

Assessment of Association of Hypocalcemia with Severity of Disease in Patients with Dengue Fever in a Tertiary Care Hospital

Jitendra Kodilkar¹, Sonal Dhamanskar², Manaswini dixit³, Sakshi Mishra⁴,
Jeetendra Singh⁵

¹Professor, Department of General Medicine, Dr. Vasant Rao Pawar Medical College, Hospital and Research Centre, Nashik-422003, India

²Junior Resident-3, Department of General Medicine, Dr. Vasant Rao Pawar Medical College, Hospital and Research Centre, Nashik-422003, India

³Final Year MBBS, Dr. Vasant Rao Pawar Medical College, Hospital and Research Centre, Nashik-422003, India

⁴Intern, Dr. Vasant Rao Pawar Medical College, Hospital and Research Centre, Nashik-422003, India

⁵Professor and Head, Department of Pharmacology, Dr. Vasant Rao Pawar Medical College, Hospital and Research Centre, Nashik-422003, India

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Corresponding author: Dr. Jeetendra Singh

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Abstract

Introduction: Dengue hemorrhagic fever (DHF) is a major cause of morbidity and mortality in tropical regions. Serum free calcium (Ca²⁺) is known to be important in cardiovascular function. We therefore evaluated association between serum Ca²⁺ level and severity of dengue.

Methodology: A cross-sectional study was carried out at a tertiary care private hospital in India. A probable case of dengue was diagnosed and classified according to World Health Organization criteria and confirmed by either IgM antibody, PCR, or NS1 antigen detection. Socio-demographic details were collected using an interviewer-administered questionnaire.

Results: The sample size was 130. The mean age was 32.1 years, and the majority were males (n = 78, 53.8%). DHF was diagnosed in 69 patients (53%). Mean serum Ca²⁺ level of the study population was 1.05 mmol/L (range 0.72–1.22). Mean serum Ca²⁺ was significantly higher in patients with dengue fever (DF) (1.04 mmol/L) than in those with DHF (1.01 mmol/L) (p < 0.05). Prevalence of hypocalcemia in DHF and DF patients was 86.9% (n = 60) and 29.7% (n = 11), respectively (p < 0.05).

Conclusions: Serum Ca²⁺ levels may significantly correlated with dengue severity. Serum Ca²⁺ levels were significantly lower and hypocalcemia was more prevalent in patients with DHF than in patients with DF. Further studies are required to determine whether hypocalcemia can be utilized as a prognostic indicator and to evaluate effectiveness of calcium therapy in prevention of dengue complications in the patients.

Keywords: Dengue, Dengue Hemorrhagic Fever, Serum Calcium.

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Introduction

Dengue is a disease spread by the *Aedes* mosquito, and it is an entity known to mankind since long. After 1960, the incidence of dengue has shown an exponential increase, with several recent outbreaks reported mainly from South Asian countries. Nearly 70% of the world's population at risk of dengue lives in the Southeast Asian and Western Pacific regions. Dengue infection and dengue haemorrhagic fever (DHF) are major causes of morbidity and mortality in the tropical regions of the world.

Dengue is a mosquito-borne viral disease that has rapidly spread in all regions of WHO in recent years. Dengue virus is transmitted by female mosquitoes mainly of the species *Aedes aegypti*. [1]

The incidence of dengue has grown dramatically around the world in recent decades. The actual numbers of dengue cases are underreported and many cases are misclassified. One recent estimate indicates 390 million dengue infections per year (95% credible interval 284–528 million), of which 96 million (67–136 million) manifest clinically (with any severity of disease). [2] Nearly 70% of the world's population at risk of dengue lives in the Southeast Asian and Western Pacific regions. Dengue infection and dengue haemorrhagic fever (DHF) are major causes of morbidity and mortality in the tropical regions of the world.

Due to this high prevalence and considerable mortality, over the last few years there has been a heightened interest in disease prevention and effective strategies for management. However, at present, the pathogenesis of dengue and its complications are not completely understood. Dengue virus (DENV) belongs to the family *Flaviviridae*, genus *Flavivirus*. It is a single-stranded

positive-sense ribonucleic acid virus with 10,700 bases. [3]

Rationale of the study: Alterations in calcium homeostasis might play a role in the pathogenesis of shock in patients with dengue infection. Researchers have postulated that autonomic dysfunction might also contribute to hypotension in dengue shock syndrome (DSS). Calcium entry via neuronal calcium channels is essential for neurotransmission; hence calcium plays an important role in the smooth functioning of the autonomic nervous system.

It has been reported that the mean total calcium levels were significantly lower in patients with DHF than in patients with uncomplicated dengue fever (DF). However, free calcium is a more useful index than total calcium and provides a better indication of calcium status.

Calcium is transported predominantly bound to serum albumin; the total calcium level, therefore, is influenced directly by the serum albumin concentration. Numerous studies have clearly demonstrated that the measurement of free calcium is the test of choice in nearly all diagnostic and treatment situations. In the present study we evaluated the association between serum free calcium level and disease severity in patients with dengue infection.

Currently, the most accepted theory is that of an abnormal or amplified immunological response occurring in a secondary infection with a different serotype than in the primary infection. [4] This result in an antibody-dependent enhancement of immunological reaction, resulting in endothelial injury, plasma leakage, reduced intravascular volume, and circulatory collapse. [5] Although no specific pathway has been identified linking known immunopathogenic events with definitive effects on

microvascular permeability, thermoregulatory mechanisms, or both, preliminary data suggest that transient disruption in the function of the endothelial glycocalyx layer occurs, which probably enhances leakage.[6] Serum calcium is known to be important in cardiac and circulatory functions. The administration of intravenous calcium has been a routine practice in resuscitation protocols for traumatic, hemorrhagic and carcinogenic shock a particle supported by the presence of hypocalcaemia and the observed beneficial effects of calcium therapy in these conditions.[7] Known cardiovascular manifestations of hypocalcaemia include hypotension, reduced myocardial function, electrocardiogram (ECG) abnormalities, and heart failure.[8] Alterations in calcium homeostasis, therefore, might play a role in the pathogenesis of shock in patients with dengue infection. Researchers have postulated that autonomic dysfunction might also contribute to hypotension in dengue shock syndrome (DSS).[9]

Aim and Objectives

Aim: The study aims to analyse whether hypocalcaemia is associated with Disease Severity in patients with Dengue.

Objectives: To identify association of hypocalcaemia with disease Severity in patients with dengue fever.

Inclusion:

- Confirmed positive cases of dengue by IgM antibody, PCR, or NS1 antigen detection
- Age ≥ 18 years
- Both sex

Exclusion:

- Pre-existing renal or hepatic dysfunction
- Known endocrine disease-causing hypocalcaemia
- unwilling to give informed consent

Methods

A cross-sectional study was carried out at a tertiary care private hospital between the time periods of June 2021 to June 2022. Cases of dengue were diagnosed and classified according to World Health Organization criteria and confirmed by IgM antibody, PCR, or NS1 antigen detection. Socio-demographic details were collected using an interviewer-administered questionnaire.

A consecutive sample of inpatients with confirmed dengue infection was recruited for the study after written informed consent was obtained. The admission register at the hospital was used as the sampling frame. Patients with hypertension, diabetes and cardiac diseases and those on anti-hypertensive/anti-arrhythmic medications, calcium supplements or any other drugs affecting calcium homeostasis were excluded, as these would have altered the blood pressure, serum calcium levels, and ECG findings.

Data Analysis:

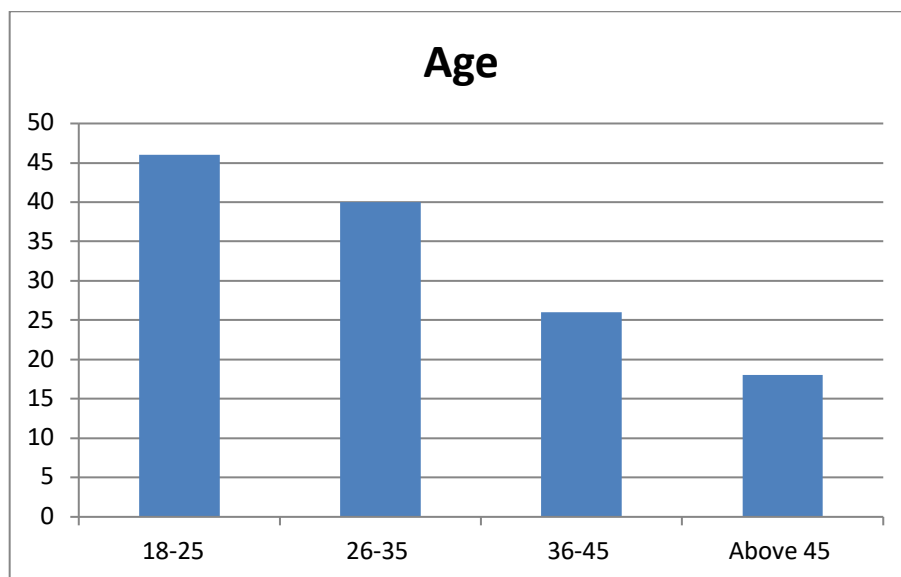
Data were analysed using SPSS version 14 (SPSS Inc., Chicago, IL, USA) statistical software package. The significance of the differences between proportions (%) and means were tested using the z-test and student's t-test or ANOVA, respectively.

Result

The sample size was 130. The mean age was 32.1 years, and the majority were males (n = 78, 53.8%). DHF was diagnosed in 69 patients (53.0%). Mean serum Ca²⁺ level of the study population was 1.05 mmol/L (range 0.72-1.22). Mean serum Ca²⁺ was significantly higher in patients with dengue fever (DF) (1.04 mmol/L) than in those with DHF (1.01 mmol/L) (p < 0.05). Prevalence of hypocalcemia in DHF and DF patients was 86.9% (n = 60) and 29.7% (n = 11), respectively (p < 0.05).

Table 1: Demographic Distribution

Age (Years)	Frequency	Percentage
18-25	46	35.3%
26-35	40	30.7%
36-45	26	20%
Above 45	18	13.8%
Total	130	100%

**Figure 1: Demographic Distribution****Discussion**

Dengue hemorrhagic fever (DHF) is a major cause of morbidity and mortality. Serum free calcium (Ca^{2+}) is known to be important in cardiac and circulatory function. We evaluated association between serum Ca^{2+} level and severity of dengue. According to the outcome of the current study, mean serum Ca^{2+} was significantly higher in patients with dengue fever (DF) (1.04 mmol/L) than in those with DHF (1.01 mmol/L) ($p < 0.05$). A significant difference was observed between mean serum calcium levels of DHF I and DHF II.

Prevalence of hypocalcemia in DHF and DF patients was 86.9% ($n = 60$) and 29.7% ($n = 11$), respectively ($p < 0.05$). However, there are currently no much randomized control trials evaluating the effectiveness of calcium

therapy in the prevention of complications in dengue infection. Hence, oral or IV calcium therapy is not routinely included in published guidelines.

Furthermore, hypocalcemia has also been demonstrated in certain cases of fever as malaria, severe meningococcal infections, and other severe acute illnesses, being associated with a poor prognosis. A similar study conducted by Kumar, Saini and Rani (2022) also has results in line with our study.

Furthermore, the exact mechanism for hypocalcemia in severe dengue infections also requires further study. Possible mechanisms include leak into the potential third spaces, disturbance in cellular transport, or changes in hormones involved in calcium metabolism [10].

Therefore, the association between hypocalcemia and the severity of dengue needs to be further evaluated. The measurement of serum calcium is not a routine practice in patients with dengue infection. Further studies are required to determine whether the presence of hypocalcemia at the onset of the illness can be utilized as a prognostic indicator to predict disease severity.

Conclusion:

Serum Ca²⁺ levels significantly correlated with dengue severity in our study. Serum Ca²⁺ levels were significantly lower and hypocalcemia was more prevalent in patients with Dengue haemorrhagic fever than in patients with Dengue fever. Further studies to be conducted to determine whether hypocalcemia can be utilized as a prognostic indicator and to evaluate effectiveness of calcium therapy in prevention of dengue complications.

References

1. <https://apps.who.int/mediacentre/factsheets/fs117/en/index.html> accessed on 20 Feb 2023
2. Bhatt S, Gething PW, Brady OJ, Messina JP, Farlow AW, Moyes CL et. al. The global distribution and burden of dengue. *Nature*; 496:504-507.
3. Murugesan A, Manoharan M. Dengue Virus. *Emerging and Reemerging Viral Pathogens*. 2020: 281–359.
4. Rothman AL, Green S, Vaughn DW. Dengue hemorrhagic fever. In: Saluzzo JF, Dodet B, editors. *Factors in the emergence of arbovirus diseases*. Paris: Elsevier. 1997; 109- 116.
5. Srikiatkhachorn A, Krautrachue A, Ratanaprakarn W, Wongtapradit L, Nithipanya N, Kalayanarooj S, Nisalak A, Thomas SJ, Gibbons RV, Mammen MP Jr, Libraty DH, Ennis FA, Rothman AL, Green S. Natural history of plasma leakage in dengue hemorrhagic fever: a serial ultrasonographic study. *Pediatr Infect Dis J*. 2007; 26: 283-290; discussion 91-92.
6. Simmons CP, Farrar JJ, Nguyen VC, Wills B. Dengue. *N Engl J Med*. 2012; 366: 1423-1432.
7. Cumming AD. The role of calcium in intravenous fluid therapy. *Arch Emerg Med*. 1993; 10: 265-270.
8. Goldstein DA. Serum Calcium. In: Walker HK, Hall WD, Hurst JW, editors. *Clinical Methods: The History, Physical, and Laboratory Examinations*, 3rd edition. Boston: Butterworth. 1990.; Chapter 143.
9. Vijayabala J, Attapaththu M, Jayawardena P, de Silva SG, Constantine G. Sympathetic dysfunction as a cause for hypotension in dengue shock syndrome. *Chin Med J*. 2012; 125: 3757-3758
10. Constantine et al. Hypocalcemia and disease severity in dengue *J Infect Dev Ctries*. 2014; 8(9):1205-1209.