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Effects of cholecalciferol supplementation and optimized calcium intakes on vitamin D status, muscle strength and bone health: A one-year pilot randomized controlled trial in adults with severe burns

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ABSTRACT

Objective: Burn patients are at risk of hypovitaminosis D and osteopenia or sarcopenia. Vitamin D pleiotropic effects may influence bone and muscle health. The aim of this pilot study was to assess effects of a cholecalciferol (VD3) supplementation and an optimized calcium (Ca) regimen on vitamin D (VD) status, bone and muscle health during sequela stage of burn injury.

Design: Monocentric randomized controlled trial.

Methods: Fifteen adults with thermal burns dating from 2 to 5 years were randomized into two groups. For 12 months, they either received a quarterly IM injection of 200,000 IU VD3 and daily oral Ca (Group D) or placebo (Group P). VD status and bone remodeling markers were assessed every 3 months. Knee muscle strength and bone mineral density were, respectively, assessed using isokinetic dynamometry and dual X-ray absorptiometry at initiation (M0) and completion (M12) of the protocol.

Results: Of all the patients, 66% presented with VD deficiency and 53% (with 3 men <40 y) were considered osteopenic at inclusion. After one year, calcidiol levels significantly increased in Group D to reach 40 (37–61) ng/ml. No significant change in bone health was observed in both groups while Group D significantly improved quadriceps strength when tested at high velocity.

Conclusions: This VD3 supplementation was safe and efficient to correct hypovitaminosis D in burn adults. When combined with optimized Ca intakes, it demonstrated positive effects on muscle health but not on bone health. A high prevalence of hypovitaminosis D and osteopenia in these patients, as well as their wide range of muscle performances, seem to be worrying when considering rehabilitation and quality of life.

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1. Introduction

Survival after burns has improved over the past last decades, but associated morbidities remain a major concern. Hypercatabolism in patients with burn surface area (BSA) >20% and prolonged immobilization are key factors in development of osteopenia and sarcopenia [1–3]. These two conditions may compromise further rehabilitation and quality of life.

The role of the skin in maintaining adequate serum vitamin D levels has been well established [4]. Burn patients, particularly those with large BSA, are an under-recognized group of hypovitaminosis D. Hypovitaminosis D may be defined as 25OH-D levels < 30 ng/ml, according to the Endocrine Society [5]. These patients avoid sun exposition because of heat intolerance and mostly because of the risk of burn scar hyperpigmentation or neoplastic degeneration [6]. Furthermore, biosynthetic function is known to be impaired after burn injury, in both burn scar and adjacent normal skin [7].

Increasingly data suggest that vitamin D has a much wider range of effects than maintaining adequate serum calcium levels [8]. It is consistent with the observation of the vitamin D receptor in several cell types, the autocrine or paracrine production of 1,25-dihydroxyvitamin D (1,25(OH)₂D, calcitriol) in several extrarenal organs, and kidney endocrine production of 1,25(OH)₂D [9]. These pleiotropic properties may influence immune response, cell proliferation, muscle performance, energy metabolism, bone strength independent of its actions on calcium absorption [10].

Data about hypovitaminosis D and osteopenia following burn are scarce [11]. Most published studies have been conducted in burn children, either during acute or rehabilitation phase. Only two prospective cohort studies have been performed in adult burn patients during acute phase after injury [12,13]. Links between vitamin D and muscle health has been more widely studied. Vitamin D may prevent muscle proteolysis and vitamin D deficiency may impair muscle force generation [14]. Furthermore, vitamin D supplementation may increase proximal muscle strength in adults experiencing vitamin D deficiency [15]. In burn literature, to the best of our knowledge, such clinical data are lacking.

The present pilot study was a randomized controlled trial conducted in Belgian adult burns. The objective was to assess the effects of a cholecalciferol supplementation and an optimized calcium regimen on vitamin D status, bone and muscle health during the recovery phase of burn injury.

2. Method

2.1. Subjects

This randomized controlled trial was conducted after approval by the local Ethics Committee of the University Hospital of Liège (ref 2012/13) and was registered in ClinicalTrials.gov database (reference NCT02092701).

Burn patients were recruited among database of two Belgian burn centers. Inclusion criteria were: age over 18 years, occurrence of injury between 2005 and 2011, burn surface area (BSA) greater than 10%. Pregnancy, renal or liver

failure, hypo or hyperparathyroidism, prior vitamin D substitution, treatment using systemic corticosteroids or anti-epileptic drugs, regular exposure to UVB (solarium), unstable cardiovascular disease or acute musculo-skeletal injury prohibiting physical exercise, were considered exclusion criteria. Eligible patients were contacted by phone call: exclusion criteria were examined and oral consent was obtained. Appointment was set for the first visit of the protocol at hospital during which consent was confirmed and signed. Patients were randomized into two groups after oral consent and before any vitamin D status assessment: a group substituted with vitamin D and calcium (Group D) and a group receiving placebo (Group P). A randomization list was generated using the online Graphpad randomizer tool (<http://graphpad.com/quickcalcs/randomN1.cfm>). Patients, investigators and laboratory technicians were blinded to the randomization process. Only the physician responsible for the present study could not be blinded because it was impossible for practical and financial reasons to get active drug and placebo with similar appearance. However, this physician was in charge of drug administration only.

2.2. Study protocol

The present trial was conducted from October 2012 to November 2013. Each patient was sent for medical visit and exams every 3 months (M0, M3, M6, M9 and M12). In the meantime, they were contacted monthly by phone to ensure the proper compliance to the protocol and the lack of any side effects.

At first visit, daily calcium intakes were evaluated using the online French GRIO tool [16] (<http://www.grio.org/calcul-apport-calcique-quotidien.php>) and physical activity was assessed using the Compendium of Physical activities [17]. Each activity, except daily common movements, was expressed as Metabolic equivalent of task (MET) per hour. The average weekly activity was evaluated at each visit.

During all the duration of the protocol, patients were asked to avoid solarium and not to change their weekly physical activity.

Patients assigned in Group D received an intramuscular injection of cholecalciferol every 3 months (quarterly regimen) at a dose of 200,000 IU (Vitamin D3 B.O.N., Bouchara Recordati, Levallois-Perret, France) and a daily oral dose of calcium carbonate (Fagron, Waregem, Belgium). According to basal daily intakes ≤500 mg or >1 g, patients received once a day a dose of calcium carbonate of 1 g or 500 mg, respectively. Patients assigned in Group P received similar placebo regimens: intramuscular injection of 1 ml of normal saline every 3 months and daily capsules containing lactose. Intramuscular injections were performed by one dedicated physician at M0, M3, M6 and M9. Quarterly IM vitamin D was chosen to maximize compliance and to avoid under-treatment. At each visit, patients were asked to bring back unused tablets of calcium or placebo to control compliance.

Vitamin D status was assessed at each visit. Bone mineral density, bone metabolism markers and muscle strength were assessed at inclusion (M0) and at completion of the protocol (M12). Visits took place in the morning or in the afternoon but

same schedule was maintained for each patient during all the duration of the protocol. At each visit, patients were asked to be fasting for last dinner or breakfast according to their appointment.

2.3. Laboratory measurements

Blood samples were collected through venous puncture. Blood was drawn into serum gel tubes, plasma heparin tube and EDTA tube (Venosafe Plastic Tubes, Terumo, Haasrode, Belgium), before being centrifugated (3500 rpm, 15 min, 4 °C). Supernatant was frozen at -80 °C and stored for later analysis.

Vitamin D metabolites (25OH-D and 1,25(OH)₂-D) and main regulators (parathormone (PTH) and fibroblast growth factor 23 (FGF23)) were measured. Serum level of 25OH-D and third generation PTH were determined using chemoluminescence (Liaison, DiaSorin, Stillwater, MN, USA). Normal ranges (NR) were, respectively, 30–80 ng/ml and 4–26 ng/l. Vitamin D deficiency was defined as 25OH-D levels <20 ng/ml and vitamin D insufficiency as 25OH-D levels of 21–29 ng/ml. In our lab, the coefficient of variation of the Liaison[®] 25OH-D assay was 6%. 1,25(OH)₂-D was assayed using chemoluminescence (iSYS automate, IDS, Boldon, UK): NR was <85 pg/ml. C-terminal FGF23 concentrations were determined with ELISA method (Immunotopics, Immunotopics International, San Clemente, CA, USA): NR was 30–176 RU/ml. Serum levels of albumin (ALB) was assayed using spectrophotometry (Cobas automate, Roche, Mannheim, Germany): NR was 38–49 g/l. Vitamin D binding protein (VDBP) concentration was determined using ELISA (R&D Systems, Minneapolis, MN, USA). Levels of calcium (Ca), phosphate (P) and creatinine were assayed with Cobas automate (Roche, Mannheim, Germany): NR were, respectively, 2.15–2.6 mmol/l, 0.74–1.51 mmol/l and 7.2–11.8 mg/l. Serum collagen type 1 cross-linked C-telopeptide (CTX), serum type 1 procollagen N-terminal (P1NP) and serum bone alkaline phosphatase (b-ALP) were assayed using chemoluminescence (iSYS automate, IDS, Boldon, UK). CTX concentration <695 ng/l and b-ALP concentration <21 µg/l were considered normal. Normal range for P1NP was 7.5–95.4 ng/ml with changes according to age and sex [18]. Serum tartrate-resistant acid phosphatase 5b (TRAP) was measured using ELISA (IDS, Boldon, UK): NR was 1.5–4.7 U/l. CTX and TRAP are bone resorption markers while P1NP and b-ALP are bone formation markers.

2.4. Bone mineral density (BMD) measurement

BMD was assessed using dual energy X-ray absorptiometry (DXA) (Discovery A, Hologic, Bedford, MA, USA) of the lumbar spine (lumbar vertebrae 1–4) and hip regions (total, shaft, trochanter, and neck). Scans were performed by the local blinded technologist and then analyzed by a blinded specialized physician. The scans were analyzed using the manufacturer's software. Absolute BMD was analyzed. Results from BMD measurements were also transformed in Z-scores and T-scores using sex-matched reference data provided by the manufacturer and standardized for Belgian patients [19]. T-score was used for postmenopausal women and men ≥50 y and Z-score was used for women and men <50 y [20]. DXA reference site was spine for patients <50 y and hip for patients

>50 y, as degenerative arthrosis may be confounder for spine measurement in patients >50 y. Osteoporosis and osteopenia were defined as a T-score, respectively, ≤-2.5 and ≤-1 according to World Health Organization guidelines. A Z-score ≤-2 was defined as “below the expected range for age”.

2.5. Dynamometry

Measurements of knee muscles strength were conducted using a Humac Norm isokinetic dynamometer (CSMI, Stoughton, MA, USA). The tested limb was chosen according to dominance (kicking leg). Before commencement of the isokinetic test, patients pedaled on stationary bike at low resistance level for 6 min (50–75 W, 60–70 rpm) and performed stretching exercises on the involved muscle groups for 5 min. Patients were then installed on the dynamometer seat with back reclined at 85°. The trunk and thigh were stabilized with belts. The knee range of motion was 100°, with the leg at initial position of voluntary maximal extension (Fig. 1). The selected isokinetic speeds were 60°/s (three repetitions of testing) and 180°/s (five repetitions of testing) in the concentric mode. These testing sequences were preceded by familiarization at 120°/s and by three submaximal trials at the selected speed. Successive testing velocities were separated by 1 min of recovery. The isokinetic testing procedure enabled the measurement of peak torque (PT in N m) and maximal work (W_{max} in joules). PT was normalized for body weight (in N m/kg). Peak torque is the maximum muscle force applied in dynamic conditions. The work performed is a more representative measure of muscle function because it takes into account the force output throughout the range of movement. All tests were performed



Fig. 1 – Isokinetic dynamometer.

under supervision of the same trained and blinded physiotherapist. Patients received the same instructions and were not allowed to look at the control screen during testing.

2.6. Statistical analysis

There was no sample size calculation as no relevant previous data were available in burn patients.

Statistical analysis was performed using Graphpad Prism (version 6.0 for Mac OSX, Graphpad Inc., San Diego, CA, USA). Because small sample size, data were analyzed using

nonparametric statistics. Evolution of laboratory measurements from M0 to M12 was considered as area under the curve (AUC) calculated via trapezoidal rule. Data were expressed as median (min-max) or counts and percentages. Unpaired data were compared using Mann and Whitney test. Paired data were compared using Wilcoxon test. Categorical variables were compared using Fisher's exact test. Correlations between different parameters were assessed using nonparametric Spearman test. A p value <0.05 was considered as significant. Bonferroni correction was applied for multiple comparisons.

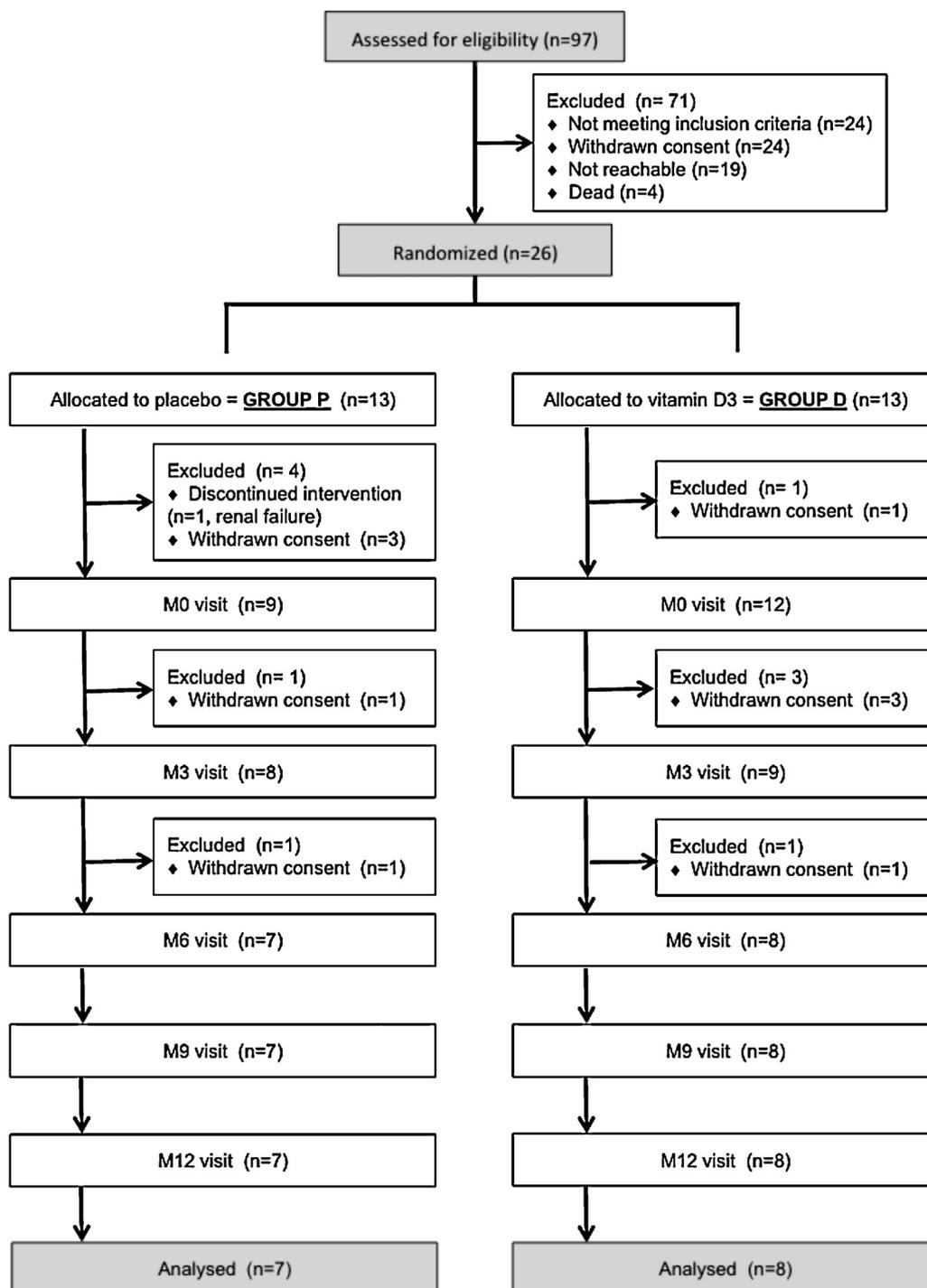


Fig. 2 – Flow chart.

3. Results

3.1. Patients

Ninety-seven patients were initially enrolled, 26 were randomized and 15 were finally analyzed (Fig. 2). Demographic data are presented in Table 1. The two groups were similar in terms of age, weight, body mass index, BSA, time since burn center discharge, in-burn center length of stay, mean weekly physical activity and basal daily Ca intakes. Despite lack of statistical significance, men tended to be more numerous than women. Most of the patients were Caucasian, only one patient in Group P was African.

Throughout the duration of the protocol, all patients followed the instructions and reported less than 5% of provided pills.

No adverse events were noted during follow-up. At mid-protocol, one patient of Group D complained of lumbar pain related to degenerative disease, which prohibited muscle testing. Analysis of muscle related data was then based on 7 patients in Group D.

3.2. Vitamin D status

At inclusion, two third of all patients presented a VD deficiency while 20% presented a VD insufficiency. Median serum 25OH-D level was 16 (7–36) ng/ml. Serum 1,25(OH)₂-D level was 37 (14–57) pg/ml. Albumin concentrations were into NR: 46 (37–49) g/l. VDBP levels were 322.8 (116.5–490.1) µg/ml. Only two patients presented VDBP levels <150 µg/ml: a Black African one and a young White European. No patients presented ion disturbances. Creatinine concentrations were into NR: 9.1 (7–11.5) mg/l).

At M0, levels of 25OH-D tended to be higher in Group D than in Group P ($p = 0.09$). In parallel, PTH levels were significantly lower in Group D than in Group P ($p = 0.01$, with statistical significance threshold after Bonferroni correction < 0.025). Levels of 1,25(OH)₂-D, FGF23, Ca and P did not statistically differ between groups. One year evolution of these parameters are presented in Table 2. Regarding total 25OH-D concentrations, Group D and Group P did not evolve similarly ($p = 0.01$). After one year of supplementation, 25OH-D increased by 111 (57–270)% in Group D while levels ranged from –14% to 100% in Group P ($p = 0.001$). In Group D at M12, 25OH-D reached 40

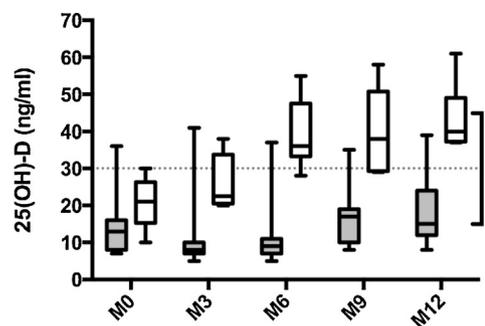


Fig. 3 – Median and ranges (min–max) of 25OH-D at each visit (M0, M3, M6, M9 and M12) in Group P (gray boxes) and Group D (white boxes). Dotted lines at 30 ng/ml 25(OH)-D represent cut-off values defining hypovitaminosis D. Evolution of 25OH-D in Group P and D from M0 to M12 was significantly different. Levels of 25OH-D at M12 in Group D were significantly higher than in Group P.

(37–61) ng/ml: it was significantly higher than in Group P ($p = 0.003$). Such concentrations were achieved after 6 months of treatment before obtaining a plateau effect (Fig. 3). No significant variation in PTH, 1,25(OH)₂-D or FGF23 was observed in both groups. No hypercalcemia was disclosed in Group D. Albumin and VDBP concentrations remained stable between M0 and M12.

3.3. Bone health

Of the 9 patients ≥ 50 y, 4 were considered osteopenic (T-score ≤ -1) and one was osteoporotic at inclusion (T-score ≤ -2.5). Hip T-score ranged from –2.9 to 0.3. BMD was 0.76 (0.519–0.929) g/cm². Two of the 6 youngest patients (<30 y) had low bone mass (Z-score at –1.7): BSA was <30% for both of them. Spine Z-score ranged from –1.7 to 2. BMD was 1.043 (0.872–1.202) g/cm². No correlation was observed between BSA and BMD.

No significant increase in BSA was detected at M12 in both groups, including in patients presenting low bone mass. At M12, BMD variation was –0.13 (–1.79 + 6.74)% in group P and 1.27 (–1.3 ± 11.7)% ($p = 0.39$).

One year evolution of bone biomarkers is presented in Table 2. No significant variation in CTX, TRAP, P1NP or b-ALP was observed in both groups.

Table 1 – Demographic data BMI: body mass index, BSA: burn surface area, LOS: length of stay, MET: metabolic equivalent of task, Ca: calcium.

Data	Group P (n = 7)	Group D (n = 8)	p Value
Sex ratio (M/F)	4/3	7/1	0.28
Age (y)	54 (29–64)	46 (25–58)	0.27
Weight (kg)	91 (55–118)	76 (48–100)	0.28
BMI (kg/m ²)	31.2 (19.3–38.1)	25.1 (18.5–31.1)	0.08
BSA (%)	30 (10–55)	32.5 (10–60)	0.71
Time since discharge (y)	3 (2–5)	2.75 (2–5)	0.69
In-burn center LOS (months)	2 (0.2–6)	3 (0.25–4)	0.38
MET-hours per week	166.8 (70–281.5)	208.2 (93.4–393.5)	0.28
Daily Ca intakes (mg)	604 (472–1947)	558 (93–1053)	0.39

Table 2 – Laboratory data at M0, M6 and M12 in Group P and Group D. Data are expressed as median and ranges (min–max). Evolution of laboratory measurements from M0 to M12 was considered as area under the curve (AUC) calculated via trapezoidal rule. AUC in Group P and Group D were compared using Mann andWhitney test (p value <0.05 was considered significant). PTH was significantly lower in Group D than in Group P ($p = 0.01$). For all the parameters, no significant variation over time was observed in both groups.

Data	Normal ranges	Group P (n = 7)			Group D (n = 8)			p Value
		M0	M6	M12	M0	M6	M12	
25OH-D	30–80 ng/ml	13 (7–36)	9 (5–37)	15 (8–39)	21 (10–30)	36 (28–55)	40 (37–61)	.0093
1,25(OH)2-D	23–109 pg/ml	39 (14–45)	36 (30–56)	44 (37–63)	36 (19–57)	51 (40–73)	57.5 (49–69)	0.295
PTH	4–26 ng/ml	41 (29–77)	47 (28–94)	36 (26–82)	23.5 (13–50)	22 (13–41)	24 (12–28)	.0003
FGF23	30–176 RU/ml	58.4 (45.3–195.7)	52.7 (37.4–110.8)	59.9 (29.6–90.7)	59.7 (40.6–66.7)	62.6 (33.2–84.1)	66 (44.1–96.1)	0.931
Ca	2.15–2.6 mmol/l	2.32 (2.23–2.4)	2.29 (2.21–2.32)	2.35 (2.29–2.4)	2.36 (2.18–2.4)	2.28 (2.21–2.36)	2.39 (2.3–2.48)	0.636
P	0.74–1.51 mmol/l	0.89 (0.71–1.16)	0.96 (0.85–1.16)	0.92 (0.7–1.22)	0.9 (0.71–1.26)	0.96 (0.71–1.06)	0.96 (0.8–1.3)	0.845
CTX	<695 ng/l	408 (224–1385)	271 (163–800)	499 (166–800)	379 (200–1001)	342 (152–679)	482 (97–1085)	0.845
TRAP	1.5–4.7 U/l	3 (2.5–5.7)	2.6 (2.5–4.1)	2.6 (2.3–4.9)	3.35 (2.6–4.8)	3.1 (2.6–3.9)	3.5 (2.4–4.1)	0.752
PTNP	7.5–95.4 ng/ml	33 (23–188)	38 (21–197)	34 (23.6–114)	40 (15–61)	42.5 (20–64)	33.6 (16–65.3)	0.761

3.4. Muscle strength

At inclusion, a great variability was observed among patients in term of isokinetic results. Hamstrings PT/BW at 60°/s ranged from 0.4 to 1.47 N m/kg, at 180°/s ranged from 0.27 to 0.99 N m/kg. Quadriceps PT/BW at 60°/s ranged from 0.69 to 3 N m/kg, at 180°/s ranged from 0.51 to 1.92 N m/kg. No significant difference was noted between groups at baseline. No correlation was found between initial PT/BW scores and BSA or physical activity.

At the end of the protocol, patients of Group P did not show any significant modification of quadriceps performances. In contrast, patients of Group D, experienced a significant improvement in quadriceps PT and W_{max} at the speed of 180°/s ($p = 0.047$ and $p = 0.03$, respectively) (Fig. 4). Quadriceps PT increased by 10.4 (–1.6 + 20)% in Group D while variation was 0 (–16.7 + 10.7)% in Group P. Similarly, quadriceps W_{max} increased by 10.64 (0–23.9)% in Group D while variation was 0 (–11.8 + 17.1)% in Group P. Quadriceps performances at low speed and hamstrings performances at both speeds did not significantly improved in either Group D or Group P.

4. Discussion

The present pilot trial is the first to consider the supplementation of hypovitaminosis D in adult burn patients in sequelar stage. Despite the low number of included patients (inherent problem in this population), this study supports efficiency of a quarterly intramuscular injection of 200,000 IU cholecalciferol to normalize VD status. The IM route was not previously described in burn research. In the present study, this route was preferred to ensure compliance and to avoid malabsorption as it may occur in about 13% of the population [21]. While adverse events have been described with high doses of PO VD3 [22,23], the present regimen of a quarterly IM injection of 200,000 IU cholecalciferol was safe: the highest 25OH-D concentration observed in this study (61 ng/ml) was far from the estimated toxic concentrations and no variations in calcium levels were disclosed. Indeed, it has been demonstrated that an IM injection of cholecalciferol produces slower increase in 25OH-D than an equivalent PO dose [24].

The combined cholecalciferol and calcium treatment tested in this trial did not increase bone mineral density nor influenced bone remodeling. Lack of marked beneficial effect of the present supplementation regimen, even in young osteopenic patients, is quite disturbing. Yet, 25OH-D reached sufficient levels in the intervention group [25]. Some hypotheses may be raised. First, 25OH-D levels normalization occurred after 6 months: six months of VD sufficiency may be inadequate to detect bone modifications. Second, DXA is a quantitative investigation of bone health, neglecting bone microstructure that is a major contributor to bone strength. In future works, it would be interesting to include qualitative evaluation when focusing on treatment of osteopenia associated with hypovitaminosis D or burn injury [26]. Finally, mild severity of burn injury or mild severity of VD deficiency may have influenced results. Indeed, it has been suggested that benefit of VD supplementation are more pronounced in patients who are markedly VD deficient [27]. Due to the limited

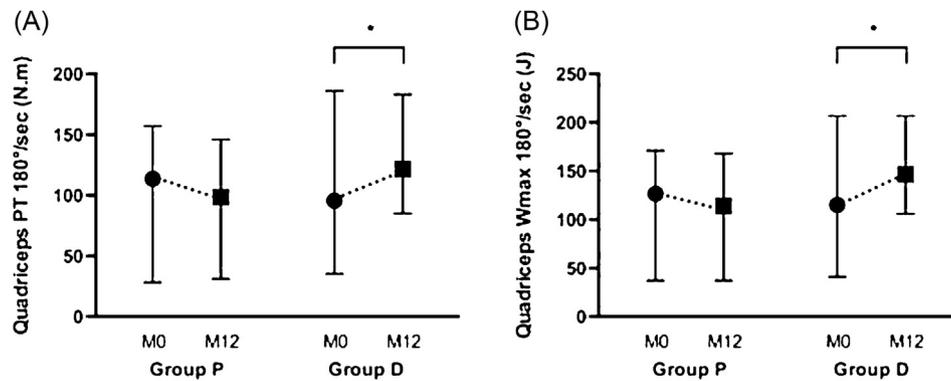


Fig. 4 – Quadriceps performances (median and ranges) at inclusion and completion of the study in Group P and Group D. PT: peak torq, W_{max} : maximal work, * $p < 0.05$.

number of included patients, association between baseline 25OH-D concentrations and response to VD supplements could not be assessed in the present study.

On the other side, the present cholecalciferol regimen, combined with optimized calcium intakes, improved quadriceps performances at rapid angular velocity. VD deficiency and critical illness myopathy may induce atrophy and reduction of type 2 muscle fibers [28,29]. Type 2 muscle fibers are fast-twitch and are recruited in activities of high intensity and short duration. These fibers are also the first to be recruited in balance, justifying the association between VD deficiency and falls in old people [30]. Thus, the present results are not surprising. Moreover, quadriceps is generally largely involved in case of burn related myopathy and sarcopenia. Improving quadriceps capability may be of high interest during rehabilitation because quadriceps is a key muscle for various functional activities.

The present study provides information about post-burn status. VDBP, which has been described as low during acute phase following burn injury [2], seems to recover during acute phase. Actin scavenging following cell trauma, reduced hepatic synthesis and hemodilution may explain the initial low levels of VDBP. However, other informations are quite worrying. There was a high prevalence of hypovitaminosis D in adult burn patients in the recovery phase. Comparison with prevalence in other populations is hazardous: age, samples collection conditions or 25OH-D assays may differ. Osteopenia was common, even occurring in young men. Osteopenia is not only the result of immobilization as bone recovery is observed after 6 months of reambulation [31]: burn induced bone loss is a clinical reality [1,2]. Finally, muscle performance measurements were extremely heterogeneous at baseline. Comparison of the present performances with those observed in healthy population or other diseases seems to be interesting but need to take into account the essential determinants of muscle performances, namely weight, gender, sex and physical activity. Different authors seem to report PT/BW of at least 2 N m/kg for quadriceps testing at 60°/s in young healthy men. Overall, it can be estimated that 15–60% of the present burn patients experienced below average muscle performances, depending on reported conditions. However, definition of such deficit for a given patient is currently unavailable. Future large

studies should aim to determine a cut-off for muscle failure, corresponding to an impaired quality of life or reduced daily living activities. Indeed, it has been demonstrated that muscle strength is a limiting factor for essential tasks for independent living such as rising from a chair [32,33]. Thus, weaker post-burn patients should be promptly recognized: they would benefit from an appropriate exercise program during rehabilitation.

Vitamin D supplementation is patently indicated in burn patients. Association of calcium supplements may have substantial interesting effects on bone turnover and bone density, even higher than VD alone [34,35]. This has to be further investigated in burn patients. If 30 ng/ml is thought to be the minimal 25OH-D level required for bone health [36], optimal level of 25OH-D for muscle is unknown [25]. Even less is known regarding VD and burn patients. Based on the present study, the issue of how much to give has been resolved, provided that acute phase has passed. However, which is the optimal timing remain key questions.

In moderate to severe burn patients, hypovitaminosis D may not be the only risk factor for osteopenia or sarcopenia. Neuroendocrine response to injury and immobilization may lead to muscle and bone loss. In this context, VD supplementation may be one approach among a global rehabilitation program including at least muscle reconditioning [37,38], dietary advises, treatment of sleep disorders and psychological support, and adjuvant drugs. Oxandrolone is the most studied anabolic drug in burn patients, is safe and has demonstrated beneficial effects in lean mass restoration [39,40]. Antiresorptive agents such as bisphosphonates have been studied in burn children with promising result both on bone [41,42] and muscle mass [43]. Place of bone anabolic drugs (such as strontium ralenate or anti-sclerostin antibody) has to be specified in burn related bone loss management.

Some limitations need to be considered in the present trial. First, Black patients were not excluded whereas they are known to have higher bone mineral density and lower VDBP levels [44]. However, the only African patient of the study was assigned in Group P so it is unlikely he influenced the present results. Second, total sun exposure was not counted up in each patient. However, all burn patients are advised to avoid sun exposure and if any, to use sunscreens. Though, sun exposure is probably a minor factor interfering with the present results.

Finally, no nutritional strategy was associated to the protocol. In particular, quantitative and qualitative protein or trace element intakes were not controlled.

In conclusion, the present randomized trial is the first report of a safe and efficient VD supplementation over one year. It allowed correction of hypovitaminosis D in burn adults, reaching 25OH-D levels considered at present as largely sufficient (i.e. 40 ng/ml). Combined with optimized calcium intakes, cholecalciferol increased quadriceps strength when tested at high velocity but did not influence bone health. Moreover, the present study highlighted worrying observations regarding post-injury condition. Hypovitaminosis D and low bone mass were highly prevalent in burn adults, even in young ones. Muscle performances were quite heterogeneous and probably insufficient. This should be of concern when designing future studies. Interest and modalities of vitamin D treatment in global burn care need to be specified, as well as its place among other anabolic strategies. Most of all, musculoskeletal status of burn patients need to be determined as soon as possible during rehabilitation phase, using quantitative and qualitative techniques, in order to provide targeted therapeutic strategies. Further studies should help to identify patients who will need an aggressive support to prevent or treat musculoskeletal complications. Ultimate goal of total burn care is not only survival but also quality of life. This requires an aggressive but also targeted and individualized rehabilitation approach.

Author's contribution

AFR, MFD, JLC, PD and EC designed research; AFR, CR and AL conducted research; AFR, MFD, DL, JLC and EC analyzed data; AFR wrote paper; MFD, DL, CR, JLC, PD and EC critically reviewed paper. All authors approved the final manuscript.

Conflict of interest statement

Each author declares that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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