Carbon Dioxide, Total
Test Code: CO2

Intended Use

For in vitro diagnostic use only

CARESIDE™ Carbon Dioxide (CO₂) cartridges are used with the CARESIDE Analyzer™ to quantitatively measure total carbon dioxide concentration in anti-coagulated whole blood, plasma or serum specimens. The CARESIDE Carbon Dioxide aids in the diagnosis and treatment of patients with respiratory and metabolic disorders associated with acid-base imbalance.

The CARESIDE Analyzer can use carbon dioxide results with sodium, potassium, and chloride results from the same specimen to calculate anion gap.

CARESIDE Carbon Dioxide, a single use cartridge, aids in specimen separation and delivers a measured volume of plasma or serum to a dry film to initiate the measurement of total carbon dioxide concentration. The film cartridge (patent pending) contains all reagents necessary to measure total carbon dioxide concentration.

Test Summary

Total carbon dioxide (CO₂) consists of bicarbonate ion (HCO₃⁻) and dissolved CO₂. Carbon dioxide is a gas, and as such occurs in very low concentrations in the blood. Bicarbonate ion as well as other ions are in equilibrium with CO₂ in the blood. The bicarbonate ion concentration in blood is related to the total carbon dioxide concentration and the pH according to the Henderson-Hasselbach equation.

The bicarbonate ion/carbonic acid pair represents the most important buffer system in blood. Clinical conditions characterized as metabolic disturbances of acid-base balance are classified as primary disturbances in bicarbonate ion concentration. Primary disturbances in the total dissolved CO₂ are characterized as respiratory disturbances. Changes in the bicarbonate ion, dissolved CO₂, or both, occur as a result of various compensatory mechanisms attempting to re-establish the normal ratio of bicarbonate ion to total dissolved CO₂.

Test Explanation

Each CARESIDE Carbon Dioxide cartridge consists of a carbon dioxide-specific multi-layer reagent film mounted in a plastic base with a hinged lid. The user introduces the anti-coagulated whole blood, serum, or plasma sample into the cartridge Sample Well, closes the lid and inserts the cartridge into the CARESIDE Analyzer.

Once loaded, the CARESIDE Analyzer scans the cartridge barcode and spins the cartridge to move the sample from the Sample Well into the cartridge channels and chambers. While spinning, the cartridge and the contained specimen are warmed to 37°C. As the cartridge continues to spin, the blood cells (if present) are separated from the plasma/serum and the cells accumulate in the separation well. Any excess sample flows into an overflow well, leaving the required volume in the metering passage.

The sample in the metering passage is automatically dispensed onto the multi-layer reagent film. In the spreading and substrate layer the sample distributes evenly on the film before it moves onto a reaction
layer where thio-NAD\(^+\) (yellow) forms in the presence of carbon dioxide.

As the cartridge spins, a photodiode measures film reflectance of light, emitted from a wavelength-specific light emitting diode (LED), over a fixed period of time. The analyzer uses the reflectance measurements and the lot-specific standard curve to calculate total carbon dioxide concentration.

**Test Principle**

The CARESIDE Carbon Dioxide test measures total carbon dioxide concentration in an enzyme-coupled reaction. Carbon dioxide in the form of bicarbonate ion combines with phosphoenolpyruvate in a reaction catalyzed by PEP-carboxylase (PEPC) to form oxaloacetate (OAC) and inorganic phosphate (PO\(_4\)^{3-}). Oxaloacetate reacts with thio-NADH (t-NADH) and hydrogen ion (H\(^+\)) in a malate dehydrogenase (MDH) catalyzed reaction to form L-malate and thiol-NAD\(^+\) (t-NAD\(^+\)).

The change of the color intensity due to the formation of thiol-NAD\(^+\), as measured by the amount of reflected light at 425 nanometers, directly relates to the specimen carbon dioxide concentration.

**Test Reaction Sequence**

\[
\begin{align*}
\text{HCO}_3^- + \text{PEP} & \xrightarrow{\text{PEPC, Mg}^{2+}} \text{OAC} + \text{PO}_4^{3-} \\
\text{OAC} + \text{t-NADH} + \text{H}^+ & \xrightarrow{\text{MDH}} \text{L-Malate} + \text{t-NAD}^+
\end{align*}
\]

**Test Element Architecture**

![Test Element Architecture Diagram](image)

**Test Cartridge Ingredients**

The active ingredients include PEP, thio NADH, magnesium, PEP carboxylase, and malate dehydrogenase.

**Test Warnings and Precautions**

- For *in vitro* diagnostic use only
- Use the total CO\(_2\) cartridge within 15 minutes of opening.
- Handle all patient samples, QC materials of human origin, and dosed test cartridges using universal precautions\(^1\).
- Protect reagent film from contamination or excessive light. Do not touch the film; touching the film can cause optical errors.
- Do not reuse cartridges.
- Specimens should be free from precipitates.

**Test Cartridge Storage & Preparation**

Cartridges are stable until the date printed on the cartridge label when stored at 2 to 8°C (36 to 46°F). Do not use if the package has been opened or damaged, or if the package seal is not intact. Unused cartridges may be returned to refrigerated storage if they are unopened.

**Test Calibration**

No user calibration is required. Each cartridge lot is factory calibrated and the calibration information is read automatically by the CARESIDE Analyzer.
Specimen Collection

The CARESIDE Carbon Dioxide cartridge is intended for use with appropriately collected whole blood, plasma or serum, and requires 90 ± 10µL sample per test cartridge.

Patient Preparation

No special preparations are required.

Whole blood
Collect whole blood in blood collection tubes containing sodium heparin. Do not use collection tubes containing citric acid. Mix anticoagulant according to manufacturer instructions allowing sufficient time for anticoagulation to occur.

Serum
Collect blood in the absence of anti-coagulant. Allow blood to fully clot at room temperature. Centrifuge the clotted blood and separate the serum from the clot promptly (within 4 hours from collection).

Plasma
Collect blood using anti-coagulant as described above (see Whole Blood, above). Promptly centrifuge the blood and separate the plasma from the cells.

Specimen Handling and Preparation

Fill vacuum tubes completely and keep specimens tightly capped to minimize loss of CO₂. Samples should be tested within 1 hour if possible in order to minimize the loss of CO₂. If immediate testing is not possible, remove serum or plasma promptly from the clot or cells. After the initial loss of CO₂ the total CO₂ consists primarily of bicarbonate ion whose concentration is relatively stable if stored at 2 to 8°C for several days². If analysis is delayed beyond 48 hours freeze, plasma or serum specimens at -20 °C or colder. Thoroughly mix any stored specimen before testing. Do not freeze whole blood.

Do not dilute specimens for routine testing. To obtain results for patients that have levels above the reportable range, dilute only serum or plasma samples. Do not dilute whole blood specimens. Use deionized CO₂-free water to perform a twofold dilution (1 part sample, 1 part diluent), and multiply the result by 2.

Test Procedures

1. Select the test to be performed on the CARESIDE Analyzer.

2. To dose the cartridge, lift the test cartridge lid by lifting up on the side clips. To the Sample Well, apply 90±10µl of sample or fill the well internally. An exact volume is not required, since the test cartridge will meter the correct volume. Do not allow the specimen to spill over the well. Any specimen outside of the well must be completely removed before proceeding.

3. Close the cartridge lid firmly until it clicks indicating complete closure.

4. Load the dosed test cartridge into the CARESIDE Analyzer and press “Confirm” (see the CARESIDE Analyzer Operator’s Manual for more information).

Quality Control

Each CARESIDE test cartridge barcode contains all required calibration information. The CARESIDE Analyzer scans the barcode, continuously performs quality control self-checks during operation, and alerts and prevents the user from proceeding if an unacceptable condition exists.

At a frequency determined appropriate by the user’s institution, evaluate analyzer performance using two levels of carbon dioxide controls. Contact CARESIDE Technical Service for a list of commercial controls compatible with the CARESIDE Analyzer. Each institution should establish quality control ranges using the CARESIDE Analyzer and monitor test performance using a Levey-Jennings chart (See CARESIDE Analyzer Operator’s Manual).

Results

Results require no user calculation and appear on the CARESIDE Analyzer screen, as they become available.

Reporting Units: mmol/L
Reportable Range: 5 to 40 mmol/L
**Expected Values (Reference Interval)**

The published reference interval for adult venous plasma or serum carbon dioxide is 23 to 29 mmol/L\(^3\).

The applicability of the above reference interval was confirmed for the CARESIDE CO\(_2\) using heparinized whole blood, heparinized plasma, and serum samples in a study of 42 ambulatory adults.

It is recommended that each institution establish values for the population they serve.

**Limitations**

A result outside the reportable range or any result not consistent with the clinical picture should be repeated to confirm.
Performance Characteristics

Detection Limit
The CARESIDE Carbon Dioxide cartridge demonstrated a detection limit of 5 mmol/L.

Accuracy
The CARESIDE Carbon Dioxide concentration agrees with a coupled enzymatic photometric method for bicarbonate (total carbon dioxide).

Precision
Human serum based controls (representing a low, middle and high carbon dioxide concentration) were tested in duplicate in at least 20 runs over a period of at least 3 days. Results are shown in the following table.

<table>
<thead>
<tr>
<th>Interferent</th>
<th>Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ascorbic acid</td>
<td>20 mg/dL</td>
</tr>
<tr>
<td>Bilirubin</td>
<td>15 mg/dL</td>
</tr>
<tr>
<td>Hemoglobin</td>
<td>300 mg/dL</td>
</tr>
<tr>
<td>Total Protein</td>
<td>15 g/dL</td>
</tr>
<tr>
<td>Triglyceride</td>
<td>3000 mg/dL</td>
</tr>
</tbody>
</table>

Additionally, the following 18 organic acids and drugs were tested and found not to interfere at clinically relevant concentrations: acetaminophen, acetoacetic acid, ammonia, benzoic acid, caproic acid, diatrizoate, ethanol, homogentisic acid, hydroxybutyric acid, hydroxyphenylpyruvate, hydroxyphenylacetic acid, hydroxyphenyllactate, ibuprofen, lactate, phenylacetate, phenylpyruvate, pyruvic acid, and salicylic acid.

Bibliography

Technical Assistance
If questions arise concerning the CARESIDE Analyzer or test cartridges, please contact CARESIDE Technical Service.

Telephone: (888) 698-CARE (2273)
(310) 338-6767
Fax: (310) 338-6789

Ordering Information
For product ordering or general information, please contact CARESIDE Customer Service.

Telephone: (888) 698-CARE (2273)
(310) 338-6767
Fax: (310) 338-6789

CARESIDE, Inc.
6100 Bristol Parkway
Culver City, CA 90230

CARESIDE Analyzer™ System

<table>
<thead>
<tr>
<th>CARESIDE Carbon Dioxide Precision</th>
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</thead>
<tbody>
<tr>
<td>Level 1</td>
</tr>
<tr>
<td>---------</td>
</tr>
<tr>
<td>Mean Conc. (mmol/L)</td>
</tr>
<tr>
<td>Within-run SD (mmol/L)</td>
</tr>
<tr>
<td>Within-run CV (%)</td>
</tr>
<tr>
<td>Total SD (mmol/L)</td>
</tr>
<tr>
<td>Total CV (%)</td>
</tr>
</tbody>
</table>

Actual precision may vary from site-to-site and from results indicated in this test insert. Each facility should verify that on-site product precision is appropriate for the site.

Interference
No significant interference was observed in anticoagulated whole blood or plasma samples when sodium heparin was used as the anticoagulant. The mean CO₂ concentration observed in 30 matched sodium heparin whole blood, sodium heparin plasma, and serum samples was identical (29 mmol/L). Also, no significant interference was observed up to the levels indicated for the following substances: