Oxygen Saturation

A Guide to Laboratory Assessment

BY SHANNON HAYMOND, PHD

Human life depends on the oxygen transport by hemoglobin. In healthy patients, the majority of molecular oxygen (O₂) is bound to hemoglobin and only a small fraction is dissolved in blood. But in patients with respiratory problems or certain metabolic and genetic disorders, the fraction of oxygenated hemoglobin can fall to dangerously low values. Therefore, laboratory assessment of oxygen saturation (SO₂)—the percentage of hemoglobin saturated with oxygen—provides an important indicator of a patient’s cardio-respiratory status and is frequently used in the emergency department, during general and regional anesthesia, and in intensive care settings.

Although the measured parameters are quite different for each, the three major analytical methods for measuring oxygen saturation—arterial blood gas analyzers, pulse oximetry, and CO-oximetry—are frequently used interchangeably by health care workers. Arterial blood gas analyzers calculate estimated oxygen saturation (O₂ sat) in a blood sample based on empirical equations using pH and PO₂ values, while pulse oximeters monitor arterial blood oxygen saturation, commonly referred to as SaO₂ or SpO₂, noninvasively by passing selected wavelengths of light through an area of the body, such as a finger. Both are measures of oxygen saturation.

The Clinical Laboratory Standards Institute (CLSI) defines O₂ sat as an estimated value based on a calculation, whereas S₂O₂ and S₂Hb refer to the arterial saturation measured spectrophotometrically. (To avoid further confusion, I will use only O₂ sat and SO₂ for the remainder of this article.) CO-oximetry, a more complex and reliable method, measures the concentration of hemoglobin derivatives in the blood, the results of which are then used to calculate various parameters such as hemoglobin derivative fractions (e.g., FO₂Hb), total hemoglobin, and oxygen saturation.

In most patients, the results—O₂ sat, SO₂, and FO₂Hb—from these three methods are virtually identical. But in cases of dyshemoglobinemia, some methods can yield misleading results, so health care professionals need to understand the differences between these methods and the limitations of each. This article will review the basics of oxygenation saturation values, so laboratory professionals can accurately assess oxygen saturation values.

An Oxygen Saturation Primer

Hemoglobin, the O₂ transport protein in blood, is comprised of four subunits: two α subunits and two non-α subunits, for example β, γ, or δ. Each subunit contains a porphyrin heme iron (Fe) moiety and seven helices. The heme Fe exists in the Fe(II) or Fe(III) oxidation state, but only the Fe(II) state is capable of binding O₂. Most clinically important dyshemoglobinemia gene mutations, such as the thalassemias, reduce the quantity of α- or β-chain synthesis in the hemoglobin subunit or lower the solubility of hemoglobin—as occurs with HBS (sickle cell hemoglobin) or HBC (hemoglobin C), for example—but gene mutations rarely alter the O₂ affinity of hemoglobin.

Hemoglobins can be divided into two classes: normal hemoglobin that is capable of binding O₂ and dyshemoglobins—hemoglobin derivatives that are incapable of binding O₂. The normal hemoglobins include oxyhemoglobin (O₂Hb) and deoxyhemoglobin (Hb), while the dyshemoglobins include carboxyhemoglobin (COHb), methemoglobin (MetHb), and sulfhemoglobin (SHb). COHb forms when a person is exposed to CO fumes and CO replaces O₂ in hemoglobin, which can result in death. SHb forms through a reaction of sulf-containing compounds with the heme moiety; cases of sulfhemoglobinemia are rare and usually result from extensive use of sulfa-containing drugs. MetHb represents the oxidized, deoxy form (Fe(III)-Hb) of hemoglobin to which O₂ cannot bind. The likely etiologies of methemoglobinemias include exposure to highly oxidizing drugs or the presence of genetic hemoglobin variants such as HbM.

To understand how hemoglobin carries and releases oxygen, the oxygen dissociation curve (ODC) serves as an important tool (Figure 1). In the lungs, where partial pressures of O₂ are high, O₂ binds to hemoglobin to form O₂Hb. Erythrocytes carrying O₂Hb then circulate in the blood and release O₂ in response to decreased partial pressures of O₂ in the tissues. The cooperativity of O₂ binding—an allosteric phenomenon whereby binding of oxygen by one hemoglobin subunit enhances the ability of the remaining subunits to bind oxygen—produces the sigmoidal shape of the curve. As O₂ binds to the second and third subunits of hemoglobin, binding increases incrementally so that the four subunits of hemoglobin all become fully saturated at the normal O₂ tension in the lungs.
lungs alveoli. The same process works in reverse in the tissues; once fully loaded hemoglobin releases one O₂ molecule, it releases the next more easily.

The ODC also explains what happens to patients when oxygen cannot bind hemoglobin. For example, in the presence of CO or when the heme Fe is oxidized to the Fe(III) state, O₂ cannot bind one of the hemoglobin subunits. These conditions decrease O₂ capacity in addition to inhibiting O₂ transport by blood. In addition, CO and oxidized heme Fe alter hemoglobin's conformation in a way that decreases the O₂ affinity for the remaining heme Fe groups, shifting the ODC to the right. The net biological effect is decreased O₂ delivery to tissues. Temperature, pH, and 2,3-DPG (2,3-diphosphoglycerate) concentration also affect the O₂ affinity of hemoglobin, shifting the ODC as shown in Figure 1.

An important aspect of the ODC is the P₅₀, the partial pressure of oxygen in the blood at which the hemoglobin is 50% saturated. The value for P₅₀—typically about 26.6 mm Hg for a healthy person—is used to calculate oxygen saturation. Temperature, pH, and 2,3-DPG concentration also affect the O₂ affinity of hemoglobin, shifting the ODC as shown in Figure 1.

Figure 1

Oxygen Dissociation Curve Of Hemoglobin

The percent saturation of hemoglobin with oxygen at different oxygen tensions is depicted by the sigmoidal curves. The P₅₀, indicated by the dashed lines, is about 27 mm Hg in normal erythrocytes. Modifications of hemoglobin function that increase oxygen affinity shift the curve to the left, whereas those that decrease oxygen affinity shift the curve to the right. Reprinted with permission from Kelley’s Textbook of Internal Medicine, 4th ed., 2000, figure 241.2. Lippincott Williams & Wilkins.

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Figure 2

Extinction Curves of Purified Hemoglobin Derivatives

Analytical Methods for Measuring Oxygen Saturation

<table>
<thead>
<tr>
<th>Device</th>
<th>Specimen</th>
<th>Measurement</th>
<th>Reported Data</th>
<th>Advantages</th>
<th>Disadvantages</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arterial Blood Gas (ABG) Analyzer</td>
<td>Blood</td>
<td>Partial pressure of oxygen dissolved in whole blood at an electrode</td>
<td>PO₂, SO₂ (O₂ sat)</td>
<td>Also measures pH and PCO₂</td>
<td>Invasive, O₂ sat may be inaccurate in hospitalized patients and if dysHb present</td>
<td>If the ABG and pulse oximetry are discrepant, an abnormal Hb may be present.</td>
</tr>
<tr>
<td>Pulse oximeter</td>
<td>Trans-cutaneous</td>
<td>Absorption at two wavelengths (660 nm and 940 nm) in blood</td>
<td>SO₂</td>
<td>Non-invasive, continuous bedside monitoring</td>
<td>Inaccurate when interfering substances are present: MetHb, certain dyes</td>
<td>When MetHb &gt;25% the pulse oximeter will read saturation of 75%–85%.</td>
</tr>
<tr>
<td>CO-oximeter</td>
<td>Blood</td>
<td>Absorption of Hb derivatives using multiple wavelengths of light</td>
<td>SO₂, FCO₂Hb, FMetHb, FCOHb, FSHb, cHb</td>
<td>Measures the concentration of Hb species</td>
<td>Invasive, performed in laboratory; not all instruments report SHb, total bilirubin</td>
<td>Most accurate method; even in cases of CO- poisoning and methemoglobinemia.</td>
</tr>
</tbody>
</table>

Table 1

**Oxygen Saturation**

**Some Common Abbreviations**

- O₂: Molecular oxygen
- O₂Hb: Oxyhemoglobin
- CO: Carbon dioxide
- HHb: Deoxyhemoglobin
- SO₂: Oxygen saturation
- COHb: Carboxyhemoglobin
- O₂Sat: Estimated oxygen saturation
- MetHb: Methemoglobin
- SₖO₂: Arterial oxygen saturation, as reported
- SₖO₂ by pulse oximeters
- SHb: Sulphhemoglobin
- HS: Sickle cell hemoglobin
- FO₂Hb: Fractional oxyhemoglobin
- HbC: Hemoglobin C
- CO₂Hb: Oxyhemoglobin concentration
- HBF: Hemoglobin F
- cHb: Total hemoglobin concentration
- FCOHb: Fractional carboxyhemoglobin

Non-Spectrophotometric Methods of Analysis

Most blood gas analyzers are comprised of a series of electrodes that provide information regarding a patient’s acid/base status (pH), respiratory function (PCO₂), and oxygenation status (PO₂). PO₂ measurements are based on changes in electrical current measured at a Clark electrode, whereas pH and PCO₂ measurements are determined from voltage changes at high-impedance electrodes. Newer, point-of-care (POC) blood gas analyzers employ dry slide technology with optical detection.

Some of the parameters measured by blood gas instruments are different than those measured by the spectrophotometric methods. The PO₂ value determined by these instruments reflects the dissolved oxygen gas in blood. The concentration of dissolved oxygen (cO₂) is linearly related to the partial pressure of oxygen in blood according to Henry’s law where the solubility constant of O₂ (aO₂) is 0.03 mL O₂/100 mm Hg: cO₂ = aO₂ × PO₂

However, the relationship between the estimated SO₂, often referred to as O₂ sat, and PO₂ may not always be linear. O₂ sat is calculated from the pH and PO₂ values and the standard ODC for oxygen saturation.

Unfortunately, this approach to calculating O₂ sat assumes a normal ODC, which is frequently not the case in hospitalized patients. To circumvent this problem, some labs customize the output of blood gas analyzers so that they do not report O₂ sat; however, some POC devices do not offer this option and could potentially mislead some health care professionals. To avoid such errors, the most recent approved guideline from CLSI discourages the use of estimated values.

Best Practice for Oxygen Saturation Measurement

Despite the limitations discussed above, measurement of the oxygen saturation of hemoglobin remains a valuable tool for assessing a patient’s respiratory status, and laboratory analysts can enhance the quality of patient care by being aware of these limitations and educating other health care workers about them. Table 1 summarizes the advantages and disadvantages of the three analytical methods for assessing oxygen saturation.

The most important point to note is that the commonly reported parameters from the three methods—O₂ sat, SO₂, and FCO₂Hb—are quite different and therefore should not be used interchangeably.

In addition, laboratory workers and other health care workers need to be aware that estimated O₂ sat values should be interpreted with caution as the algorithms used to calculate this value assume that the patient has normal O₂ affinity, normal 2,3-DPG concentration, and no dyshemoglobinemia or hemoglobinopathy.

Finally, it is important to note that SO₂ has limited value as the sole indicator of oxygen status in cases of suspected dyshemoglobinemia, because the presence of dyshemoglobins affects O₂ capacity and affinity but not saturation. In these cases, information from CO-oximetry is necessary. In addition to the FCO₂Hb, CO-oximeters should be programmed to report the various dyshemoglobin fractions in addition to FCO₂Hb, as FCO₂Hb alone has limited utility.

**SUGGESTED READINGS**


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