The Effect of Bolus Vitamin D₃ Supplementation on Distal Radius Fracture Healing: A Randomized Controlled Trial Using HR-pQCT

Frans L Heyer,¹,²,³ Joost JA de Jong,⁴,⁵ Paul C Willems,⁶,⁷ Jacobus J Arts,⁶,⁷,⁸ Sandrine G P Bours,⁶,⁹ Sander M J van Kuijk,¹⁰ Judith A P Bons,¹¹ Martijn Poeze,¹,² Piet P Geusens,⁶,⁹,¹² Bert van Rietbergen,⁵,⁷,⁸ and Joop P van den Bergh¹,⁹,¹²,¹³

¹NUTRIM School for Nutrition and Translational Research in Metabolism, Maastricht University, Maastricht, The Netherlands
²Department of Surgery, Maastricht University Medical Center, Maastricht, The Netherlands
³Department of Surgery, VieCuri Medical Center Venlo, Venlo, The Netherlands
⁴MHeNs School for Mental Health and Neuroscience, Maastricht University, Maastricht, The Netherlands
⁵Department of Radiology and Nuclear Medicine, Maastricht University Medical Center, Maastricht, The Netherlands
⁶CAPHRI School for Public Health and Primary Care, Maastricht University, Maastricht, The Netherlands
⁷Department of Orthopedic Surgery, Maastricht University Medical Center, Maastricht, The Netherlands
⁸Department of Biomedical Engineering, Eindhoven University of Technology, Eindhoven, The Netherlands
⁹Department of Rheumatology, Maastricht University Medical Center, Maastricht, The Netherlands
¹⁰Department of Clinical Epidemiology & Medical Technology Assessment, Maastricht University Medical Center, Maastricht, The Netherlands
¹¹Central Diagnostic Laboratory, Maastricht University Medical Center, Maastricht, The Netherlands
¹²Faculty of Medicine and Life Sciences, Hasselt University, Hasselt, Belgium
¹³Department of Internal Medicine, VieCuri Medical Center Venlo, Venlo, The Netherlands

ABSTRACT
Vitamin D is an important factor in bone metabolism. Animal studies have shown a positive effect of vitamin D₃ supplementation on fracture healing, but evidence from clinical trials is inconclusive. A randomized controlled trial was performed to assess the effects of vitamin D₃ supplementation on fracture healing using HR-pQCT-based outcome parameters. Thirty-two postmenopausal women with a conservatively treated distal radius fracture were included within 2 weeks postfracture and randomized to a low-dose (N = 10) and a high-dose (N = 11) vitamin D intervention group receiving a 6-week bolus dose, equivalent to 700 and 1800 IU vitamin D₃ supplementation per day, respectively, in addition to a control group (N = 11) receiving no supplementation. After the baseline visit 1–2 weeks postfracture, follow-up visits were scheduled at 3–4, 6–8, and 12 weeks postfracture. At each visit, HR-pQCT scans of the fractured radius were performed. Cortical and trabecular bone density and microarchitectural parameters and microfinite element analysis–derived torsion, compression, and bending stiffness were assessed. Additionally, serum markers of bone resorption (CTX) and bone formation (PINP) were measured. Baseline serum levels of 25OHD₃ were <50 nmol/L in 33% of all participants and <75 nmol/L in 70%. Compared with the control group, high-dose vitamin D₃ supplementation resulted in a decreased trabecular number (regression coefficient β: −0.22; p < 0.01) and lower compression stiffness (B: −3.63; p < 0.05, together with an increase in the bone resorption marker CTX (B: 0.062; p < 0.05). No statistically significant differences were observed between the control and low-dose intervention group. In conclusion, the bolus equivalent of 700 U/day vitamin D₃ supplementation in a Western postmenopausal population does not improve distal radius fracture healing and an equivalent dose of 1800 IU/day may be detrimental in restoring bone stiffness during the first 12 weeks of fracture healing. © 2021 The Authors. Journal of Bone and Mineral Research published by Wiley Periodicals LLC on behalf of American Society for Bone and Mineral Research (ASBMR).
Introduction

Fracture healing consists of a complex series of events aimed at restoring the mechanical function of the affected bone region. Concepts such as the four-phase model, beginning with inflammation, followed by soft- and hard-callus formation ending with long-term remodeling have been used as a framework to understand these cellular and molecular processes. In addition, the so-called diamond model has been developed to integrate the various therapeutic factors that affect fracture healing. Animal studies have reported a positive effect of vitamin D3 on fracture healing as measured by radiographic imaging, histology, and mechanical testing. In a clinical setting, retrospective studies have shown that patients with a delayed-union have lower 25OHD levels compared with normally healing fracture patients, and that 25OHD deficiency is common in nonunion patients. Furthermore, a clinical study investigating combined vitamin D3 and calcium supplementation showed increased callus BMD in proximal humerus fractures.

In this randomized controlled trial (RCT), the effect of early vitamin D3 supplementation on distal radius fracture healing was assessed using HR-pQCT and microfine element analysis (μFEA) as primary outcome measurement. These techniques have been shown to be capable of quantifying changes in bone density, structure, and biomechanical properties during distal radius fracture healing, while also being associated with clinical outcome, facilitating a high-resolution and objective method of assessing the intervention.

We hypothesized that a bolus equivalent of 700 IU/d of vitamin D3 would enhance fracture healing and that a higher equivalent dose of 1800 IU/d could be even more beneficial, especially in vitamin D-deficient patients.

Participants and Methods

Study design

This single-blind RCT was conducted at the Maastricht University Medical Center (MUMC) in the Netherlands. Early supplementation of vitamin D3 during fracture healing was compared with a control group receiving no intervention. Study visits were scheduled 1–2 weeks (visit 1), 3–4 weeks (visit 2), 6–8 weeks (visit 3), and 12 weeks (visit 4) postfracture. The study protocol, approved by the institutional ethics committee (file number NL33512.068.10), was submitted to the Dutch Trial Register (NTR) and filed under registration number NTR3821. All participants provided written informed consent before enrolling in the study.

Participants

Women aged 50 years and older presenting at the emergency room of the MUMC with a distal radius fracture, receiving cast immobilization, were screened for inclusion. Patients requiring surgical treatment were excluded because of the effect of metal implants on the primary outcome measures. Patients with a known systemic or metabolic bone disorder, such as hyperthyroidism, hyperparathyroidism, chronic kidney disease (with an estimated glomerular filtration rate <30 mL/min/1.73 m²), sarcoidosis, or an active inflammatory disease (rheumatoid arthritis, inflammatory bowel disease) were also excluded. Final exclusion criteria were use of oral glucocorticoids in the past 12 months, malignant disease in the past 12 months, previous (bone) surgery at the current fracture site, a neuromuscular or neurosensory condition, severe concurrent joint involvement, or the inability to provide informed consent.

In addition to the study procedures described below, all study participants were invited to participate in the screening program for osteoporosis at the local fracture liaison service, as per the national guidelines. This screening included laboratory tests for metabolic bone disorders and DXA of the lumbar spine and femur.

Intervention

Clinically, vitamin D status is determined by the measurement of serum 25-hydroxyvitamin D (25OHD) concentration. Previous studies have shown a comparable effect of daily versus monthly doses of vitamin D3 (cholecalciferol) on serum 25OHD levels, although it is still unknown if a sudden increase of 25OHD serum levels could, independent of absolute serum levels, contribute to the negative effects observed by others with a 500,000 IU bolus dose.

To achieve full compliance, liquid vitamin D3 50,000 IU/mL (Fagron BV), was administered orally at the first and third study visits in two bolus doses. Participants enrolled in the low-dose group received 0.6 mL or 30,000 IU once per 6 weeks (equivalent to 700 IU daily) versus 1.5 mL or 75,000 IU once every 6 weeks (equivalent to 1800 IU daily) in the high-dose group.

If there was an indication for antosteoporosis treatment, standard supplementation with 800 IU vitamin D3 daily was started after the end of this study.

Randomization

Block randomization was performed by an independent member of the orthopedic trial bureau using a computer-generated randomization list (SPSS 16.0; IBM Corp). Patients were allocated using sequentially numbered envelopes to either a control group (N = 10) receiving no early vitamin D3 supplementation, a low-dose intervention group (vitamin D3 700 IU daily), or a high-dose intervention group (vitamin D3 1800 IU daily). No placebo was used in the control group. Participants were asked to report any use of over-the-counter vitamin D supplements.

During data collection and image and statistical analyses, the investigator performing these tasks was blinded for the intervention assignment.
Measurements

HR-pQCT scanning

At each visit, the fractured radius was scanned using a first-generation HR-pQCT scanner (XtremeCT; Scanco Medical AG). In addition, the contralateral side was scanned at the first and last visit using the same scan protocol to detect an effect of the intervention on nonfractured bone. This protocol, similar to previous studies, consisted of two consecutive stacks of 9 mm using the standard in vivo settings as prescribed by the manufacturer (82-μm isotropic voxel size, X-ray tube voltage 60 kVp and tube current 0.900 mA, 100 ms integration time, 750 projections/180°). The acquired image thus comprised an 18 mm/220 slices long region of the distal radius. Use of the standard reference point on the articular surface of the distal radius is not feasible in case of a fracture. Therefore, scan offset was set at 3.0 mm from the proximal edge of the lunate (Fig. 1).17

The cast was usually removed at or shortly before the third study visit, 6–8 weeks postfracture. To avoid bias of the subsequent scans,27 the cast was preserved and temporarily replaced around the wrist during the later scans. To facilitate the lower arm with cast and minimize motion artifacts, a custom carbon holder with inflatable cushion (Pearltex AG, Schlieren, Switzerland) was used.17

All scans were quality-graded according to Pialat and colleagues.28 Scans with significant motion artifacts (i.e., grade 4 or 5) were repeated once. Only scans with grade 1–3 were used for the analyses.

Image analyses

HR-pQCT images were analyzed by a single investigator blinded for the randomization using μFEA in addition to the standard evaluation method, both provided by the manufacturer (Scanco Medical AG) as described previously.17 Briefly, after semiautomatic contouring of the periosteal boundary, segmentation of the mineralized tissue was achieved using a Laplace-Hamming filter (epsilon 0.5 and cutoff frequency 0.4) with normalization (range, 0–1000) and global thresholding (threshold 400). BMDs (mgHA/cm²) were determined for the trabecular, cortical, and total region of interest. A 3D ridge extraction method was used to assess trabecular number (1/mm) and derive trabecular thickness (mm) and separation (mm).29

Microfinite element analyses

Segmented images were used to create μFE models with brick elements similar in size to the 82-μm isotropic voxels. Assigned material properties were a Young’s modulus of 10 GPa and a Poisson’s ratio of 0.3.30 The μFE models were subjected to four virtual load cases to assess bone stiffness: a high-friction compression test (prescribed displacement of 1% length change in axial direction), a rotation test around the longitudinal axis (0.01 rad), and two rotational tests around the sagittal and transversal axis (0.01 rad). The latter two were combined to calculate estimated bending stiffness.17

Bone turnover makers

Nonfasting venous blood samples were taken in the morning (before noon) at each visit and before administering the cholecalciferol at visits 1 and 3. From these samples, the serum marker of bone formation PINP and serum marker for bone resorption CTX were measured using chemiluminescence immunometric assays on the IDS-iSYS instrument (Immunodiagnostic Systems, PLC).

Functional outcome: PRWE

Study participants completed the Dutch version of the Patient-Rated Wrist Evaluation (PRWE)31,32 at each visit. This 15-item questionnaire has been validated to assess functional outcome after distal radius fractures.33 The PRWE consists of a pain domain (5 questions) and a function domain (10 questions), resulting in a combined score ranging from 0 (no pain/disabilities) to 100 (worst pain/disabilities).

Additional data

Medical history (previous surgeries and/or fractures, medication) and baseline characteristics (age, height, and weight) were collected at the first visit. In addition, data from the fracture liaison service were extracted: daily calcium intake (including supplements), smoking status and alcohol intake, DXA results (lumbar spine and proximal femur BMD and T scores), and baseline 25OHD serum levels. From the blood sample collected at visit 4, 25OHD response after the intervention was also measured using a chemiluminescence immunometric assay on the IDS-iSYS instrument (Immunodiagnostic Systems, PLC).

Sample size

PS power and sample size calculator software (version 3.0.14) was used to calculate sample size (http://biostat.mc.vanderbilt.edu/wiki/Main/PowerSampleSize). Limited data on clinical fracture healing trials using HR-pQCT and μFEA were available at the time of study inception. It was estimated that 0.5 kN constituted a clinically relevant difference between control and intervention groups. A power of 80% and a significance level α = 0.05 yielded a sample size of 10 subjects per group: 30 in total.
Statistical analysis

Baseline characteristics were presented as median with interquartile range (IQR; continuous data) or number with percentage (dichotomous data). Outcome measures during follow-up were compared between the control and intervention groups using generalized estimating equations (GEEs). This statistical model is able to handle the longitudinal nature of this study, as well as the missing data as a result of discarding the scans of insufficient quality. The GEE model was adjusted for baseline serum 25OHD and for the baseline measurement of each respective outcome: The follow-up measurements of trabecular number were corrected for the baseline trabecular number. The two HR-pQCT measurements of the contralateral wrist were compared using the Wilcoxon signed rank test. All statistical analyses were performed in SPSS 24.0 (IBM Corp).

Results

From June 2013 until May 2016, 32 participants were enrolled in the study (Fig. 2). A significant part of the screened patients \( N = 54 \) were not included because of unforeseen reasons: A number of patients were from different regions and received further treatment and follow-up elsewhere, some had accompanying fractures requiring surgery (e.g., hip fractures) or were judged to be unable to participate successfully in the scans and follow-up schedule because of tremors (Parkinson disease) or the high amount of eldercare they received.

From the included patients, one withdrew informed consent after the first study visit, and another dropped out following surgical intervention because of a secondary dislocation of the fracture. Both patients were replaced with additional inclusions in accordance with the amended study protocol, where they were assigned to the group randomization from the participant they replaced, to realize 10 subjects per group.

Baseline characteristics of the groups were similar (Table 1), with the exception of the number of patients with a fracture requiring reduction. Serum levels of 25OHD were <50 nmol/L in 30%–40% and <75 nmol/L in 60%–90% of patients (see Table 1 for the specific values for each group). None of the study participants reported over-the-counter vitamin D supplement use during participation.

Microarchitecture and BMD

The HR-pQCT-based BMD and microarchitectural parameters are presented in Fig. 3. Despite randomization, a baseline difference
was observed that was considered clinically relevant, e.g., a mean baseline trabecular density of 179 ± 1.8 (mean with SE) for the control group versus 156 ± 9.6 and 162 ± 8.0 for the low- and high-dose groups, respectively. The baseline was incorporated in the statistical models for adjustment.

Longitudinal changes showed first an increase in trabecular BMD during fracture healing, followed by a decrease. No statistically significant differences were observed with regard to microarchitectural or BMD parameters between the control group and the low-dose intervention group (all p values >0.05). In the high-dose vitamin D3 group, a decreased trabecular number was observed (β coefficient = −0.22; 95% CI, −0.36 to −0.08; p value = 0.002) and a correspondingly increasing trabecular separation (β coefficient = 0.05; 95% CI, 0.009 to 0.096; p value = 0.018), compared with the control group.

Adjustment for BMD at the lumbar spine, femoral neck, or total hip as assessed with DXA did not change these results.

**Microfinite element analyses**

No statistically significant differences were observed between the control group and the low-dose intervention group (p values between 0.48 and 0.85; Table 2). In the high-dose group a decreased compression stiffness was observed compared with the control group (β coefficient = −3.63; 95% CI, −6.76 to −0.50; p value = 0.023).

Measurements of the contralateral radius revealed no changes between baseline and 12 weeks postrupture in the nonfractured distal radius (p values between 0.1 and 1), although the number of usable scans was limited caused by the presence of severe motion artifacts (Fig. 2).

**Serum markers**

Median serum 25OHD 12 weeks postrupture was 61 nmol/L (IQR, 42–72) for the control group, 70 nmol/L (IQR, 66 to 81) and 81 nmol/L (IQR, 70 to 95) for the low- and high-dose vitamin D3 intervention groups, respectively. Compared with the control group, both intervention groups had a higher 25OHD level at visit 4 (low dose: p value = 0.016; high dose: p value <0.001).

Longitudinal analyses showed an increased level of CTX during the first 3–6 weeks post-fracture in the high-dose vitamin D group (Fig. 4) compared with the control group (β coefficient = 0.062; 95% CI, 0.0004–0.12; p value = 0.048), whereas no difference between the control and low-dose group was observed. No statistically significant differences were detected for the bone resorption marker PINP (p values >0.05).

**Functional outcome**

No differences in PRWE score were found between the control group and the low- or high-dose intervention groups during the study period (p value = 0.4 and 0.2 respectively).

**Discussion**

In this study, early bolus supplementation of vitamin D3 after a distal radius fracture did not result in enhanced fracture healing as assessed using HR-pQCT nor in improved patient reported outcomes. Remarkably, a decreased trabecular number and compression stiffness and an increase of the serum marker of bone resorption CTX was observed in the high vitamin D3 dose group compared with the control group.

The longitudinal changes of HR-pQCT and μFEA parameters during fracture healing seen in this study match the pattern we
described earlier in our observational studies: First an increase in (trabecular) bone density is seen, followed by a decrease corresponding to the formation and remodeling of the fracture callus, with increasing bone stiffness from 3–4 weeks postfracture onward.17,26

An interesting observation is that compression stiffness differs between the high dose and the control group, but torsion and bending stiffness do not. The latter two are predominantly determined by the cortical perimeter, whereas the trabecular compartment contributes primarily to compression stiffness.35 Because the trabecular compartment features a large surface area to bone ratio as compared with the cortex, it is possible that metabolic changes (such as fracture healing influenced by 25OHD) can have more impact in the trabecular compartment.

Fig 3. Longitudinal HR-pQCT–derived bone density, microarchitectural, geometric, and biomechanical parameters. Data presented as mean ± SEM. SEM, standard error of the mean. *Statistically significant different from control group.
Biomechanical parameters should thus be drawn cautiously. Part of the explanation for the lower trabecular number (Fig. 3) can be observed to persist during follow-up. Nevertheless, statistical correction of 1 g in the RCT by Doetsch and colleagues, whereas in our study the dietary daily calcium intake was on average <750 mg and no supplementation was provided. Nevertheless, the patients in our study did not show a significant negative calcium balance as indicated by the normal PTH levels (Table 1). Another explanation is the baseline 25OHD level: In a vitamin D-deficient population such as the study by Doetsch and colleagues (with a baseline 25OHD level of 10.2 ± 5.9 nmol/L), vitamin D3 supplementation could increase BMD in fracture callus, whereas the relatively normal 25OHD levels (~40 nmol/L) in our population could preclude such an effect. The baseline levels of 25OHD could also be related to our observation that a higher dose of vitamin D3 supplementation could be detrimental to fracture healing. Although unanticipated, this finding is in accordance with a recently published RCT by Burt and colleagues. In that study, a (nonfracture) population with 75–80 nmol/L serum 25OHD at baseline received 400, 4000, or 10,000 IU vitamin D3 supplementation a day. Their findings included a dose-dependent negative effect on BMD, with a higher dose of vitamin D3 resulting in a more pronounced decrease in volumetric BMD as assessed with HR-pQCT.

The mechanism underlying the possible detrimental effect of high-dose vitamin D3 supplementation on bone remains unclear, but several hypotheses have been suggested. For instance, animal studies have shown that a high dose of vitamin D3 can result in upregulation of 25-hydroxyvitamin D24-hydroxylase (also known as CYP24), the enzyme responsible for catabolizing the biologically active form of vitamin D, 1,25-dihydroxyvitamin D3. This mechanism could protect the organism from vitamin D toxicity. However, a study where different dosing regimens of vitamin D3 were compared, a monthly dose of 45,000 IU showed a steady increase of serum 25OHD when assessed at 1, 7, and 28 days after administration. Although the higher dose used in our study could theoretically result in a transient drop of serum 25OHD between the measurements at baseline...
and 12 weeks, further evidence to support this hypothesis is warranted.

The supplementation regimen of a bolus dose once per 6 weeks is also an item of discussion. This method of supplementation was chosen to ensure full compliance and corresponds to 60,000 and 150,000 IU/d in the intervention groups, corresponding to a daily dose of 700 and 1800 in the low- and high-dose groups, respectively. This was based on the national guidelines, although other guidelines recommend 1500–2000 IU/d.

However, a bolus dose is not similar to the daily equivalent dose. Studies have shown detrimental effects of monthly bolus doses of cholecalciferol on falls. In our study, although the bolus regimen is relatively short compared with Bischoff-Ferrari and colleagues (3 months vs 1 year), it is currently unclear whether the detrimental effect in the 1800 IU/d equivalent group is the result of the 6-week bolus dose or the higher supplementation itself is responsible. Future studies should address this issue, but there is increasing data in support of daily dosing regimens for cholecalciferol supplementation.

Studies on the effect of vitamin D supplementation in the treatment of osteoporosis focus on the increased intestinal calcium absorption as the mechanism of action. However, the direct effects of vitamin D on bone tissue are probably of greater importance in the current study. The vitamin D receptor (VDR) on osteoblasts controls the expression of RANKL. This important osteoclastic factor binds on its receptor (RANK) on osteoclasts, stimulating proliferation and thus enhancing bone resorption. A similar mechanism was also found in chondrocytes, indicating that this pathway could be important in both the early as well as in the later stages of fracture healing. Indeed, the observed increase of the serum marker of bone resorption CTX in this study (Fig. 4) supports this hypothesis. Furthermore, no differences between the groups were detected in PINP levels, the marker for bone formation, supporting the hypothesis that the decrease in bone strength is caused by vitamin-D mediated increased resorption.

Nevertheless, the intricacies of VDR signaling in osteoblasts and the subsequent indirect effects on osteoblast activity remain to be elucidated.

With respect to the bone turnover markers, it is known that CTX is influenced by a circadian rhythm, as well as by food intake. Therefore, guidelines recommend using morning overnight fasting samples for CTX monitoring. However, considering the challenging logistics caused by the multiple procedures during study visits (regular clinical follow-up, blood sample collection, HR-pQCT scanning) combined with the average age of our population, we anticipated that adding the fasting requirement to the study protocol would result in a lower inclusion rate and higher loss to follow-up. Unfortunately, this introduces a preanalytical variability that invalidates the comparison of CTX levels to other studies. Nevertheless, because the samples were taken under similar nonfasting conditions during the same time of the day in follow-up visits, the serial measurements of CTX are usable in our study design.

The strengths of this study include the randomized design with two distinct dosing regimens, where patients in the intervention groups were administered vitamin D3 supplementation in 6-week bolus doses during study visits, thus ensuring full patient compliance. Furthermore, outcome measures were assessed with a blinded, detailed evaluation using HR-pQCT with μFEA. These outcome measures focus on the target of the intervention: bone density, structure, and strength, and are more precise than fracture healing quantification using conventional X-ray imaging. Although in the clinical setting, patient-reported outcome measures such as the PRWE are important, in this study we were primarily interested in the direct action of vitamin D3 supplementation on the healing bone and chose to power the trial on this outcome measurement.

An important limitation of this study is the lack of significant serum 25OHD deficiency at baseline in all groups (mean of approximately 60 nmol/L), although we expected our study to include a vitamin D deficient population based on our previous work showing that a substantial proportion of patients with a fracture has 25OHD levels below 50 nmol/L. The healthy user bias may be a likely explanation for this finding. In combination with the required follow-up regimen of four visits in 12 weeks, the inclusion and exclusion criteria likely resulted in the selection of a relatively healthy subset of postmenopausal women presenting...
with a distal radius fracture. As a result, the findings in our study are limited to a predominantly 25OHD nondeficient population.

Second, the control group had less severe fractures compared with the intervention groups, as indicated by the need for fracture reduction (one in the control group vs five in each intervention group) and the baseline bone stiffness of the fracture region (Fig. 3). Although a correction for baseline was used in the statistical model, it cannot be discounted that the difference in baseline has an effect on the range of observed changes during fracture healing. Future trials should consider more strict inclusion criteria regarding fracture type or facilitate stratification by baseline bone stiffness in the analyses. Furthermore, based on the osteoporosis screening after completion of this trial, a low BMD (T score <−2.5) was present in 8 out of 20 patients in the intervention groups and in none of the control group.

Finally, the number of patients in each group was low, although the high-resolution outcome measurements ensured adequate power to detect an effect in bone microarchitecture and estimated strength. Two currently active RCTs investigating vitamin D3 supplementation in fracture healing will provide further evidence on this interesting topic.50,51

In conclusion, this RCT did not show a beneficial effect of early bolus vitamin D3 supplementation during distal radius fracture healing on bone density, microarchitecture, or bone stiffness based on HR-pQCT scans in a nonvitamin D-deficient patient group. A possible detrimental effect of a high-dose vitamin D3 bolus dose was observed, requiring further investigation. Because of the small sample size and limitations as discussed, these conclusions should be regarded as preliminary findings.

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Author Contributions

Frans Heyer: Data curation; formal analysis; investigation; project administration; resources; validation; visualization; writing—original draft; writing—review & editing. Joost de Jong: Conceptualization; investigation; methodology; project administration; resources; software; supervision; writing—review & editing. Paul Willems: Conceptualization; investigation; methodology; resources; supervision; writing—review & editing. Sandrine Bours: Conceptualization; investigation; methodology; project administration; resources; software; supervision; writing—review & editing. Martijn Poeze: Funding acquisition; methodology; resources; supervision; writing—review & editing. Piet Geusens: Conceptualization; funding acquisition; methodology; resources; supervision; writing—review & editing. Bert van Rietbergen: Conceptualization; methodology; resources; software; supervision; writing—review & editing. Judith Bons: Data curation; investigation; resources; writing—review & editing. Martijn Poeze: Funding acquisition; methodology; resources; supervision; writing—review & editing. Joop Bergh: Conceptualization; formal analysis; funding acquisition; methodology; project administration; resources; supervision; validation; writing—original draft; writing—review & editing.

Conflict of Interest

Bert van Rietbergen is a consultant for Scanco Medical AG. J J Arts is a board member of the workgroup Biotechnology of the Dutch Orthopedic Association (NOV). All other authors declare that they have no conflicts of interest.

Peer Review

The peer review history for this article is available at https://publons.com/publon/10.1002/jbmr.4311.

Data Availability Statement

Research data are not shared at this time, due to follow-up studies still ongoing.

References

1. Einhorn TA. The cell and molecular biology of fracture healing. Clin Orthop Relat Res. 1998;355 Suppl:S7-S21.
2. Morgan EF, Hussein AI, Einhorn TA. Biomechanics of fracture healing. In Bilezikian JP, Bouillon R, Clemens T, et al., eds. Primer on the Metabolic Bone Diseases and Disorders of Mineral Metabolism. 9th ed. Hoboken, NJ: Wiley-Blackwell; 2013 pp 109-114.
3. Schindeler A, McDonald MM, Bokpo P, Little DG. Bone remodeling during fracture repair: the cellular picture. Semin Cell Dev Biol. 2008;19:459-466.
4. Giannoudis PV, Einhorn TA, Marsh D. Fracture healing: the diamond concept. Injury. 2007;38(suppl 4):S3-S6.
5. Giannoudis PV, Einhorn TA, Schmidmaier G, Marsh D. The diamond concept-open questions. Injury. 2008;39(suppl 2):S5-S8.
6. Vannucci L, Brandi M-L. Healing of the bone with anti-fracture drugs. Expert Opin Pharmacother. 2016;17:2267-2272.
7. Dawson-Hughes B, Mithal A, Bonjour J-P, et al. IOF position statement: vitamin D recommendations for older adults. Osteoporosis Int. 2010;21:1151-1154.
8. Bours SPG, van Geel TACM, Geusens PP, et al. Contributors to secondary osteoporosis and metabolic bone diseases in patients presenting with a clinical fracture. J Clin Endocrinol Metab. 2011;96:1360-1367.
9. Gorter EA, Krijnen P, Schipper IB. Vitamin D deficiency in adult fracture patients: prevalence and risk factors. Eur J Trauma Emerg Surg. 2016;42:369-378.
10. Delgado-Martínez AD, Martínez ME, Carrascal MT, Rodríguez-Avial M, Munuera L. Effect of 25-OH-vitamin D on fracture healing in elderly rats. J Orthop Res. 1998;16:650-653.
11. Omeroğlu H, Atše Y, Akkuş O, Korkusuz F, Biçimoğlu A, Akkaş N. Biochemical analysis of the effects of single high-dose vitamin D3 on fracture healing in a healthy rabbit model. Arch Orthop Trauma Surg. 2018;116:271-274.
12. Fu L, Tang T, Miao Y, Hao Y, Dai K. Effect of 1,25-dihydroxy vitamin D3 on fracture healing and bone remodeling in ovariectomized rat femora. Bone. 2008;44:893-898.
13. Gorter EA, Krijnen P, Schipper IB. Vitamin D status and adult fracture healing. J Clin Orthop Trauma. 2017;8:34-37.
14. Brumbaugh PF, Speer DP, Pitt MJ. 1 Alpha, 25-dihydroxyvitamin D3 a metabolite of vitamin D that promotes bone repair. Am J Pathol. 1982;106:171-179.
15. Connor DP, Monla YT, Earthman TP. Metabolic and endocrine abnormalities in patients with nonunions. J Orthop Trauma. 2007;21:557-570.
16. Doetsch AM, Faber J, Lynnerup N, Wätjen I, Bliddal H, Danneskiold-Samsøe B. The effect of calcium and vitamin D3 supplementation on the healing of the proximal humerus fracture: a randomized placebo-controlled study. Calcif Tissue Int. 2004;75:183-188.
17. de Jong JJ, Willems PC, Arts JJ, et al. Assessment of the healing process in distal radius fractures by high resolution peripheral quantitative computed tomography. Bone. 2013;64:65-74.

18. Meyer U, de Jong JJ, Bours SG, et al. Early changes in bone density, microarchitecture, bone resorption, and inflammation predict the clinical outcome 12 weeks after conservatively treated distal radius fractures: an exploratory study. J Bone Miner Res. 2014;29:2065-2073.

19. Heyer FL, de Jong JJA, Willems PC, et al. Long-term functional outcome of distal radius fractures is associated with early post-fracture bone stiffness of the fracture region: an HR-pQCT exploratory study. Bone. 2019;127:510-516.

20. de Jong JJ, Lataster A, van Rietbergen B, et al. Distal radius plate of CFR-PEEK has minimal effect compared to titanium plates on bone parameters in high-resolution peripheral quantitative computed tomography: a pilot study. BMC Med Imaging. 2017;17:18.

21. Dutch Society for Rheumatology. Guideline on Osteoporosis and Fracture Prevention, 3rd revision [Internet]. [Utrecht]. 2011. Available from: https://richtlijndatabase.nl/richtlijn/osteoporose_en_fractuurpreventie_-_startpagina.html

22. Giustina A, Adler RA, Binkley N, et al. Controversies in vitamin D: summary statement from an international conference. J Clin Endocrinol Metab. 2019;104:234-240.

23. Ish-Shalom S, Segal E, Salganik T, Raz B, Bromberg IL, Vieth R. Comparison of daily, weekly, and monthly vitamin D3 in ethanol dosing protocols for two months in elderly hip fracture patients. J Clin Endocrinol Metab. 2008;93:3430-3435.

24. Binkley N, Gemar D, Engelke J, et al. Evaluation of ergocalciferol or cholecalciferol dosing, 1,600 IU daily or 50,000 IU monthly in older adults. J Clin Endocrinol Metab. 2011;96:981-988.

25. Sanders KM, Stuart AL, Williamson EJ, et al. Annual high-dose oral vitamin D and falls and fractures in older women: a randomized controlled trial. JAMA. 2010;303:1815-1822.

26. de Jong JJ et al. Fracture repair in the distal radius in postmenopausal women: a follow-up 2 years Postfracture using HRpQCT. J Bone Miner Res. 2016;31:1114-1122.

27. de Jong JJA, Heyer FL, Arts JJC, et al. Effect of a cast on short-term reproducibility and bone parameters obtained from HR-pQCT measurements at the distal end of the radius. J Bone Joint Surg Am. 2016;98:356-362.

28. Pialat JB, Burghardt AJ, Sode M, Link TM, Majumdar S. Visual grading of motion induced image degradation in high resolution peripheral computed tomography: impact of image quality on measures of bone density and micro-architecture. Bone. 2012;50:111-118.

29. Hildebrand T, Laib A, Müller R, Dequeker J, Rüegsegger P. Direct three-dimensional morphometric analysis of human cancellous bone: microstructural data from spine, femur, iliac crest, and calcaneus. J Bone Miner Res. 1999;14:1167-1174.

30. Pistoia W, van Rietbergen B, Lochmüller EM, Lill CA, Eckstein F, Rüegsegger P. Estimation of distal radius failure load with micro-finite element analysis models based on three-dimensional peripheral quantitative computed tomography images. Bone. 2002;30:842-848.

31. MacDermid JC, Turgeon T, Richards RS, Beadle M, Roth JH. Patient rating of wrist pain and disability: a reliable and valid measurement tool. J Orthop Trauma. 1998;12:577-586.

32. Brink SM, Voskamp EG, Houpt P, Emmelot CH. Psychometric properties of the patient rated wrist/hand evaluation - Dutch language version (PRWHE-DLV). J Hand Surg Eur. 2009;34:S56-S57.

33. Goldhahn J, Angst F, Simmen BR. What counts: outcome assessment after distal radius fractures in aged patients. J Orthop Trauma. 2008;22:S126-S130.

34. Twisk JWR. Chapter 3: Continuous Outcome Variables. Applied Longitudinal Data Analysis for Epidemiology (pp. 16–50). Cambridge, England: Cambridge University Press; 2013.

35. Boussein ML, Karakis D. Bone geometry and skeletal fragility. Curr Osteoporos Rep. 2006;4:49-56.

36. Burt LA, Billington EO, Rose MS, Raymond DA, Hanley DA, Boyd SK. Effect of high-dose vitamin D supplementation on volumetric bone density and bone strength. JAMA. 2019;322:736-745.

37. Dawson-Hughes B, Harris SS. High-dose vitamin D supplementation: too much of a good thing? JAMA. 2010;303:1861-1862.

38. Beckman MJ, Johnson JA, Goff JP, Reinhardt TA, Beitz DC, Horst RL. The role of dietary calcium in the physiology of vitamin D toxicity: excess dietary vitamin D blunts parathyroid hormone induction of kidney 1-hydroxylase. Arch Biochem Biophys. 1995;319:535-539.

39. Holick MF, Binkley NC, Bischoff-Ferrari HA, et al. Evaluation, treatment, and prevention of vitamin D deficiency: an endocrine society clinical practice guideline. J Clin Endocrinol Metab. 2011;96:1911-1930.

40. Bischoff-Ferrari HA, Dawson-Hughes B, Orav EJ, et al. Monthly high-dose vitamin D treatment for the prevention of functional decline: a randomized clinical trial. JAMA Intern Med. 2016;176:1-10.

41. Ginde AA, Blatchford P, Reese K, et al. High-dose monthly vitamin D for prevention of acute respiratory infection in older long-term care residents: a randomized clinical trial. J Am Geriatr Soc. 2017;65:496-503.

42. Martineau AR, Jolliffe DA, Hooper RL, et al. Vitamin D supplementation to prevent acute respiratory tract infections: systematic review and meta-analysis of individual participant data. BMJ. 2017;356:i6583.

43. Griffin G, Hewison M, Hopkin J, et al. Perspective: Vitamin D supplementation prevents rickets and acute respiratory infections when given as daily maintenance but not as intermittent bolus: Implications for COVID-19. Clin Med (Lond). 2021;21(2):e144-e149. https://doi.org/10.7861/clinmed.2021-0035.

44. Lieben L, Carmeliet G. The delicate balance between vitamin D, calcium and bone homeostasis: lessons learned from intestinal- and osteocyte-specific VDR null mice. J Steroid Biochem Mol Biol. 2013;136:102-106.

45. Kim S, Yamazaki M, Zella LA, Shevde NK, Pike JW. Activation of receptor activator of NF-kappaB ligand gene expression by 1,25-dihydroxyvitamin D3 is mediated through multiple long-range enhancers. Mol Cell Biol. 2006;26:6469-6486.

46. Masuyama R, Stockmans I, Torrekens S, et al. Vitamin D receptor in chondrocytes promotes osteoclastogenesis and regulates FGF23 production in osteoblasts. J Clin Invest. 2006;116:3150-3159.

47. Szulc P, Naylor K, Hoyle NR, Eastell R. Leary ET; National Bone Health Partnership Advisory Group. The role of vitamin D in the development of osteoporosis. Osteoporos Int. 2017;28:2541-2556.

48. Boudes P. Drug compliance in therapeutic trials a review. Control Clin Trials. 1998;19:257-268.

49. Govender S, Cismona C, Genant HK, et al. Recombinant human bone morphogenetic protein-2 for treatment of open tibial fractures: a prospective, controlled, randomized study of four hundred and fifty patients. J Bone Joint Surg Am. 2002;84-A:2123-2134.

50. FAITH-2 Investigators, Slobogean GP, Sprague S, et al. Fixation using alternative implants for the treatment of hip fractures (FAITH-2): patients randomized controlled trial in young femoral neck fracture patients. J Bone Joint Surg Am. 2002;84-A:2123-2134.

51. Sprague S, Bzovsky S, Connelly D, et al. Study protocol: design and rationale for two months in elderly hip fracture patients. J Clin Endocrinol Metab. 2011;96:1911-1930.

52. Sprague S, Bzovsky S, Connelly D, et al. Study protocol: design and rationale for a pilot multi-centre 2 randomized controlled trial to determine optimal vitamin D3 supplementation strategies for acute fracture healing. Pilot Feasibility Stud. 2019;5:135.