VITAMIN D STATUS IN CRITICALLY ILL CHILDREN

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ABSTRACT

Background:

Hypovitaminosis D is an independent risk factor for cardiovascular disease, muscle weakness, impaired metabolism, immune dysfunction and compromised lung function. Hypovitaminosis D is common in critically ill adults and has been associated with adverse outcomes. The prevalence of hypovitaminosis D and its significance in critically ill children are unknown.

Methods:

We performed a prospective study to determine the prevalence of hypovitaminosis D in 316 critically ill children, and examined its association with physiological and biochemical variables, length of PICU stay and hospital mortality.

Results:

The prevalence of hypovitaminosis D ($25(\text{OH})\text{D}_3 < 50 \text{ nmol/L}$) was 34.5%. Hypovitaminosis D was more common in postoperative cardiac patients than general medical ICU patients (40.5% vs 22.6%; $p = 0.002$), and the cardiac patients had a higher inotrope score ($2.5 [1.9-3.3]$ vs. $1.4 [1.1-1.9]$, $p = 0.006$). Additionally, ionized calcium within the first 24 hours was lower in patients with $25(\text{OH})\text{D}_3 < 50 \text{ nmol/L}$ ($1.07 [0.99-1.14]$) compared to patients with normal vitamin D3 ($1.17 [1.14-1.19]$, $p = 0.02$). Hypovitaminosis D was not associated with a longer PICU stay or increased hospital mortality.

Conclusion:

Hypovitaminosis D is common in critically ill children, and is associated with higher inotropes in the post-operative cardiac population, but not with PICU length of stay or hospital survival.

INTRODUCTION

The role of Vitamin D in metabolism and homeostasis in the general population is well established, but there is now a growing interest in its potential association with overall health outcomes, and in acute, and critical illness.¹ The key metabolic role of vitamin D is in calcium and phosphate homeostasis, acting on 3 main target tissues: intestine, kidney and bone. However, there is now extensive literature highlighting the importance of the vitamin D pathway as a modulator of a variety of fundamental cellular processes. By regulating the expression of more than 200 genes, including those influencing cell growth, 1 α ,25-dihydroxyvitamin D₃ plays an important role in the proliferation, maturation and death of cells.¹⁻⁴

It is now well-recognised that low levels of vitamin D are very common in apparently healthy adults and children⁵, and in chronically ill patients. Several studies have identified vitamin D deficiency as an independent risk factor for cardiovascular disease, muscle weakness, impaired glucose metabolism, and immune dysfunction as well as compromised lung and endothelial function.⁶⁻¹⁰ Furthermore, in the general adult population vitamin D deficiency has been shown to be independently associated with increased all-cause mortality.¹¹

Vitamin D₃ is primarily produced through the action of ultraviolet B activity on cutaneous provitamin-D (7-dehydrocholesterol). Vitamin D₃ is a prohormone which is metabolized by the liver to 25-hydroxyvitamin D₃ (25(OH)D₃), which has a half-life of approximately 15 days. Although it is an inactive precursor to the active 1 α ,25-dihydroxyvitamin D₃ (1,25(OH)₂D₃) (produced in the kidneys), 25(OH)D₃ levels are generally used to reflect an individual's overall vitamin D status.^{12,13}

Several recent studies have investigated vitamin D status in acutely ill patients, and have revealed a high prevalence of hypovitaminosis D in adult intensive care patients, which was associated with adverse outcomes.¹⁴⁻¹⁶ The significance of vitamin D status in Paediatric Intensive Care Unit (PICU) patients is currently unclear. The aim of this study is to investigate the prevalence of hypovitaminosis D and its association with outcome in critically ill children requiring intensive care admission.

METHODS

Patients and Clinical Data

This prospective study was approved by the Ethics Committee of The Royal Children's Hospital Melbourne. We measured serum 25(OH)D₃ in 316 children on admission to a mixed cardiac and general tertiary PICU between 1st July 2010 and 1st March 2011. Patients with 22q11 deletion (which is routinely screened for in our cardiac population) and those with chronic liver, renal or metabolic bone disease were not included in this study.

Patient demographic and clinical data were recorded, including age, gender, weight, nature of admission (planned / unplanned), principal diagnosis, PIM2¹⁷ probability of death, RACHS¹⁸ risk of death (for cardiac patients), duration of ventilation, ICU length of stay, ICU outcome and hospital length of stay. The following cardiovascular complications were also documented: arrhythmia, hypotension (mean arterial pressure < 5th centile for age), hypertension (mean arterial pressure > 95th centile for age), inotropic score¹⁹, duration of vasoactive support, need for extracorporeal life support and cardiac arrest. Details of any proven intercurrent or acquired infection were recorded, as were the following metabolic derangements: hyperglycemia (> 10 mmol/L), hypoglycemia (< 3 mmol/L) and serum lactate > 1.8 mmol/L. Finally, for all patients the ionized and total calcium at admission and serum phosphate were routinely measured and the lowest ionized serum calcium within the first 24 hours documented. Additionally, total dose of calcium supplementations (mmol/kg) were recorded during the first 24 hours of intensive care stay.

Laboratory assays

25(OH)D₃ levels were drawn from indwelling arterial or central venous catheters, alongside other routine blood tests. Serum concentration of 25-Hydroxyvitamin D [25(OH)D₃] were measured using the immunoassay analyzer Roche Cobas[®] E 601 (Roche Diagnostics Ltd.). The reference range for this assay is 50 – 160 nmol/L (20 – 64 ng/mL; conversion factor x 0.4)). Serum calcium and phosphate on admission were measured with the Vitros[®] 5600 analyser (OrthoClinical Diagnostics). Ionized calcium (mmol/L), pH, lactate (mmol/L), bicarbonate (mmol/L) and blood glucose (mmol/L) on admission and during the first 24 hours were measured with the Siemens Rapidlab[®] 1265 (Siemens Healthcare Diagnostics Inc).

Definition of Hypovitaminosis D

Hypovitaminosis D was defined as a serum 25-hydroxyvitamin D₃ concentration of less than 50 nmol/L (or less than 20 ng/mL).²⁰⁻²²

Statistical methods

Statistical analysis was performed using STATA v.11 (STATA Corp., College Station, TX, USA). Results are described as numbers (%) for categorical data, median values (interquartile range) or geometric means (95% confidence interval) for continuous variables. Where appropriate continuous variables were transformed into their natural logarithm before used in the Student's t-test to compare the data of vitamin D deficient and sufficient patients. Associations between categorical variables and groups of patients with different serum 25(OH)D₃ concentrations were analyzed using the Chi-squared test.

RESULTS

Total Population

Three hundred and sixteen children were included in the study, which was carried out during the winter of 2010 through to early spring of 2011. Demographic data and clinical characteristics are shown in table 1. The prevalence of hypovitaminosis D (25(OH)D₃ < 50 nmol/L) in the cohort as a whole was 34.5%. 214 (67.7%) of the children were Caucasian, and the remainder: Asian (20.2%), Indigenous (3.8%) or other (8.3%). There was no difference in vitamin D status between ethnic groups, nor was there any seasonal variation in vitamin D levels (see Fig. 1). Hypovitaminosis D was more common in cardiac than non-cardiac patients (40.5% and 22.6% respectively, $p = 0.002$). The cardiac patients were more likely to be intubated and to require vasoactive drugs, had a shorter ICU stay and shorter duration of ventilation than the non-cardiac group.

General medical patients (Table 2)

This patient group comprised 106 children, of whom 24 (22.6%) had a serum 25(OH)D₃ level of less than 50 nmol/L, and 5 (4.7%) had a level less than 30 nmol/L. 78.3% of the general medical patients were unplanned admissions and 21.7% were planned post-operative admissions. Sixty-eight children (64.2%) suffered from chronic disease. These included neurological or neuromuscular disease (35.3%), genetic syndrome (25%) and cancer (20.6%). Hypovitaminosis D was not associated with age, weight, gender, the presence of chronic disease, need for mechanical ventilation, hypotension or hypertension, need for vasoactive support, ICU or hospital length of stay, a higher admission PIM2 score or death.

There was no association between the admission levels of calcium or phosphate and Vitamin D status. However, the lowest measured ionized calcium within the first 24 hours was significantly lower in children with $25(\text{OH})\text{D}_3 < 50 \text{ nmol/L}$ (1.07 [0.99-1.14]) compared to those with normal vitamin D_3 (1.17 [1.14-1.19], $p = 0.02$). The prevalence of hypoglycemia, hyperglycemia or lactic acidosis was not related to vitamin D status, and there was no association between vitamin D status and the presence of culture-positive infections.

Postoperative cardiac patients (Table 3)

Two hundred and ten children were included in this patient group. Eighty-five (40.5%) were found to be vitamin D deficient of whom 12 (5.7%) had a serum $25(\text{OH})\text{D}_3$ level of less than 30 nmol/L. There was no association between admission vitamin D_3 and age, weight, gender, PIM 2 score, duration of ventilation, ICU or hospital length of stay, RACHS probability of death, and overall hospital mortality. 164 of the postoperative cardiac patients (78.1%) received inotropic and/or vasopressor support, and the need for, or the duration of vasoactive drug therapy was not associated with vitamin D status. However, the inotrope score and the total dose of dobutamine (our first line agent after cardiac surgery) during the first 24 hours were higher in patients with hypovitaminosis D. These patients required more dobutamine within the first 24 hours (3.51 [2.68-4.56] mg/kg) compared to vitamin D sufficient children (1.99 [1.47-2.68] mg/kg, $p = 0.005$), and had a higher inotrope score (2.5 [1.9-3.3] vs. 1.4 [1.1-1.9], $p = 0.006$). There was no relationship between vitamin D status and the incidence of hypotension or hypertension, the incidence of any post-operative arrhythmia, or the need for extracorporeal support.

There was no relationship between vitamin D status on admission and total calcium and phosphate levels, and the lowest measured ionized calcium within the first 24 hours was similar in the two groups. However, the proportion of children who required calcium supplementation within the first 24 hours was higher in patients with hypovitaminosis D (51.3% vs. 38.2%, $p = 0.027$). The occurrence of hypoglycemia and hyperglycemia, lactic acidosis or proven infection was related to vitamin D status.

DISCUSSION

This study has shown hypovitaminosis D is present in more than one third of children on admission to PICU. It is more common in the cardiac than the non-cardiac population, in whom this was found in over 40%, compared to 22% of non-cardiac patients. Of note, none of our patients had 22q11.2 deletion, as this is routinely measured in all patients with congenital heart

disease presenting at our institution. Our findings differ to those published to date in the adult population, in that we did not see an association between Vitamin D status and survival, nor did we see an association between vitamin D status and duration of mechanical ventilation or ICU stay. In children with congenital heart disease, hypovitaminosis D was associated with greater levels of inotropic support, but was not associated with surgical complexity.

The prevalence of hypovitaminosis D in our population of critically ill Australian children was 34.5%. This is lower than in the adult ICU population in which vitamin D deficiency has been reported to range from 38-79%^{15,23}, but substantially higher than the reported prevalence of 18% in Mansbach's population-based study of healthy American children,⁵ and 8% in pre-pubertal Australian children.^{24,25} The published prevalence of vitamin D deficiency in American and Australian adolescents ranges from 29% to 68%.^{21,26-28} However, given that the median age of our cohort was less than 3 years, it is reasonable to suggest that hypovitaminosis D in children requiring PICU is more common than in their healthy peers. This would be particularly applicable to patients with cardiac disease, in whom the majority were infants, and would be deemed to have chronic disease burden. The associated vitamin D deficiency can be associated with a lack of sun exposure, chronic nutritional deficiency, as well as subtle impairment of hepatic and renal function. A recent study from our own institution demonstrated low vitamin D levels in 54.9% of chronically ill (non-cardiac) patients or disabled children who were not inpatients.²⁹ We did not observe this in our cohort, though this is somewhat limited by the heterogeneous nature of 'chronic disease' and relatively small numbers.

Vitamin D deficiency has recently been shown to be independently associated with all cause mortality in some cohorts of critically ill adults¹⁴⁻¹⁶ while other investigators have not found this relationship.^{23,30} We saw no association between vitamin D status and actual or predicted (PIM 2) mortality. Given that the overall mortality in the PICU is far lower than in the adult ICU population and that our study was not sufficiently powered to pick up differences in survival, we also applied duration of ventilation and length of ICU stay as additional outcome measures. However, we still did not demonstrate any relationship between vitamin D status and these surrogate measures of ICU outcome.

We were interested to examine the association between vitamin D status and the incidence of proven infection. Vitamin D plays a physiological role in regulating innate and adaptive immunity, and in the modulation of T-and B-cell activity, cytokine production and the expression of antimicrobial peptides.³¹⁻³³ Animal data have shown that severe vitamin D deficiency leads to reduced lung function,³⁴ and the Third National Health and Nutrition examination survey

showed a strong association between vitamin D levels and lung function as well as the risk for upper respiratory tract infections in the general population.^{34,35} Data from adult critical care populations is conflicting, in that some studies demonstrate a clear association between vitamin D deficiency and culture-positive infection³⁶, whereas others do not^{23,37}. A few studies have associated vitamin D deficiency with severity of disease in infants and children with acute lower respiratory infection.^{38,39} Muhe identified vitamin D deficiency as an important predisposing factor for pneumonia in children in developing countries.⁴⁰ We did not identify an association with the incidence of infection or the need for ventilation. Roth et al did not find an association between vitamin D deficiency and severity of presentation of bronchiolitis, as defined by the need for hospitalisation. While we would suggest caution in extrapolating from non-ICU studies, or from studies in adult populations it is important to note that in our cohort severe vitamin D deficiency (less than 30nmol/l) was rare, proven infection rates were relatively low, and the majority of our patients (80%) were ventilated on, or shortly after PICU admission. Thus, we cannot with confidence rule out a subtle association in our relatively limited study population.

The association between vitamin D status and inotrope score in the post-operative cardiac patients is a novel finding. Ventricular dysfunction and heart failure are common in children with rickets,^{41,42} and myocardial dysfunction, heart failure and sudden cardiac death have also been associated with vitamin D deficiency.^{9,12} It is now known that vitamin D has both genomic and non-genomic effects in the heart, and influences cardiac systolic and diastolic function through its interaction with cardiac receptors (VDR), and that disruption of VDR leads to cardiac hypertrophy through interaction with the renin-angiotensin system^{43,44}. Although we did not routinely assess cardiac function or other markers of systemic oxygen delivery in our post-operative cardiac patients, there was no difference in their PIM-2 and RACHS scores between the vitamin D deficient and sufficient patients. It is therefore possible that our findings may indicate an association with myocardial dysfunction and impairment of oxygen delivery, which could be systematically addressed in future studies.

Our study confirmed a relationship between ionized calcium and vitamin D levels. In the general PICU population, the lowest measured ionised calcium level was lower in children with hypovitaminosis D. This is in keeping with studies in adult ICU patients. While this relationship was not quite the same in post-operative cardiac patients, the total calcium replacement was higher in those with low levels. It is important to note however that the threshold for treating hypocalcemia in cardiac patients is much lower than in non-cardiac patients, (generally around

1.10-1.20 in our practice), so a true untreated ‘nadir’ of untreated ionised calcium would rarely be seen in the cardiac group. Thus, total calcium replacement would be a surrogate for this in the cardiac patients.

Limitations

An important limitation to this current study is that there is no current consensus on a specific (or different) definition for Vitamin D deficiency in infants and young children, and that our data (similar to many published studies in other patient groups) does not include measurement of parathyroid hormone. Moreover, we (and others) have measured blood levels of 25(OH)D₃ rather than the active form – 1,25 dihydroxyvitamin D₃, and it is possible that its overall activity may be different in the critically ill population. Indeed, given these uncertainties we have chosen to apply the term ‘hypovitaminosis D’ to our population rather than ‘vitamin D deficiency’. On the basis of the available evidence, several authors have stated that similar to adults, the 25(OH)D₃ concentration for infants and children should be ≥ 50 nmol/L, which is the basis for the definition in our current study. However, it should be noted that other investigators have suggested that the cut-off should be lowered to 37.5nmol/L.⁴⁵ An additional note of caution, relates to the reliability of the currently available vitamin D assays. While there is no absolute consensus on whether immunoassays (as used in our study) are superior or inferior to high-performance liquid chromatography techniques, there is known to be poor reproducibility between the two methods.⁴⁶ Another limitation is the relatively small sample size and lack of a control population – which limited our ability to conclude with confidence some important issues, including the association with infection or severity of lung disease. Moreover, for cardiac patients in particular, a pre-operative vitamin D level would have been helpful, to address any confounding impact of CPB and fluid shifts on post-operative vitamin D level. Moreover an objective measure of cardiac performance, alongside total duration of inotropic support would undoubtedly be of interest in drawing further conclusions regarding our observation of increased inotropic needs in the deficient group. Finally, we did not investigate serial levels of vitamin D, particularly in patients requiring prolonged ICU stay, nor did we assess in detail the parathyroid axis in our patients. However, given our current observations, future studies could address all of these questions.

CONCLUSION

Hypovitaminosis D is common in critically ill children in the PICU, particularly in infants and children with heart disease. Hypovitaminosis D was associated with hypocalcemia in the non-cardiac population, and increased need for calcium replacement in the cardiac population. In

contrast with studies in adults, we did not demonstrate an association between vitamin D status and survival or PICU length of stay, but did find a strong relationship with early post-operative inotropic needs in the cardiac population. Further evaluation of the paediatric population is warranted, particularly with respect to children with congenital heart disease.

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Legend to Figure 1

Graph showing Vitamin D levels by month. *Note that Australian 'winter' months are June, July and August, and the 'summer' is December, January February. The shortest winter days in Melbourne (June) have around 10 daylight hours, and longest summer days (December) have approximately 14 daylight hours.*

Table 1:**Baseline characteristics of the population**

Variable	Total cohort (<i>n</i> =316)	General medical patients (<i>n</i> =106)	Postoperative cardiac patients (<i>n</i> =210)
Age, months	16.5 [3.1-75.2]	32.3 [10.2-129.1]	7.1 [2.2-56.1]*
Weight, kg	9.5 [4.7-21.4]	13.0 [7.3-25.0]	7.0 [4.2-17.0]*
Gender			
male, n (%)	186 (58.9)	69 (65.1)	117 (55.7)
female, n (%)	130 (41.1)	37 (34.9)	93 (44.3)
Admission			
emergency, n (%)	103 (32.6)	83 (78.3)	20 (9.5)*
elective	213 (67.4)	23 (21.7)	190 (90.5)*
PIM2 probability of death, %	2.2 [1.5-4.0]	3.4 [1.1-5.5]	2.0 [1.5-2.8]
RACHS probability of death, %			5.6 [3.8-14.4]
ICU stay, hrs	46.8 [23.7-135.3]	66.5 [30.5-172.9]	42.5 [23.2-93.6]*
Died, n (%)	10 (3.2)	6 (5.7)	4 (1.9)
Hospital stay, hrs	245.1 [143.7-513.0]	356.8 [174.8-862.8]	212.5 [129.0-383.3]*
Intubation, n (%)	276 (87.3)	71 (67.0)	205 (97.6)*
Intubation, hrs	18.6 [7.9-69.4]	15.6 [0-84.3]	19.2 [10.9-56.5]*
Respiratory support, n (%)	291 (92.1)	85 (80.2)	206 (98.1)*
Respiratory support, hrs	21.6 [10.5-82.1]	32.1 [4.6-116.6]	19.9 [11.3-59.8]*
Inotropic/vasopressor support, n (%)	202 (63.9)	38 (35.8)	164 (78.1)*
25(OH)D, nmol/l	56.5 [44.0-70.0]	65.5 [52.0-84.3]	52.0 [41.3-64.0]*
normal (≥ 50 nmol/l), n (%)	207 (65.5)	82 (77.4)	125 (59.5)*
deficient (< 50 nmol/l), n (%)	109 (34.5)	24 (22.6)	85 (40.5)*
Serum Calcium ^a , mmol/l	2.43 [2.23-2.70]	2.30 [2.10-2.45]	2.52 [2.31-2.88]*
Lowest ionised Calcium ^b , mmol/l	1.17 [1.12-1.22]	1.17 [1.10-1.22]	1.17 [1.13-1.21]
Serum Phosphate ^a , mmol/l	1.77 [1.49-1.96]	1.49 [1.25-1.86]	1.83 [1.61-1.99]*

Values of variables are expressed as number (percentage) or median [interquartile range]

^a on admission to the Intensive Care Unit

^b during the first 24 hours in the Intensive Care Unit

* $p < 0.05$ [Medical vs Cardiac patients]

PIM2 – Paediatric Index of Mortality Score 2

RACHS – Risk Adjustment for Congenital Heart Surgery

Table 2:**Characteristics and p-values for general medical intensive care patients**

Variable	25(OH)D ₃ < 50 nmol/l (n=24)	25(OH)D ₃ ≥ 50 nmol/l (n=82)	p-value
Age, months	29.1 [11.5-73.7]	24.1 [16.3-35.5]	0.70
Weight, kg	16.3 [11.5-23.1]	13.5 [8.2-16.3]	0.32
Male, n (%)	16 (66.7)	53 (64.6)	0.85
PIM2 probability of death, %	2.3 [1.4-3.8]	3.2 [2.4-4.2]	0.27
ICU stay, hr	58.0 [35.9-94.6]	75.9 [58.6-98.5]	0.34
Died, n (%)	1 (4.2)	5 (6.1)	0.72
Hospital stay, hr	428.4 [265.1-699.2]	387.6 [295.9-507.8]	0.71
Intubation, n (%)	13 (54.2)	56 (68.3)	0.42
Intubation, hr	36.6 [19.9-67.4]	46.1 [29.4-72.2]	0.54
Respiratory support, n (%)	18 (75.0)	67 (82.7)	0.47
Respiratory support, hr	52.5 [30.6-89.1]	45.1 [29.1-69.4]	0.65
Inotropic/vasopressor support, n (%)	10 (41.7)	28 (34.2)	0.50
Serum calcium ^a , mmol/l	2.16 [1.96-2.37]	2.31 [2.24-2.38]	0.17
Lowest ionised calcium ^b , mmol/l	1.07 [0.99-1.14]	1.17 [1.14-1.19]	0.02*
Serum Phosphate ^a , mmol/l	1.61 [1.41-1.81]	1.51 [1.41-1.61]	0.38
Lactate ^b >1.8 mmol/l	14 (58.3)	39 (47.6)	0.35
Glucose ^b < 3 mmol/l	1 (4.2)	5 (6.1)	0.72
Glucose ^b > 10 mmol/l	8 (33.3)	18 (22.0)	0.25
Infection, n (%)	5 (20.8)	16 (19.5)	0.89
Hypotension, n (%)	10 (41.7)	28 (34.2)	0.50
Hypertension, n (%)	4 (16.7)	11 (13.4)	0.69
Arrhythmia, n (%)	2 (8.3)	5 (6.1)	0.70
Cardiac arrest, n (%)	2 (8.3)	5 (6.1)	0.70

Values of variables are expressed as number (percentage) or geometric mean [95% confidence interval]

^a on admission to the ICU

^b during the first 24 hours in the ICU

* p < 0.05

PIM2 – Paediatric Index of Mortality 2

RACHS – Risk Adjustment for Congenital Heart Surgery

Table 3: Characteristics and p-values for postoperative cardiac surgical patients

Variable	25(OH)D ₃ < 50 nmol/l (n=85)	25(OH)D ₃ ≥ 50 nmol/l (n=125)	p-value
Age, months	9.2 [5.5-15.5]	8.7 [5.9-12.7]	0.84
Weight, kg	10.2 [8.2-12.7]	8.2 [7.0-9.5]	0.10
Male, n (%)	43 (50.6)	74 (59.2)	0.22
PIM2 probability of death, %	2.3 [2.0-2.7]	2.4 [2.1-2.8]	0.60
RACHS probability of death, %	6.3 [4.6-8.5]	7.0 [5.4-9.1]	0.55
ICU stay, hr	50.4 [40.4-62.2]	50.9 [42.1-61.6]	0.91
Died, n (%)	2 (2.4)	2 (1.6)	0.70
Hospital stay, hr	232.8 [200.3-273.1]	257.2 [221.4-298.9]	0.37
Intubation, n (%)	83 (97.7)	122 (97.6)	0.98
Duration of intubation, hr	24.5 [18.9-31.5]	26.3 [21.1-32.8]	0.67
Respiratory support, n (%)	83 (97.7)	123 (98.4)	0.70
Duration of respiratory support, hr	25.8 [19.7-33.4]	29.4 [23.3-37.3]	0.44
Inotrope/vasopressor support, n (%)	65 (76.5)	99 (79.2)	0.64
Inotrope/vasopressor support, hr	35.9 [26.3-49.4]	31.8 [23.1-43.4]	0.56
Inotrope score	2.5 [1.9-3.3]	1.4 [1.1-1.9]	0.005*
Serum Calcium ^a , mmol/L	2.52 [2.43-2.62]	2.60 [2.52-2.69]	0.20
lowest ionised Calcium ^b , mmol/L	1.16 [1.14-1.17]	1.17 [1.15-1.18]	0.38
Serum Phosphate ^a , mmol/L	1.85 [1.77-1.94]	1.81 [1.76-1.86]	0.38
Calcium supplementation ^b , n (%)	40 (47.1)	40 (32.0)	0.027*
Lactate ^b >1.8 mmol/l	51 (60.0)	76 (60.8)	0.91
Glucose ^b < 3 mmol/l	4 (4.7)	9 (7.2)	0.46
Glucose ^b > 10 mmol/l	18 (21.2)	31 (24.8)	0.54
ECMO, n (%)	4 (4.7)	5 (4.0)	0.63
Infection, n (%)	11 (12.9)	11 (8.8)	0.34
Hypotension, n (%)	21 (24.7)	38 (30.4)	0.37
Hypertension, n (%)	8 (9.4)	17 (13.6)	0.36
Arrhythmia, n (%)	13 (15.3)	13 (10.4)	0.29
Cardiac arrest, n (%)	5 (5.9)	6 (4.8)	0.73

Values of variables are expressed as number (percentage) or geometric mean [95% confidence interval]

^a on admission to the ICU

^b during the first 24 hours in the ICU

* p < 0.05

Fig. 1