

# When Is It Appropriate to Order an Ionized Calcium?

Laura M. Calvi and David A. Bushinsky

Department of Medicine, University of Rochester School of Medicine and Dentistry, Rochester, New York

## ABSTRACT

Convincing evidence demonstrates that ionized calcium and not total calcium is the physiologically relevant component of blood calcium. Direct measurement of ionized calcium, however, is limited by difficulties in accurate analysis, lack of standardization, and need for special handling, all resulting in increased cost; therefore, strategies have been developed to estimate ionized calcium from total calcium adjusted for levels of albumin, measurements that are more available and relatively inexpensive. This commentary compares the advantages and limitations of direct or calculated determinations of ionized calcium. Also examined are available data illustrating the settings in which measurement of ionized calcium is preferred and, in some cases, necessary for clinical decision-making.

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The physiologic importance of calcium is far-reaching, with fundamental and distinct but interdependent intracellular and extracellular activities. Intracellular calcium is a crucial regulator of numerous cellular events, including muscle contraction, signaling, hormone secretion, glycogen metabolism, and cell division.<sup>1</sup> Extracellular calcium not only provides a steady supply of calcium for intracellular use but also plays an important role in clotting and membrane integrity.<sup>1</sup> In mammals, nearly all body calcium resides within the mineral phases of bone, contributing to the mechanical properties of the skeleton as well as providing a reservoir for extracellular ions. Soluble extracellular calcium, including intravascular calcium, constitutes approximately 0.1% of the total body calcium content.<sup>1,2</sup>

As with other ionized constituents found in body fluids, the measurement of total blood calcium fails to reveal its varied chemical forms and the portion that is present as free ions, the so-called ionized calcium.<sup>3</sup> Initial dialysis experiments by Rona and Takahashi<sup>4</sup> demonstrated that total serum calcium

could be separated into diffusible and nondiffusible fractions. Of these, the protein-bound fraction represents 30 to 55%, diffusible ionic complexes (e.g., bicarbonate, citrate, sulfate, phosphate lactate)<sup>5</sup> comprise approximately 5 to 15%, and approximately 50% is freely ionized.<sup>6</sup> Most of the protein-bound calcium is complexed to albumin, with the remainder binding to globulins.<sup>5</sup> Experiments by Moore<sup>6</sup> and McLean and Hastings<sup>7,8</sup> confirmed that ionized calcium accounts for the biologically active form of serum calcium and subsequently demonstrated the crucial role of ionized calcium in the calcium homeostasis of healthy individuals and patients with parathyroid abnormalities.

Homeostatic mechanisms relying on parathyroid hormone (PTH) and vitamin D have evolved to defend the narrow physiologic range of extracellular and intravascular calcium.<sup>1</sup> Identification of the calcium-sensing receptor as the principal control mechanism for PTH secretion by the parathyroid glands in response to fluctuation of ionized calcium further supports the pivotal role of this

fraction of circulating calcium in calcium homeostasis.<sup>9</sup>

Because ionized calcium is the most important physiologic component of calcium and is controlled by stringent endocrine regulation, strategies either to measure it directly or to estimate it from measurements of total calcium have emerged. Both methods, however, have limitations that must be understood for appropriate interpretation of calcium levels in the clinical setting.

The initial method for direct measurement of ionized calcium was based on a bioassay with obvious limitations of applicability to clinical practice.<sup>7</sup> While recognizing the ideal importance of directly measuring ionized calcium, McLean and Hastings<sup>8</sup> developed an alternative nomogram to derive estimates of ionized calcium from total calcium and protein measurements. Deriving ionized calcium, however, is only an approximation based on several assumptions and is affected by numerous variables in addition to protein, including pH, magnesium, citrate, and albumin-to-globulin ratios.<sup>8</sup> Because 1g/dl albumin binds approximately 0.8 mg/dl calcium, ionized calcium is estimated typically from measurements of total calcium and albumin. For correction for hypoalbuminemia, 0.8 mg/dl (0.2

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**Correspondence:** Dr. Laura Calvi, Department of Medicine, University of Rochester School of Medicine and Dentistry, 601 Elmwood Avenue, Box 693, Rochester, NY 14642. Phone: 585-275-2901; Fax: 585-273-1288; E-mail: [laura\\_calvi@urmc.rochester.edu](mailto:laura_calvi@urmc.rochester.edu)

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mmol/L) must be added to the total calcium measurement for each 1-g/ml decrease in albumin concentration below the normal 4.0 g/dl.<sup>5</sup> The binding of calcium to albumin is also affected by extracellular fluid pH. Acidemia decreases calcium binding to protein, with consequent increases in ionized calcium as a fraction of total calcium. In patients with perturbations of extracellular fluid pH, each 0.1 decrease in pH increases ionized calcium by approximately 0.2 mg/dl (0.05 mmol/L).<sup>5</sup>

Precision in ionized calcium measurement was revolutionized after the introduction of ion-selective electrodes<sup>10</sup>; however, the clinical application of this technique was initially limited and delayed by its cost, susceptibility to errors, need to prevent CO<sub>2</sub> losses from the sample, and control of pH.<sup>6</sup> Advances in technology for direct measurement of ionized calcium have decreased the cost and improved its availability in the clinical setting since the 1980s.<sup>11</sup> A number of limitations remain, however, particularly in the outpatient setting, including the technical challenge of equipment maintenance, frequent electrode replacement with associated downtime, and redundancy of instrumentation and personnel, all leading to increased costs. In addition, measurement standardization is lacking.

The technical issues with direct measurement of ionized calcium relating to analytical performance, standardization, sample handling, and cost continue to plague its application to the outpatient setting.<sup>12</sup> Numerous studies, however, have identified specific clinical situations in which direct measurement of ionized calcium is clearly superior to its calculation from total calcium and albumin, even with corrections for pH. Specifically, reports suggest that ionized calcium is superior in identifying calcium disturbances in patients receiving transfusions with citrated blood; in critically ill patients; and in patients with the late stages of chronic kidney disease (CKD), hyperparathyroidism, and hypercalcemia of malignancy.<sup>11</sup>

In critically ill surgical patients, corrected total calcium measurements

poorly correlate with hypocalcemia.<sup>13–15</sup> In this clinical setting, hypoalbuminemia, acidemia, acute elevations of free fatty acid concentrations, and lipid infusions during parenteral nutrition may result in poor correlation of total calcium with direct measurements of ionized calcium.<sup>16–18</sup> Hypocalcemia is common in intensive care units, where corrected serum calcium levels fail to classify accurately as many as 40% of cases of hypocalcemia.<sup>19</sup> No factors could be identified to determine any subgroup of patients in which corrected total levels would accurately estimate ionized calcium.<sup>19</sup> It is interesting that despite abundant literature advising ionized calcium measurements in the critical care setting, surgical practitioners still rely heavily on corrected serum calcium levels.<sup>19</sup>

In the later stages of CKD, pH and albumin fluctuations may also alter relative calcium fractions unpredictably. Although the Kidney Disease Outcomes Quality Initiative (K/DOQI) guidelines recommend the use of albumin-corrected total calcium, published algorithms do not accurately predict ionized calcium.<sup>20–22</sup> Direct measurements of ionized calcium, which are rarely done in this patient population, are important for optimal clinical decision-making.<sup>11,20,23</sup> In particular, hypercalcemia may be overdiagnosed when total calcium and albumin measurements are used to estimate ionized calcium, leading to potentially inappropriate clinical choices regarding the use of vitamin D and its analogues, cinacalcet, or calcium-containing phosphate binders.<sup>20</sup> In patients with CKD, additional studies comparing the direct measurement of ionized calcium with that of estimated ionized calcium using published algorithms are clearly warranted. If we elect not to measure ionized calcium directly in this patient population, perhaps more accurate algorithms can be developed, similar to those now used to estimate glomerular filtration.

Citrate also binds calcium, lowering the ionized calcium and inhibiting blood coagulation.<sup>24</sup> Direct measurements of ionized calcium are routinely necessary in patients treated with continuous

venovenous hemofiltration, especially when citrate is used as the anticoagulant.<sup>25</sup> In this instance, ionized calcium must be measured not only in the systemic circulation but also in relation to the dialyzer to determine adequacy of anticoagulation and to detect citrate toxicity.<sup>26–28</sup> Because direct determinations of citrate are rarely performed, it is not possible to correct for the reduction of ionized calcium caused by the binding of calcium to citrate; in this setting, it is imperative that ionized calcium be measured directly.

Ionized calcium may also have greater diagnostic accuracy in hyperparathyroidism, hypercalcemia of malignancy, and neonatal hypocalcemia.<sup>11,29</sup> Although ionized calcium is more sensitive than albumin-corrected total calcium in the diagnosis of hypercalcemia of malignancy,<sup>30</sup> the clinical usefulness of this measurement is unclear. In at least one study,<sup>31</sup> slightly increased ionized calcium levels did not predict the development of frank hypercalcemia in patients with solid malignant tumors.

Even when symptomatic, total calcium in primary hyperparathyroidism may be normal or only intermittently elevated, and, not surprising, ionized calcium in this setting is superior to total calcium measurements.<sup>32–34</sup> Moreover, in a case series of 25 patients with surgically demonstrable parathyroid adenomas associated with hyperparathyroidism and normal total calcium, direct measurement of ionized calcium was more sensitive than estimation of ionized calcium based on total calcium.<sup>35</sup> Given the superiority of direct measurements of ionized calcium in identifying patients with primary hyperparathyroidism compared with estimates based on corrected total calcium, it is likely that estimates based on total calcium will be similarly inaccurate in identifying patients with CKD and secondary and tertiary hyperparathyroidism.

In a case series of 33 patients with hyperparathyroidism in the setting of multiple endocrine hyperplasia type 1 (MEN1), derivation of ionized calcium based on measurement of total calcium and albumin alone underestimated the

diagnosis compared with direct measurement of ionized calcium.<sup>36</sup> This false-negative result is particularly noteworthy because hypercalcemia is typically the presenting manifestation of MEN1 and is often used as a screen for asymptomatic adults in affected families.<sup>37</sup>

In conclusion, abundant evidence establishes the importance of ionized calcium in several pathologic conditions. Although its direct measurement remains costly and technically challenging, the algorithms to predict ionized calcium from total calcium have not proved accurate, especially in patients with complex illness. In the critical care setting, ionized calcium should be the routine measurement as well as where procedures such as continuous venovenous hemofiltration mandate the direct measurement of ionized calcium. In the outpatient setting, estimating ionized calcium from measurements of total calcium and albumin remain more feasible and less costly; however, direct measurement of ionized calcium is now suggested in several ambulatory conditions, including patients in the later stages of CKD as well as in patients with suspected hyperparathyroidism and MEN1. With time, the number of these conditions will almost certainly expand and measurements of ionized calcium, the physiologically active component of total calcium, will become the routine, preferred method for determining the level of calcium in all patients. This beneficial evolution in a clinical measurement should lead to demonstrable improvements in patient care.

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## DISCLOSURES

None.

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