



Calcium and magnesium in human toenails do not reflect bone mineral density

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Abstract

Nail mineral composition is influenced by several physiological and pathological processes. Potentially, nails could be used to monitor alterations in the level of incorporation of specific elements produced by nutritional abnormalities, disease states or chronic exposure to toxic agents. The purpose of this study was to investigate whether the calcium and magnesium content in nail clippings, as measured by instrumental neutron activation analysis (INAA), correlates with bone mineral density (BMD), as measured by quantitative microdensitometry (QMD), and therefore could be interesting as a screening instrument for osteoporosis. The study involved 220 women, who participated in a breast cancer screening project (the DOM-project) in Utrecht, the Netherlands. The correlations found between Ca and Mg measurements and bone mineral densities were very low (correlation coefficients ranging from 0.03 to 0.18). It is concluded that Ca and Mg measurements in nail clippings by INAA cannot be used for screening purposes in the prevention of osteoporosis.

Keywords: Osteoporosis; Nails; Calcium; Magnesium; Instrumental neutron activation analysis; Bone mineral density

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1. Introduction

Osteoporosis and its accompanying fractures constitute a major cause of morbidity, mortality and medical expense [1]. Once bone mass has decreased to the point where fractures occur, therapeutic options are limited. As a consequence, interest has shifted from treating patients with osteoporosis to the prevention of osteoporosis. To offer effective and acceptable preventive strategies, it is necessary to select high-risk groups. The methods mostly used to select persons at high risk for osteoporosis are bone mass measurements, which are usually elaborate and expensive.

Nail mineral composition is influenced by several physiological and pathological processes [2]. However, because of the slow rate of growth, the elemental composition is not expected to be affected by transient factors which perturb the serum mineral levels. Therefore, the mineral content of nail clippings might be a reliable indicator of the long-term pattern of mineral metabolism.

Calcium is an important mineral in bone metabolism: 99% of the total calcium content of the body is located in the skeleton. A deficient dietary calcium intake has often been mentioned as a causal factor for osteoporosis [3]. The role of magnesium in osteoporosis is still under investigation, but several authors have postulated that magnesium deficiency could play a role in the pathophysiology of osteoporosis [4]. Calcium and magnesium are deposited in both bone and nails. By the technique of instrumental neutron activation analysis (INAA) it is possible to determine the concentrations calcium and magnesium in nails. The method is fast, accurate and non-destructive.

The aims of this study were first to investigate whether there is a correlation between calcium and magnesium content in human toenails (as measured by INAA) and bone mineral density (as measured by quantitative microdensitometry; QMD) and, secondly, if such a correlation exists, we wished to determine whether these measurements could be used for screening purposes in the prevention of osteoporosis.

2. Subjects and methods

The data and biological material used in this study were obtained from women, born between 1911 and 1941, who participated in a breast cancer screening program (the DOM project) in Utrecht, the Netherlands [5]. Part of the population agreed to take part in a study on osteoporosis. Both in 1984 and 1989 bone density measurements were made in the second phalanx of the index finger [6,7], height and weight were measured and toenail clippings were collected. In addition, the women filled out questionnaires that provided information about variables supposed to have an association with osteoporosis. For 220 women both nail clippings and bone density measurements from 1984 and 1989 were available.

2.1. Determination of calcium and magnesium in toenails by INAA

INAA was applied to determine the Ca and Mg concentrations. The analyses were performed using the facilities of the Interfaculty Reactor Institute of the Delft Uni-

versity of Technology, the Netherlands [8]. The analysis was based on the measurement of the short half-life radionuclides ^{49}Ca (half-life 8.7 min) and ^{27}Mg (half-life 9.5 min), as produced by neutron activation.

To remove adjacent dirt and surface contaminations, the toenails were cleaned prior to analysis. The cleaning procedure consisted of subsequent treatment during 15 min with acetone, with distilled water for 10 min and acetone again for 15 min in an ultrasonic bath. All reagents were of ultra-pure quality. The samples were freeze-dried for 15 h (overnight) to obtain specimens of comparable moisture content. Humidity variations during weighing were taken into account.

The toenails were irradiated for 10 min at a thermal neutron flux of 5×10^{16} neutrons/s per m^2 . The batches held two toenail samples, two neutron flux monitors, a blank and a sample of a reference material for quality control. After a decay of 15 min, the γ -ray spectrum of the induced radioactivity was measured for 15 min with an automated γ -ray spectrometer equipped with a well-type germanium detector.

The accuracy of the method was checked by analysis of certified reference materials. NBS-1572 'Citrus leaves' was selected. Evaluation of the analyses of 30 samples showed mean concentrations for Ca and Mg, of, respectively, $6.13 \pm 0.12 \times 10^3$ mg/kg (certified value $5.8 \pm 0.3 \times 10^3$ mg/kg) and $3.32 \pm 0.03 \times 10^4$ mg/kg (certified value $3.15 \pm 0.09 \times 10^4$ mg/kg).

2.2. Measurement of bone mineral density by QMD

QMD was performed on the mid-phalanx of the second digit by means of a standardized postero-anterior (PA) radiograph of the right hand and a lateral (Lat) radiograph of the right index finger with an aluminium wedge as calibration standard. At the diaphysis of the mid-phalanx, optical density was measured with a scanning device and compared with the optical density of the reference wedge. PA and Lat measurements were combined and bone mineral density (BMD) was calculated in $\text{mm Al equivalent/mm}^3$. The method has been shown to be precise (coefficient of variation $< 2\%$). It gives an estimate of the integral bone density in the peripheral skeleton [6,7].

2.3. Statistical analysis

The parameters were tested for normal distribution and Pearson's correlation

Table 1
Characteristics of the study population in 1984 ($n = 220$)

	Mean	(S.D.)	Range
Age (years)	54.6	(9.1)	42.9–72.8
Height (cm)	165.1	(5.7)	152.0–183.5
Weight (kg)	70.0	(9.9)	50.5–121.0
Quetelet Index (kg/m^2)	25.7	(3.6)	18.0–42.4
Age at menopause ^a (years)	47.6	(5.7)	29.0–58.0

^aOne hundred and forty-four women were postmenopausal, 76 women were premenopausal in 1984.

Table 2
Concentrations of calcium and magnesium in ppm (mg/kg) in the toenail clippings of 199 women, measured in 1984 and 1989

	Mean	(S.D.)	Range
Ca (1984)	914.5	(373.8)	261.0-2230.0
Ca (1989)	851.5	(337.3)	119.0-2110.0
Mg (1984)	126.7	(60.3)	47.2-385.0
Mg (1989)	123.5	(66.3)	10.0-539.0

coefficients were calculated. Mean bone mineral densities in 1984 and 1989 and Ca and Mg contents in 1984 and 1989 were compared using a paired Student's *t*-test. All analyses were made with the SPSS/PC⁺ software package (version 4.0.1, SPSS Inc., Chicago, IL, USA).

3. Results

The characteristics of the study population in 1984 are presented in Table 1. The mean age of the women was 54.6 years. The postmenopausal age (and status) is important, because of enhanced bone loss in the first postmenopausal years [9]. Four outliers (extremely high Ca and Mg values) were excluded from the analysis as were 17 nail clippings in which it was impossible to measure the Ca or Mg concentration for technical reasons. One hundred and ninety-nine paired samples gave reliable results. A detailed examination of the excluded cases did not reveal any differences from the study cases.

Table 2 shows the mean values (\pm S.D.) of the calcium and magnesium concentrations in the nail clippings (derived from the women in 1984 and 1989). Correlations between the first and second Ca measurement and between the first and second Mg measurement were rather low ($r = 0.43$, $r = 0.23$, respectively). The paired-sample *t*-test showed a significant difference between the first and second Ca measurement ($P < 0.05$), while no difference was found between the two Mg measurements.

Table 3 shows the bone mineral densities as measured by QMD at the diaphysis of the second phalanx of the index finger. There was a strong correlation between the first and second bone mass measurement ($r = 0.85$; $P < 0.001$). The bone mass declined significantly (paired sampled *t*-test $P = 0.01$).

Table 3
Mean bone mineral density (BMD; mm Al eq./mm³) in the index fingers of 199 women, measured in 1984 and 1989

	Mean	(S.D.)	Range
BMD 1984	0.525	(0.07)	0.339-0.713
BMD 1989	0.518	(0.06)	0.358-0.704

Table 4
Correlation coefficients (Pearson) between the calcium and magnesium concentrations and the bone densities (BMD) in 1984 and 1989

	BMD 1984	BMD 1989
Calcium 1984	0.18*	0.14
Magnesium 1984	0.11	0.13
Calcium 1989	n.a.	0.10
Magnesium 1989	n.a.	0.03

* $P < 0.05$; n.a., not available.

Table 4 gives correlation coefficients between calcium and magnesium concentrations and bone densities in 1984 and 1989. None of the correlation coefficients exceeded 0.2.

As the assessment of the degree of bone loss is equally as important as the detection of low bone mass, correlation coefficients between the elements Ca and Mg and the difference in bone density between 1984 and 1989 (' Δ BMD') are given. These correlation coefficients are also given for the tertile of women with the highest bone loss ('tertile highest bone loss'; see Table 5).

4. Discussion

Screening a population for osteoporosis by means of analysis of toenail clippings would be an ideal method. Nails are easy to sample and store and INAA has shown to be an accurate, non-destructive method to measure several elements in nails. A number of reports have shown altered levels of specific elements in nails from individuals with specific pathologies, including cystic fibrosis [10], Alzheimer's disease [11] and chronic uraemia [12].

As especially calcium and probably magnesium play an important role in bone metabolism, a decreased or increased calcium or magnesium concentration in toenails could tell us something about the condition of our bones.

To investigate whether it is possible to identify high-risk groups for osteoporosis by means of nail analysis, the correlations between nail parameters (calcium and magnesium concentrations) and a bone parameter (bone mineral density) were

Table 5
Correlation coefficients (Pearson) between the Ca and Mg concentrations in 1984 and the 'difference in bone density' (Δ BMD) and 'tertile highest bone loss' (1984–1989)

	Δ BMD ($n = 199$)	Tertile highest bone loss ($n = 66$)
Calcium 1984	-0.07	-0.12
Magnesium 1984	0.02	-0.24

studied. Table 4 showed (cross-sectionally) very low correlations both in 1984 and 1989. Therefore, Ca and Mg concentrations in nails seem not to be valuable measurements in detecting low bone mass. As one could postulate that the concentrations of Ca or Mg may be indicative of future changes in bone density, correlation coefficients between the Ca and Mg concentrations and the change in bone density, especially bone loss, were studied.

Again, very low (non-significant) correlation coefficients were found. We examined the possibility of a higher correlation between bone density and the combined values of Mg and Ca, but the correlations remained low (data not shown). Furthermore, there was only a moderate correlation between the calcium measurement in 1984 and 1989 ($r = 0.43$). We conclude that the calcium or magnesium content in toenail clippings, as measured by INAA, does not reflect the bone mineral density. This argues against the use of this method as a screening instrument for osteoporosis.

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