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|---|---|
| Title: | Pulse oximeter accuracy during stable hypoxia plateaus |
| Device Names: | Nihon Kohden Pulse oximeter and sensors |
| Regulatory Sponsor: | Nihon Kohden Corporation |
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# Contents

**Abbreviations and Definitions of Terms**

| | |
|---|---|
| $SpO_2$ | Peripheral capillary oxygen saturation |
| $SaO_2$ | Arterial oxygen saturation |
| FDA | Food and Drug Administration |
| ISO | International Organization for Standardization |
| MST | Monk Skin Tone |
| ITA | Individual Topology Angle |
| BMI | Body Mass Index |
| CPAP | Continuous Positive Airway Pressure |
| COHb | Carboxyhemoglobin |
| MetHb | Methemoglobin |
| ctHb | Total hemoglobin concentration |
| ECG | Electrocardiogram |
| CRF | Case Report Form |
| VSRG | Vital Sign Research Group |
| PI | Principal Investigator |
| IRB | Institutional Review Board |
| SaDE | Serious Adverse Device Effect |
| UADE | Unanticipated Adverse Device Effect |
| Rms | Root mean square |
| ABL | Radiometer ABL |
| DUT | Device Under Test |
| NK | Nihon Kohden |

**Aims**

This study aims to test the accuracy of Nihon Kohden pulse oximeters in range of arterial $HbO_2$ saturations from 100 down to 70%. This is done by comparing the pulse oximeter reading during a brief, steady-state hypoxia plateaus with a gold-standard measurement of blood oxyhemoglobin saturation, i.e., an arterial blood sample processed in a multi-wavelength hem oximeter. This study is designed in accordance with ISO and FDA regulatory standards. Nihon Kohden may submit the data obtained to the FDA for device clearance.

**Reference**

- ISO 80601-2-6:2024(E). DIS
- ISO 14155:2020
- Pulse Oximeters for Medical Purposes - Non-Clinical and Clinical Performance Testing, Labeling, and Premarket Submission Recommendations (FDA Guidance draft)

**Subjects**

Enrollment

At least 48 subjects (up to 56 subjects) will be enrolled.
According to ISO/DIS 80601-2-61:2024, it is required that 25% of all subjects be allocated to each MST category (1–4, 5–7, and 8–10). To ensure at least 24 subjects in each category, the study is designed to enroll a total of at least 96 subjects. We aim for a total of 98 subjects by combining 50 cases from previous research with 48 new cases. To account for an anticipated dropout rate of approximately 15%, we plan to recruit 56 subjects for the current study.

This study does not include socially vulnerable individuals as study subjects.

Clinical Validation Study

$SpO_2$ data

Subjects will have an average of 20 +/-4 $SaO_2$-$SpO_2$ data pair with a maximum range of 16-30 data points and 90% of participants will have at least one usable data point below 85% and at least 69% of subjects will have one usable data pair between 70-80%.

$SaO_2$ data

When two ABLs are determined to be valid, the $SaO_2$ reference value is the average of the $SaO_2$ measurements of the two ABLs. The validity of ABL measurements is determined by the following procedure:

- Check if the difference between two ABL readings is within 2%
- If the difference between two ABL readings is within 2%, the readings of two ABLs are valid.
- Readings of two ABL differed by more than 2% saturation will be excluded from the analysis.
- If only one ABL returns the value, the value will be assumed to be correct.

The skin tone distribution of subjects will be divided into 3 cohorts based on the Monk Skin Tone scale and ITA (Individual Topology Angle) determined at the dorsal distal phalanx.

1) At least 30% of subjects will be Monk Skin Tone (MST) 1-4 with an ITA less than 30
2) At least 30% of subjects will be MST 5-7 and have an ITA between -30 and 30

3) At least 30% of subjects will be MST 8-10 with an ITA less than -30 (with one half of MST 8-10 subjects having an ITA of less than -50)
4) In each MST group, at least 40% of participants are male, and at least 40% of participants are female.

At least 33% of each sex will be included in the study.

Inclusion Criteria
1. The subject is male or female, aged 18 to 50.
2. The subject is in good general health with no evidence of any medical problems.
3. The subject is fluent in both written and spoken English.
4. The subject has provided informed consent and is willing to comply with the study procedures.

Exclusion criteria:
1. The subject has a BMI above 40
2. The subject has blood pressure above 160 systolic or 95 diastolic
3. The subject has a known history of heart disease, lung disease, kidney or liver disease.
4. Diagnosis of asthma, sleep apnea, or use of CPAP.
5. Subject has diabetes.
6. Subject has a clotting disorder.
7. The subject has a hemoglobinopathy or history of anemia, per subject report or the first blood sample, that in the opinion of the investigator, would make them unsuitable for study participation. (COHb<3%, MetHb＜2%, ctHb＞10 g/dL).
8. The subject has any other serious systemic illness.
9. The subject has smoked in the last 12 hours.
10. Any injury, deformity, or abnormality at the sensor sites that in the opinion of the investigators would interfere with the sensors working correctly.
11. The subject has a history of fainting or vasovagal response.
12. The subject has a history of sensitivity to local anesthesia.
13. The subject has a diagnosis of Raynaud's disease.
14. The subject has unacceptable collateral circulation based on exam by the investigator (Allen's test).
15. The subject is pregnant, lactating or trying to get pregnant.
16. Subjects who have uneven skin tones at the forehead or the sensor site.
17. The subject is unable or unwilling to provide informed consent or is unable or unwilling to comply with study procedures.
18. The subject has any other condition, which in the opinion of the investigators would make them unsuitable for the study.

**Investigational Device**
This study is to evaluate the performance of the following pulse oximeter sensors shown in Table 1.
Table 1. Study devices

| Device name | Model name |
|---|---|
| Pulse oximeter | OLV-4202 |
| $SpO_2$ probe | TL-271T3 |
| $SpO_2$ probe | TL-273T3 |
| $SpO_2$ probe | TL-220T |

| | |
|---|---|
| $SpO_2$ probe | TL-631T |
| $SpO_2$ probe | TL-201T_BASE1 |
| $SpO_2$ probe | TL-201T_BASE3 |
| $SpO_2$ Connection Cord | JL-400T |

Sensor Placement
Table 2 and 3 shows how the sensors will be placed on the study subjects.

Table 2. Sensor placement on the right-hand side

| Probe | Subject 1, 4, 7, 10, 13, 16, 19, 22, 25, 28, 31, 34, 37, 40, 43, 46, 49, 52, 55 | Subject 2, 5, 8, 11, 14, 17, 20, 23, 26, 29, 32, 35, 38, 41, 44, 47, 50, 53, 56 | Subject 3, 6, 9, 12, 15, 18, 21, 24, 27, 30, 33, 36, 39, 42, 45, 48, 51,54 |
|---|---|---|---|
| TL-201T_BASE1 | Index | Middle | Ring |
| TL-201T_BASE3 | Middle | Ring | Index |
| TL-220T | Ring | Index | Middle |

Table 3. Sensor placement on the left-hand side

| Probe | Subject 1, 4, 7, 10, 13, 16, 19, 22, 25, 28, 31, 34, 37, 40, 43, 46, 49, 52, 55 | Subject 2, 5, 8, 11, 14, 17, 20, 23, 26, 29, 32, 35, 38, 41, 44, 47, 50, 53, 56 | Subject 3, 6, 9, 12, 15, 18, 21, 24, 27, 30, 33, 36, 39, 42, 45, 48, 51, 54 |
|---|---|---|---|
| TL-271T3 | Index | Middle | Ring |
| TL-273T3 | Middle | Ring | Index |
| TL-631T3 | Ring | Index | Middle |

Co-oximeter:
3 ABL-90 multi-wavelength oximeter

ECG monitor
A 3-lead ECG will be taken on each subject with the sponsor Control instrument. The electrodes for the sponsor Control instrument will be the L-150X disposable ECG electrodes.

Safety of the investigational device
Safety tests have been conducted for the probe (TL-201T-BASE1, TL-201T_BASE3, TL-631T3, TL-220T), and the validity of the device's performance has been confirmed. The sensor probes (TL-271T3) have been approved in Japan and by the FDA in the US for measuring pulse oximetry. In addition, The OLV-4200 series has been approved in Japan as a pulse oximeter. Based on the above, we consider that there are no issues with the use of the investigational device and related equipment in this study.

Device Accountability
The investigational device used in the study is a single monitor identifiable by its serial number. The disposable pulse oximeter probes are designed for single-patient use, and the lot number will be recorded on each data collection form.

**Procedures**
Subject skin tone will be measured and recorded by a calibrated colorimeter at the forehead, inner bicep and the are closest to the measurement site of the pulse oximeter. MST and ITA scores shall be determined for each subject.

Any nail bed coverings such as acrylic nails, gel-based manicures, nail jewelry, nail polish or long nails will be removed prior to testing.

A local anesthetic is injected around the radial artery, and a 22-gauge catheter is inserted in the radial artery. Pulse oximeters/device under test will be attached to the fingers, Reference oximeter probes may be placed on non-testing fingers, these probes include a Nonin 7500 and Masimo-Rad 97. ECG electrodes will be placed on the subject to monitor the subject's heart rate. A quality control check of ECG monitor is performed before use. Sensors under test placement and serial numbers will be documented for each subject. Subjects will be placed in a seated semi-reclined position. Subjects will breathe a nitrogen-air-carbon dioxide mixture to produce the desired level of hypoxemia. Stable, safe and controlled hypoxia is obtained by breath-by-breath analysis of respiratory gas using an CWE gas analyzer that permits the inspired gas mixture to be adjusted to achieve the desired degree of blood oxygen saturation.

We will reduce subjects $SpO_2$ to about the 90s, 80s and 70s, collecting four samples in each plateau where a blood sampling is taken, and the DUT measurement will be recorded simultaneously. This equals 12 blood samples; we will do this twice to achieve 24 data pair samples, for a total of at least 960 data pairs. Before desaturation, a blood sample is taken to ensure the subject meets the inclusion and exclusion criteria equaling 25 blood samples. At each plateau (90s, 80s, and 70s), in order to ensure that the arterial blood sample measured is from a stable state, a discarded blood sample (which will not be analyzed) is collected before the first blood sampling. Each level of saturation is held for up to 3 minutes. Arterial blood samples are obtained from the radial artery catheter at least 20 seconds apart when $SpO_2$ levels are stable. Before sampling, the gas operator will keep the subject at a stable blood oxygen level for 45 seconds to ensure that the test pulse oximeter readings are stable. During the test, a stable plateau is defined as a change in $SpO_2$ levels being no greater than 2%. $SaO_2$ readings will also be reviewed during the test to determine the stability of the plateau. $SaO_2$ will be measured using three sets of co-oximeters (ABL-90 multi-wavelength oximeter) that have undergone quality control and calibration by VSRG prior to use. The operator then changes the inspired air concentration to attain the next desired level of hypoxia. This takes about 10-15 minutes. Each desaturation run is terminated by a breath of 100% $O_2$ followed by room air. Two desaturation runs together enable obtaining a total of 20-25 blood samples. Up to 30 samples may be obtained from each subject. A self-calibrated Radiometer ABL-90 multi-wavelength oximeter determines the saturation of each arterial blood sample. The target saturation levels shall satisfy ISO and FDA standards for testing, which is 70% to 100%, with an approximate even distribution of samples in each of the decadal ranges of 70%-80%, 80%-90%, and above 90%. Each Monk Skin Tone and ITA cohorts shall contribute approximately equal number of data points in each $SaO_2$ decade range.

The study takes about an 1 hour and 15 minutes of each subject's time. Total expected duration of the study is 10 days.

If the subject attests to being a smoker, the subject will be asked to abstain from smoking for 12 hours prior to the study. If the subject has a carboxyhemoglobin above 3% after taking the first blood sample the subject may be given 100% oxygen for about 20 minutes to lower their carboxyhemoglobin levels.

**Statistics**

The number of subjects and comparisons (paired-pulse oximeter readings and arterial saturation values) is determined by current FDA and ISO guidance requirements [2]. In this type of study, some subjects may drop out; some readings can be lost due to motion or other interference, and occasionally, some may not consent. During analysis, plateau stability will be defined as a no-greater-than change of 2.% $SaO_2$ between blood samples.

The following demographic data will be collected on the subjects:

- Gender (male, female, other)
- Age
- Skin tone by Monk score determined by 3 observers. In the case of disagreement, when 2 of 3 values are the same, use that value. If there is disagreement between all observers, the middle value will be used.
- Height (cm)
- Weight (kg)
- BMI
- Dominant hand (left or right)
- ITA / Monk skin tone values at the device under test site, forehead and inner bicep
- Race
- Ethnicity
- Thickness of the measurement site (finger)

The following CRFs will be used to record data and information:

- Subject demographics CRF: Subject information will be recorded.
- Adverse event CRF: An adverse event related or not related to the test devices will be recorded.
- Subject withdrawal CRF: Any subject withdrawal after the subject signs informed consent will be recorded.
- Device deficiency CRF: A deficiency occurred to the test devices will be recorded.
- Protocol deviation CRF: Any deviation from the defined procedures as outlined in the protocol including mistake in recording of blood draw time will be recorded.

**Data Analysis**

Endpoints are ARMs and Bias.

The blood analysis data are provided in all cases, including the $SaO_2$, MetHb, COHb and Hgb concentration. After all subjects' data have been collected, data management and statical analysis will be conducted by VSRG . ABL devices will be set to display the upper limit at 100%. The $SaO_2$ readings will not be provided to the sponsor until the device under test readings are provided to Vital Signs Research Group. The data analysis report will follow ISO 80601-2-61:2024. A summary of the data analysis is as follows:

- A Table of the oximeter readings versus corresponding blood $SaO_2$ values for each data pair and each cohort along with heart rate, heart rate from the device under test, assessment for stability for each arterial sample, all removed data points and justification, and perfusion readings from the device under test.
- A modified Bland-Altman plot for each instrument or instrument/probe combination of the bias between the oximeter reading and the $SaO_2$ measured by the hemoximeter.
- A bland Altman plot of the hemoximeter vs the device under tests readings
- Linear regression equations and plots for the bias of each instrument across the range of 70% to 100% $SaO_2$
- Tables of the root mean square error or bias, its standard deviation, standard error, 95% confidence interval, maximum and minimum and root mean square error, all computed both overall and in each decadal range tested.
- A table of the demographics of the subject population.
- For each subject a mean bias will be calculated in the ranges of 70%-85% and 85% to 100%.
- A differential bias in the ranges of 70% to 85% and 85% to 100% will be reported by regressing each subject's individual mean bias in each of the two $SaO_2$ ranges vs ITA.
- A graphical representation of the regression to determine differential bias will be provided along with a table.
- Using the linear regressions, estimate the differential bias between the values at an ITA of -50 and +50.
- Tables of the root mean square error or bias, its standard deviation, standard error, 95% confidence interval, maximum and minimum and root mean square error, all computed both overall and in each decadal range tested for each MST group.
- Tables of the root mean square error or bias, its standard deviation, standard error, 95% confidence interval, maximum and minimum and root mean square error, all computed both overall and in each decadal range tested for each sex.

$SpO_2$ data will be reduced using the ARMS method and accuracy will be reported as the ARMS deviation between the co-oximeter and the test instruments for all points gathered from each sensor.

$$A_{rms} = \sqrt{\frac{\sum_{i=1}^{n} (SpO_{2i} - S_{Ri})^2}{n}}$$

The analysis is limited to participants who complied entirely with the study protocol, including fulfilling all inclusion and exclusion criteria, undergoing the specified procedures, and completing all required assessments.

Success criteria is as follows:

| Primary outcome measure | |
|---|---|
| $SpO_2$ accuracy | Rms of within ±3 [%$SpO_2$] (70 to 100 [%]) |
| Secondary outcome measure | |
| $SpO_2$ accuracy | Rms of within ±2 [%$SpO_2$] (80 to 100 [%])<br>Rms of within ±3 [%$SpO_2$] (70 to 80 [%]) |

Missing data will not be supplemented. If missing data occurs, the reason must be documented. The number of incomplete pairs and ratio of incomplete pairs will be calculated.

Data that meets the following conditions will be also thrown out:

- The CO-oximeter experienced error conditions
- Two CO-oximeter readings differed by more than 2% saturation
- Data points from CO-oximeter at a plateau which differ by more than 2% from the average of the others
- Data points from CO-oximeter which are the only ones at a plateau

Demographic characteristics will be summarized using standard descriptive summaries (e.g. means and standard deviations for continuous variables such as age and percentages for categorical variables such as gender).

**Subject Safety**

Pulse oximeters are considered non-significant risk medical devices. The LED light energy utilized in typical test measurements is within the same range as other cleared marketed devices and introduces no further risks. An LED light emits light that passes through the tissue. A light detector then measures how much light was absorbed by the tissue. Based on the absorbance ratio of different light wavelengths, the device calculates the oxygen saturation.

**Risks and Benefits**

Breathing a very low oxygen mixture may cause dizziness and might cause loss of consciousness for a few seconds. It may make one feel very short of breath during the test and for a few seconds afterwards. This study will not seek to reach saturations below 70%. Hypoxia may cause tachycardia and increased blood pressure during the test. A much more severe and prolonged lack of oxygen could cause brain injury or death, but the duration and depth of hypoxia are limited by the test protocol to short intervals. The needle catheter used to take blood may hurt when inserted despite local anesthesia, and there may be a black and blue spot afterward. A vasovagal response due to the catheter placement is possible. It is remotely possible the artery might be damaged or clot or a tendon sheath near it be injured by the needle, resulting in some soreness. These risks are unlikely in healthy human subjects with an arterial line for such a short duration. There are also risks of burns and skin problems from poor blood circulation.

Subjects will not receive any direct personal benefit from participation in this study.

**Risk Mitigations**

Subjects are all monitored with accurate FDA approved reference oximeters and continuous end-tidal gas analysis to prevent the risk of more profound hypoxia than desired. The subject's heart rate and blood pressure will be monitored throughout the study. Investigators are experienced anesthesiologists adept at assessing breathing and in maintaining appropriate airway conditions. The lab has all resuscitation equipment immediately available.

A risk of burns and skin problems due to poor blood circulation can be avoided by not placing $SpO_2$ probes too tightly.

**Informed Consent**

Written informed consent is obtained before any study procedures. In discussions with the study coordinator before the day of the study, potential subjects will be offered the consent form to review. On the day of the study subjects are given the consent form which they read and sign if they wish to participate. A study doctor is present to answer questions.

Only subjects clearly able to understand and read English will be enrolled. Subjects will be asked if they have any questions and are told they can withdraw at any time.

**Adverse event reporting**
Adverse events and serious adverse events will be assessed throughout the study by the PI, or research nurse designated by the PI. If any adverse event occurs during the study, appropriate medical treatment will be provided to the subject by a medical staff at the laboratory. Nihon Kohden may suspend or prematurely terminate the study when there are serious violations and deviations from the GCP which could adversely affect subject's safety and correct data recording.

Adverse Effects will be collected throughout the study on the source document worksheet and reported on the CRF. Information reported on the CRF will include:

- Adverse Effect diagnosis term to be determined by investigator.
- A detailed description of the event.
- The date of event onset.
- The relatedness of the event to the device as determined by investigator.
- Seriousness.
- Actions taken as a result of the event.
- The outcome of the event.
- The date the investigator was first informed of the event.

Unanticipated adverse device effect (UADE) and serious adverse device effects (SADEs) will be reported to the sponsor and IRB, as applicable, as soon as the PI becomes aware, but in no event later than 10 working days after the investigator first learns of the effect.

**Device Deficiencies**
All device deficiencies related to the investigational device will be documented throughout the duration of the clinical investigation. Information reported on the CRF will include:

- Date device deficiency discovered
- Date device deficiency occurred
- Description of the device deficiency
- Date of final observation
- A device deficiency occurred during use on the subject.
- An adverse event related to device deficiency occurred.
- Description of the action taken

**Monitoring**
It is the responsibility of the sponsor to ensure proper monitoring of the study per regulations. Appropriately trained personnel or delegates appointed by the sponsor will perform study monitoring at the study center to ensure that the study is conducted in accordance with the protocol, the Clinical Trial Agreement, and applicable regulatory requirements. The sponsor must, therefore, be allowed access to the subject's records as requested per the Informed Consent Form and Clinical Trial Agreement.

Monitoring visits will be conducted throughout the clinical study. Remote monitoring is acceptable.

The frequency of monitoring visits will occur based on subject enrollment for each test, duration of the study, study compliance, findings from previous monitoring visits and any suspected inconsistency in data that requires investigation. Regulatory documents (e.g., Informed Consent Form, IRB/EC approval letters and Clinical Trial Agreements, etc.) will be reviewed at the study center.

Monitoring visits may be conducted periodically to assess site study progress, the investigator's adherence to the protocol, and regulatory compliance, including but not limited to IRB review and approval of the study, maintenance of records and reports, and review of source documents against subject CRFs. Monitors facilitate site regulatory and study compliance by identifying findings of non-compliance and communicating those findings along with recommendations for preventative/corrective actions to site personnel. This may be done in collaboration with the study management and the local field personnel, if available. Communication with the site personnel occurs during and after the visit via a written follow-up letter. Monitors may work with study personnel to determine appropriate corrective action recommendations and to identify trends within the study or at a particular center. Study closure visits will be conducted via telephone, letter, or on-site at each enrolling study center.

**Termination of the Study**

The test will be discontinued when a subject requests to withdraw from the study, or the PI will terminate the desaturation procedure and/or a subject's participation in the study because of the reasons including: 1) the subject experiences unexpected oxygen desaturation or 2) the subject loses consciousness.

If the principal investigator or sponsor determines that a significant violation of ISO 14155:2020, this protocol, or continued non-compliance or breach of the clinical investigation agreement constitutes a serious impediment to the proper conduct of the clinical investigation, the trial will be suspended. Such cases include:

1. Concerns regarding the safety of the subjects
2. Inability to collect appropriate data
3. Discovery of significant falsification in records or reports
4. Major deviations or non-compliance with the clinical investigation protocol (excluding cases where it is necessary to avoid imminent danger to the subject or for other unavoidable medical reasons)

Either the principal investigator or the sponsor will notify the ethics committee of the suspension. The investigator will promptly inform the subjects of the suspension and take appropriate medical and other necessary measures.

**Data Storage**
Identifiable subject information is always stored securely following all applicable rules and regulations. Consent forms and other study related documents are retained following VSRG data retentions policy.

**Amendments to the Protocol**
Any amendments or revisions to the protocol shall be made in accordance with the Nihon Kohden Standard Operating Procedure "NKC03010-180038: Preparation and Revision of Clinical Investigation Plans and Case Report Forms". Such changes require prior approval from the Ethics Committee. The study shall not proceed under the amended plan until such approval has been obtained.

**Statement of Compliance**
This study will be conducted in compliance with the following:

- ISO 80601-2-6:2024(E). DIS
- ISO 14155:2020
- Pulse Oximeters for Medical Purposes - Non-Clinical and Clinical Performance Testing, Labeling, and Premarket Submission Recommendations (FDA Guidance draft)
- Declaration of Helsinki (2024 revision)