}H]19-norD$_2$ and [^{3}H]1,25(OH)$_2$D$_3$ in parathyroid glands, kidney, intestine, and bone marrow were very similar (Figure 2), although greater peak accumulation of 1,25(OH)$_2$D$_3$ than 19-norD$_2$ was observed in the parathyroid glands. These findings indicated that the low calcemic activity of 19-norD$_2$ could not be attributed to rapid clearance or to reduced accessibility to the intestine and bone.

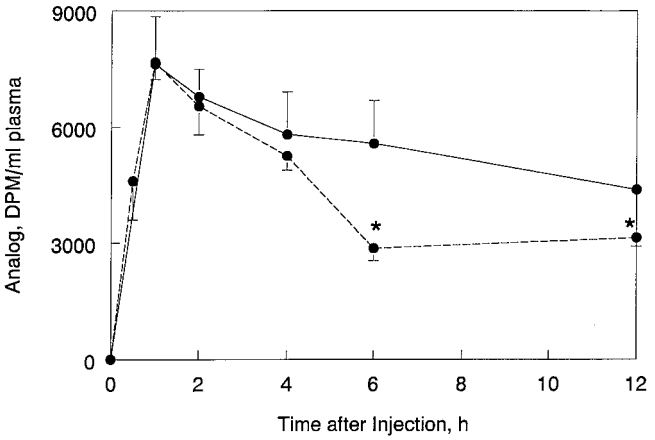

Figure 1. Plasma levels of [^{3}H]1,25(OH)$_2$D$_3$ and [^{3}H]19-norD$_2$ after a single intraperitoneal injection. Normal rats were injected with 0.5 mCi of [^{3}H]1,25(OH)$_2$D$_3$ (solid line) or [^{3}H]19-norD$_2$ (dashed line), and blood samples taken at the specified times were analyzed by HPLC for the parent tritiated compound. Mean $\pm$ SD (n = 4). *$P <$ 0.05 19-norD$_2$ *versus* 1,25(OH)$_2$D$_3$ by *t* test.

Calcemic Activities after Single Injections of 19-norD$_2$ and 1,25(OH)$_2$D$_3$

To examine further the calcemic actions of 19-norD$_2$ in these tissues, we measured the effects of the compounds on intestinal calcium transport and bone calcium mobilization using the vitamin D–deficient rat model. The lower basal rate of calcium transport in vitamin D–deficient rats permits a better assessment of stimulation by vitamin D compounds. Two d before treatment, the vitamin D–deficient rats were placed on a vitamin D–deficient, calcium-deficient diet, which allowed for the assessment of bone resorption by the increase in serum calcium. The rats were given single intraperitoneal injections of 19-norD$_2$ or 1,25(OH)$_2$D$_3$ at doses of 60 or 600 pmol. After 24 h, intestinal calcium transport was measured by the isolated duodenal loop method in which ^{45}Ca is introduced into the duodenal loop and ^{45}Ca uptake into the blood is measured 10 min later. As shown in Figure 3, after a single intraperitoneal injection, 19-norD$_2$ and 1,25(OH)$_2$D$_3$ elicited similar dose-dependent increases in calcium transport. Increments in serum calcium were also similar (Figure 4), indicating that 19-norD$_2$ was as potent as 1,25(OH)$_2$D$_3$ in stimulating bone resorption.

Calcemic Activities of 19-norD$_2$ and 1,25(OH)$_2$D$_3$: 7-D Time Course

The equivalent calcemic activities of 19-norD$_2$ and 1,25(OH)$_2$D$_3$ after a single injection seemed to disagree with the lower calcemic activity of 19-norD$_2$ observed in other,

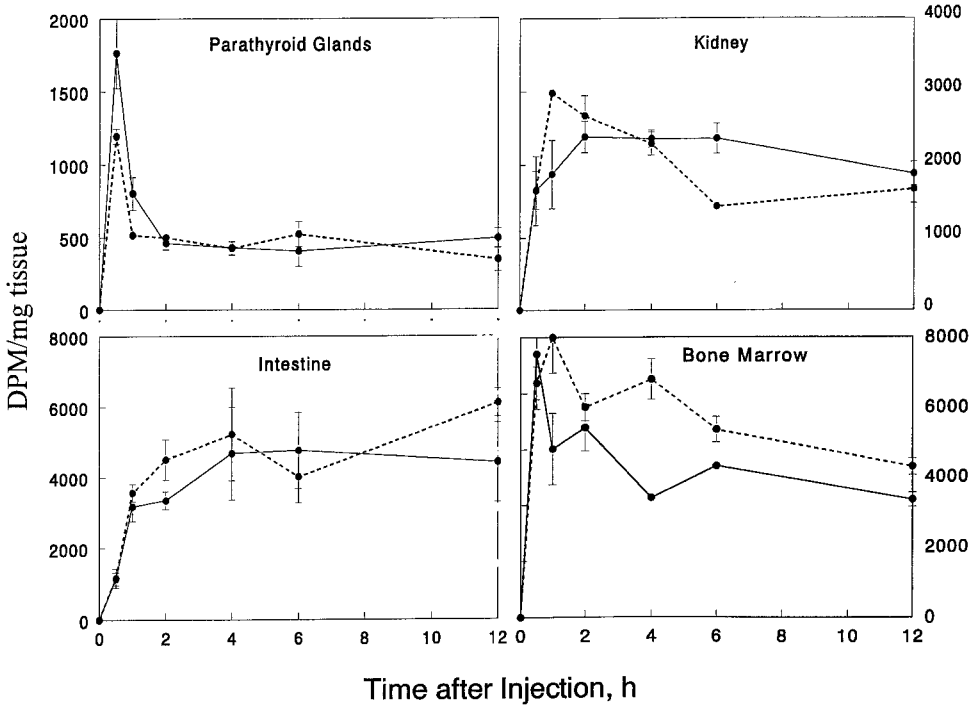

Figure 2. Tissue localization of [^{3}H]1,25(OH)$_2$D$_3$ and [^{3}H]19-norD$_2$ after a single intraperitoneal injection. Normal rats were injected with 0.5 mCi of [^{3}H]1,25(OH)$_2$D$_3$ (solid lines) or [^{3}H]19-norD$_2$ (dashed lines). Tissue samples obtained at the specified times were dissolved in tissue solubilizer, and the tritium content was determined as described in the Materials and Methods section. Mean ± SD (n = 4).

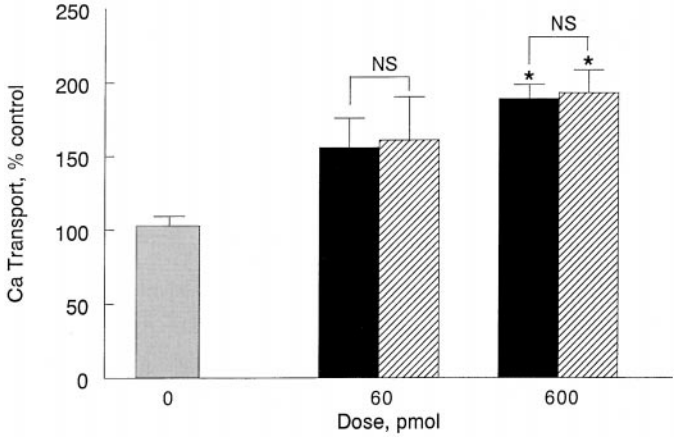

Figure 3. Intestinal calcium transport after a single intraperitoneal injection of 1,25(OH)$_2$D$_3$ or 19-norD$_2$. Vitamin D–deficient rats were injected intraperitoneally with vehicle (▨) or the specified dose of 1,25(OH)$_2$D$_3$ (■) or 19-norD$_2$ (▨). After 24 h, the rate of calcium transport was determined by the *in situ* duodenal loop method. Data are expressed as mean ± SEM (n = 6). *P < 0.05 *versus* untreated rats.

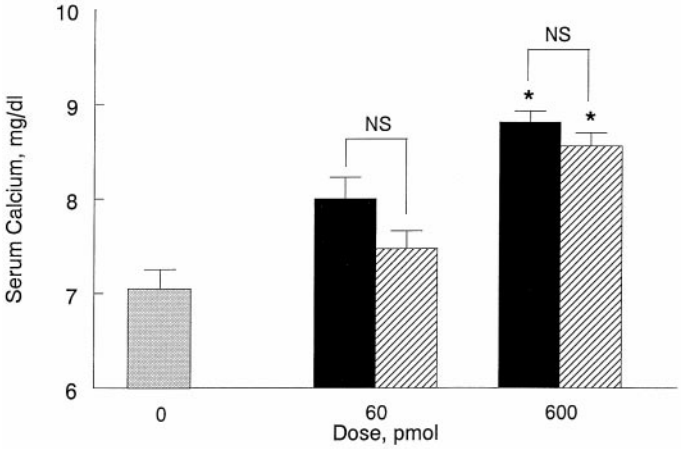

Figure 4. Bone calcium mobilization after a single intraperitoneal injection of 1,25(OH)$_2$D$_3$ or 19-norD$_2$. Vitamin D–deficient rats were fed a vitamin D– and calcium-deficient diet for 2 ds and then injected intraperitoneally with vehicle (▨) or the specified dose of 1,25(OH)$_2$D$_3$ (■) or 19-norD$_2$ (▨). Serum calcium was measured 24 h later as an assessment of bone mobilization. Data are expressed as mean ± SD (n = 6). *P < 0.05 *versus* untreated rats.

more chronic studies. Therefore, a detailed time course of the calcemic response to 19-norD$_2$ was performed. Normal rats received intraperitoneal injections of 240 pmol of 19-norD$_2$ or 1,25(OH)$_2$D$_3$ every other day for 7 d. Serum calcium was measured 24 h after each injection. Figure 5 shows that 19-norD$_2$ and 1,25(OH)$_2$D$_3$ produced the same increment in serum calcium after the first injection. However, with subsequent injections, a divergence in the serum calcium curves was observed. By 5 and 7 d, the serum calcium levels were significantly different.

Calcemic Activities after Seven Daily Injections of 19-norD$_2$ and 1,25(OH)$_2$D$_3$

To determine whether the divergence in the calcemic response is due to differential effects of 19-norD$_2$ and

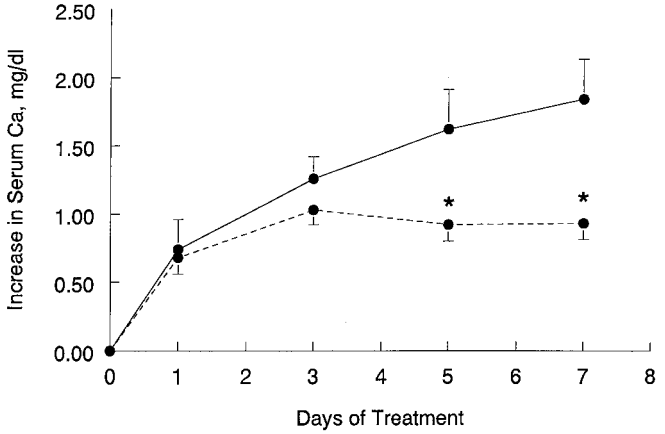

Figure 5. Divergence of the calcemic activities of 1,25(OH)$_2$D$_3$ and 19-norD$_2$ with time. Normal rats were given daily injections of 240 pmol of 1,25(OH)$_2$D$_3$ (solid line) or 19-norD$_2$ (dashed line). Blood was taken from the tail vein, and total serum calcium was measured. Data are expressed as the increase in serum calcium over day 0. Mean ± SD (n = 4). *P < 0.05 19-norD$_2$– *versus* 1,25(OH)$_2$D$_3$-treated rats.

1,25(OH)$_2$D$_3$ on intestinal calcium transport and/or bone mobilization, vitamin D–deficient rats were placed on a vitamin D– and calcium-deficient diet and given seven daily intraperitoneal injections of 600 pmol of 19-norD$_2$ or 1,25(OH)$_2$D$_3$. Intestinal calcium transport was assayed 24 h after the final injection, and serum calcium was measured as a cumulative assessment of bone mobilization. With 7 d of treatment, 19-norD$_2$ elicited smaller increases in intestinal calcium transport and bone mobilization (Figure 6) than 1,25(OH)$_2$D$_3$.

Pharmacokinetics after Seven Daily Injections of 19-norD$_2$ and 1,25(OH)$_2$D$_3$

The reason for the lower calcemic activity of 19-norD$_2$ with chronic treatment was unclear, but a change in pharmacokinetics was a possibility. Therefore, normal rats were given daily intraperitoneal injections of 600 pmol of 19-norD$_2$ or 1,25(OH)$_2$D$_3$ for 7 d. On the last day, the dose was spiked with 0.5 mCi of the tritiated form of each compound. Serum levels and tissue accumulation of the tritium was determined over a 24-h period. Figure 7 shows that 19-norD$_2$ was cleared slightly faster than 1,25(OH)$_2$D$_3$, as seen after a single injection (Figure 1). However, Figure 8 shows that the time courses of localization of the two compounds to the intestine and bone were similar, despite the different calcemic activities. Thus, the lower calcemic actions of 19-norD$_2$ in the intestine and bone after chronic treatment could not be attributed to a change in its pharmacokinetics.

Intestinal VDR Levels after 7-D Treatment with 19-norD$_2$ or 1,25(OH)$_2$D$_3$

At least one explanation for the decreased response to 19-norD$_2$ with chronic treatment is that the analog downregulates the VDR. To assess this possibility, we measured the intestinal VDR content in the normal rats that received 240 pmol of 19-norD$_2$ or 1,25(OH)$_2$D$_3$ every other day for 1 wk (see Figure 5). The levels of intestinal VDR, assessed by binding assay, were not different for the 19-norD$_2$–treated and 1,25(OH)$_2$D$_3$-treated rats (203 ± 38 *versus* 183 ± 18 fmol/mg protein, respectively). Thus, the altered response to 19-norD$_2$ did not seem to involve changes in the VDR levels.

Discussion

Vitamin D analogs with more desirable biologic profiles than that of 1,25(OH)$_2$D$_3$ are under development for the treatment of various clinical disorders (18). Three analogs have now been approved for secondary hyperparathyroidism in patients with renal failure: 19-norD$_2$ and 1α-(OH)D$_2$ in the

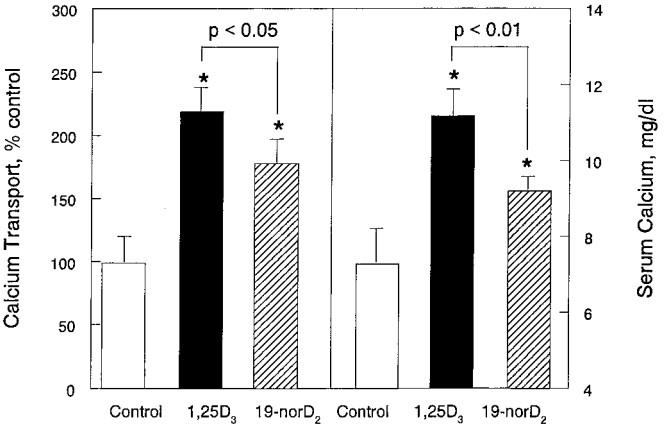

Figure 6. Intestinal calcium transport and bone mobilization after 7 d of treatment. Vitamin D–deficient rats on a calcium-deficient diet were given seven daily injections of vehicle (□) or 600 pmol of 1,25(OH)$_2$D$_3$ (■) or 19-norD$_2$ (▨). Intestinal calcium transport was measured 24 h after the last injection, and the serum calcium was measured as an assessment of bone mobilization. Mean ± SEM (n = 12). *P < 0.05 *versus* untreated rats.

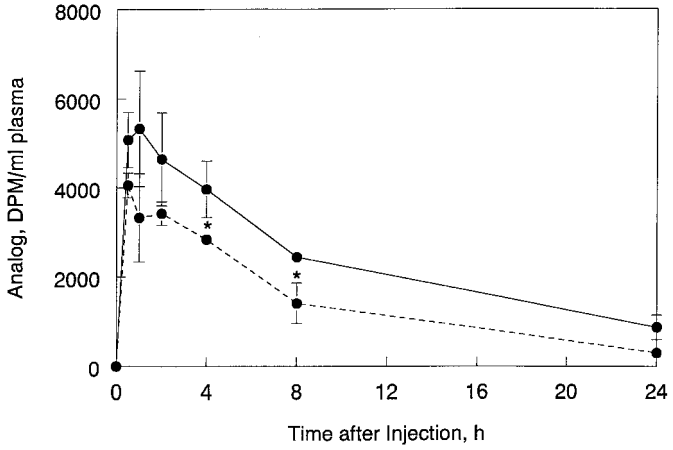

Figure 7. Plasma clearance of [^{3}H]1,25(OH)$_2$D$_3$ and [^{3}H]19-norD$_2$ after 7 d of treatment with 1,25(OH)$_2$D$_3$ or 19-norD$_2$. Normal rats were given six daily injections of 600 pmol of 1,25(OH)$_2$D$_3$ (solid line) or 19-norD$_2$ (dashed line). A final 600-pmol injection of each compound contained 0.5 mCi of tritiated tracer. Blood samples taken at the specified times were analyzed by HPLC for the parent tritiated compound. Mean ± SD (n = 4). *P < 0.05 19-norD$_2$ *versus* 1,25(OH)$_2$D$_3$ by t test.

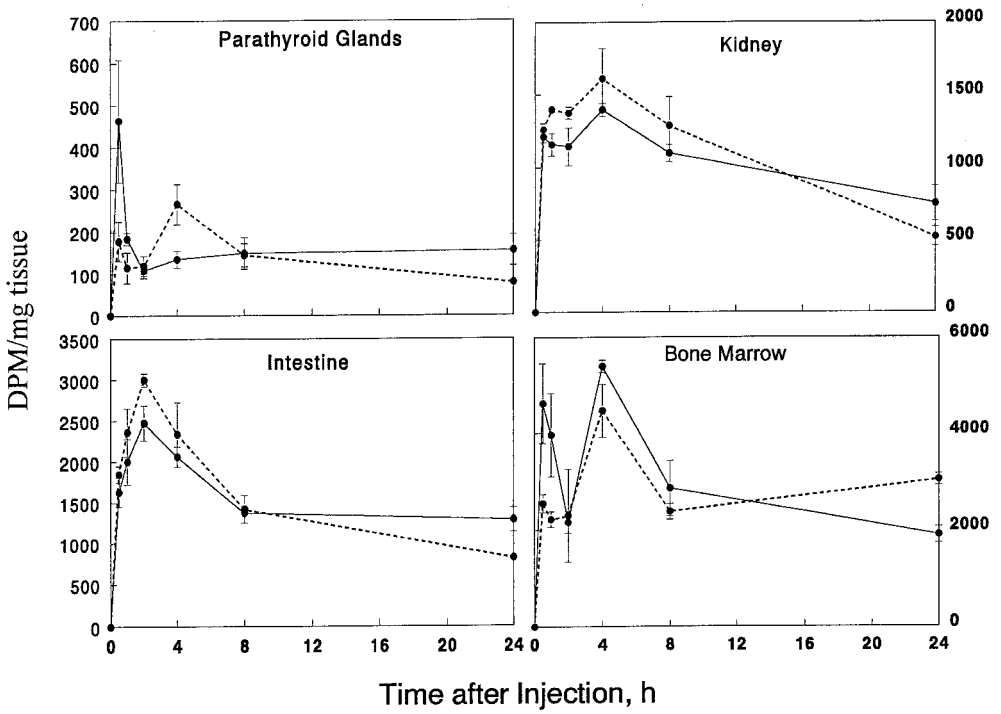

Figure 8. Tissue localization of [^{3}H]1,25(OH)$_2$D$_3$ and [^{3}H]19-norD$_2$ after 7 d of treatment with 1,25(OH)$_2$D$_3$ or 19-norD$_2$. Normal rats were given six daily injections of 600 pmol of 1,25(OH)$_2$D$_3$ (solid lines) or 19-norD$_2$ (dashed lines). A final 600-pmol injection of each compound contained 0.5 mCi of tritiated tracer. Tissue samples obtained at the specified times were dissolved in tissue solubilizer, and the tritium content was determined as described in the Materials and Methods section. Mean ± SD.

United States and OCT in Japan. The selectivity of these compounds on the parathyroid glands seems to be due to their lower calcemic activity rather than to enhanced potency to suppress PTH.

The mechanisms that are responsible for the low calcemic activity of these vitamin D analogs are under investigation. The best characterized analog to date is OCT. This compound has a 500-fold lower affinity than 1,25(OH)$_2$D$_3$ for the serum vitamin D binding protein, leading to more rapid clearance and shorter residence time in target tissues (13,14,16). At the same time, the decreased interaction with DBP allows greater accessibility to target cells, which results in higher peak tissue levels (14,16). The shorter residence time of OCT in the target tissue produces a more transient stimulation of intestinal calcium transport and bone mobilization than 1,25(OH)$_2$D$_3$ (16). However, despite the more rapid disappearance of OCT from the parathyroid glands (14), this analog elicits a prolonged suppression of PTH (19). Altered pharmacokinetics likely play a key role in the low calcemic activities of other vitamin D analogs with low DBP affinity, such as calcipotriene (Dovonex, Leo Pharmaceutical Products, Ballerup, Denmark), which is approved for the treatment of psoriasis (20,21).

The present study demonstrates that the lower calcemic activity of 19-norD$_2$ cannot be attributed to altered pharmacokinetics. The DBP affinity of 19-norD$_2$ is only three times lower than that of 1,25(OH)$_2$D$_3$, and its clearance rate and tissue localization are not different from that of 1,25(OH)$_2$D$_3$. This is true with both acute and chronic administration of the analog. The similar effects on intestinal calcium transport and bone mobilization after a single intraperitoneal injection of 19-norD$_2$ and 1,25(OH)$_2$D$_3$ were consistent with the nearly identical localization of the compounds in intestine and bone. However, despite similar pharmacokinetics at the end of 7 d of treatment, 19-norD$_2$ elicited smaller increases in intestinal calcium transport and bone mobilization than 1,25(OH)$_2$D$_3$.

These findings suggest that with chronic treatment, the responses to 19-norD$_2$ in the intestine and bone are diminished. The mechanism for this apparent resistance is unclear. One possibility is that chronic treatment induces rapid intracellular catabolism of 19-norD$_2$ in the intestine and bone, thereby reducing the availability of the analog to the vitamin D receptor. In fact, we have noted that 19-norD$_2$ is catabolized slightly more rapidly than 1,25(OH)$_2$D$_3$ in primary cultures of mouse bone marrow (22), but the difference is small and probably cannot fully account for the differential effects of 19-norD$_2$ and 1,25(OH)$_2$D$_3$ in that system. Determining the relative rates of catabolism of 19-norD$_2$ and 1,25(OH)$_2$D$_3$ in target tissues *in vivo* is very difficult, if not impossible. However, analysis of the tritium present in the intestine revealed very little tritiated metabolites of either compound; greater than 90% of the tritium was parent 19-norD$_2$ or 1,25(OH)$_2$D$_3$, indicating that the tissue content of the injected compounds was not different.

Other possible explanations for the apparent induced resistance to 19-norD$_2$ were investigated. VDR content was not differentially affected by 7 d of administration of 19-norD$_2$ *versus* 1,25(OH)$_2$D$_3$. A previous study in which 19-norD$_2$ and

1,25(OH)$_2$D$_3$ were administered for 2 mo demonstrated lower intestinal VDR content in the 19-norD$_2$–treated uremic rats (23). However, this difference was not evident after 1 wk in the present study. VDR functions as a heterodimer with retinoid X receptor (RXR) (24). It is not known whether 19-norD$_2$ downregulates RXR, and there is no evidence that vitamin D compounds regulate RXR expression. Some vitamin D analogs have been shown to bind to the VDR in a different manner than 1,25(OH)$_2$D$_3$, inducing in the receptor unique conformations that likely affect the function of the receptor (25–28). Whether 19-norD$_2$ produces an altered VDR conformation has not been investigated, but the similar activities of 19-norD$_2$ and 1,25(OH)$_2$D$_3$ *in vitro* and *in vivo* after a single injection make this possibility unlikely.

We previously showed that chronic treatment with 19-norD$_2$ leads to a decrease in endogenous 1,25(OH)$_2$D$_3$ levels, presumably through effects on both synthesis and degradation of the natural vitamin D hormone. Although the time course for this suppression of endogenous 1,25(OH)$_2$D$_3$ by 19-norD$_2$ has not been examined, earlier studies with the analog OCT demonstrated that the decrease was evident by 24 to 48 h after injection (29). Thus, endogenous 1,25(OH)$_2$D$_3$ levels in the 19-norD$_2$–treated rats would be expected to decrease just before the divergence of the calcemic actions of 19-norD$_2$ and 1,25(OH)$_2$D$_3$ (Figure 5). In addition, the low levels of 1,25(OH)$_2$D$_3$ in the vitamin D–deficient rats would be expected to fall even further with 19-norD$_2$ treatment. Our observation that the calcemic activities of 19-norD$_2$ in the intestine and bone decrease with chronic administration would be consistent with the hypothesis that 19-norD$_2$ is not capable of supporting all of the actions of 1,25(OH)$_2$D$_3$ required for stimulation of intestinal calcium transport and bone mobilization. 19-norD$_2$ is known to bind well to the VDR and to mimic all of the genomic responses of 1,25(OH)$_2$D$_3$, but its nongenomic activity has not been tested. It is possible that depletion of endogenous 1,25(OH)$_2$D$_3$ levels reduces the activation of the membrane receptor for 1,25(OH)$_2$D$_3$ (30–32), thus reducing the activity of key pathways required for the calcemic responses in the bone and intestine. The apparent lack of a requirement of the nongenomic actions to suppress PTH may explain the therapeutic advantage of 19-norD$_2$ in the treatment of secondary hyperparathyroidism. Additional studies are necessary to test this hypothesis.

The lack of a role for pharmacokinetics in the low calcemic activity of chronically administered 19-norD$_2$ suggests the possibility that the disparate calcemic actions of 19-norD$_2$ and 1,25(OH)$_2$D$_3$ observed *in vivo* may be reproduced *in vitro*. Recent data obtained in mouse bone marrow cultures indicated that 19-norD$_2$ was less active than 1,25(OH)$_2$D$_3$ in stimulating *in vitro* bone resorption (22), and this differential effect seemed to be dependent on time of incubation. Determining whether the mechanism for the reduced bone resorption by 19-norD$_2$ *in vitro* is responsible for the diminished resorbing activity of the analog *in vivo* will require a clearer understanding of the factors involved.

In summary, we found that acute administration of 19-norD$_2$ produces the same calcemic responses in the intestine and bone as 1,25(OH)$_2$D$_3$ but that with more chronic treatment, 19-norD$_2$ becomes less calcemic than 1,25(OH)$_2$D$_3$. The mechanism that is responsible for this seeming induced resistance to the analog is not clear but seems not to involve pharmacokinetics or lower levels of the VDR. Therefore, it is important to establish which target tissues and genes may be differentially affected by acute *versus* chronic treatment with 19-norD$_2$. However, the high activity of 19-norD$_2$ in the parathyroid glands with chronic treatment allows this vitamin D analog to suppress PTH selectively, providing a new agent for the treatment of secondary hyperparathyroidism in patients with chronic renal failure.

Acknowledgments

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