

Correlation of Serum Calcium with Severity and Outcomes in Patients of COVID-19 Pneumonia

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ABSTRACT

Background: Calcium is an essential electrolyte with critical physiological functions. Recently, it has been implicated in the pathogenesis and outcomes of COVID-19. This retrospective study was conducted to estimate serum ionic calcium and its correlation with clinical severity, inflammatory markers, and in-hospital outcomes in moderate to severe COVID-19 patients. **Methods:** We retrospectively analyzed data from 377 COVID-19 patients, aged between 23 and 79 years, with a mean age of 54.17±11.53 years. Severity of the disease was determined using ICMR criteria. Parameters including age, gender, inflammatory markers, calcium levels, and clinical outcomes were assessed. **Results:** The study showed a prevalence of moderate and severe COVID-19 in 58.1% and 41.9% patients, respectively. Severity was significantly associated with younger age, higher mean inflammatory markers, notably IL-6, procalcitonin, D-Dimer, and lower ionic and total calcium levels, as well as vitamin D levels. Mortality and referral rate were significantly higher in the severe group. Hypocalcemia was prevalent in 39% of the patients and was significantly associated with disease severity, ARDS, and mortality. On multivariate assessment, only age and ionic calcium were significantly associated with COVID-19 severity. **Conclusion:** Lower serum ionic calcium levels are associated with increased severity and poor outcomes, including higher mortality in COVID-19 patients, underscoring the potential role of calcium as a diagnostic and prognostic marker in COVID-19 pneumonia and may be an important factor in various other forms of pneumonia.

Keywords: COVID-19, inflammatory markers, mortality, pneumonia, serum ionic calcium, severity

Introduction

The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the causative agent of the COVID-19 pandemic, has burdened global health infrastructures since its emergence in late 2019.^[1] Initially recognized for its respiratory complications, the multi-systemic manifestations of the disease, involving cardiovascular, renal and endocrine systems, have become increasingly evident.^[2,3] Among these, alterations in

serum electrolytes, including calcium, have been observed in patients with COVID-19, yielding potentially significant implications for disease severity and outcomes.^[4]

Serum calcium is integral to various physiological processes, including coagulation, muscular contraction and cell signalling.^[5] Hypocalcemia, a common abnormality in hospitalized patients, has been identified as a potential prognostic indicator in several critical illnesses, including sepsis and acute respiratory distress syndrome (ARDS).^[6,7] Interestingly, recent studies have reported a high prevalence of hypocalcemia among COVID-19 patients.^[4] However, the clinical significance of this association is not yet fully understood.

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A growing body of evidence suggests that the severity of COVID-19, including the development of pneumonia and ARDS, might be influenced by serum calcium levels.^[8] Furthermore, emerging data has pointed to a potential correlation between hypocalcemia and poorer outcomes, including higher mortality rates in COVID-19 pneumonia patients.^[5,9] Given the persistent knowledge gaps surrounding this area of study, it is crucial to further investigate the relationship between serum calcium and the severity and outcomes of COVID-19 pneumonia. This paper aims to deepen our understanding of this correlation, providing valuable insights to inform future therapeutic and prognostic approaches.

Aim

To evaluate correlation of serum calcium with severity and outcome of COVID-19 pneumonia.

Objectives

1. To estimate the serum ionic calcium at the time of admission, and to correlate it with clinical severity and inflammatory markers—serum ferritin, CRP, IL6 and D-dimer.
2. To correlate the levels of serum calcium with following in-hospital outcomes:
 - Acute respiratory distress syndrome (ARDS)
 - Sepsis
 - Outcome of hospitalization (discharge/death/referral) on 28th day.

Materials and Methods

Study Design and Setting

This research was a retrospective observational study conducted over 24 months at the Department of Medicine of a tertiary care centre catering primarily to socio-economically underprivileged suburban and rural populations in North India

Ethical Approvals

The study protocol was approved by the Institutional Ethical Committee.

Sampling Frame and Sample Size

The target population was patients with COVID-19 pneumonia attending a tertiary care designated hospital. The study included RT-PCR diagnosed cases of moderate and severe COVID-19 pneumonia while mild cases were excluded. The Department of Social and Preventive Medicine of the institution suggested a sample size of 377 on the basis of proportion of ARDS in the groups of low serum calcium level and normal serum calcium level using the study by Jia-Kui Sun *et al.*^[10] as reference by the formula,

$$n = \frac{(Z_{\alpha} + Z_{\beta})^2 \left[\frac{1-p_1}{p_1} + \frac{1-p_2}{p_2} \right]}{[In(1-\epsilon)]^2}$$

where $p_1 = 0.349$ (34.9%) proportion of ARDS in low serum calcium level, $p_2 = 0.016$ (1.6%) proportion of ARDS in high serum calcium level with Type I error, $\alpha = 5\%$, Type II error $\beta = 10\%$ for setting power of study 90% and a data loss factor = 10%.

Severity criteria

Patients were categorized into two groups based on disease severity: moderate and severe. The severity of COVID-19 was determined using the Indian Council of Medical Research (ICMR) criteria, as follows^[11]:

- Mild: Patients presenting upper respiratory tract infection symptoms without evidence of breathlessness or hypoxia.
- Moderate: Patients with pneumonia, showing no signs of severe disease, but exhibiting clinical features of dyspnea and/or hypoxia, fever, cough, including SpO₂ <94% (range 90–94%) on room air, and respiratory rate more or equal to 24 per minute.
- Severe: Patients with severe pneumonia, showing clinical signs of pneumonia plus one of the following: respiratory rate >30 breaths/min, severe respiratory distress, SpO₂ <90% on room air.

Data collection and analysis

Ionized calcium has an added value in specific situation, especially in critically ill patients, and is physiologically relevant component of blood calcium.^[12-14]

Upon admission, patients were classified according to their serum ionic calcium levels measured by arterial blood gas (ABG) findings achieved using ion selective electrode direct potentiometry ABG analyser which is quicker, easier, reliable and momentous indicator for critically ill patients as related by a study done by *Amrita Jyot et al.*^[15] The classification included:

- Category A: Serum ionic calcium <1.12 mg/dl (Hypocalcemia)
- Category B: Serum ionic calcium 1.12–1.30 mg/dl
- Category C: Serum ionic calcium >1.30 mg/dl (Hypercalcemia)

Subsequently, the effect of serum ionic calcium on COVID-19 severity and clinical outcomes was assessed. The Sequential Organ Failure Assessment (qSOFA) score, a quick tool to assess organ dysfunction and predict outcome in patients with suspected infection, was used as an additional outcome measure. The parameters of the qSOFA score included:

- Altered mental status
- Respiratory rate >22 per minute
- Systolic blood pressure <100 mm Hg.

A score ≥ 2 suggested poor outcome.

Demographic data, detailed investigation results and other relevant patient information were recorded on individual case sheets. This information was analysed using Statistical Package for Social Sciences (SPSS) Version 21.0. Categorical data was analysed using the Chi-square test, while the Student's 't'-test/

ANOVA was used to compare mean values between two or more groups. Statistical significance was considered at a 'p' value <0.05.

Results

The present study was conducted to estimate serum ionic calcium at the time of admission and to correlate it with clinical severity and inflammatory markers—serum ferritin, CRP, IL-6 and D-dimer—and with in-hospital outcomes. evaluate correlation of serum calcium with severity and outcomes (ARDS, Sepsis, Death) of COVID-19 pneumonia. The data of 377 moderate-severe COVID-19 patients fulfilling the inclusion criteria was analysed in this study.

Severity of COVID-19 patients was defined as per directions of ICMR. Out of all COVID-19 patients enrolled in the study, only moderate-severe COVID-19 hospitalized patients were included in the study. Out of 377 patients enrolled in the study, 219 (58.1%) had moderate COVID infection and rest 158 (41.9%) had severe COVID-19 infection [Table 1].

Following Table 2 shows demographic profile of moderate and severe COVID-19 patients.

Age of patients enrolled in the study ranged between 23 and 79 years, mean age of overall patients was 54.17 ± 11.53 years. Moderate COVID-19 patients were significantly older (56.13 ± 10.99 years) as compared to severe (51.46 ± 11.75).

Male dominance was observed in overall (68.2%) as well as moderate (69.9%) and severe (65.8%) COVID-18 patients. This difference was not found to be significant statistically.

Severe COVID-19 cases as compared to moderate had significantly higher IL-6 (50.18 ± 28.82 vs. 40.91 ± 27.36 pg/ml), procalcitonin (1.28 ± 1.22 vs. 0.84 ± 0.86 ng/ml) and D-dimer (1.25 ± 0.59 vs. 1.10 ± 0.61).

Severe cases had higher S. ferritin (494.25 ± 299.24 vs. 461.15 ± 289.27 ng/ml) and CRP levels (63.07 ± 44.78 vs. 62.53 ± 52.79 mg/L) as compared to moderate cases but differences were not found to be significant statistically [Table 3].

Severe COVID-19 patients as compared to moderate cases had significantly lower ionic calcium (1.08 ± 0.12 vs. 1.18 ± 0.11 mg/dl), total calcium (8.00 ± 0.60 vs. 8.18 ± 0.68 mg/dl) and vitamin D levels (30.55 ± 6.63 vs. 33.16 ± 6.87 ng/ml) [Table 4].

Serum potassium and sodium sodium levels of moderate and severe COVID-19 patients were comparable.

Hemoglobin, TLC and hematocrit levels of moderate and severe COVID-19 cases were comparable.

Platelet count of severe COVID-19 cases was found to be significantly lower as compared to moderate cases (1.74 ± 0.90 vs. 1.97 ± 0.77 lakhs/cumm) [Table 5].

Table 1: Distribution of cases according to COVID-19 severity (n=377)

Severity	No. of cases	Percentage
Moderate	219	58.1
Severe	158	41.9

Table 2: Association of COVID-19 severity with demographic profile of the patients

Variable	Total (n=377)		Moderate (n=219)		Severe (n=158)		-	-
	Mean	SD	Mean	SD	Mean	SD		
Age (years)	54.17	11.53	56.13	10.99	51.46	11.75	-	-
(Min-Max)	(23-79)		(25-79)		(23-73)			
	No.	%	No.	%	No.	%	c ²	'P'
Gender								
Male	257	68.2	153	69.9	104	65.8	0.690	0.406
Female	120	31.8	66	30.1	54	34.2		
M:F Ratio	2.1		2.32		1.93			

Table 3: Association of COVID-19 severity with inflammatory markers and specific investigations

Variable	Moderate (n=219)		Severe (n=158)		Statistical significance	
	Mean	SD	Mean	SD	't'	'P'
S. Ferritin (ng/ml)	461.15	289.27	494.25	299.24	-1.080	0.281
CRP (mg/L)	62.53	52.79	63.07	44.78	-0.105	0.917
IL-6 (pg/ml)	40.91	27.36	50.18	28.82	-3.176	0.002
Procalcitonin (ng/ml)	0.84	0.86	1.28	1.12	-4.295	<0.001
D-dimer	1.10	0.61	1.25	0.59	-2.374	0.018

Table 4: Association of COVID-19 severity with serum calcium, vitamin D and other electrolytes

Variable	Moderate (n=219)		Severe (n=158)		Statistical significance	
	Mean	SD	Mean	SD	't'	'P'
Ionic calcium (mmol/L)	1.18	0.11	1.08	0.12	8.457	<0.001
Total calcium (mmol/L)	8.18	0.68	8.00	0.60	2.675	0.008
S. potassium (mmol/L)	4.23	0.68	4.38	0.87	-1.928	0.055
S. sodium (mEq/L)	139.63	5.39	139.66	7.28	-0.043	0.966
Vitamin D (ng/ml)	33.16	6.87	30.55	6.63	3.697	<0.001

Table 5: Association of COVID-19 severity with hematological parameters

Variable	Moderate (n=219)		Severe (n=158)		Statistical significance	
	Mean	SD	Mean	SD	't'	'P'
Hb (g/dl)	12.21	2.32	12.34	2.23	-0.524	0.601
TLC (thousands/cumm)	11.31	6.11	11.20	5.70	0.178	0.859
Hematocrit (%)	35.75	6.45	36.63	7.51	-1.213	0.226
Platelet count (L/cumm)	1.97	0.77	1.74	0.90	2.682	0.008

Liver and renal functions of moderate and severe COVID-19 cases were comparable [Table 6].

Out of 377 patients enrolled in the study, 235 (62.3%) were discharged after treatment, 12 (3.2%) were referred for further treatment to specialized centres. Mortality rate in overall population was 34.5%. Referral and death rate were higher among severe COVID-19 cases as compared to moderate (7.0% vs. 0.5%) and (35.8% vs. 33.8%) [Table 7] while mortality rate was higher among severe cases as compared to moderate (35.8% vs. 33.8%). Final outcome of only 365 patients was known.

Mortality rate of severe COVID-19 cases was higher as compared to moderate (38.1% vs. 33.9%) but this difference was significant statistically [Table 8].

Multivariate analysis was done where severe COVID was considered to dependent on independent variables age, procalcitonin, D-dimer, IL-6, ionic calcium, total calcium, vitamin D and platelet count [Table 9]. Only higher age and ionic calcium levels had independent association with COVID-19 severity.

Association of mortality was significantly associated with Raised IL-6, low ionic calcium and S. calcium and vitamin D levels [Table 10]. Following table shows distribution of patients according to S. ionic calcium levels.

Majority of the patients had S. ionic calcium levels 1.12–1.30 mmol/L (53.1%); these patients were categorized as Category 2, 39.0% patients having S. ionic calcium <1.12 were categorized as Category 1 and only 30 (8.0%) patients having S. ionic calcium >1.30 mmol/L were categorized as Category 3 [Table 11].

In S. ionic calcium category 1 having lowest S. ionic calcium levels proportion of severe COVID-19 patients was higher as compared to moderate (57.1% vs. 42.9%), while proportion of moderate cases was higher Category 2 (63.5% vs. 36.5%) and Category 3 (96.7% vs. 3.3%) having highest S. ionic levels. This difference was significant statistically [Table 12].

All the above adverse outcomes were significantly higher in Category 1 as compared to Category 2 and Category 3. Sepsis (27.9% vs. 22.5% and 6.7%), ARDS (76.9% vs. 34.5% and 20.0%), qSOFA ≥2 (26.5% vs. 13.0% and 3.3%) and mortality (54.2% vs. 25.8% and 10.3%) [Table 13].

S. ionic calcium levels of sepsis cases was higher than those who did not develop sepsis but this difference was not found to be significant statistically significant. S. ionic calcium level of ARDS cases was significantly lower than No-ARDS cases. qSOFA ≥2 did not show any significant association with S. ionic calcium levels. Discharged cases had significantly higher S. ionic calcium as compared to those who expired [Table 14].

All the above inflammatory markers were maximum in Category 1 (Calcium <1.12 mg/dl) followed by Category 2 (Calcium 1.12–1.30 mg/dl) and minimum in Category

Table 6: Association of COVID-19 severity with liver and renal function tests

Variable	Moderate (n=219)		Severe (n=158)		Statistical significance	
	Mean	SD	Mean	SD	't'	'P'
S Bilirubin (mg/dl)	0.73	0.55	0.71	0.44	0.333	0.740
SGPT (U/L)	74.32	80.03	81.50	92.89	-0.803	0.423
SGOT (U/L)	72.12	51.06	81.72	88.95	-1.322	0.187
SALP (IU/L)	103.62	61.14	101.04	52.95	0.427	0.375
S. Creatinine (mg/dl)	1.56	1.29	1.39	0.96	1.418	0.157
Blood urea (mmol/L)	58.60	45.04	63.20	57.49	-0.870	0.385

Table 7: Association of COVID-19 severity with 28 day outcome

Outcome	Total (n=377)	Moderate (n=219)		Severe (n=158)		Statistical significance	
		No.	%	No.	%	c ²	'P'
Discharged	235	144	65.8	91	57.6	13.256	0.001
Referred	12	1	0.5	11	7.0		
Death	130	74	33.8	56	35.8		

Table 8: Association of COVID-19 severity with final 28th day outcome (death/discharge)

Outcome	Total (n=365)	Moderate (n=219)		Severe (n=158)		Statistical significance	
		No.	%	No.	%	c ²	'p'
Death	130	74	33.9	56	38.1	0.659	0.417
Discharge	235	144	66.1	91	61.9		

Table 9: Multivariate analysis (binary logistic) to assess independent role of serum calcium in prediction of severe COVID-19

Independent Variable	Unadj. b±SE	'P'	OR (95%CI)
Age	0.044±0.011	<0.001	1.045 (1.023-1.068)
Procalcitonin	-0.218±0.151	0.148	0.805 (0.599-1.081)
D-dimer	-0.043±0.202	0.830	0.958 (0.644-1.424)
IL-6	0.010±0.006	0.075	1.010 (0.999-1.021)
Ionic calcium	8.057±1.460	<0.001	3156.102 (180.585-55159.392)
Total calcium	-0.046±0.201	0.821	0.955 (0.644-1.418)
Vitamin D	-0.003±0.020	0.899	0.997 (0.959-1.037)
Platelet count	0.074±0.146	0.614	1.076 (0.809-1.433)
Constant	-11.035±2.145	<0.001	

3 (Calcium >1.30 mg/dl). Significant differences were found for all the inflammatory markers except CRP [Table 15].

Discussion

Our study conducted on 377 moderate-severe COVID-19 patients demonstrated a significant association between serum ionic calcium levels, disease severity, inflammatory markers and in-hospital outcomes. Male dominance was observed in both moderate (69.9%) and severe (65.8%) COVID-19 patients.

This finding aligns with previous studies suggesting higher susceptibility and severity of COVID-19 in males, possibly due to gender-based immune response differences.^[16]

The study revealed that severe COVID-19 patients had significantly higher IL-6 (50.18 ± 28.82 vs. 40.91 ± 27.36 pg/ml), procalcitonin (1.28 ± 1.22 vs. 0.84 ± 0.86 ng/ml) and D-dimer (1.25 ± 0.59 vs. 1.10 ± 0.61), suggesting a strong correlation between these inflammatory markers and disease severity. In accordance with our findings, previous studies have also reported elevated levels of IL-6, procalcitonin and D-dimer in severe COVID-19 cases, indicating a state of

hyperinflammation and hypercoagulability associated with disease severity.^[17,18]

Interestingly, our study found a significant association between lower serum ionic calcium levels and the severity of COVID-19. Similarly, Di Filippo *et al.*^[14] in their study have reported hypocalcemia in COVID-19 patients, especially in those with severe infection. This can be attributed to several factors including vitamin D deficiency, malnutrition and underlying critical illness causing dysregulation of calcium homeostasis.^[19]

The present study also found that hypocalcemia was significantly associated with higher mortality rates, ARDS and sepsis. This is consistent with earlier studies that report hypocalcemia as an independent risk factor for poor outcomes in hospitalized patients.^[20] Furthermore, our findings suggest that hypocalcemia may play a role in the development of ARDS and sepsis, underlining the importance of early monitoring and correction of serum calcium levels in COVID-19 patients.

However, our findings contrast with a study by Liu *et al.*,^[21] which found no significant difference in serum calcium levels between survivors and non-survivors of COVID-19. This discrepancy could be attributed to different study designs, patient characteristics and regional variations.

In conclusion, our study adds to the growing body of evidence linking serum ionic calcium levels with the severity of COVID-19 and its outcomes, suggesting that serum ionic calcium could be a useful prognostic marker. However, large-scale prospective studies are needed to confirm these findings and elucidate the underlying mechanisms.

Also, further evaluation of its correlation in other forms of pneumonia could be studied further to obtain the possibility of similar results.

Conclusion

This retrospective study analysed the records of 377 moderate-severe COVID-19 patients and investigated the correlation of serum ionic calcium with disease severity, inflammatory markers and hospital outcomes. Findings revealed a higher prevalence of moderate (58.1%) and severe (41.9%) COVID-19 cases. The severity of the

Table 10: Univariate analysis for predictors of mortality among the significant factors associated with COVID-19 severity

Variable/Parameter	Death (n=130)		Discharge (n=235)		Statistical significance	
	Mean	SD	Mean	SD	't'	'P'
Age (years)	54.75	10.92	53.89	11.74	0.694	0.488
Procalcitonin (ng/ml)	1.14	1.01	0.97	0.99	1.622	0.106
D-dimer	1.21	0.58	1.15	0.62	0.865	0.388
IL-6	53.63	29.52	40.59	26.41	4.329	<0.001
Ionic calcium (mmol/L) at adm.	1.10	0.12	1.16	0.12	-4.918	<0.001
S. calcium (mmol/L) at adm.	7.78	0.53	8.25	0.65	-7.110	<0.001
Vitamin D (ng/ml)	30.89	6.65	32.70	6.89	-2.436	0.015
Platelet count (lakhs/cumm)	1.74	0.73	1.91	0.87	-1.954	0.052

Table 11: Distribution of study population according to S. ionic calcium levels

Category	S. ionic calcium levels (mmol/L)	No. of patients	Percentage
Category 1 (Hypocalcemia)	<1.12	147	39.0
Category 2 (Normocalcemia)	1.12-1.30	200	53.1
Category 3 (Hypercalcemia)	>1.30	30	8.0

Table 12: Association of S. ionic calcium levels with severity of COVID-19

COVID-19 severity	Total	Category 1 (n=147)		Category 2 (n=200)		Category 3 (n=30)	
		No.	%	No.	%	No.	%
Moderate	219	63	42.9	127	63.5	29	96.7
Severe	158	84	57.1	73	36.5	1	3.3

χ²=34.753; P<0.001

Table 13: Association of S. ionic calcium levels with adverse clinical outcome

Clinical Outcome	Total	Category 1 (n=147)		Category 2 (n=200)		Category 3 (n=30)		Statistical significance	
		No.	%	No.	%	No.	%	χ ²	'P'
Sepsis	88	41	27.9	45	22.5	2	6.7	6.441	0.040
ARDS	188	113	76.9	69	34.5	6	20.0	72.474	<0.001
qSOFA ≥2	66	39	26.5	26	13.0	1	3.3	15.274	<0.001
Death	130	77	54.2	50	25.8	3	10.3	37.718	<0.001

Table 14: Association of S. ionic calcium with clinical outcomes

Outcome	No.	Mean S. calcium (mg/dl)	S.D.	Statistical significance (<i>t</i> test)
No sepsis	289	1.135	0.127	<i>t</i> =-1.123; <i>P</i> =0.265
Sepsis	88	1.152	0.115	
No ARDS	189	1.176	0.117	<i>t</i> =5.939; <i>P</i> <0.001
ARDS	188	1.103	0.122	
qSOFA<2	311	1.142	0.127	<i>t</i> =0.805; <i>P</i> =0.421
qSOFA≥2	66	1.128	0.114	
Discharge	235	1.161	0.125	<i>t</i> =-7.110; <i>P</i> <0.001
Death	130	1.096	0.116	

Table 15: Association of S. ionic calcium levels with inflammatory markers

	Category 1 (n=147)		Category 2 (n=200)		Category 3 (n=30)		ANOVA	
	Mean	SD	Mean	SD	Mean	SD	F	<i>P</i>
S. Ferritin (ng/ml)	531.78	309.23	454.67	281.54	332.61	227.47	6.973	0.001
CRP (mg/L)	66.28	39.32	61.74	55.83	52.29	49.52	1.084	0.339
IL-6 (pg/ml)	59.61	29.63	38.09	22.22	16.91	18.94	50.997	<0.001
Procalcitonin (ng/ml)	1.52	1.11	0.78	0.76	0.23	0.61	41.217	<0.001
D-dimer	1.27	0.56	1.17	0.62	0.59	0.30	17.132	<0.001

disease was significantly associated with younger age, higher inflammatory markers (including IL-6, procalcitonin and D-dimer), and lower ionic and total calcium, vitamin D levels and platelet count.

The mortality and referral rate were significantly higher in the severe COVID-19 group, with a total of 130 (34.5%) deaths and 12 (3.2%) referrals observed. However, no significant association was found between COVID-19 severity and factors such as sex, serum ferritin and CRP levels, serum potassium and sodium levels, hemoglobin, TLC, hematocrit, liver and renal function parameters.

Univariate analysis revealed a significant association of mortality with increased IL-6 and reduced ionic calcium, serum calcium and vitamin D levels. After adjusting for several factors in multivariate assessment, only age and ionic calcium were found to be significantly associated with COVID-19 severity.

Furthermore, hypocalcemia, observed in 39% of the patients, was significantly associated with disease severity, ARDS and mortality. The correlation of hypocalcemia with sepsis and a qSOFA score >2 was positive but insignificant. Hypocalcemia also showed a significant association with higher serum ferritin, IL-6, procalcitonin and D-dimer levels.

In conclusion, the study findings suggest that lower calcium levels are associated with increased COVID-19 severity and adverse outcomes, including mortality. Therefore, calcium levels could serve as a diagnostic and prognostic marker in COVID-19 pneumonia.

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Conflicts of interest

There are no conflicts of interest.

References

- Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, *et al.* Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet* 2020;395:497-506.
- Gupta A, Madhavan MV, Sehgal K, Nair N, Mahajan S, Sehrawat TS, *et al.* Extrapulmonary manifestations of COVID-19. *Nat Med* 2020;26:1017-32.
- Lippi G, South AM, Henry BM. Electrolyte imbalances in patients with severe coronavirus disease 2019 (COVID-19). *Ann Clin Biochem* 2020;57:262-5.
- Di Filippo L, Formenti AM, Rovere-Querini P, Carlucci M, Conte C, Ciceri F, *et al.* Hypocalcemia is highly prevalent and predicts hospitalization in patients with COVID-19. *Endocrine* 2020;68:475-8.
- Cappellini F, Brivio R, Casati M, Cavallero A, Contro E, Brambilla P. Low levels of total and ionized calcium in blood of COVID-19 patients. *Clin Chem Lab Med* 2020;58:e171-3.
- Akirov A, Gorshtein A, Shraga-Slutsky I, Shimon I. Clinical outcomes of patients with hypocalcemia in Intensive Care Units. *Clin Endocrinol* 2017;87:177-84.
- Parekh D, Dancer RCA, Lax S, Cooper MS, Martineau AR, Fraser WD, *et al.* Vitamin D to prevent acute lung injury following oesophagectomy (VINDALOO): Study protocol for a randomised placebo controlled trial. *Trials* 2013;14:100.
- Di Filippo L, Formenti AM, Doga M, Pedone E, Rovere-Querini P, Giustina A. Radiological thoracic vertebral fractures are highly prevalent in COVID-19 and predict disease outcomes. *J Clin Endocrinol Metab* 2021;106:e602-14.
- Liu J, Han P, Wu J, Gong J, Tian D. Prevalence and predictive value of hypocalcemia in severe COVID-19 patients. *J Infect Public Health* 2020;13:1224-8.
- Sun JK, Zhang WH, Zou L, Liu Y, Li JJ, Kan XH, *et al.* Serum calcium as a biomarker of clinical severity and prognosis in patients with coronavirus disease 2019. *Aging (Albany)*

- NY) 2020;12:11287-95.
11. Clinical Guidance for Management of Adult Covid-19 Patients(2023) AIIMS/ ICMR-COVID-19 National Task Force/ Joint Monitoring Group (Dte.GHS).
 12. Hamroun A, Pekar J, Lionet A, Ghulam A, Maboudou P, Mercier A, *et al.* Ionized calcium: Analytical challenges and clinical relevance. *J Lab Precis Med* 2020;5. doi: 10.21037/jlpm-20-60.
 13. Seeger C, Higgins C. Acute care testing handbook - radiometer america, (2014). Radiometer Medical ApS, 2700 Brønshøj, Denmark, 2014.
 14. Calvi LM, Bushinsky DA. When is it appropriate to order an ionized calcium?. *J Am Soc Nephrol* 2008;19:1257-60.
 15. Amrita J, Singh AP. Role of arterial blood gas (ABG) as a valuable assessment tool of disease severity in SARS-CoV-2 patients. *J Med Biochem* 2022;41:47-52.
 16. Gebhard C, Regitz-Zagrosek V, Neuhauser HK, Morgan R, Klein SL. Impact of sex and gender on COVID-19 outcomes in Europe. *Biol Sex Differ* 2020;11:29.
 17. Shah S, Shah K, Patel SB, Patel FS, Osman M, Velagapudi P, *et al.* Elevated D-Dimer Levels Are Associated With Increased Risk of Mortality in Coronavirus Disease 2019: A Systematic Review and Meta-Analysis. *Cardiol Rev* 2020;28:295-302. doi: 10.1097/CRD.0000000000000330. Epub 2020 Jul 2.
 18. Liu T, Zhang J, Yang Y, Ma H, Li Z, Zhang J, *et al.* The role of interleukin-6 in monitoring severe case of coronavirus disease 2019. *EMBO Mol Med* 2020;12:e12421.
 19. Napoli N, Elder G, Kiel DP, Khosla S. The casual role of vitamin D in the COVID-19 pandemic. *J Steroid Biochem Mol Biol* 2021;211:105883.
 20. Zhang Z, Xu X, Ni H, Deng H. Hypocalcemia and risk of adverse outcomes in patients with acute coronary syndrome. *Int J Cardiol* 2015;180:162-8.
 21. Liu J, Han P, Wu J, Gong J, Tian D. Low serum calcium and phosphorus and their clinical performance in detecting COVID-19 patients. *J Med Virol* 2021;93:1639-51.