

Glucose Meter Evaluations – Capillary Blood Testing

Introduction

The design of certain blood glucose meters restricts their use to test only fresh human capillary whole blood; these meters do not display accurate patient results with either low- or high-oxygenated blood samples. In order to assess the performance and inaccuracy of these devices, one of two possible protocols must be performed: (1) human subject donors must be recruited so that fresh, capillary blood can be drawn and tested or (2) fresh venous blood must be drawn and must be adjusted to ‘capillary-equivalent’ oxygen partial pressures (PO_2). The purpose of this document is to provide information and recommendations for conducting glucose meter performance evaluations and testing these glucose meters with capillary or ‘capillary-equivalent’ blood samples.

PO_2 -Adjusted Venous Blood

Adjusting the PO_2 of venous blood to mimic ‘capillary-equivalent’ oxygen tensions requires considerable skill and expertise; blood gas equipment that is costly, complex, and inherently unstable; access to barometer readings and localized barometric pressure; and knowledge of many pre-analytical and analytical factors that can affect blood gases.

Clearly, this is not a practical option for many clinicians and non-laboratorians.

Therefore, human subject recruitment and testing capillary blood directly from skin punctures is the preferred method. However, [Appendix A](#) provides a materials list and suggested testing procedure if appropriate skills and equipment exist and if human subject testing cannot be performed.

Note: The results from PO_2 adjusted venous blood testing must be limited to laboratory evaluation purposes only – not for patient testing.

Human Subjects:

Informed consent and protocol documents should be reviewed and approved by an institutional review board or human subject ethics committee prior to the commencement of the study. Each enrolled subject should be aware of the study risks and benefits and must agree to and sign an Informed Consent document.

For glucose meter evaluation studies, between 40 and 100 human subjects should be enrolled depending on the statistical plan. The study population should be described and recruited using criteria from the Statement for Reporting Studies of Diagnostic Accuracy (STARD). If possible, both users and trained personnel should perform the meter testing.

Examples of possible exclusion/inclusion criteria are:

- Disease state (e.g. Type 1, Type 2, Gestational, pre-diabetic, or non-diabetic)
- Severity of disease (e.g. A1c values)
- Blood hematocrit level
- Patient-specific interferences (i.e. oxygen or drug therapy, poor peripheral perfusion, edematous puncture sites, recent blood transfusion, and/or fed state)
- Age
- Willingness to undergo skin punctures, phlebotomy, and/or testing procedures
- Language and comprehension (i.e. ability to follow manufacturer's instructions)

Reference Method:

Comparative methods that can be traced to materials or methods of higher order should be used as the reference method. For glucose, this means that the reference method should be designed to test glucose in human serum or plasma – not human whole blood. The reference method should be checked to be stable and to be within quality control limits. It is highly recommended that that accuracy of the reference method be checked with appropriate National Institute of Standards and Technology (NIST) reference materials (e.g. SRM 965a). The total error (bias plus imprecision) of reference methods for glucose should not exceed $\pm 8-10\%$.

Storage and Handling of Reagents:

Manufacturer's instructions for storage and handling should be followed. Usually, this means storage of reagent test strips and control solution at room temperature below 30° C (86° F). Keep away from heat and direct sunlight. Do not refrigerate or freeze. Obtain enough test strips for control solution tests as well as duplicate tests for each subject sample. Protect test strips in the bottle both before and during testing by following manufacturer's instructions.

Testing Environment:

The testing should take place in a room that has controlled temperature and humidity according to specifications in manufacturer's instructions.

Training:

Clinical and other testing personnel who will perform the glucose meter testing should be trained to the device limitations, manufacturer's instructions, safety practices, and to the test protocol. Personnel testing the reference (plasma) glucose, centrifuge, and the blood hematocrit must be trained to the safe and correct use of these devices and equipment.

Test Procedure:

1. Follow safety guidelines per institutional requirements. Wear safety equipment (gloves, protective eyewear, and lab coat, etc.) as appropriate.
2. Prepare device according to manufacturer's instructions (e.g. calibration code, control solution test within range, correct sample application, etc.). Check that the device is with control solution range if the manufacturer recommends this test.
3. Prepare patient – ensure that skin site is clean and dry prior to skin puncture. Follow Clinical and Laboratory Standards Institute (CLSI) guidelines (e.g. NCCLS H4-A3) for safe skin punctures, blood collection, and blood handling.
4. Test blood hematocrit
5. Test glucose meter in duplicate with capillary blood. Apply the blood according to manufacturers instructions. If possible, both users and trained personnel should perform the meter testing.
6. Within 5 minutes, a portion of the same sample should be centrifuged and the plasma tested in duplicate with the reference method.
7. If necessary, samples may be spiked so that the meter data is distributed across reportable range. Care should be taken to not affect hematocrit or PO₂ due to this spiking process.

Data analysis

1. Check that the blood hematocrit is within the range for the meter or else excluded
2. Check that duplicate reference measurements are within 4% or 0.22 mmol/L (4 mg/dL) or else excluded.
3. Check that the data spans the reportable range for the glucose meter.
4. Compare individual meter results to the mean of reference duplicates

Acceptance Criteria

CLSI and ISO acceptance criteria are: 95% of the data are clinically accurate.

Clinically accurate data are defined as data within ± 15 mg/dL for glucose < 75 mg/dL or $\pm 20\%$ for glucose ≥ 75 mg/dL.

References:

Bossuyt PM, Reitsma JB, Bruns DE, Gatsonis CA, Glasziou PP, Irwig LM, et al. The STARD Statement for Reporting Studies of Diagnostic Accuracy: Explanation and Elaboration. *Clin Chem* 2003; 49(1): 7-18.

Bossuyt PM, Reitsma JB, Bruns DE, Gatsonis CA, Glasziou PP, Irwig LM, et al. Towards complete and accurate reporting of studies of diagnostic accuracy: the STARD initiative. *Clin Chem* 2003; 49(1): 1-6.

NCCLS. Point-of-Care Blood Glucose Testing in Acute and Chronic Care Facilities; Approved Guideline – Second Edition. NCCLS document C30-A2 (ISBN 1-56238-471-6). NCCLS, 940 West Valley Road, Suite 1400, Wayne PA 19087-1898 USA, 2002.

ISO 15197 In vitro diagnostic test systems - Requirements for blood-glucose monitoring systems for self-testing in managing diabetes mellitus. First edition dated 2003-05-01. ISO 15197:2003(E) International Standards Organization, Geneva, Switzerland.

SKUP. Report on OneTouch Ultra, SKUP/2005/39. Scandinavian evaluation of laboratory equipment for primary health care. NOKLUS, Division of General Practice, University of Bergen, 5009 Bergen, Norway. [www.SKUP.nu]

Kristensen G, Nerhus K, Thue G, Sandberg S. Standardized evaluation of instruments for self-monitoring of blood glucose by patients and a technologist. *Clin Chem* 2004; 50(6): 1068-71.

NCCLS H4-A4 Procedures and devices for the collection of diagnostic blood specimens by skin puncture; approved standard - fourth edition. (www.CLSI.org)

Appendix A

Protocol for Adjusting the PO₂ of Venous Whole Blood to “Capillary-Equivalent” PO₂

Purpose:

The purpose of this protocol is to describe a procedure for modifying the PO₂ of venous whole blood to that PO₂ of ‘capillary-equivalent’ whole blood. This protocol may be used for laboratory evaluation of ‘capillary-blood-only’ devices.

Note #1: The results from PO₂ adjusted venous blood testing must be limited to laboratory evaluation purposes only – not for patient testing.

Materials:

- Sodium or lithium heparin tubes containing 5-6 mL of fresh human whole blood from a five or more donors with hematocrit within manufacturer’s limitations and within 12-hours of phlebotomy.
- Blood gas analyzer and supplies (per blood gas analyzer manufacturer’s instructions)
- Syringes (with caps) and blunt needles
- Barometer (or access to local barometric pressure)
- Concentrated ($\geq 8,000$ mg/dL) glucose spiking solution, if needed
- Safety equipment (gloves, protective eyewear, and lab coat, etc. as appropriate)

Note #2: The PO₂ of room air at sea level is ~160 mmHg [$760 \times .21 = 159.6$ mmHg]. The PO₂ of freshly drawn venous blood is ~30-40 mmHg. The goal of this procedure is to create a blood sample with a “capillary-equivalent” PO₂ of between 60-70 mmHg. If your altitude and barometric pressure are different, adjust these values as appropriate.

Note #3: Make all glucose, hematocrit, or other blood sample adjustments prior to beginning this procedure. Minimize the exposure of the blood to air - do not agitate, rock, or excessively expose the blood sample to room air. Blood sample PO₂ is extremely sensitive to blood sample manipulation and must be performed immediately prior to applying the sample to the test strip.

Procedure:

Step	Action	Rationale
1	Follow safety guidelines per institutional requirements. Wear safety equipment (gloves, protective eyewear, and lab coat, etc.) as appropriate.	
2	Follow manufacturer's instructions for the Blood Gas Analyzer. Calibrate blood gas analyzer (using slope and calibration gases if appropriate) and check system for accuracy and stability. Ensure that the expected gas values are calculated using the local barometric pressure. Calibrate and maintain blood gas instrument per CLSI recommendations and manufacturers instructions.	Blood Gas System check
3	Carefully draw at least 3-mL of venous blood into a syringe and measure the sample PO ₂ . Be careful not to excessively aerate or induce bubbles into the sample.	Initial check of sample PO ₂
4	If the PO ₂ is > 70 mmHg, the blood sample cannot be used.	The PO ₂ in the blood is above the 60-70 mmHg capillary target
5	If the PO ₂ is < 60 mmHg, carefully draw a small amount of air (10-20% by volume) into the syringe. The volume of air in this headspace depends on the PO ₂ found in Step 2 – a larger air-to-blood ratio should be used if the initial PO ₂ is low; a smaller air-to-blood ratio should be used if the initial PO ₂ is higher.	The PO ₂ in the blood is below the capillary target and must be exposed to a very small amount of air to increase blood sample PO ₂ .
6	The PO ₂ should be adjusted carefully and checked frequently until it is found to be within the range of 60-70 mmHg. Then, expel all air from the syringe, seal it, and keep the blood remaining in the syringe airtight.	Mixing blood with atmospheric air will increase blood sample PO ₂ .
7	Briefly open the syringe to measure the PO ₂ of the blood with a blood gas analyzer. Immediately replace the syringe cap to keep the balance of the blood in the syringe airtight.	Subsequent check of modified sample PO ₂
8	Repeat steps #4-6 until the final PO ₂ equals 65 ± 5 mmHg.	Venous blood with a PO ₂ of 65 ± 5 mmHg is 'capillary-equivalent' blood

9	When target PO ₂ is confirmed by blood gas measurement, immediately apply the blood sample to the glucose test strip. In-between tests, protect the blood in the syringe from any additional exposure to air.	Prolonged exposure to air may increase the PO ₂ above capillary levels.
10	The sample must be used immediately after preparation. Repeat Steps #1-8, as needed, for multiple samples.	Erythrocyte metabolism will lower sample PO ₂ and blood glucose if timing is not controlled. Exposure to atmospheric air will increase sample PO ₂ if not controlled.
11	Both meters and reference methods must be tested in duplicate – return to the procedure and follow recommended testing and data analysis procedures found in this document.	CLSI NCCLS C30-A2 recommendations

References for Appendix A:

Mahoney JJ, et al. Arterial Blood Gas Analysis, Chapter 9 In: Respiratory Care: A Guide to Clinical Practice, 4th Edition. Burton GG et al, editors. 1997. Lippincott-Raven Publishers, Philadelphia, PA USA.

CLSI C46-A. Blood Gas and pH Analysis and Related Measurements; Approved Guideline. [Available at <http://www.clsi.org>].

Sacks DB. Carbohydrates – Chapter 24 In: Tietz Textbook of Clinical Chemistry 1999;WB Saunders Company, Philadelphia, PA 19106.

Davis C et al. Determination of the partial pressures of Oxygen in Capillary samples and Venous whole blood samples from syringes and Vacutainers [abstract]. Clin Chem 1995;41(6): S201.