Bicarbonate Testing

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Introduction

This scientific bulletin is intended to provide clinicians with an overview of the latest evidence-based thinking/recommendations regarding the monitoring of serum bicarbonate levels in patients with End-stage Renal Disease (ESRD). Concerns regarding the accuracy of serum bicarbonate testing results—as well as practical tips that can help to ensure accurate, reliable values—are discussed in detail.

The accurate measurement of serum bicarbonate values is central to the assessment/treatment of metabolic acidosis. A number of parameters can affect bicarbonate values. Awareness on the part of the clinician and staff regarding these issues can ensure that accurate results are obtained and that these results are correctly interpreted.

Metabolic Acidosis: A Common Complication of End-stage Renal Disease

Metabolic acidosis is an abnormally increased hydrogen ion concentration in the blood, and is often seen in patients with ESRD receiving maintenance dialysis therapy.[1-8] The degree of acidemia is commonly measured by serum bicarbonate levels. Low serum bicarbonate concentrations in a maintenance dialysis patient almost always indicate metabolic acidosis.[6]

Chronic acidosis can have severe consequences, and is increasingly considered to be a cause of morbidity and possibly even mortality among dialysis patients.[2-5,7,8] Deleterious effects associated with chronic acidosis include muscle wasting, bone disease, impaired growth, abnormal hormone secretion (growth and thyroid hormone), impaired insulin sensitivity, impaired vitamin D synthesis, hypertriglyceridemia, progression of renal failure, and increased beta-2 microglobulin accumulation.[3-8]

Normalization of a patient's pre-dialysis or stabilized serum bicarbonate concentration can be achieved by increasing the basic anion concentration in the dialysate and/or by administering oral bicarbonate.[6,7] Thus, regular monitoring and treatment, as delineated in the KDOQI guidelines is recommended.[2,3,6]

Concerns Surrounding Serum Bicarbonate Testing

There are several concerns in relation to serum bicarbonate testing, relating to blood collection techniques, shipping, laboratory practices and workflow, and apparent discrepancies between results obtained from separate laboratories.[6,8] Each of these concerns is addressed in detail below.

Blood Collection Techniques

Blood collection techniques can influence measured serum bicarbonate values.[6] Most notably, the presence of fibrin clots in the sample and/or inadequate sample volume can lead to falsely low reported results.

If a serum sample is received by the testing laboratory with fibrin clots, these clots must be removed prior to bicarbonate testing to avoid blocking the sample probe. This exposes the sample to air for a short period of time, which can result in CO_2 loss (see *"Effect of Time to Analysis,"* page 5) and may decrease reported values.

Clinicians and staff can help avoid fibrin clots by:

- Ensuring there is *not* excessive heparin in the sample
- Ensuring that the sample is not prematurely centrifuged (e.g., prior to complete clot formation)

A related concern is sample volume. If the Vacutainer® SST™ tube utilized for sample collection is received by the testing laboratory with an inadequate volume of serum, the sampling probe can be clogged by the SST™ gel; thus, these "short" samples must be aliquoted into sample cups. This aliquoting increases the exposure of the sample to air and can be associated with decreased reported bicarbonate values.

Table 1

National Kidney Foundation's Kidney Disease Outcomes Quality Initiative (KDOQI™): Serum Bicarbonate Testing Recommendations[6]

BACKGROUND	SELECTED SERUM BICARBONATE-RELATED RECOMMENDATIONS
The National Kidney Foundation's Kidney Disease Outcomes Quality Initiative (KDOQI) guidelines provide evidence-based clinical practice guidelines for chronic kidney disease and its related complications. These guidelines have been extensively adopted in the United States and abroad. Complete guidelines are available at **www.kidney.org/professionals/kdoqi/guidelines_updates/doqi_upex.html**	"Most trials report that normalizing the predialysis or stabilized serum bicarbonate concentrations is beneficial for protein, amino acid and bone metabolism, and protein-energy nutritional status. Thus, the serum bicarbonate should be monitored regularly at monthly intervals and correction of metabolic acidemia by maintaining serum bicarbonate at or above 22 mmol/L [mEq/L] should be a goal of the management of individuals undergoing maintenance dialysis."

Bottom Line

Proper sample collection techniques can ensure the absence of fibrin clots in the sample and adequate sample volume, thus mitigating concerns regarding exposure to air during clot removal/aliquoting of samples. A full tube, without heparin contamination, allowed to clot completely prior to centrifugation will assure accurate and consistent bicarbonate results.

Shipping Concerns

Most dialysis centers send patient samples (including samples utilized for bicarbonate testing) to central laboratories specializing in the unique handling, testing, and reporting requirements of the dialysis patient population. There has been some concern regarding the validity of bicarbonate testing results associated with shipped samples (specifically, those shipped by air), with published[9] and unpublished reports suggesting that shipping is associated with falsely decreased bicarbonate values. These studies, however, are affected by intra-laboratory instrument and workflow discrepancies.[8,10] In addition, data from two controlled studies conducted in our (Spectra) laboratories show that when comparisons are carried out in a single laboratory or between laboratories with identical instruments and workflows, the mode by which specimens are transported has no effect on the accuracy of the bicarbonate measurement.[8]

The first of these studies was conducted to test the effects of shipping (via next-day commercial air cargo or FedEx® air carriers) and time-in-transit on CO_2 values.[8] Three specimen tube samples were drawn from each of thirty healthy volunteers working at either the Spectra Laboratories New Jersey or California facility. One sample from each volunteer was analyzed on-site; the remaining two samples were shipped cross-country, one by next-day commercial air cargo and the other by FedEx® air carriers. Results are summarized in Table 2; no significant differences were seen among specimens ($p>0.05$ for all comparisons).

A second, larger study was also conducted comparing bicarbonate values reported from ESRD patient samples shipped via ground versus air; this study allowed us to control for any potential differences between patient samples and those obtained from healthy volunteers.[8] Data files from Spectra Laboratories generated during a single month (May 2000) were stratified based on whether the source sample arrived via ground (n=7,060) or air (n=55,168) transport.

The results of this study can be seen in Table 3. Specimens transported by air were associated with slightly higher CO_2 levels ($p<0.0001$. Note that a *lower* value would be expected in the case of a compromised sample). This degree of difference is not clinically significant, and falls within the bounds of normal analytical variation.[8]

Table 2

Effect of Shipping Blood Specimens Via Next-day Commercial Air Cargo or FedEx® Air Carriers on Serum Bicarbonate Values[8]

SPECTRA LABORATORIES (NJ)			
	BASELINE	COMMERCIAL AIRLINE	FedEx®
Mean CO_2 (mEq/L) ± SD	26.8 ± 1.38	26.9 ± 1.91	26.3 ± 2.12
SPECTRA LABORATORIES (CA)			
	BASELINE	COMMERCIAL AIRLINE	FedEx®
Mean CO_2 (mEq/L) ± SD	26.9 ± 2.21	27.1 ± 1.44	27.8 ± 1.72

Baseline values were obtained on site (at the sending lab) $p>0.05$ for all comparisons. Testing was funded by Spectra Laboratories, Inc.

Table 3

Comparison of Serum Bicarbonate Values Obtained From Blood Specimens Transported Via Ground Transport Versus Air Transport to a Single Laboratory[8]

	n	Mean CO_2 (mEq/L) ± SD
Ground Transportation	7,060	20.7 ± 3.78
Air Transportation	55,168	20.8 ± 3.72

$p<0.0001$

Bottom Line

Although some concern has been expressed in regards to air transport potentially resulting in falsely lowered bicarbonate values,[6,9] studies in our laboratories indicate that when samples are analyzed utilizing identical instrumentation and workflows, the mode by which specimens are transported has no significant effect on the accuracy of the bicarbonate measurement.[8]

Effect of Time to Analysis

It is well recognized that a given laboratory's practices/workflow can have a substantial impact on the accuracy of serum bicarbonate values.[6,8] Indeed, some consider practice/workflow variations to be more important than any other variable in terms of influencing the accuracy of a given bicarbonate measurement. The KDOQI guidelines specifically address this issue, stating that "introduction of air into the collection tube, the technique of removal of blood for assay, and long delays in the measurement can each adversely affect the results," and "for more accurate values, blood should not be allowed to have contact with air [and] delays in processing of the samples should be avoided."[6]

We have also analyzed the rate of loss of CO_2 from patient samples left standing in an open analyzer cup for one hour (N=48).[8] Here, mean CO_2 levels fell from 19.9 mEq/L to 17.4 mEq/L, a rate of loss of –2.5 mEq/L/hr. In order to minimize sample exposure to air and thus the chance for falsely decreased measured bicarbonate values, Spectra Laboratories has implemented a series of unique and effective specimen handling processes. Efforts to minimize exposure of samples to ambient air include decapping each SST tube just prior to analysis, as well as the utilization of chemistry instruments that can take the sample directly from the collection tube.

Effect of Assay Methods

Serum bicarbonate (as total CO_2) is generally measured in one of two ways: via enzymatic analysis or ion-selective electrode analysis. Approximately two thirds (3578-5365) of laboratories in a College of American Pathologists (CAP) survey used the enzymatic method, with the other third (1703-5365) using the ion-selective electrode method.

Proficiency survey data provided by the CAP clearly demonstrate that differences between laboratories "on the order of 2 to 4 mEq/L" may be method-related. Table 4 shows results from two recent sample surveys (survey "CHM-06" and survey "CHM-07"). All these results are from enzymatic methods. Ion specific electrode methods on these survey samples yielded results intermediate between the highest and lowest values shown here. (Note: Survey materials are generally not human serum and thus this data should be viewed with caution. However, differences among methods seen in survey data are usually indicative of differences observed with patient samples.)

Table 4

Mean CO_2 Values From CAP Proficiency Survey Data, Showing Differences in Values Among Laboratories Using Different Analytical Methods

	Survey Sample "CHM-06" (N=5365)		Survey Sample "CHM-07" (N=5409)	
Peer Group*	n	Mean CO_2 (mEq/L) ± SD	n	Mean CO_2 (mEq/L) ± SD
Highest	1058	25.6 ± 1.1	1079	15.6 ± 0.9
Lowest	232	22.0 ± 1.2	212	13.0 ± 0.8

*Laboratories using instrumentation

Bottom Line

Laboratory practices and workflow parameters can have a substantial effect on time-to-analysis of samples and thus reported bicarbonate values. Spectra Laboratories has implemented highly effective quality control measures aimed at minimizing exposure to ambient air and thus preserving sample integrity.

Bottom Line

Different assay methods can result in different reported bicarbonate values. The same laboratory and method of analysis should be used for serial measurements.[6]

Summary

- In patients with End-stage Renal Disease, chronic metabolic acidosis can have far-reaching negative effects.[1-8]
- Serum bicarbonate (total CO_2) is an important evaluation tool; low serum bicarbonate concentrations in maintenance dialysis patients are almost always indicative of metabolic acidosis.[6]
- The National Kidney Foundation's Kidney Disease Outcomes Quality Initiative (KDOQI) guidelines recommend serum bicarbonate levels be measured on a regular basis, and that if levels fall below 22 mmol/L, treatment be initiated.[6]
- Adjustment of the dose of bicarbonate administered during the dialysis treatment should be considered in patients who consistently exhibit serum bicarbonate levels below 22 mmol/L.
- A number of technical concerns can be associated with serum bicarbonate testing; these include the effects of blood collection techniques, shipping concerns, the effects of time to analysis, and the effects of assay methodology.
- An understanding of how these variables can impact measured serum bicarbonate levels can help clinicians obtain accurate, consistent results, and better interpret these results.

For more information, visit the Spectra Laboratories website at www.spectra-labs.com

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