

Article

Total Serum Calcium and Recovery after Rehabilitation in Patients with Stroke

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Abstract: Calcium deficiency is frequently observed in stroke survivors, but no data exist concerning the relationship between calcium and rehabilitation outcome in patients with stroke. Therefore, we examined in a group of subacute stroke patients if changes in the Barthel Index after a rehabilitation treatment were associated with blood parameters related to calcium status. We retrospectively explored serum calcium status (total calcium, ionized calcium), serum total protein status, and serum albumin percentage in 30 subacute stroke patients admitted to our rehabilitation center. Patients underwent a 6-week rehabilitation treatment (each session lasting 45 min, 2 sessions/day, 6 days/week). Overall, 26.7% of patients had total calcium levels below the reference range, whereas 100% of patients had ionized calcium in the reference range. Total protein and albumin were below the reference range in about 77% and 67% of patients, respectively. We found that only total calcium was correlated with the change from baseline of the Barthel Index (BI) ($\rho = 0.466$, $p = 0.009$). A multiple linear regression model confirmed that in our sample the total calcium significantly predicted the change from baseline of the Barthel Index ($F_{5, 24} = 4.074$, $p = 0.008$, adj. $R^2 = 0.346$). This study suggests a possible connection between serum calcium status and total protein status of stroke patients undergoing rehabilitation treatment and rehabilitation outcomes. Further investigations are necessary to confirm the importance of testing serum calcium status of patients at admission in a rehabilitation unit for an eventual supplementation or a dietary personalized program.

Keywords: calcium; stroke; rehabilitation; nutrition

1. Introduction

Stroke is the first cause of disability and the second largest cause of death worldwide [1]. Patients after stroke have a very heterogeneous clinical spectrum together with dissimilar and often incomplete recovery of motor function in response to rehabilitation treatment [2].

Malnutrition is frequently observed in stroke survivors, and a recent meta-analysis identified a plethora of potential risk factors for this condition [3]. Malnutrition should be monitored in patients with stroke: in fact, it can significantly influence the outcome of post-stroke rehabilitation, leading to a poor recovery [4].

A consequence of malnutrition is calcium deficiency. Calcium is a key essential nutrient that is fundamental in the human body; it is involved in vascular contraction, vasodilatation, muscle functions, nerve transmission, intracellular signaling, and hormonal secretion [5]. Calcium levels in the

human body are strictly controlled, as are other essential metals, and regulated by three hormones: parathormone, calcitonin, and Vitamin D [6].

If levels of calcium are low for long periods, people may develop dry and scaly skin, brittle nails, and coarse hair. Muscle cramps involving the back and legs are common. These symptoms disappear if the calcemia status is restored. For a good level of serum calcium, a controlled diet together with adequate level of Vitamin D are generally sufficient. A period of hypocalcemia status that is too long can affect the brain and cause neurologic or psychologic symptoms, such as confusion, memory loss, delirium, depression, and hallucinations, as well as osteoporosis problems. This condition can be affected by other causes, such as a low level of parathyroid hormone (hypoparathyroidism), pancreatitis, low magnesium levels, or certain drugs [6]. For this reason, adequate calcium levels are important for all subjects but have to be particularly controlled during ageing, especially for bone health.

A very recent study on 84 patients with acute ischemic stroke was performed to determine if admission calcium status was associated with stroke outcomes after systemic thrombolysis [7]. Patients with low admission calcium who received systemic thrombolysis more frequently, required also endovascular therapy. This result could represent unique thrombosis physiology, which deserves further investigation. While limited by small numbers, these patients with low calcium also showed a trend towards worse observed clinical outcomes [7].

A study on 659 patients with an ischemic stroke showed that higher serum calcium levels measured at 72 to 96 h from insult was associated with lesser stroke severity and better 3-month functional and independence outcome [8]. In this study, the highest quartile calcium levels were associated with a higher score on the Barthel Index scale—a measure of independence in activities of daily living—3 months after the acute event. However, to the best of our knowledge, the relationship between calcium levels and the outcome of a rehabilitation intervention was not investigated.

In light of the above, in a group of subacute stroke patients admitted to our rehabilitation facility for a rehabilitation treatment of 6 weeks, we examined if changes in independence score measured by the Barthel Index were associated with the following blood parameters: total and ionized calcium serum levels and two other nutrition status blood parameters, in particular albumin and total protein.

2. Materials and Methods

2.1. Subject

We retrospectively explored serum calcium status (total calcium, ionized calcium), serum total protein status, and serum albumin percentage in a group of 30 subacute stroke patients admitted to our rehabilitation center. Inclusion criteria were a first event (ischemic or hemorrhagic stroke, verified by MRI or CT), age between 40 and 85 years, and time since stroke less than six months. All participants gave informed consent according to the Declaration of Helsinki. The institutional Ethics and Experimental Research Committee of the Fondazione Don Carlo Gnocchi approved the study protocol on March 19, 2019 (FDG_19.3.2019).

2.2. Rehabilitation Intervention

Patients underwent a rehabilitation treatment for 6 weeks, each session lasting 45 min, with a frequency of 2 sessions a day, 6 days/week. Each day, one session aimed to restore lower limb functions through sensorimotor stimulation; passive, active-assisted, and active mobilizations; exercises for muscle strength recovery; stretching; functional and task-oriented training; proprioceptive exercises; postural passages and transfers; sitting and standing training; motor coordination and balance training; walking training; and activities of daily living recovery training. The second session, instead, targeted the upper limb by means of therapeutic tasks focused on functional improvement, sensorimotor reorganization, and spasticity inhibition obtained through task-oriented exercises that included reaching and grasping movements and activities of daily living (e.g., transfers, dressing, brushing and combing hair, etc.).

2.3. Rehabilitation Outcome

The changes in functional independence in personal activities of daily living were assessed using the modified Barthel Index (BI), which is an ordinal scale used to measure performance in activities of daily living [9]. It assesses the ability of an individual with a neuromuscular or musculoskeletal disorder to take care of himself or herself and consists of 10 items evaluating both personal care (feeding, dressing, and hygiene) and mobility activities (transferring, walking or wheeling).

Each performance item is rated on this scale with a number of points assigned to each level or ranking. Possible values range from 0 to 100, with lower scores representing a greater dependency. Specifically, we evaluated the BI at admission (T0) and after the rehabilitation treatment (T1), and we used the change from baseline ($\Delta BI = BI_{T1} - BI_{T0}$) as the rehabilitation outcome.

2.4. Biochemical Analyses

Blood analyses in all patients were collected and tested within 1 week from admission to our rehabilitation unit through an external laboratory following medical prescription.

Total calcium values were analyzed with the Arsenazo III colorimetric method; it employed a metallochrome indicator: (3,6-bis ((E)-(2-Arsonophenyl)diazanyl)-4,5-dihydroxynaphthalene-2,7-disulfonic acid) 2,7-Bis (2-arsonophenylazo)-1,8-dihydroxynaphthalene-3,6-disulfonic Acid) that changes color when complexed to the calcium ion under physiological conditions [10].

Ionized calcium measurements were calculated from total calcium and total protein measurements following the formula proposed by Pfitzenmeyer [11].

Total proteins were determined with the Biuret method, a commonly test employed in biochemical laboratory for detecting the presence of peptide bonds, through a copper (II) ion which forms a coordination colored complex in alkaline medium. The percentage of albumin was calculated after performing serum protein capillary electrophoresis. In this technique, there is no solid matrix and proteins are separated by strong electroendosmotic forces. A high current is applied and negatively charged proteins such as albumin try to move towards the anode. Liquid buffer flows toward the cathode and drags proteins with a weaker charge [12].

2.5. Statistical Analysis

Descriptive statistics were used to summarize sample characteristics and biochemical variables. To investigate the relationship between biochemical variables and the improvement in performance in activities of daily living, as measured by the ΔBI , we used the Spearman's rank-order correlation coefficient. Biochemical variables that were significantly correlated with the outcome (ΔBI) were then included in a multivariate regression analysis, together with demographic and clinical characteristics, to identify possible independent predictors of recovery. *p* values lower than 0.05 were considered significant. Statistical analysis was performed using SPSS (IBM SPSS Statistics for Windows, Version 25.0, IBM Corp, Armonk, NY, USA).

3. Results

3.1. Demographics, Clinical Characteristics, and Biochemical Status

Demographics and clinical characteristics at baseline of the enrolled sample are reported in Table 1. The total calcium level was below the reference range in 26.7% of patients (mean values 9.0 ± 0.5 mg/dL; reference range: 8.8–10.6 mg/dL; Figure 1A), while all patients (100%) showed ionized calcium in the reference range (mean values of 5.0 ± 0.2 mg/dL; reference range: 4.5–5.3 mg/dL; Figure 1B). Total protein (mean values 6.3 ± 0.6 g/dL; reference range: 6.6–8.3 g/dL) and albumin (mean values $52.7 \pm 5.4\%$; reference range: 55.8–66.1%) were below the reference range in about 77% and 67% of patients, respectively (Figure 1C,D). After the rehabilitation treatment, patients improved their performance in activities of daily living, as showed by a statistically significant increase in their Barthel Index score (mean changes 21.6 ± 15.6 , $p < 0.001$).

Table 1. Baseline characteristics of the sample (n = 30).

Baseline Characteristics	Mean ($\pm$ SD) or n (%)
Age (years)	66.4 $\pm$ 12.2
Sex	
Men	18 (60.0%)
Women	12 (40.0%)
Index stroke type	
Ischemic	21 (70.0%)
Hemorrhagic	9 (30.0%)
Affected side	
Right	12 (40.0%)
Left	18 (60.0%)
Comorbidities	
Hypertension	22 (73.3%)
Type 2 Diabetes	10 (33.3%)
Dislipidemia	6 (10%)
Hearth disease	7 (23.3%)
Dysphagia	7 (23.3%)
Language impairment	7 (23.3%)
Neglect syndrome	6 (20.0%)
Days from index stroke to enrollment	90.5 $\pm$ 41.2
Modified Barthel Index (0–100)	25.3 $\pm$ 15.3

3.2. Relationship between Biochemical Status and Rehabilitation Outcome

The univariate analysis showed that only total calcium was correlated with the change from baseline of the BI (Spearman's $\rho = 0.466$, $p = 0.009$), whereas ionized calcium, total protein, and albumin were not (Table 2).

Table 2. Correlation between biochemical variables and changes in performance in activities of daily living, as measured by the changes of the Barthel Index (Δ BI). Values in bold indicate a p -value lower than 0.05.

Biochemical Variables	Δ BI	
	Spearman's ρ	p -Value
Total calcium	0.466	0.009
Ionized calcium	0.278	0.137
Total protein	0.278	0.136
Albumin	-0.095	0.623

Values in bold indicate a p -value lower than 0.05.

Finally, the multiple linear regression model (Table 3) significantly predicted the change from baseline of the Barthel Index ($F_{5, 24} = 4.074$, $p = 0.008$, adj. $R^2 = 0.346$). Among the covariates, only total calcium level significantly predicted the outcome.

Table 3. Multiple regression analysis.

Independent Variables	Unstandardized Coefficients		Standardized Coefficients	t	p -Value
	B	SE			
Total calcium	10.989	5.298	0.366	2.074	0.049
Time since stroke	-0.031	0.072	-0.083	-0.435	0.667
Age	-0.412	0.225	-0.325	-1.833	0.079
Type of stroke	7.504	6.201	0.226	1.210	0.238
Barthel Index (baseline)	-0.316	0.161	-0.314	-1.961	0.062
Constant	-41.947	57.915			

SE: Standard Error. Values in bold indicate a p -value lower than 0.05.

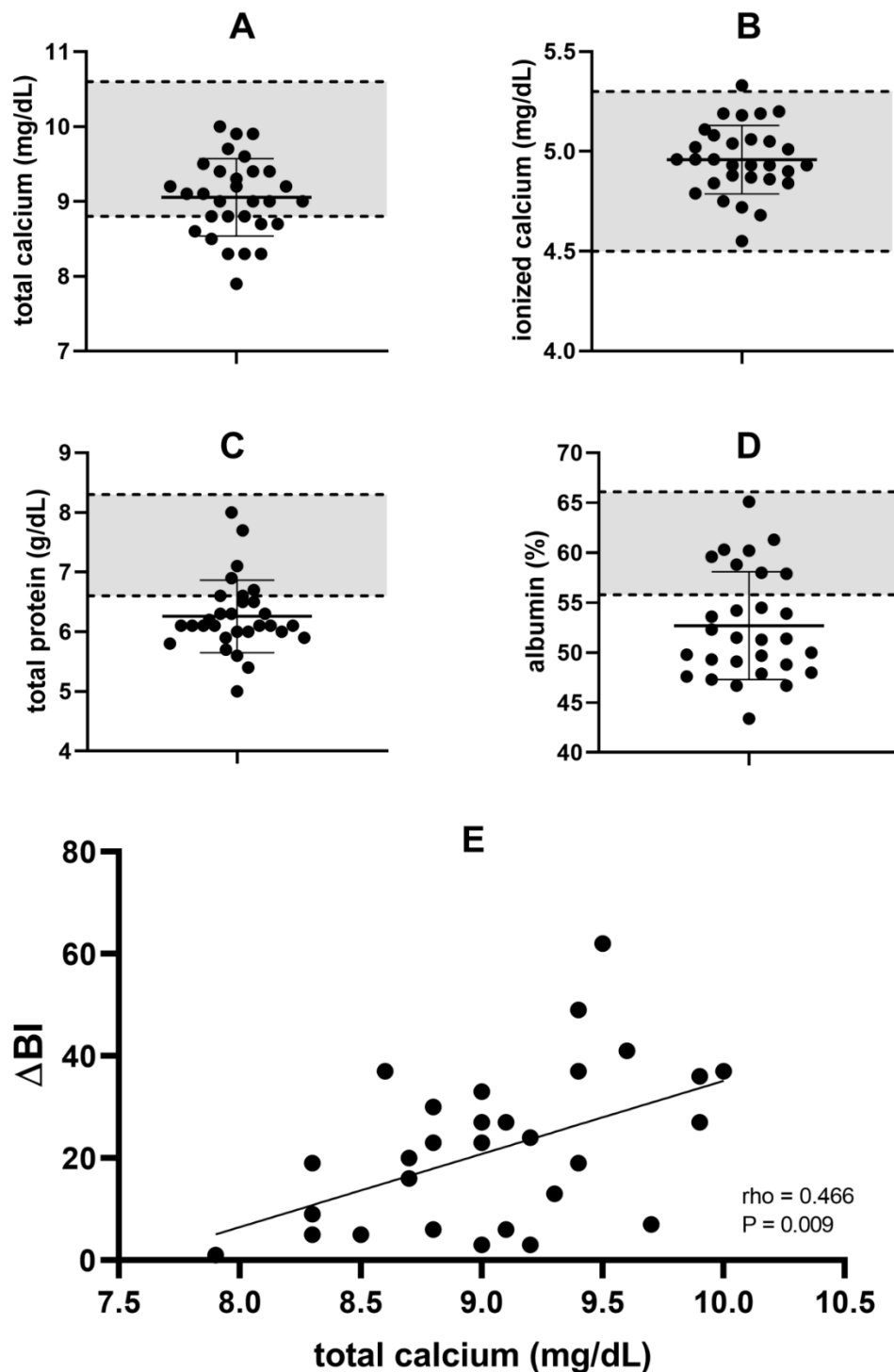


Figure 1. Dot plots (with mean values and error bars) of (A) total calcium, (B) ionized calcium, (C) total protein, and (D) albumin percentage. Reference ranges are identified by grey areas enclosed by dashed lines. (E) Scatterplot of the change from baseline of the Barthel Index (Δ BI) after 6 weeks of treatment versus the total calcium at baseline (ρ = Spearman's rank-order correlation coefficient).

4. Discussion

The principal result of this retrospective explorative study is that in stroke survivors undergoing rehabilitation, higher serum levels of total calcium at baseline significantly predict a better recovery in terms of disability, as measured by the change from baseline of the Barthel Index. This result is in

line with the study from Ovbiagele et al., which reported that in 659 subjects elevated serum calcium levels measured 72 to 96 h after ischemic stroke predicted a greater independence score on the Barthel Index scale measured 3 months after the acute event [8]. This result was confirmed by the same research group on 237 ischemic stroke patients, evaluated for calcium levels within 24 h from ictus [13]. They found that higher total serum calcium associated with lesser admission stroke severity and better functional outcome at discharge [13].

In our study, although the sample was smaller, we found the same association between serum calcium levels and ability in daily activity before and after 6 weeks of rehabilitation treatment, within 3 months from stroke onset. Moreover, in our group of subacute post-stroke patients calcium levels were almost all into the range of normality, but we found that patients with a higher total calcium profile improved better, probably because these patients had an enhanced general musculoskeletal condition and bone health status.

The studies on serum calcium levels and its role in recovery after stroke are very limited; most of the studies are focused on cellular calcium and its involvement in ischemia [14] or on the correlation between levels of calcium and stroke insult [15], as in a very recent study on patients with acute ischemic stroke performed to determine if admission calcium status associated with stroke outcomes after systemic thrombolysis [7].

Further studies probably are necessary to deepen our understanding of this topic and clarify the connection between clinical outcome and biochemical indices of calcium status.

For example, in light of the investigation of the general calcium status in patients, it is also important to test Vitamin D, which regulates calcium levels. A very recent meta-analysis showed that Vitamin D serum concentrations—specifically 25(OH) vitamin D—had a positive association with post-stroke recovery [16,17]. Unfortunately, we did not have the possibility to test 25(OH) vitamin D measurements, which would have better clarified this topic.

Another significant result of our study is that in the group of patients analyzed, serum total protein levels and serum albumin percentages were below the reference ranges (Figure 1C,D). It is known that patients with stroke have low compliance regarding the prescribed diet, have poorer eating habits often due to dysphagia, and generally have low dietary quality. An old observational study reported that poor nutritional status on admission predicts poor outcomes after stroke [18]. We did not find a correlation between protein serum levels and recovery outcome, but this result highlights a compromised general nutritional status of patients when admitted to our rehabilitation center.

A further study suggested that intensive nutritional supplements improved outcomes in stroke rehabilitation, as assessed with measures of motor function [19]; more recently, a supplementation of amino acids in post-stroke patients has been suggested [20].

Considering these factors, in post-stroke patients undergoing a rehabilitation program, the nutritional status, which includes mineral status (calcium), vitamin levels (in particular 25(OH) vitamin D), and protein levels, should be accurately screened in rehabilitation centers, following appropriate nutritional guidelines [18].

This study is only a small explorative investigation aimed to confirm the importance of the measurement of calcium status in rehabilitation programs on the basis of the correlation with recovery outcome. In fact, a limitation of this study is the small sample size, and therefore our results should be confirmed in a wider sample. Moreover, the scarce levels of total protein and albumin percentage suggest a compromised nutritional status, prompting a need to deepen our understanding of the overall nutritional status of patients hospitalized after stroke and undergoing rehabilitation treatment.

5. Conclusions

This preliminary explorative study suggests a possible connection between serum calcium status of stroke patients undergoing rehabilitation treatment and rehabilitation outcome and deserves further investigations to confirm the importance of controlling calcium status together with Vitamin D, both at admission and during the rehabilitation program. For this reason, clinicians should assess calcium

levels at admission in the rehabilitation centers and during the rehabilitation treatment for an eventual supplementation or a dietary program. Moreover, the very low levels of protein serum status found in almost all patients suggest that a special glance at the overall nutritional status should be taken into consideration in a post-stroke rehabilitation program, which should be planned with a more personalized and tailored approach.

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