Calcium absorption and excretion in patients treated with verapamil

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1 The effect of verapamil on the intestinal absorption of calcium was studied using a single isotope technique. Serum calcium and urinary excretion of calcium in the urine were followed in nine patients during treatment with verapamil for 2 months.

2 A dose of 80–120 mg (three times daily) resulted in a significant reduction of systolic and diastolic blood pressure.

3 There was no change in the intestinal calcium absorption (α) expressed as the fraction of given activity absorbed per hour ($\alpha = 0.82 \pm 0.19$ vs $\alpha = 0.83 \pm 0.25$; $\pm$ s.d., NS) or of the excretion of calcium in urine (Ca/creatinine ratio 0.35 ± 0.20 vs 0.31 ± 0.33 ; NS).

4 Serum calcium was not significantly different before and during treatment ($2.43 \text{ mmol l}^{-1} \pm 0.10$ vs $2.47 \text{ mmol l}^{-1} \pm 0.14$; NS).

5 This study demonstrates that verapamil, at doses recommended for clinical use, does not seem to affect the intestinal absorption of calcium, the serum calcium concentration or the excretion of calcium in urine.

Keywords calcium absorption calcium excretion hypertension
intestinal absorption verapamil

Introduction

Verapamil is a calcium channel blocker extensively used in the treatment of cardiac arrhythmias, angina pectoris and essential hypertension. Experiments on rats have shown that verapamil reduces the *in vitro* intestinal uptake of calcium (Wrobel & Michalska, 1977; Sjöden *et al.*, 1983; Pento & Johnson, 1983), while the intestinal absorption of calcium *in vivo* is enhanced (Goligorsky *et al.*, 1984). Because of the lack of knowledge about the effect of verapamil on human calcium homeostasis, this study was undertaken to establish whether verapamil affects calcium absorption and excretion in patients.

Methods

Subjects and study design

Ten patients with mild to moderate hypertension (WHO Stage I–II) or moderate angina pectoris were admitted to the study. One female patient failed to complete the study because of complaints about side effects (malaise and constipation). Therefore nine patients (seven males and two females, mean age 59 years, range 45–75 years) completed the study and are included in this report. For details concerning the individual patients see Table 1. The patients were either newly diagnosed or considered poorly controlled with existing treatment, or were having side

Table 1 Clinical characteristics of the patients. Their previous treatments were discontinued 3 weeks before starting the present study

Patient	Sex	Age (years)	Diagnosis	Previous treatment
1	M	59	Essential hypertension	—
2	M	62	Essential hypertension	—
3	M	63	Essential hypertension Rheumatoid arthritis	—
4	M	45	Essential hypertension	—
5	M	63	Essential hypertension	Prazosin
6	M	53	Essential hypertension	—
7	M	48	Angina pectoris	Nitroglycerin
8	F	75	Angina pectoris Hypertension NUD	Nitroglycerin Thiazides
9	F	65	Hypertension NUD	Thiazides

effects with their present medication. None of the patients had previously been treated with calcium blockers.

Patients with conditions known to influence calcium homeostasis and intestinal calcium absorption such as intestinal malabsorption, previous intestinal resection, hyperthyroidism, hyperparathyroidism, renal dysfunction, sarcoidosis, epilepsy or steroid treatment were excluded.

This study was approved by the Ethics Committee at the Karolinska Institute, Huddinge Hospital, and informed consent was obtained from all patients.

After a period of at least 3 weeks without drug treatment, serum and single urine specimens were obtained from the patients after overnight fast. The intestinal calcium absorption tests were performed in the morning hours starting at 08.30–09.00 h with the patients fasting for at least 12 h. The patients were then given verapamil (Isoptin® Knoll), at a dose of 80 mg, three times daily. After 2 weeks, the blood-pressure was reexamined and the dose adjusted if necessary to 120 mg three times daily. The treatment was maintained for 2 months at what time blood and single urine specimens were collected after overnight fast. The patients were allowed to take their morning dose of verapamil together with a glass of water 1 h before starting the calcium absorption test. The intestinal absorption tests were again started at the same time between 08.30–9.00 h.

Laboratory technique

The intestinal absorption of ^{45}Ca was measured

after oral administration of 250 ml of 2.0 mM Ca^{2+} solution (CaCl_2) containing 5 μCi ^{45}Ca . Blood was drawn at regular intervals 0–120 min after ingestion of the solution and the activity of ^{45}Ca was determined in duplicate with a liquid scintillation counter (Pye-Unicam 2900). By means of computer analysis the hourly fractional rate of radiocalcium absorption (α) was calculated (Lindgren *et al.*, 1978). The mathematical model calculating α was adopted from Marshall & Nordin (1969).

S-Ca, s-P, serum alkaline phosphatase (ALP), serum creatinine, u-Ca, u-P and urinary creatinine were measured by standard automated techniques (Gindler & King, 1972; Bartels & Roijers, 1975; Anon, 1974; Bergman & Öhman, 1980) and serum immunoreactive parathyroid hormone (iPTH) by radioimmunoassay directed towards the midportion molecule (PTH Omega 53–68, Cambridge Medical Diagnostics Inc, USA). Urinary hydroxyproline (HPL) was measured by acid cation resin exchange (Hypronosticon®, Organon Teknika, 2300 Turnhout, Belgium) and urinary cAMP was measured by competitive protein binding (Amersham International, Amersham, UK). To determine patient compliance, the concentrations of verapamil and norverapamil in serum were measured by gas chromatography (Anderson *et al.*, 1986).

Statistics

For statistical analysis of differences between groups, two-tailed paired *t*-tests were performed. Statistical significance was set at $P < 0.05$.

Results

Treatment with verapamil for 2 months resulted in significant reductions of supine systolic (157 ± 11 mm Hg vs 147 ± 16 mm Hg, $P < 0.05$) and diastolic (102 ± 10 mm Hg vs 88 ± 5 mm Hg, $P < 0.001$) blood-pressure showing that adequate doses were used. This effect was also present after 2 weeks of treatment. The serum creatinine was not affected by verapamil treatment (Table 2). The serum calcium (Table 2) and the urinary calcium excretion (Table 3), as well as serum and urinary phosphate, were unaffected by verapamil treatment. The serum iPTH level increased slightly during treatment as did the excretion of cAMP in urine, but these increases were not statistically significant (Tables 2 and 3). Alkaline phosphatase and urinary hydroxyproline also increased slightly but insignificantly (Tables 2 and 3).

The average hourly fractional rate of radio-calcium absorption (α) did not change with verapamil treatment (Table 4). However, individual patients showed variable responses (Figure 1). Thus although the serum levels of verapamil (293.3 ± 233.4 ng ml⁻¹) and its metabolite nor-verapamil (230.1 ± 180.7 ng ml⁻¹) were well within the therapeutic range (Storstein *et al.*, 1981), no consistent change of calcium absorption was noted.

Discussion

In view of the fact that verapamil has pronounced effects on cardiac and smooth muscle cells (Singh

Table 4 Effect of verapamil on the intestinal absorption of calcium (α)

Treatment	α
-Verapamil	0.82 ± 0.19
+Verapamil	0.83 ± 0.25

α is expressed as the fraction of the given activity absorbed per hour.

Figure 1 The intestinal absorption of calcium (α) before and after 2 months of treatment with verapamil. α is expressed as the fraction of the given activity absorbed per hour.

et al., 1978) as well as on hormonal secretion (Rubin *et al.*, 1981), and that animal data indicated an inhibition of the intestinal calcium absorption (Sjödén *et al.*, 1983), it was important to determine the effect on human calcium balance.

Table 2 Effect of verapamil on serum variables.

Treatment	S-Ca (mmol l ⁻¹)	S-P (mmol l ⁻¹)	S-iPTH (μ g l ⁻¹)	S-ALP (μ kat l ⁻¹)	S-creatinine (μ mol l ⁻¹)
-Verapamil	2.43 ± 0.10	0.94 ± 0.18	1.22 ± 1.05	2.98 ± 1.15	89.5 ± 15.3
+Verapamil	2.47 ± 0.14	0.96 ± 0.19	1.37 ± 0.51	3.50 ± 1.18	91.0 ± 13.3
	NS	NS	NS	NS	NS

Values are given as means $\pm$ s.d.

Table 3 Effect of verapamil on urinary variables

Treatment	U-Ca/ creatinine (mmol mmol ⁻¹)	U-P/ creatinine (mmol mmol ⁻¹)	U-HPL/ creatinine (μ mol mmol ⁻¹)	U-cAMP/ creatinine (nmol mmol ⁻¹)
-Verapamil	0.35 ± 0.20	1.29 ± 0.48	11.4 ± 4.1	3.8 ± 2.9
+Verapamil	0.31 ± 0.33	1.34 ± 0.75	14.1 ± 5.3	5.2 ± 2.4
	(9) NS	(9) NS	(8) NS	(7) NS

Values given as means $\pm$ s.d. Number within brackets = *n*.

We have chosen to study the intestinal calcium absorption by using the ^{45}Ca isotope technique. This procedure has been described previously (Marshall & Nordin, 1969). In short labelled calcium is assumed to be absorbed from the intestine at a rate α , proportional to the residual concentration. The labelled calcium is mixed with the calcium in plasma and extracellular fluid, from which it is removed into bone and excreted into the urine and faeces. Fairly accurate estimates of the intestinal calcium absorption from simple analysis of plasma radioactivity measurements can be done. Bullamor *et al.* (1970) found a correlation coefficient of 0.93 between true calcium absorption measured with calcium balance technique and calcium absorption estimated using ^{45}Ca isotope technique. The reproducibility of the ^{45}Ca intestinal absorption test (1 s.d.) was $\pm 12\%$ (Nordin *et al.*, 1985).

It has also been shown that α rapidly increased following doses with $1\alpha\text{OHD}_3$ (Peacock *et al.*, 1974). In the same patients, the intestinal absorption of calcium returned almost to normal after a few days without treatment. Therefore comparisons of α in the same individual seems valid if experimental circumstances are well controlled.

The patients in this study were told not to change their dietary habits. No change in serum creatinine was noted and all creatinine values were in the normal range.

The study period of 2 months was chosen so that enough time was allowed to have the patients in steady state conditions and so that blood pressure regulation was stable. Considerations were furthermore given to have the study period short enough so that seasonal variations in vitamin D status were minimized.

The present study demonstrated that with 2 months treatment of verapamil in doses that significantly reduced BP no inhibition of the intestinal absorption of calcium could be demonstrated. There were also no significant changes of serum and urinary calcium or serum and urinary phosphorous during verapamil treatment.

The finding that calcium absorption was unchanged could have two explanations; either the calcium absorption is not influenced by verapamil or else calcium absorption is impaired by verapamil but secondary hyperparathyroidism follows and normalizes calcium absorption. We found that the mean value for serum iPTH in-

creased as did the urinary excretion of cAMP. These values were not statistically different from pre-treatment values but, nevertheless, the observed trends may be of biological significance. On the other hand no change in s-Ca was noted as would have been expected in the case of secondary hyperparathyroidism. It is however important to recognize that since our study consists of only nine individuals, the small sample size might lead to a statistical type II error, i.e. a significant change might not be detected due to the small sample number (Freiman *et al.*, 1978).

We also found an increase in the mean values for ALP and u-HPL, although these values did not differ significantly from pre-treatment levels. ALP is considered to reflect the activity of the osteoblasts on bone formation, while u-HPL is a marker of bone resorption by the osteoclasts (Nordin, 1984). Our results may indicate an increase in bone turnover by verapamil.

The effect on bone tissue could be due to an enhancement of PTH production which in turn would increase the production of $1.25(\text{OH})_2\text{D}_3$, the metabolically active form of vitamin D_3 (Sjödén, 1985). *In vitro* Sommerville & Keye (1984) found that verapamil inhibited the release of calcium from bone, although this seemed to be due to an inhibition of cell calcium transport rather than to a decrease of bone resorption.

With age, the intestinal absorption of calcium declines (Bullamor *et al.*, 1970) and this fact has been suggested as one cause for the occurrence of senile osteoporosis. A negative calcium balance and reduced levels of $1.25(\text{OH})_2\text{D}_3$ have been reported in postmenopausal osteoporosis (Gallagher *et al.*, 1979). During the last decades the number of hip fractures in Sweden (Johnell *et al.*, 1984) and other countries in western Europe (Baker, 1980) has increased more than the age-related increase. The intake of various drugs has been discussed as a risk factor for hip fracture. A recent study indicated however that drugs were not involved in the rising incidence of hip fracture (Rashiq & Logan, 1986).

Verapamil has been shown to inhibit the intestinal uptake of calcium *in vitro*. However verapamil does not decrease the intestinal absorption of calcium in humans. This calcium transport blocker therefore seems safe in patients as calcium balance is not negatively affected by verapamil. However, further studies on the effects by verapamil on bone metabolism may be warranted.

References

- Anderson, P., Bondeson, U. & de Faire, U. (1986). Pharmacokinetics of verapamil in patients with hypertension. *Eur. J. clin. Pharmac.*, **31**, 155–163.
- Anon. (1974). Recommended methods for the determination of four enzymes in blood. *Scand. J. clin. lab. Invest.*, **33**, 291–306.
- Baker, M. R. (1980). An investigation into the secular trends in the incidence of femoral neck fracture using Hospital Activity Analysis. *Publ. Health London*, **94**, 368–374.
- Bartels, P. C. & Roijers, A. F. M. (1975). A kinetic study on the influence of the parameters in the determination of inorganic phosphate by the molybdenum blue reaction. *Clin. Chim. Acta.*, **61**, 135–144.
- Bergman, A. & Öhman, G. (1980). Effect of detergent on kinetic Jaffé-method assay of creatinine. *Clin. Chim. Acta*, **26**, 1729–1732.
- Bullamore, J. R., Wilkinson, R., Gallagher, J. C., Nordin, B. E. C. & Marshall, D. H. (1970). Effect of age on calcium absorption. *Lancet*, **ii**, 535–537.
- Freiman, J. A., Chalmers, T. C., Smith, H. & Kuebler, R. R. (1978). The effect of beta, the type II error and sample size in the design and interpretation of the randomized control trial. Survey of 71 'negative' trials. *New Engl. J. Med.*, **299**, 690–694.
- Gallagher, J. C., Riggs, B. L., Eisman, J., Hamstra, A., Arnaud, S. A. & DeLuca, H. F. (1979). Intestinal calcium absorption and serum vitamin D metabolites in normal subjects and osteoporotic patients. *J. clin. invest.*, **64**, 729–736.
- Gindler, E. M. & King, J. D. (1972). Rapid colorimetric determination of calcium in biologic fluids with methylthymol blue. *Am. J. clin. Path.*, **58**, 376–382.
- Goligorsky, M. S., Chaimowitz, C., Shany, S., Rapoport, J. & Sharony, Y. (1984). Verapamil improves defective intestinal calcium absorption in uremia. *Adv. exp. Biol. Med.*, **178**, 153–161.
- Johnell, O., Nilsson, B., Obrant, K. & Sernbo, I. (1984). Age and sex patterns of hip fracture—changes in 30 years. *Acta orthop. Scand.*, **55**, 290–292.
- Lindgren, U., Lindholm, S. & Sarby, B. (1978). Short-term effects of 1-alpha-hydroxy-vitamin D₃ in patients on corticosteroid treatment and in patients with senile osteoporosis. *Acta med. scand.*, **204**, 89–92.
- Marshall, D. H. & Nordin, B. E. C. (1969). Kinetic analysis of plasma radioactivity after oral ingestion of radiocalcium. *Nature*, **222**, 797.
- Nordin, B. E. C. (1984). *Metabolic bone and stone disease*, pp 214–217. London: Churchill Livingstone.
- Nordin, B. E. C., Robertson, A., Seamark, R. F., Bridges, A., Philcox, J. C., Need, A. G., Horowitz, M., Morris, H. A. & Deam, S. (1985). The relation between calcium absorption, serum dehydroepiandrosterone, and vertebral mineral density in postmenopausal women. *J. clin. Endocrinol. Metab.*, **60**, 651–657.
- Peacock, M., Gallagher, J. C. & Nordin, B. E. C. (1974). Action of 1 α -hydroxy vitamin D₃ on calcium absorption and bone resorption in man. *Lancet*, **i**, 385–389.
- Pento, J. T. & Johnson, M. E. (1983). The influence of verapamil on calcium transport and uptake in segments of rat intestine. *Pharmacology*, **27**, 343–349.
- Rashiq, S. & Logan, R. F. A. (1986). Role of drugs on fractures of the femoral neck. *Br. med. J.*, **292**, 861–863.
- Rubin, R. P. (1981). Actions of calcium antagonists in secretory cells. In *New perspectives on calcium antagonists*, ed. Weiss, G. B., pp. 147–158. Bethesda: American Physiological Society.
- Singh, B. N., Ellrodt, G. & Petere, C. T. (1978). Verapamil: a review of its pharmacological properties and therapeutic uses. *Drugs*, **15**, 169–197.
- Sjödén, G., Jarnagan, K. & DeLuca, H. F. (1983). Inhibition of oxygendependent calcium ion transport in rat intestine by verapamil while phosphate ion transport is unaffected. *Calc. Tiss. Int.*, **Suppl** 35, A32.
- Sjödén, G. (1985). Effects of vitamin D. A comparison of 1 α OHD₂ and 1 α OHD₃. *Acta Orthop. Scand.*, **56**, suppl 217: 1–84.
- Somerville, P. J. & Kaye, M. (1984). Effect of calcium-sodium exchange and calcium channel inhibition on calcium release from bone in an *in-vitro* bone perfusion system. *Adv. exp. Biol. Med.*, **178**, 223–231.
- Storstein, L., Midtbø, K., Hals, O. & Myhre, E. (1981). Antihypertensive effect of verapamil in relation to plasma concentrations of verapamil and its active metabolite nor-verapamil. *Curr. Ther. Res.*, **29**, 112–119.
- Wrobel, J. & Michalska, L. (1977). The effect of verapamil on intestinal calcium transport. *Eur. J. Pharmac.*, **45**, 385–387.

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