Method Comparison/User Evaluation of the i-SENS Self-Monitoring Blood Glucose / β-Ketone System (CareSens PRO GK BT)

Protocol: BGM-2205084

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Version History

Version No.	Version Date	Details
1	19 May 2022	Initial version.
2	27 Jul 2022	Updated name of product to CareSens PRO GK BT. Clarified the QC solutions used with the Randox reference equipment. Included a hypoglycemia/hyperglycemia reference image within the study Questionnaire and removed Note 2.

Investigator Signature

I confirm that I have read this protocol, I understand it, and I will work according to this protocol. I will also work consistently with the ethical principles that have their origin in the Declaration of Helsinki and that are consistent with good clinical practices and the applicable laws and regulations. I will accept the monitor's overseeing of the trial. I will abide by the publication plan set forth in my agreement with i-SENS, Inc. I confirm that if I or any of my staff are members of the ethics review board, we will abstain from deliberation and voting on this protocol.

Investigator's Signature

Date of Signature
(DD MMM YYY)

Investigator's Name (print)

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ABBREVIATIONS

AE	Adverse Event
BG	Blood Glucose
CRF	Case Report Form
DD	Device Deficiency
GCP	Good Clinical Practice
HCP	Health Care Professional
ICF	Informed Consent Form
ID/GC/MS	Isotope Dilution Gas Chromatography Mass spectrometry
IRB	Institutional Review Board
JCTLM	Joint Committee for Traceability in Laboratory Medicine
NCCLS	National Committee for Clinical Laboratory Standards
PI	Principal Investigator
RX Imola	Randox Imola D-3-Hydroxybutyrate (Ketone) analyzer (Randox Laboratories)
SAE	Serious Adverse Event
SMBG	Self-Monitoring of Blood Glucose
UADE	Unanticipated Adverse Device Effect
YSI	YSI 2300 Stat Plus Glucose Analyzer (YSI Incorporated)

1. Background Information

The CareSens PRO GK BT Blood Glucose/Blood β-Ketone Monitoring System is used for the quantitative measurement of the glucose/β-ketone level in capillary whole blood as an aid in monitoring diabetes management effectively at home. The CareSens PRO GK BT Blood Glucose/Blood β-Ketone Monitoring System includes the Meter, Blood Glucose Test Strips/Blood β-Ketone Test Strips, a user's manual, a quick reference guide, batteries, lancets, a lancing device and a carrying case. Two levels of control solutions (control level A and B) for glucose test and two levels of control solutions (control level A and B) for β-ketone test are also associated with the test system.

This in vitro diagnostic system requires 0.4 μL of blood and delivers results in 5 seconds for the glucose test and 0.5 μL of blood and delivers results in 8 seconds for the β-ketone test. It contains a meter memory of up to 1000 test results with time and date. The system has an optional strip expiration date indicator, hypoglycemia / hyperglycemia indicator and adjustable alarm features.

The CareSens PRO GK BT meter can calculate and display the averages of total test results, pre-meal test results, post-meal test results and fasting test results, from the past 1, 7, 14, 30 and 90 days if the appropriate information has been provided to the meter. The meter is designed to minimize code related errors in monitoring by using a no-coding function.

The clinical trial will assess the accuracy and usability of the meter in untrained subjects with or without diabetes or pre-diabetes.

The clinical trial will follow the requirements described in the FDA Guidance for Self-Monitoring Blood Glucose (SMBG) Test Systems for Over-the-Counter Use (Section VI. PERFORMANCE EVALUATION FOR SMBGs, Section C) for glucose, and CLSI EP09-A3 Measurement Procedure Comparison and Bias Estimation Using Patient Samples for β-ketone.

2. Study Objectives and Purpose

The method comparison and user evaluation study are designed for the assessment of both system accuracy in the hands of the intended users as well as other aspects to support lay use, such as a labeling assessment and usability.

Objectives

- To determine if untrained people with or without diabetes or pre-diabetes can obtain acceptable glucose and β-ketone results from fingerstick capillary blood.
- Glucose: 95% of all glucose results in the study should be within ±15% of the comparator results across the entire claimed measuring range of the device and 99% of all SMBG results within ±20% of the comparator results across the entire claimed measuring range of the device.
- β-ketone: 95% of the individual ketone results in this study should be within ±0.3mmol/L at concentration < 1.5mmol/L and within ±20% at concentration ≥ 1.5mmol/L of the comparator results.

3. Study Design

A draft device labeling (which have undergone a Flesch and Kincaid reading assessment), is given to the study subjects and no other training or assistance from a trained study staff/technician is provided to the user.

A minimum of 10 test strip vials/packages covering a minimum of 3 test strip lots are included in the test. The test strips will have undergone typical shipping and handling conditions from the site of manufacture to a U.S. site prior to being used in the study.

Taking into consideration of a "worst-case scenario", the strips used in the method comparison/user evaluation shall have undergone a container shipping by sea (approximately 60 days) and its shipping and handling records shall be kept.

Study testing procedures are conducted by trained study staff/technicians familiar with all aspects of the protocol, documentation, forms, use of the investigations and commercially available SMBG, data collection and GCP. All training will be documented and filed.

Glucose

Following the FDA SMBG Guidance, at least 350 evaluable and different subjects, which meet the inclusion/exclusion criteria, are to be included in the user evaluation of the Self-Monitoring Blood Glucose Test Systems (i.e. SMBG). These results will be compared to results from the reference equipment (YSI STAT Plus 2300 glucose analyzer). From the minimum of 350 evaluable subjects, at least 10 fingerstick capillary blood samples shall be < 80 mg/dL and >250 mg/dL respectively. Therefore the number of subjects may exceed 350 until the specified glucose concentration ranges are filled.

β-ketone

The test results are to be analyzed based on the CLSI EP09-A3 guideline through method comparison analysis using capillary blood samples. The performance is evaluated with a minimum of 350 capillary blood samples and compared to results from the reference equipment (Randox Imola D-3-Hydroxybutyrate analyzer). From the minimum of 350 evaluable subjects, at least 10 fingerstick capillary blood samples shall be ≥1.5 mmol/L.

4. Selection and Withdrawal of Subjects

4.1 Subject Recruitment

Recruiting will be done by the PI and trained study staff at the clinical site.

4.2 Informed Consent

Consent will be obtained before the study commences at the clinical site by the PI and trained study staff. In obtaining and documenting informed consent, the investigator must comply with applicable regulatory requirement(s) and adhere to ICH GCP and the requirements in the Declaration of Helsinki before any study-related activity.

The investigator or designee must give the subject verbal and written information about the study and the procedures involved in a form that the subject can read and understand. The subjects must be fully informed of their rights and responsibilities while participating in the study as well as possible disadvantages.

The investigator or designee must ensure the subject has ample time to come to a decision whether or not to participate in the study. Study subjects will be given the opportunity to ask questions. A voluntary, signed and personally dated informed consent must be obtained from the subject before any study-related activity.

The responsibility for seeking informed consent must remain with the investigator, but the investigator may delegate the task to a properly trained person, in accordance with local requirements. The written informed consent must be signed and personally dated by the person who obtained the informed consent before any study-related activity.

If information becomes available that may be relevant to the subject's willingness to continue participating in the study, the investigator must inform the subject in a timely manner, and a revised written subject information must be provided, and a new informed consent must be obtained. The informed consent describes the study, what is expected of the subject during the study, its potential risks, the subject's privacy rights under HIPAA (Health Insurance Portability and Accountability Act of 1996), and the details of participation.

4.3 Inclusion/Exclusion Criteria

The subjects enrolled are assigned a screening number and will be assessed for study subject inclusion/exclusion criteria.

Inclusion Criteria:

- Males and females, 18 years of age and older
- People with type 1 diabetes, type 2 diabetes, pre-diabetes and no diabetes (self- reported)
- Able to speak, read and understand English (subjects must demonstrate ability to read a sentence from a page of the draft device labeling instructions (user's manual) to qualify for the study)
- Willing to complete all study procedures
- Has read, understood, and signed the Informed Consent Form

Note: The study group consists of naive SMBG users and non-naive SMBG users. At least 10% of study participants should be naive for SMBG, including subjects without diabetes.

Exclusion Criteria:

- Hemophilia or any other bleeding disorder
- Works for a medical laboratory, hospital, other clinical setting or a medical device company that involves training on or clinical use of blood glucose meters
- Physical, visual or neurological impairments as determined by the investigator or designee that would make the subject unable to perform self-testing (reason for exclusion will be clearly documented by investigator or designee directly on the subject disposition form)
- A condition, which in the opinion of the investigator or designee, would put the subject or study conduct at risk (reason for exclusion will be clearly documented by investigator or designee on the subject disposition form).

Note 1: In the event of a physical, visual or neurological impairment that makes it difficult for a subject to complete the questionnaire, the subject's verbal responses will be accepted and should be appropriately documented as such within the subject records form.

Note 2: Evaluable data from all eligible subjects in the study will be included even if more than 350 samples are collected; no eligible subject data will be excluded from the data analysis. (See section 5.8.3. Data Evaluability).

Upon meeting the inclusion/exclusion criteria, a unique subject number is assigned. The trained study staff will collect self-reported subject demographics, diabetes (or pre-diabetes) history, self-monitoring of blood glucose/β-ketone history, education background and other information relevant to the study and to the ability to use a meter.

4.4 Study Termination and Subject Withdrawal Criteria

Subjects may be withdrawn from the study at their own request for any reason or at the discretion of the PI (or designee) for one of the following reasons:

- Adverse Event [See section 7]
- Illness of a subject
- Subject non-compliance with protocol requirements
- Other, at the discretion of the investigator

In such cases, the subject will be withdrawn from further study participation. Data collected from withdrawn subjects will be analyzed and results will be retained for safety assessments. If available, data will be retained in analyses related to the objectives of the study unless there is a valid reason to believe that the data may be biased, incorrect, or confounded.

5. Materials and Methods

5.1 Materials and Equipment

- CareSens PRO GK BT meters
- Test strips (glucose/ β-ketone): at least 3 lots, at least 10 vials/packages
- CareSens PRO GK BT user's manual, and quick reference guide
- Control solutions: CareSens PRO (glucose) level A and level B and KetoSens (β-ketone) level A and B
- Spare batteries for meters
- A commercially approved SMBG meter, control solution(s), and user's manuals
- YSI 2300 STAT Plus glucose analyzer (YSI)YSI reagent and YSI supplies
- Randox Imola D-3-Hydroxybutyrate (Ketone) analyzer (Rx Imola)
- Randox reagent and Randox supplies
- Thermo-hygrometer
- Micro centrifuge

- Tenderlett™ or tenderlett-like single use lancing device (single device for each subject)
- Lancing devices and lancets (single device for each subject)
- Heparinized collection tubes
- Micro hematocrit tubes, sealant, micro hematocrit tube reader
- Syringes and needles
- Pipettes
- Alcohol wipes
- Gauze
- Band-Aids
- Eppendorf tubes
- Kim wipes
- Clorox Germicidal wipes (EPA#: 67619-12)
- Non-latex gloves
- Supplies to treat hypoglycemia
- Laboratory coats
- Safety goggles
- Biohazard Sharps container

5.2 Preparation and Administration

Study materials will be provided by the sponsor and shipped, or hand carried to the clinical site by the sponsor (or sponsor representative).

The applicable serial and lot numbers for materials and equipment should be recorded upon receipt.

Storage

The investigational device must be stored in an area that is secure and accessible to trained study staff only.

Dispensing of Study Investigational Device and Materials

Dispensation of the investigational device and study materials will be performed by the individuals responsible for conducting the study at the clinical site per the Delegation of Authority Log.

Return of Study Investigational Device and Materials

All study related materials will be collected by the sponsor (or sponsor representative) upon completion of the study.

Device Accountability

It shall be the sponsor's responsibility to ensure that an accurate record of investigational devices issued and returned is maintained.

All investigational devices will be retrieved at the conclusion of the study. The devices are assumed to be contaminated and will require to be handled as such (Standard precautions).

The devices will undergo disinfection per validated disinfection procedure prior to further analysis.

5.3 Preparatory Steps by the Trained Study Staff

Prior to first use, the meters will have times and dates preset to the time zone of use.

5.3.1 Labeling

The meters and strip lot vials/packages shall bear a label in accordance with Title 21 CFR 812.5 with the following information:

"CAUTION-Investigational device. Limited by United States law to investigational use."

5.3.2 Safety and Disinfection procedures

1) Only the trained study staff will perform disinfection procedures.
2) The meters will be used by multiple subjects in this study; therefore the meters must be disinfected between subjects.
   ① Open the cap of the Clorox Germicidal Wipes container and pull out 1 towelette and close the cap.
   ② Wipe the entire surface of the meter 3 times horizontally and 3 times vertically using one towelette to pre-clean blood and other body fluids.
   ③ Dispose of the used towelette in a trash bin.
   ④ Pull out 1 new towelette and wipe the entire surface of the meter 3 times horizontally and 3 times vertically using a new towelette to remove blood borne pathogens.
   ⑤ Dispose of the used towelette in a trash bin.
   ⑥ Allow exteriors to remain wet for 1 minute, and then wipe the meter using a Kim wipe.
3) The meters must also be disinfected after use by the trained study staff prior to shipment back to i-SENS and at any time a meter becomes contaminated with blood.
4) A new lancing device will be provided to each subject. At the end of the subject's visit, the lancing device will be discarded or given to the subject to keep.
5) The lancets used in the study are single-use and will be disposed of in a biohazard sharps container after use.
6) Trained study staff will wear disposable non-latex exam gloves during study procedures where the risk of transmission of disease is present. These procedures include performing blood glucose and β-ketone measurements, sample processing, and laboratory procedures involving blood and plasma

samples. Trained study staff in contact with the subjects will use a new pair of gloves for each subject and will change gloves between subjects.

7) Trained study staff will record all the meter results on the appropriate form. If the meter has turned off and results are difficult to see, the meter memory will be reviewed, and the results will be obtained from the meter memory.

8) Disposable bench top covers to cover the work areas where body fluids and risk of disease transmission are present must be changed between subjects. Hard surfaces will be disinfected between subjects.

9) A commercially approved device may be used for determining the subject's glucose/β-ketone level during the study as needed for safety reasons or to ascertain the glucose/β-ketone level for study reasons. The meter will be disinfected between subjects following the manufacturer's disinfecting instructions.

10) All the CareSens PRO GK BT meters used in the study will be tested using control solutions, Level A and B of CareSens Pro control solution for the glucose and level A and level B of KetoSens control solution for the β-ketone and recorded at the beginning and at the end of each clinical study day and at any time it is suspected that the meter and test strips are not working properly. If the results are out of range, troubleshooting will be performed (see the user's manual) and the control test will be repeated (a total of three (3) attempts per control test are allowed). If the meter tests are out of range after troubleshooting is complete, the meter will be removed from the study. The meters that failed control solution testing and were removed from the study will be recorded and reported. This will be documented on the Device Deficiency Form.

5.3.3 Temperature and Humidity Monitoring

The trained study staff will measure the temperature and humidity in the testing area(s) three times per day: prior to first subject of the day, halfway through the day, and after all subjects completed that day. This will be done using a thermometer/hygrometer supplied by the clinical site and the results will be recorded on the appropriate form.

5.4 Subject Evaluation Procedure

Each subject is provided with all the materials needed for blood glucose/β-ketone self-testing.

1) The subjects are given sufficient time to review the user's manual and the quick reference guide. No training is provided by the trained study staff.

   Note: The subjects should obtain their own fingertip capillary sample and perform a test using only the draft device labeling as instructions (user's manual and the quick reference guide). No other training or prompting should be provided to the subject and no assistance should be given from the trained study staff. The subjects should be sequestered in different stations that they cannot observe or be influenced by the testing technique of other subjects or trained study staff.

2) The subject washes his/her hands with warm water and soap, rinses and dries well.

3) The subject lances his/her finger using a lancing device and performs a blood glucose test with a glucose test strip on the meter. Three successful fingersticks/lances are allowed maximum (actual lances on the finger) to get a meter result. The lancet should be changed after each actual fingerstick. The trained study staff records the times of the meter tests, results, number of successful lances, and marks whether the subject completed all steps on the CRF.

4) Immediately within 5 minutes of the first evaluable meter reading, the trained study staff collects more blood for YSI and hematocrit measurement (approx. 300-350 µL) using a Tenderlett TM or tenderlett-like single use lancing device. A total of up to 3 additional lancings are allowed to collect blood in the collection tubes. Centrifuge blood within 10 minutes of sample collection and conduct YSI within 20 minutes of centrifugation. Maximum of 35 minutes to complete from evaluable meter result.After measuring blood glucose, the subject lances his/her finger and performs a β-ketone test with a β-ketone test strip on the same meter. The trained study staff /technician records the times of the meter tests, results, number of successful lances, and marks whether the subject completed all steps on the CRF.

   Note: The meter automatically recognizes the measuring strip when each strip is inserted. If the KetoSens strip is inserted, KETONE will appear on the result window.

5) Immediately within 5 minutes of the first evaluable meter reading, the trained study staff collects more blood for Rx Imola measurement (approx. 500~550 µL) using a Tenderlett TM or tenderlett-like single use lancing device. A total of up to 3 additional lancings are allowed to collect blood in the collection tubes. Centrifuge blood within 10 minutes from sample collection and conduct Rx Imola measurements within 20 minutes of centrifugation. Maximum of 35 minutes to complete from evaluable meter result.

6) Collect more blood and fill two micro hematocrit tubes for hematocrit test. The blood for hematocrit test can be taken from one of fingersticks already done.

   Note: The subject's hematocrit measurement should be within 20-60% in order for the result to be considered evaluable.

7) For any occurrence of meter errors, the subject should follow the instructions in the user's manual.

8) The time of each test, meter results, any occurrence of meter error codes is recorded by the trained study staff on CRF.

9) Each evaluable fingerstick meter results are compared to the capillary YSI results and Rx Imola results. (More than one comparator measurement results averaged for each sample)

   NOTE: If plasma cannot be measured immediately using YSI and Rx Imola within 15 mins after blood collection (in case of analyzer correction or device malfunction), store the plasma separated from the blood collection tube refrigerated.

For the study, multiple meters will be used and strips of at least 10 vials/packages over at least 3 lots will be used and rotated.

Table 1. Blood Testing Schematic

Fingerstick testing	
• Select strips for subject blood testing (Lot A, Lot B and Lot C rotated) • Record time of 1st test	
Glucose First lancing	**Subject test**
• Study Staff records time of test, result of glucose and if subject completed each step properly (check boxes on CRF), and if subject encountered any problems.	
Tenderlett or similar (Study Staff) stick	**For YSI testing**
• Immediately following meter test (≤5 minutes), study staff lances subject fingertip • Record time of lancing • Collect blood in Sodium Heparin tube (approx. 300-350 µL)	

• Centrifuge blood and record time (should be within 10 minutes of sample collection) • Test the sample and record time and result of glucose on CRF	
β-ketone Second lancing	**Subject test**
• Study Staff records time of test, result of β-ketone and if subject completed each step properly, and if subject encountered any problems.	
Tenderlett or similar (Study Staff) stick	**For Rx Imola testing**
• Immediately following meter test (≤5 minutes), study staff lances subject fingertip • Record time of lancing • Collect blood in Sodium Heparin tube (approx. 500 ~ 550μL) • Centrifuge blood and record time (should be within 10 minutes of sample collection) • Test the sample and record time and result of glucose on CRF	
Hematocrit Testing	
• Fill 2 micro hematocrit tubes and record time and result of hematocrit	
Record meter reading or meter error codes or subject errors as appropriate	

Note: Trained study staff should use a new pair of gloves for each subject and should change gloves between subjects.

5.5 System Check: Meter Control Tests

All the meters will be tested using control solutions, Level A and B of CareSens Pro control solution for the glucose and level A and level B of KetoSens control solution for the β-ketone at the beginning and end of each study day and the results will be recorded. If the results are out of range, troubleshooting will be performed (see the user's manual) and the test will be repeated (a total of three (3) attempts per control test are allowed). If the QC results are out of range after troubleshooting is complete, the meter will be removed from the study. The meters who failed control solution testing and were removed from the study will be reported. This will be documented on the Device Deficiency Form.

5.6 Operation of YSI Glucose Analyzer

5.6.1 YSI Control Tests

The YSI Analyzer auto calibrates every 5 samples or 15 minutes, whichever comes first. If the YSI fails to calibrate correctly, the trained study staff will troubleshoot the system and perform a new calibration. The glucose results for the blood samples analyzed previous to the failed calibration will be rejected.

For quality control testing on YSI Analyzer, the YSI control solutions and the 180 (2747) standard will be run at the beginning and end of the study day or at any time the YSI needs to be restarted. Each quality control solution is required to be within its stated range and 180 (2747) standard in order to continue to use the analyzer in the study. In addition, the calibration shift should not have a difference of more than 2% and the probe current should be less than 6 nA before using the YSI Analyzer. If not, the trained study staff/technician will troubleshoot the analyzer and repeat the quality control testing. If quality control solution testing continues to fall out of range, the glucose concentration values for the blood samples analyzed previous to the failed control solution results will be rejected.

The YSI will be maintained and operated according to the instructions in the manufacturer's operating manual. The YSI will be set up and appropriate maintenance performed before the study begins. The YSI clock will be set to 24-hour format using an accurate time device (such as site central clock or atomic clock).

Trained study staff will keep a maintenance log, including daily operational checks, maintenance, and membrane changes.

5.6.2 Glucose Testing of Subject Plasma Samples

YSI 2747 standard is run before each subject sample cycle, and it needs to be within the acceptable range.

The subject capillary plasma samples will be tested in duplicate. If the average of replicates 1&2 do not fall within range (within 4% for readingsabove100mg/dL or 4 mg/dL for samples less than or equal to 100mg/dL) the sample will be run a third time and the 2 average replicate readings that fall within range (4% for readings above 100mg/dL or 4 mg/dL for samples less than or equal to 100mg/dL) will be recorded as evaluable.

In general, duplicates will be averaged for each subject. If the sample is insufficient or missing and have only a single YSI value, it will be used as the YSI determination. If none of the 3 average replicates fall within (4% for readings above 100mg/dL or 4 mg/dL for samples less than or equal to 100mg/dL) will be recorded as non-evaluable. If the YSI autocals between subject sample duplicates, then another YSI 2747 standard will be run before completing the subject sample cycle.

5.7 Operation of Rx Imola β-ketone Measurement

5.7.1 Rx Imola Control

Rx Imola ketone analyzer will be calibrated using Ranbut, Ranbut Cal and distilled water at least once a week. Ranbut is prepared by putting 10 ml of R1a solution in R1b container. The solution can be used for one week after preparation. If FAIL is displayed during calibration, the trained study staff/technician will troubleshoot the system and perform a new calibration. If calibration continues to fail, reproduce the Ranbut solution and perform a new calibration. Rx Imola will also be calibrated when QC is failed or Ranbut used for sample measurements is replaced.

For quality control testing on Rx Imola analyzer, QC lv 2 and QC lv 3 of Rx control solution will be run at the beginning and end of the study or at any time the Rx Imola needs to be restarted. Each quality control solution is required to be within its stated range in order to continue to use the analyzer in the study. If quality control solution testing continues to fall out of range, the β-ketone concentration values for the blood samples analyzed previous to the failed control solution results will be rejected.

The Rx Imola will be maintained and operated according to the instructions in the manufacturer's operating manual. The Rx Imola will be set up and appropriate maintenance performed before the study begins.

Trained study staff will keep a maintenance log, including daily operational checks, maintenance, and calibration solution changes.

5.7.2 β-ketone Testing of Subject Plasma Samples

The subject capillary plasma sample will be tested in duplicate using Rx Imola analyzer. If the measured values differ by > 0.075 mmol/L at ketone < 1.5 mmol/L or > 5% at ketone ≥ 1.5 mmol/L, the sample will be run a third time and the 2 average replicate readings that fall within range will be recorded as evaluable. In general, duplicates will be averaged for each subject. If the sample is insufficient or missing and has only a single Rx Imola value, it will be used as the Rx Imola determination. If none of the 3

average replicates fall within > 0.075 mmol/L at ketone < 1.5 mmol/L or > 5% at ketone ≥ 1.5 mmol/L for readings, it will be recorded as non-evaluable.

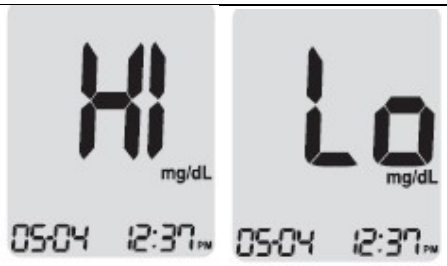
5.8 Subject Questionnaire and Other Information

After all subject testing has been completed, the study participants will be provided a sample meter that displays a hypoglycemia/hyperglycemia reading. Following this, they are asked to complete a usability questionnaire. The trained study staff will administer a questionnaire to the subjects and check for completeness of the subject's responses. In the event of a physical, visual or neurological impairment that makes it difficult for a subject to complete the questionnaire, a subjects' verbal responses will be accepted and recorded.

5.8.1 Questionnaire: Evaluation of Instructions for Use – User's Manual – Quick Reference Guide -ease of use of the system

The questionnaire will include statements for which a numerical score or rating will be provided by subjects (1=Very Difficult, 2=Fairly Difficult, 3= Neither Easy nor Difficult, 4=Fairly Easy, 5=Very Easy). The frequency distribution of the scores will be tabulated. In addition, the percent of subjects scoring 3 or more will be calculated for each statement along with exact 95% confidence intervals.

	Questions	Very Difficult	Fairly Difficult	Neither Easy nor Difficult	Fairly Easy	Very Easy
1	Understanding the overall instructions in the User's manual and Quick reference guide is…	1	2	3	4	5
2	Following the instructions in the User's manual on how to run a blood sugar and ketone test is…	1	2	3	4	5
3	Performing a blood test with this meter is…	1	2	3	4	5
4	Seeing and reading the meter display is…	1	2	3	4	5
5	Viewing my blood sugar and ketone test results on the meter is…	1	2	3	4	5
6	Handling the meter is…	1	2	3	4	5
7	Handling the test strips is…	1	2	3	4	5
8	Following the instructions on how to insert a test strip in the meter is…	1	2	3	4	5
9	Understanding the indicator of hypoglycemia/hyperglycemia is …	1	2	3	4	5

						
10	Understanding the error message displayed on the meter screen is…	1	2	3	4	5
11	Changing the test mode from blood sugar to ketone is…	1	2	3	4	5
12	Recognizing that the test mode has changed is…	1	2	3	4	5

Note 1 – If the subject does not experience an error message during his/her testing, an error message (e.g. Er1 – A used test strip was inserted) will be demonstrated by the trained study staff after the subject completes all fingerstick testing

5.8.2 Demographic, Diabetes History, Hematocrit, and Subject Questionnaire

Descriptive statistics for demographics, diabetes history, and subject questionnaires will be calculated. Histograms will be constructed where appropriate.

Hematocrit will be measured in duplicates and averaged for each subject where available. The subject's hematocrit measurement should be within 20-60% in order for the subject to be considered evaluable and be used for blood glucose measurement comparisons. The mean, median, minimum, maximum, and standard deviation, will be computed for average hematocrit determinations.

5.8.3 Data Evaluability

Blood glucose/β-ketone data will be considered not evaluable for the following reasons:

- Subjects without a hematocrit result or a hematocrit result outside of 20-60% range.
- The readings from the subject meter tests that the subject feels were incorrectly completed. (If the subject states that the test s/he performed was completed incorrectly, and repeats the test, then the new reading will be used as the subject's evaluable result.)
- Failure to lance the subject's fingertip using the Tenderlett device or Tenderlett-like device within 5 minutes of the subject's first fingerstick self- test.
- Failure to separate the plasma from the red blood cells within 10 minutes of Tenderlett blood collection.
- Failure to measure YSI/Rx Imola reading within 2 hours after separating plasma from the red blood cells.

6. Risk / Benefit Assessment

6.1 Biohazard to trained Study staff/technicians

The only risk to trained study staff conducting the study is the risk of transmission of blood borne pathogens from the handling of blood from study subjects of unknown health conditions. This risk is very small and is mitigated by the use of safety equipment such as gloves, eyewear and lab coats, and by following standard precautions when handling blood samples.

6.2 Biohazard to Subjects

This study presents minimal risk to the subject. To reduce the risk of blood borne pathogen transmission, each study subject will receive a disinfected meter and a disposable lancing device specific for their use only in the study. The subjects will receive a supply of disposable lancets for use with the meter. Tenderlett or tenderlett-like lancing devices are for single use only.

6.3 Risks and discomfort to subjects

This is a low-risk study. These risks are similar to those normally incurred when a person lances his or her finger with a sterile lancet and to having diabetes.

Hypoglycemia/Hyperglycemia or hyperketonemia

Since subjects may have diabetes, there is a risk they will experience a hypoglycemic/ hyperglycemic or hyperketonemia event during the time they are at the study site. If the study test result on the CareSens PRO GK BT indicates a low or high result, they should not use the result to make any therapeutic decisions as CareSens PRO GK BT is intended for investigational use only. They will be advised to continue to use their own meter to monitor their blood glucose and ketone.

Fingerstick

The fingerstick is performed using lancing devices. Pain, redness, bruising and discomfort may occur. Local irritation or infection, fainting, nerve injury or continued bleeding may occur, but this is unlikely. Subjects should be instructed to report such events to the investigator or trained study staff.

6.4 Benefits

There is no direct benefit from participating in this study. A potential benefit associated with the study may be the sense of well-being gained by contributing to the development of improved or new blood glucose monitoring systems, which may be beneficial to people with diabetes.

7. Adverse Event Management and Medical Device Deficiency Reporting

The procedures to be performed under this protocol are considered to be low risk.

During the clinical study, the clinical research site will determine if any adverse events or device deficiencies have occurred. The PI is responsible for reporting all Serious Adverse Events to the sponsor within 24 hours of learning of the event. All adverse events will be monitored until they are resolved or stable.

7.1 Definitions

Adverse Event (AE)

Any clinically significant undesirable experience (sign, symptom, illness, or other medical event) occurring in a patient that appears or worsens during a clinical study. A clinically significant event is any event (sign, symptom, lab/imaging abnormality, or diagnosis) that is noteworthy enough to merit documentation in standard medical records (e.g. History and Physical, Progress notes, clinic visit notes, etc.).

Adverse Device Effect (ADE)

An ADE is an adverse event (AE) that is related to or associated with the study device.

Serious Adverse Event (SAE)

A SAE is defined as an adverse event that is fatal, life-threatening, results in persistent or significant disability or incapacity, requires medical or surgical intervention to prevent permanent impairment or damage, or results in initial (in-patient) hospitalization or prolongation of hospitalization. Life-threatening is defined as the substantial risk of dying at the time of the adverse event (AE) or suspect that continued use of the device would result in the subject's death.

Serious Adverse Device Effect (SADE)

A SADE is a serious adverse event (SAE) that is related or associated with the study device.

Device Deficiency (DD)

A DD is inadequacy of a medical device with respect to its identity, quality, durability, reliability, usability, safety or performance.

7.2 Assessment of Adverse Event

7.2.1 Severity of Adverse Events

The following definitions for rating severity of adverse events may be used:

- Mild: Awareness of signs or symptoms, but easily tolerated; are of minor irritant type; causing no loss of time from normal activities; symptoms would not require medication or a medical evaluation; signs and symptoms are transient.
- Moderate: Discomfort severe enough to cause interference with usual activities, requiring treatment but not extended hospitalization or intensive care for the subject.

- Severe: Incapacitating, causing inability to do work or usual activities; signs and symptoms may be of systemic nature or require medical evaluation and/or treatment, requiring additional hospitalization or intensive care (prolonged hospitalization).

7.2.2 Relationship of Adverse Event to the Study Device

The principal investigator will categorize the relationship of the event to the investigational device as follows:

- Not related: AE is due to an underlying disease state or concomitant medication, or therapy not related to the study device.
- Probably not related: AE has minimum or no temporal relationship to the investigational device and/or a more likely alternative etiology exists.
- Possibly related: AE has a strong temporal relationship to the investigational device or surgical procedure and alternative etiology is equally or less likely compared to the potential relationship to the investigational device.
- Probably related: AE has a strong temporal relationship to the investigational device and another etiology is unlikely.
- Definitely Related: Causal relationship has been established and documented.

7.2.3 Anticipated Adverse Device Effects

The following events have been identified as possible adverse device (lancing) effects:

- Bleeding
- Bruising around the lancing site
- Pain or discomfort
- Redness
- Irritation
- Infection

7.2.4 Anticipated Adverse Events

The following events have been identified as anticipated adverse events:

- Hypo/Hyperglycemia
- High/Low hematocrit range (Normal hematocrit range: 45-52% for men and 37-48% for women other than during pregnancy)
- Syncope or near syncope

7.2.5 Unanticipated Adverse Device Effect (UADE)

Due to the low-risk nature of this device, an UADE is not expected to occur. However, an (UADE) is defined as any serious adverse effect on health or safety or any life-threatening problem or death caused by, or associated with the device, if that effect, problem, or death was not previously identified in nature, severity, or degree of incidence in the investigational plan, or any other unanticipated serious problem associated with the device that relates to the rights, safety, or welfare of subjects.

The principal investigator will determine the severity, or the extent of the event was classified as at a minimum, possibly related to the device, the event will be documented as an UADE. If the event is classified as an UADE, the investigator will submit to the IRB and Sponsor, a report of any unanticipated adverse device effect occurring during the investigation as soon as possible, and in no event later than 10 business days after the investigator learns of the effect.

For studies on products that have not yet been cleared for market, it is important that all adverse events be documented and included in the study file.

7.2.6 Events related to hypoglycemia and hyperglycemia

Hypoglycemia and hyperglycemia can be a common occurrence with subjects who have diabetes. If it is found that a subject has a hypoglycemic event (as determined by the PI), during a study visit, the subject will treat themselves per their usual routine with or without assistance by the site staff if applicable hypoglycemic and hyperglycemic events are considered anticipated and will not be recorded on the Adverse Event Form unless they become serious. Subjects who are hypo- or hyperglycemic may continue with fingerstick testing if they so choose, and if the PI determines that it is safe. Duration of the subject's study visit may be prolonged.

7.3 Procedure for Reporting an Adverse Event

Adverse events will be documented during this study by completing the Adverse Event Form. During the study visit, adverse events will be recorded by the trained study staff and evaluated. The nature of each event and date of onset, outcome, course, maximum intensity and action taken with respect to treatment should be established. Details of any corrective treatment should be recorded on the AE Form. The Investigator should follow-up on the status of subjects experiencing an ongoing adverse event until the event has been resolved, or until the condition has stabilized.

- The investigator or designee will notify the Study Manager or Monitor within 24 hours from awareness of any Serious Adverse Event that occurred during the study. The sponsor will promptly review all information relevant to the safety of the investigational device
- The investigator or designee must notify the sponsor and reviewing IRB by phone or fax of Unanticipated Serious Device-Related Adverse Events within 24 hours of learning of the event, followed by a written report within 10 working days after learning of the event.
- Upon receipt of a report of an UADE by the Study Manager or Monitor, the report will be immediately forwarded to i-SENS.
- The sponsor must report Unanticipated Adverse Device Effects (UADE) to the FDA, all participating investigators, and reviewing IRBs within 10 working days after the sponsor first receives notice of the event.

- Regardless of the above definitions, any additional adverse experience that the investigator considers serious, and/or of concern in relationship to the study must be documented and reported to the Sponsor's Study Manager or Monitor within 24 hours.

7.4 Procedure for Reporting a Device Deficiency

All device deficiencies related to the identity, quality, durability, reliability, safety or performance of an investigational medical device shall be documented throughout the clinical investigation and appropriately managed by the sponsor. Device deficiencies that did not lead to an adverse event but could have led to a medical occurrence

- if either suitable action had not been taken,
- if intervention had not been made, or
- if circumstances had been less fortunate,

shall be documented accordingly and reported to the sponsor.

All device deficiencies have to be reviewed and determined whether they could have led to an adverse effect (AE) and/or unexpected adverse device effect (UADE). All device deficiencies must be documented accordingly and reported to the sponsor. Device deficiencies that could have led to a serious AE and/or UADE, shall be reported to the sponsor within 24 hours of awareness.

7.5 Emergency contact details for reporting serious adverse events and serious adverse device effects.

Any SAE, including events resulting in death, due to any cause (related or unrelated to the study device), that may occur during a clinical study must be reported immediately (within 24 hours of awareness) to the sponsor via the Electronic Data Capture (EDC) system. If the EDC system is unavailable the site should notify the study manager.

8. Regulatory

8.1 Declaration of Helsinki

This study will be conducted in accordance with the Declaration of Helsinki. This set of ethical principles was created to ensure that the rights and safety of clinical trial subjects is the priority in all clinical research. All individuals who take part in the clinical trial are responsible for following the principles in the Declaration of Helsinki.

8.2 Investigational Review Board (IRB) Approval

An IRB must review this protocol, the ICF, and any other supporting study documents which impact subject safety, prior to starting the study. The clinical study site may not begin the study until the IRB has provided a written and dated approval letter that identifies the version/date of the protocol and ICF to the investigator. Any amendments to the protocol will need sponsor and IRB approval before continuing with the study.

The principal investigator and/or the sponsor will submit any reports or updates the IRB/EC may require during the course of the study.

Any reportable adverse events will be submitted to the IRB/EC by the principal investigator and/or designee.

8.3 Informed Consent Requirement

Each subject must provide informed consent before she/he can participate in the study. The informed consent process fully apprises the subjects of the risks and benefits to them and to society for participating in the study. The ICF will clearly state that designated trained study staff will be able to view the subject's medical records. The ICF will also state that i-SENS representatives may observe some subjects as part of study staff training, study monitoring or for troubleshooting problems with the investigational device.

If the subject understands and agrees to participate in the study, she/he will sign the ICF. All subjects will be given a copy of the signed and dated ICF. If a subject has a question about his/her rights, he/she may contact a member of the IRB at any time during or after study participation.

8.4 Study Documentation Procedures

All subject information and data will be documented directly on study worksheets and entered into the EDC. The investigator will keep study records for a minimum of five years.

8.5 Study Monitoring

The study will be monitored by CRO and sponsor. The Study Manager/Study Monitor prior to the study start will complete a monitoring plan. The Study Manager or designee will conduct a study initiation visit, at least 1 monitoring visit, and a close out visit (note: the latter two visits may occur within a continuous timeframe).

The CRO and sponsor may observe some of the subjects during their study visit as part of study monitoring. This will be done under supervision of the investigator. The CRO and sponsor will maintain subject confidentiality and will not interfere with the rights and safety of human subjects, or bias study conduct.

8.6 Investigator's Report of Study Closure

The Study Monitor will send a letter to the site informing them that the study is closed. The study will be considered closed when all the required data has been acquired for data analysis.

The Investigator or designee will submit a report summarizing subject disposition and other study details, as appropriate, to the Study Manager and the reviewing IRB at completion of the study. This report will be completed within 3 months of the study closure date.

9. Acceptance Criteria

Glucose

95% of all glucose results in the study should be within ±15% of the comparator results across the entire claimed measuring range of the device and 99% of all SMBG results within ±20% of the comparator results across the entire claimed measuring range of the device.

β-ketone

95% of the individual ketone results in this study should be within ±0.3mmol/L at concentration < 1.5mmol/L and within ±20% at concentration ≥ 1.5mmol/L of the comparator results.

10. Data Presentation and Data Analysis Method

For data analysis, a linear regression study will be performed using Passing-Bablok analysis with confidence bands for the regression line (95%). Passing-Bablok analysis is a robust analysis method for it takes account of extreme data points, and it does not assume a normally distributed measurement error, therefore is reliable when testing equality of biometrical evaluation. Accuracy will be determined based on:

PD (Percent Difference) = 100 * |T-R| / R ≤ 15 %.

When T= device result and R= Laboratory reference method result.

Also a bias plot will be produced and the accuracy will be presented in an error interval tables.

Table 2. Summary of data within specified mg/dL of the comparator method for glucose concentrations across the entire range

Within ±5%	Within ±10%	Within ±15%	Within ±20%
X/Y (%)	X/Y (%)	X/Y (%)	X/Y (%)

Table 3. Summary of data within specified mmol/L of the comparator method for β-ketone concentrations across the entire range

For β-ketone concentration < 1.5 mmol/L		
Within ±0.15 mmol/L	Within ±0.225 mmol/L	Within ±0.3 mmol/L
X/Y (%)	X/Y (%)	X/Y (%)

For β-ketone concentration ≥ 1.5 mmol/L		
Within ±10%	Within ±15%	Within ±20%
X/Y (%)	X/Y (%)	X/Y (%)

Descriptive statistics for demographics, diabetes history, and subject questionnaires will be calculated. Histograms will be constructed where appropriate.

In addition, the number of capillary samples with concentrations <80 and ≥250 mg/dL for glucose and ≥ 1.5 mmol/L for ketone will be reported.

Any missing, unused and spurious data will be noted with justifiable reasons (e.g. missing sample)

REFERENCE

[1] FDA Self-Monitoring Blood Glucose Test Systems for Over-the-Counter Use, (2020)

[2] CLSI EP9-A3 (Measurement Procedure Comparison and Bias Estimation Using Patient Samples; Approved Guideline-Third Edition.

[3] NCCLS Publication NRSCL, I-A Development of Definitive Methods for the National Reference System for the Clinical Laboratory. Approved Guideline; National Committee for Clinical Laboratory Standards: Wayne, PA (1991).

[4] White, VE et al. The Accurate Determination of Serum Glucose by Isotope Dilution Mass Spectrometry – Two Methods: Biomed, Mass Spectrum. Vol 9 pp. 395-405 (1982)

[6] Bablok, W. and Passing, H. (1985). Application of statistical procedures in analytical instrument testing. *Journal of Automatic Chemistry*. Vol.7, No.2 pp.74-79.