A Novel Hemoglobin, Bonn, Causes Falsely Decreased Oxygen Saturation Measurements in Pulse Oximetry

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BACKGROUND: A 4-year-old boy and his father exhibited low oxygen saturation measured transcutaneously by pulse oximetry, a finding that could not be confirmed by arterial blood gas analysis. Both patients exhibited slight hemolysis in their blood, and the boy had a microcytic anemia. There was no evidence of hypoxemia or methemoglobinemia. Despite the normal results from the arterial blood gas analysis, a right-to-left-shunt was assumed in the boy until a cardiology examination excluded this diagnosis. Sleep apnea syndrome was suspected in the father and treated with nocturnal positive pressure respiration based on the low oxygen saturation values obtained with pulse oximetry. Only after consultation with our laboratory was a hemoglobin variant suspected and investigated.

METHODS: We performed hemoglobin protein analysis by HPLC, electrophoretic separation, and spectrophotometry and DNA sequence analysis of the α-globin gene.

RESULTS: Both HPLC chromatographic separation and alkaline electrophoresis revealed a unique hemoglobin peak. In both patients, α-globin gene sequencing revealed a mutation resulting in a histidine-to-aspartatic acid substitution at position α87. The low oxygen saturation measurement by pulse oximetry was due to hemoglobin Bonn oxymoglobin having an absorption peak at 668 nm, near the 660 nm measured by pulse oximeters.

CONCLUSION: Hemoglobin Bonn is a novel hemoglobin variant of the proximal α-globin that results in falsely low oxygen saturation measurements with pulse oximetry.

A 4-year-old boy being treated for Morgagni hydatid had elective surgery under general anesthesia. Intraop-
brother and the son’s sister. The laboratory was contacted, leading to identification of a hemoglobin anomaly in both father and son.

In both patients, chromatographic separation of hemoglobin using the Variant II HPLC (Bio-Rad) showed a distinct peak at 0.67–0.70 min, immediately before the HbA1c peak. Gel electrophoresis was performed with Hydrasys (Sebia) using a commercially available method (Hydragel Hemoglobin). Alkaline electrophoresis revealed an unknown band cathodally before the HbA0 peak.

Genomic DNA was isolated by the use of the QIAmp DNA Blood Kit (Qiagen). PCR amplification of the respective products and bidirectional automated sequence analysis was performed as recently described (1). PCR primers were designed to selectively amplify the human \( \alpha \)- and \( \beta \)-globin genes. The forward primer (\( \alpha1 \)-2F, 5’-CCAAGGCACCTCAAC CGT-3’; \( \alpha1 \)-2R, 5’-GCCATCTCGCCCTCGACC-3’). In father and son, sequencing of the \( \alpha \)-globin gene revealed a point mutation c.C299G (Genbank acc. no. NM_000558) in exon 2 of the hemoglobin, alpha 1 (HBA1) gene, resulting in an H87D exchange. Spectrophotometric measurement of the pretreated capillary blood using the LS 500 spectrophotometer (Lange) revealed an additional absorption maximum of oxyhemoglobin at 668 nm, while the absorption curve of deoxyhemoglobin remained inconspicuous (Fig. 1).

Pulse oximetry is commonly used for rapid measurement of pulse rate and oxygen saturation. Because of their absorption spectra, however, hemoglobin variants and some derivatives of normal hemoglobin may interfere with pulse oximeter measurement (2, 5). For example, hemoglobins Köln and Cheverly also produce falsely-low oxygen saturation values by pulse oximetry (6, 7).

In commonly applied pulse oximeters, absorption is measured at 660 m and 940 nm, where the largest differences in absorption of oxyhemoglobin and deoxyhemoglobin can be found. Hemoglobin Bonn has an additional oxyhemoglobin absorption maximum at 668 nm, resulting in a falsely-low estimation of oxygen saturation (Fig. 1) (4, 9).

Despite normal oxygen saturation findings from the blood gas analyzer, diagnoses were made in both father and son that resulted in relatively complex and cost-intensive workups or therapy as well as considerable psychological stress. The father eventually received therapeutically incorrect treatment (nocturnal positive pressure respiration for suspected sleep apnea syndrome). Only upon contacting the laboratory was a hemoglobin anomaly considered.

Hemoglobin Bonn can be easily identified by chromatography, electrophoresis, or spectrophotometry. With the identification of this and other hemoglobins that interfere with pulse oximeter measurement, complex and expensive examinations might be avoided.

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