Clinical Study Protocol

Feasibility Study for VCool™ Intranasal Cooling System in Healthy Volunteers

Sponsor:	NeuroIntact, Inc. 1100 Wicomico Street, Suite 330 Baltimore, MD 21230
Protocol Number:	00080424
Version:	2.0
Date:	25 November 2025
Device Name:	VCool™ Intranasal Cooling System
Development Phase:	Pilot Study

This clinical study will be conducted according to the protocol and in compliance with ISO 14155:2020, Clinical investigation of medical devices for human subjects, good clinical practice (GCP); 21 CFR 812 Investigational Device Exemptions, ICH Harmonized Guideline GCP E6(R3) and with other applicable regulatory requirements.

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TABLE OF ABBREVIATIONS

ADE	Adverse Device Effect
AE	Adverse Event
BSAS	Bedside Shivering Assessment Scale
°C	Degrees Celsius
CFR	Code of Federal Regulations
CRF	Case Report Form
CT	Computed Tomography
FDA	Food and Drug Administration
GCP	Good Clinical Practice
ICF	Informed Consent Form
ICH	International Conference on Harmonization
ID	Identification
IDE	Investigational Device Exemption
IFU	Instructions For Use
IRB	Institutional Review Board
ISO	International Organization for Standardization
MAE	Major Adverse Event
PI	Principal Investigator
SADE	Serious Adverse Device Effect
SAE	Serious Adverse Event
TEAE	Treatment Emergent Adverse Event
TTM	Targeted Temperature Management
UADE	Unanticipated Adverse Device Effect

KEY ROLES AND RESPONSIBILITIES

Contact Information

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PROTOCOL APPROVAL PAGE

Protocol Title	Feasibility Study for VCool Intranasal Cooling System in Healthy Volunteers
Protocol Number	00080424
Protocol Version	2.0
Date of Issue	01 August 2024
Sponsor Name	NeuroIntact, Inc.
Address	1100 Wicomico Street, Suite 330 Baltimore, MD 21230

This clinical study protocol was subjected to critical review. The information it contains is consistent with current knowledge of the risks and benefits of the investigational product, as well as with the moral, ethical, and scientific principles governing clinical research and will be conducted according to the protocol and in compliance with ISO 14155:2020, Clinical investigation of medical devices for human subjects, good clinical practice; 21 CFR 812 Investigational Device Exemptions, ICH Harmonized Guideline Good Clinical Practice (GCP) E6(R3) and with other applicable regulatory requirements.

Sponsor:

Name: Bryan Nicholson

Title: President & CEO

Signature: _____

Date: _____

INVESTIGATOR PROTOCOL AGREEMENT

Protocol Title	Feasibility Study for VCool Intranasal Cooling System in Healthy Volunteers
Protocol Number, Version	00080424, Version 2.0

By my signature, I

1. Agree to conduct the study in accordance with the relevant, current protocol and will only deviate from the protocol when necessary to protect the safety, rights, or welfare of the participant.
2. Agree to personally conduct or supervise the described investigation. While the PI may delegate one or more functions to an associate or sub-investigator, the PI retains overall responsibility for proper conduct of the study.
3. Agree to inform any patients that the study product is being used for investigational purposes and I will ensure that the requirements relating to obtaining informed consent in 21 CFR Part 50 and Institutional Review Board (IRB) review and approval in 21 CFR Part 56 are met.
4. Agree to report to the Sponsor adverse experiences that occur during the investigation in accordance with 21 CFR 812.150. I have read and understand potential risks and side effects of the study product.
5. Agree to ensure all associates, colleagues, and employees assisting in the conduct of the study are informed about their obligations in meeting the above commitments.
6. Agree to maintain adequate and accurate records.
7. Ensure that an IRB that complies with the requirements of 21 CFR Part 56 will be responsible for the initial and continuing review and approval of the clinical investigation. I agree to promptly report to the IRB all changes in the research activity and all unanticipated problems involving risks to human participants and others. I will not make any changes in the research protocol without consent from the Sponsor and will not institute those changes in the research protocol until after approved by the Sponsor and IRB, except where necessary to eliminate apparent immediate hazards to human participants.
8. Agree to comply with all other requirements regarding the obligations of clinical investigators and all other pertinent requirements in 21 CFR Part 812.
9. Agree to participate in an appropriate training program as part of the study initiation.
10. Agree to allow staff of the Sponsor and its authorized representatives, as well as representatives from regulatory agencies, to review, inspect and copy any documents pertaining to this clinical investigation.

This document contains confidential information belonging to the Sponsor, and therefore may not be disclosed to any other person or entity without the prior written permission of the Sponsor unless such disclosure is required by law or regulation or required for the execution of the PI's responsibilities pertaining to this study.

Investigator Signature
I have read and understand the contents of the clinical protocol including this Statement of Investigator Compliance. I agree to follow and abide by the guidelines set forth in this document.

_____
Principal Investigator Name (print)

_____ _____

Principal Investigator Signature Date

1 PROTOCOL SYNOPSIS

Protocol Title:	Feasibility Study for VCool Intranasal Cooling System in Healthy Volunteers
Protocol Number:	00080424
Version:	2.0
Date:	01 August 2024
Development Phase:	Pilot Study
Sponsor:	NeuroIntact, Inc. 1100 Wicomico Street, Suite 330 Baltimore, MD 21230
Study Objective:	The objective of this study is to evaluate the performance and safety of the VCool Intranasal Cooling System in healthy adult volunteers. The study is designed to gather preliminary data regarding air flow rates and time required to reduce core body temperature to 35.5℃ and maintain the temperature between 35℃ and 36℃ for one hour after cooling.
Number of Sites / Number of Participants:	The study will be conducted at one site in Maryland. A total of 10 participants need to complete the study.
Primary Endpoints:	1. To confirm the VCool System can lower core body temperature to 35.5℃ within 2 hours. 2. To confirm the VCool System can maintain a core body temperature between 35℃ and 36℃ for one hour after cooling. 3. To demonstrate the differential impact of VCOOL system between core body and brain temperature
Secondary Endpoint:	1. To assess the shivering experienced by participants based on the 4-point Bedside Shivering Assessment Scale (BSAS).
Safety Evaluation:	All treatment emergent adverse events (TEAEs) reported by participants will be tabulated. Each adverse event (AE) will be evaluated by the Investigator in terms of seriousness, severity (mild, moderate, severe) and relationship (not related, possibly related, probably related, definitely related) to the study device.
Study Design:	This is a single-center, prospective, open label, feasibility study in healthy adult participants. Participants will be monitored for up to 2 hours while using the VCool System to determine if the target temperature of 35.5℃ is reached. If the target core body temperature is reached within 2 hours, the participant will be monitored for an additional hour to determine if the VCool System can maintain the participant's core body temperature between 35℃ and 36℃ for one hour.

	During the study, the participants will have their blood pressure and temperature monitored and will be assessed for shivering and adverse events.
Study Duration:	The overall study participation for each participant will be one visit lasting approximately 4 hours. Study enrollment is anticipated to be completed within two weeks.
Study Device:	VCool Intranasal Cooling System
Proposed Indication for Use:	The VCool Intranasal Cooling System is intended for temperature reduction in adult patients where cooling is indicated.
Eligibility Criteria:	Participants who meet all the inclusion criteria and none of the exclusion criteria are eligible for enrollment in the study. **Inclusion Criteria** 1. Male or female ages 18 to 55. 2. An Institutional Review Board (IRB) approved informed consent is signed and dated prior to any study-related activities. 3. Baseline core body temperature between 36.8°C and 37.5°C. 4. Have the ability to understand the requirements of the study and are willing to comply with all study procedures. 5. In the opinion of the Investigator, are able to participate in the study. **Exclusion Criteria** 1. History of cardiovascular, respiratory, or metabolic disorder 2. Any contraindication to undergoing Magnetic Resonance Imaging (MRI) 3. Pregnant 4. Severe peripheral vascular disease 5. History of Raynaud's disease 6. Currently experiencing a respiratory infection 7. Chronic rhinosinusitis 8. History of sickle cell disease 9. History of cold agglutinin disease 10. History of cryoglobulinemia 11. Known deviated septum or nasal deformity 12. History of nosebleeds or a nosebleed within past 24 hours

	13. Use of antipyretics or other medications affecting body temperature within the past 24 hours.

2 INTRODUCTION

2.1 Background

Inducing normothermia or mild hypothermia is known to have therapeutic benefits in various medical conditions where neuronal damage can occur, including stroke and cardiac arrest patients [1-9]. Among numerous benefits, cooling, known as Targeted Temperature Management (TTM), has been shown to reduce cerebral edema and neurogenic fever. Numerous studies have been completed inducing normothermia or mild hypothermia in healthy volunteers using different cooling technologies including surface and intravascular approaches with no serious adverse events [10-14].This study is the first to use thermoelectrically cooled air delivered through the nasal passages via nasal cannula to reduce core body temperature.

The VCool Intranasal Cooling System has been previously studied in a porcine animal model which demonstrated safety and efficacy [15]. In this study, swine were cooled and met the primary outcome of effective cooling (34°C from 38.1°C) There was no secondary short or medium evidence of injury in CT imaging of the animal's snout anatomy, CT perfusion of the brain, or histological analysis of the brain and snout.

2.2 Study Objective

The objective of this study is to evaluate the performance and safety of the VCool Intranasal Cooling System (VCool) in healthy adult volunteers. The study is designed to gather preliminary data regarding air flow rates and time required to reduce core body temperature to 35.5℃ and maintain the temperature between 35℃ and 36℃ for one hour after cooling. Additional data will be obtained on whether intranasal cooling results has a differential temperature impact on core body temperature as compared to brain temperature.

The data collected will help to develop automated cooling models and the control system algorithm by examining participant cooling response as a function of air flow rate and time the air is supplied to the participants.

2.3 Risk and Benefit Assessment

2.3.1 Potential Risks

The only known study-related risk is the use of a nasal cannula to deliver cold air to the participant. The potential risks associated with this delivery are:

Nosebleed

Shivering

Irritation of the nasal passages

Skin irritation at the site of the cannula

Nasal congestion

Over cooling

Participants will be de-identified using a study specific number at the time of enrollment. All Case Report Forms (CRFs) will be labeled with the de-identified participant identification (ID) number. No identifiable participant identification information will be provided to NeuroIntact.

Information collected for this study is confidential. However, officials of the Food and Drug Administration (FDA) or other regulatory agencies may research records during the ordinary course of carrying out their functions. A Sponsor representative may also inspect the research records to make certain the study data is accurate. The Investigator, regulatory agencies, and the Sponsor will protect the confidentiality of the records.

2.3.2 Risk Mitigation

The protocol has been designed to minimize the risks to study participants enrolled in the study. In general:

- Prior to study enrollment the Investigator must ensure the participant meets the inclusion/exclusion criteria
- Each participant will be fully informed about the conditions of study participation, and the risks and potential benefits of the study procedure and asked to provide an executed Informed Consent prior to being enrolled in the study
- Institutional Review Board (IRB) review and approval of the study protocol, Informed Consent Form, and any other participant-facing documents will be obtained prior to study initiation.
- The participant's core temperature will be continuously monitored with direction to adjust the VCool if the core temperature is <35°C for a sustained period of 5 minutes, to mitigate overcooling of the participant
- If the participant develops a nosebleed, the VCool will be stopped
- Participants will be assessed for shivering and if severe according to the Bedside Shivering Assessment Scale, the VCool flow and temperature will be adjusted

2.3.3 Potential Benefits

The information obtained in this study will not benefit the study participants directly but may aid future patients requiring cooling of body temperature.

2.4 Study Rationale

The two leading categories of TTM devices include surface cooling systems and intravascular cooling systems, both of which have disadvantages. Surface systems limit patient access, are difficult to transport, and can cause skin irritation and shivering. Intravascular systems require an invasive procedure and can lead to infection. Both approaches are also intolerable to awake patients. The VCool Intranasal Cooling System is anticipated to overcome these issues as well as cool patients rapidly and be easy to use.

3 STUDY DEVICE

3.1 Device Description

The VCool Intranasal Cooling System is intended to cool the core body temperature of patients for whom cooling is medically indicated. The VCool System produces a flow of thermoelectrically cooled air from an external source (hospital compressed wall medical air or oxygen). The air flow is delivered from the VCool System to the patient via an insulated air delivery tube. The air delivery tube is insulated to keep the air cooled. A two-prong nasal cannula is attached to the delivery tube and placed in the patient's nostrils. Cooled air enters the nasal passages and exits freely through the nose and mouth. The nasal prongs do not create a seal at the nostrils and therefore do not create excess airway pressure. The VCool System does not utilize invasive catheters, surface cooling pads, or any cooling agents. The VCool System is not used to deliver respiratory support.

The VCool is a small device weighing less than 25 pounds. There is an internal power source, a Peltier (thermoelectric cooling unit), air tubing, and thermistors. The system can deliver air flow rates of up to 40 liters per minute set to a temperature of 5°C at the end of the cannula. All parts of the system are constructed of biocompatible materials and designed for electrical safety. In this study, the VCool will be connected to a medical grade air compressor which will supply the air for cooling. A commercially available nasal cannula will be used to deliver the cooled air.

3.2 Indications for Use

The VCool Intranasal Cooling System is intended for temperature reduction in adult patients where cooling is medically indicated.

4 INVESTIGATIONAL PLAN

4.1 Study Design

This is a single-center, prospective, open label, feasibility study in healthy adult participants. Eligible participants who have signed an informed consent will be enrolled in the study. The participant will be assigned a unique ID number. After obtaining relevant demographic information, testing will ensue.

Baseline vital sign measurement (heart rate, temperature, blood pressure, respiratory rate) will be obtained. The participant will then undergo continuous MRI of the brain to include sequences that enable thermography. Baseline imaging sequences will take approximately 30 minutes. The the Investigator or designee will turn on the VCool System, while the VCool remains outside of the MRI suite, for up to two hours or until the target temperature of 35.5℃ is reached. Body temperature will be continuously measured using a MRI compatible thermometer. During the cooling period and once endpoint is reached the the participant will undergo repeated MRI with thermography. Once study endpoint is reached, the participant will be taken off of VCool and MRI thermography. If the target core body temperature is reached before 2 hours, the participant will be monitored until two hours are completed to determine if the VCool System can maintain the participant's core body temperature between 35℃ and 36℃ for one hour.

During the study, the participants will have their temperature monitored and will be assessed for shivering and adverse events.

Study participation is one visit lasting approximately 4 hours.

4.2 Study Site

The study will be conducted at the University of Maryland School of Medicine HSF III research facility.

4.3 Participant Enrollment

Participants will be screened for inclusion in the study to ensure that 10 participants complete the study.

5 STUDY ENDPOINTS

5.1 Primary Endpoints

1. To confirm the VCool System can lower core body temperature to 35.5℃ within 2 hours.

2. To identify whether cooling with the VCool System results in a gradient between brain and core body temperature.

3. To confirm the VCool System can maintain a core body temperature between 35℃ and 36℃ for one hour after cooling.

5.2 Secondary Endpoint

4. To assess the shivering experienced by participants based on the 4-point Bedside Shivering Assessment Scale (BSAS).

5.3 Safety Considerations

All treatment emergent adverse events (TEAEs) reported by participants will be tabulated.

Each adverse event (AE) will be evaluated by the Investigator in terms of seriousness, severity (mild, moderate, severe) and relationship (not related, possibly related, probably related, definitely related) to the study device.

6 STUDY POPULATION

6.1 Inclusion Criteria

Participants are eligible for enrollment if all the inclusion criteria are met:

1. Male or female ages 18 to 55

2. An Institutional Review Board (IRB) approved informed consent is signed and dated prior to any study-related activities

3. Core body temperature between 36.8℃ and 37.5℃

4. Have the ability to understand the requirements of the study and are willing to comply with all study procedures

5. In the opinion of the Investigator, are able to participate in the study

6.2 Exclusion Criteria

Participants are not eligible for enrollment if any of the exclusion criteria apply:

1. History of cardiovascular, respiratory, or metabolic disorder.
2. Any contraindication to undergoing Magnetic Resonance Imaging (MRI)
3. Pregnant
4. Severe peripheral vascular disease
5. History of Raynaud's disease
6. Currently experiencing a respiratory infection
7. Chronic rhinosinusitis
8. History of sickle cell disease
9. History of cold agglutinin disease
10. History of cryoglobulinemia
11. Known deviated septum or nasal deformity
12. History of nosebleeds or a nosebleed within past 24 hours
13. Use of antipyretics or other medications affecting body temperature within the past 24 hours

6.3 Study Duration

Each participant's involvement in the study will be one study visit lasting approximately 4 hours. No follow-up or subsequent participant involvement is required. Study enrollment is expected to be completed within 8 weeks.

7 STUDY PROCEDURES

7.1 Pre-screening

Potential participants will be pre-screened by a designated member of the study team to determine if the potential participant meets any of the exclusionary criteria based on medical history. If the participant is determined to be potentially eligible for participation, they will be scheduled for a screening visit at the study site.

7.2 Screening

7.2.1 Informed Consent

Study staff trained in basic human research subjects protection will explain the study protocol, procedures, and objectives to the volunteer and obtain written informed consent for participation in the study prior to performing any study procedures, in accordance with the requirements outlined in the Code of Federal Regulations (CFR) Title 45, Part 46 and Title 21, Part 50. This will be done by reviewing all the information contained in the informed consent form (ICF), allowing the volunteer ample time to review the form, ask questions, and have their questions answered to the satisfaction of the volunteer. The ICF will be presented to the volunteer in a language that is fully comprehensible to them.

Volunteers wishing to participate will be asked to sign and date the ICF and will be provided with a copy for their records. The original will be maintained with the participant's study records.

7.3 Study Enrollment

7.3.1 Participant Identification

If the volunteer is willing to participate in the study and has signed and dated the ICF, they will be assigned a unique ID number. The ID number is a 2-digit number assigned sequentially (i.e., 01, 02, 03). The ID number will be written on each page of the CRF, and all other documents related to the participant.

The site will maintain a screening and enrollment log for participant identification, to which all consented participants will be added. A participant is considered enrolled when they have signed the ICF. If during the screening process a participant fails to fulfill any of the eligibility criteria, this will be documented, and the participant will be classified as a screen failure. Volunteers will continue to be screened until 10 participants have completed the study.

The Principal Investigator (PI) or designee at each site will maintain a master log that will correlate each participant ID number to the participant's name. The log will remain with the PI and will be confidential.

7.3.2 Participant Stipend

Participants will be issued a stipend of 200$ for their participation in the study.

7.4 Eligibility

The inclusion and exclusion criteria will be reviewed. Participants are eligible for participation in the study if all the inclusion criteria are met and none of the exclusion criteria are met. If the participant fails to fulfill any of the eligibility criteria, this will be documented. The PI or designee will sign and date the CRF indicating agreement with the determination of eligibility.

If the participant fails to fulfill any element of the inclusion and exclusion criteria, the participant will be classified as a screen failure and will not be advanced any further into the clinical study.

7.5 Baseline Data

The following demographic information will be obtained and documented on the CRF for all enrolled participants:

Age

Sex

Race

Ethnicity

Height (inches)

Weight (pounds)

Wipe the lateral forehead with an alcohol wipe. Place the 3M SpotOn® zero-heat-flux temperature sensor on the participant's forehead.

Record the patient temperature (°C) using the 3M SpotOn® zero-heat-flux forehead sensor, the Braun ThermoScan 7 tympanic sensor, and an MRI compatible axillary sensor.

Remove the 3M SpotOn® zero-heat-flux forehead sensor and the Braun ThermoScan 7 tympanic sensor before escorting to MRI suite.

7.6 Procedure

7.6.1 VCool System Set Up

Attach the disposable delivery tube to the VCool System

Install the gas inlet filter on the back with the filter bowl vertical (glass side down)

Attach the air hose to the gas inlet filter

Connect the power cord to a grounded outlet

Turn the power switch to "ON"

Connect the RS232 cable to the VCool System and to the laptop computer

Turn on the computer

Select the following:

TC-48-20 program.

COM5

Communications

Set Point and set value to -19.0

Controller options

Save data to file

Go to file Explorer, select LabView Data

Select New and enter participant identification number as file name

Save data to file

Turn on gas and set flow meter to 35 liters per minute.

Confirm temperature is set to -10°C

Confirm gas temperature setting at the end of the delivery tube is -5°C

Attach nasal cannula to the delivery tube

7.6.2 MR Imaging procedure

- Participants will undergo MRI at the HSF3 research facility at the University of Maryland, Baltimore (UIMB). Each imaging session will last approximately 90 minutes. The protocol will include high-resolution anatomical MRI for structural reference and tissue segmentation; MR Spectroscopy (MRS) to measure metabolite and water resonance frequencies for temperature estimation; MR Thermometry to measure phase changes over time to track temperature dynamics; Perfusion MRI to measure cerebral blood flow (CBF); as well as other structural and functional imaging sequences. These sequences will be repeated at multiple time points during the controlled cooling period to monitor temporal changes in temperature, perfusion, and tissue properties.

7.6.3 Participant Set Up and Monitoring

Have the participant lie on their back in an inclined position and place the nasal cannula on the participant, securing the straps around their head

Record the temperature from both temperature sensors and record the time

Record the flow rate

Initiate the cooling and record the time. The induction phase for participant cooling can take up to 2 hours.

Record the core body temperature every 2 minutes from both temperature sensors and record the time. The core body temperature is monitored throughout the study.

If the core body temperature is <35℃ for 5 minutes:

1. Remove nasal cannula, if temperature starts to rise after 2 minutes keep cannula off until temperature gets back to 35℃. Then reapply nasal cannula and resume treatment.

2. If temperature continues to fall below 35℃ after 2 minutes, cover patient with electric blanket that has been pre heated on standby. Keep blanket on until temperature gets back to 35℃.

Assess for shivering every 10 minutes using the BSAS. Record the results and time. The BSAS is recorded throughout the study.

Bedside Shivering Assessment Scale

0	None
1	Mild: Shivering localized to neck/thorax
2	Moderate: Intermittent involvement of the upper extremities and/or thorax
3	Severe: Generalized shivering or sustained upper and/or lower extremity shivering

The flow can be increased to 40 liters per minute after 10 minutes

Record any VCool System adjustments to flow rate or temperature and record the new flow rate and temperature and time of adjustment. Record the reason for the adjustment.

Assess the participant for adverse events (AEs) throughout the study

If the participant has a nosebleed, discontinue the study and note the time.
Monitor patient at first sign of shivering

- If **Mild,** cover patient with regular blanket.
- If **Moderate or severe –** discontinue cooling treatment.

Continue to record the temperature until either temperature sensor reaches 35.5℃ or the VCool System has been used for 2 hours. Note the time the target temperature was achieved.

If the participant's core body temperature does not reach 35.5℃ in 2 hours, the study is concluded

- Remove the nasal cannula from the participant and dispose of it properly
- Monitor the participant's core body temperature until it returns to baseline. Blankets can be applied to help with warming.
- Remove the axillary thermometer

If the participant's core body temperature reaches 35.5℃, proceed to the next steps

Maintain the target temperature of 35.0℃ to 36.0℃ for 1 hour:

Adjust flow rate by 5 liters per minute from baseline every 2 minutes

- If temperature falls below 35℃., reduce flow rate by 5 liters per minute every 2 minutes until temperature rises back to 35℃
- If temperature rises above 36℃, increase flowrate 5 liters per minute every 2 minutes until temp falls below 36℃

Remove the nasal cannula and the air delivery tube and dispose of them properly

Once out of the MRI suite, record the patient temperature (℃) using the 3M SpotOn® zero-heat-flux forehead sensor, the Braun ThermoScan 7 tympanic sensor

Monitor the participant's core body temperature until it returns to baseline. Blankets can be applied to help with warming.

Remove the 3M SpotOn® zero-heat-flux forehead sensor and the Braun ThermoScan 7 tympanic sensor before escorting to MRI suite.

7.6.4 VCool System Shut Down

Turn the power switch on the VCool System to "OFF"

Turn off the flow meter

Remove the medical air hose from the flow meter

Disconnect the power cord

Wipe the exterior of the VCool System using the provided Caviwipes

Air delivery tube and cannula are disposable and should be discarded

8 EARLY PARTICIPANT WITHDRAWAL OR DISCONTINUATION

Participants may discontinue the study due to consent withdrawal or withdrawal due to an AE. The Investigator may elect at any time to discontinue a participant for any reason if such a decision is in the participant's best medical interest or it is determined the participant is not

compliant with the protocol. If a participant discontinues the study for any reason, or is withdrawn by the Investigator, the primary reason for termination or discontinuation should be documented on the CRF.

9 EARLY STUDY END

If the study is ended prematurely or suspended:

- The Sponsor or representative will promptly inform the Investigators of the termination or suspension and the reasons and inform the regulatory authority(ies) (where required by applicable regulatory requirements)
- The IRBs will be promptly informed and provided with the reasons(s) for termination or suspension by the Sponsor or by the Investigator

If the Investigator (or IRB) terminates or suspends the investigation without prior agreement of the Sponsor:

- The Investigator will promptly inform the Sponsor and the IRB, provide a detailed written explanation of the termination or suspension, and, if feasible, designate another qualified Investigator to be trained and continue follow-up of already-enrolled participants. The new Investigator/site would be requalified by the Sponsor or representative. Participants would be re-consented.
- The Investigator will inform the institution (where required by applicable regulatory requirements
- The Investigator will promptly inform the participants and ensure appropriate therapy and follow-up for the participants
- The Sponsor or representative will inform the regulatory authority(ies) (where required by applicable regulatory requirements)

10 ADVERSE EVENTS

The Investigator is responsible for the detection and documentation of events meeting the definition of an AE or SAE as provided in this protocol. Only treatment emergent AEs and SAEs, occurring after randomization through the end of the study visits will be recorded. All AEs, including SAEs that occur during the participant's study participation, must be documented in the participant's source document and CRF.

10.1 Definition of an Adverse Event

An adverse event is any untoward medical occurrence, associated with the use of an investigational product, whether or not considered related to the investigational product. An AE is any unfavorable and unintended sign, symptom or disease temporally associated with the use of an investigational product and does not imply any judgement about causality. AEs will be considered study-related if the event follows a reasonable temporal sequence from a study-

related procedure or device and could readily have been produced by the study procedure or device.

An event that emerges during treatment having been absent pre-treatment or worsens relative to the pre-treatment state should be reported as a TEAE.

Laboratory or functional test abnormalities generally are not considered AEs unless they are associated with clinical signs or symptoms or require medical intervention.

An AE includes:

- Exacerbation of a pre-existing illness
- Increase in frequency or intensity of a pre-existing episodic event or condition
- Condition detected or diagnosed after investigational product placement even if present prior to the start of the study
- Continuous persistent disease or symptoms present at Baseline that worsen following the start of the study

Adverse events occurring to any study participant during study participation will be reported by the site to the study Sponsor or representative by means of the AE CRF. The AE CRF includes severity, seriousness, action taken, causality and outcome of each event. The Medical Monitor will determine if the AE meets the threshold for a Major Adverse Event (MAE), will verify UADE status, and will determine the relatedness of the event to the device and/or procedure.

All adverse events will be coded and summarized by tabulating the number and percentage of participants reporting each event (AE, SAE, and UADE) for the safety population. An AE that is related to the device is called an Adverse Device Effect (ADE).

10.2 Definition of a Serious Adverse Event

A serious adverse event (SAE) or serious adverse device effect (SADE) is any AE that resulted in any of the following outcomes:

- Death
- Life-threatening -the participant was, in the opinion of the Investigator, at immediate risk of death from the AE as it occurred
- Disability or incapacity
- Inpatient hospitalization* or prolongation of existing hospitalization
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered an SAE when, based upon appropriate medical judgement, they may jeopardize the participant and may require medical or surgical intervention to prevent one of the outcomes listed in this definition
- Fetal distress, fetal death, or congenital abnormality

*Planned hospitalization for an existing condition is not an SAE. Hospitalization is defined as greater than 24 hours in the hospital.

An SAE that is related to the device is called a serious adverse device effect (SADE).

10.3 Severity of Adverse Events and Serious Adverse Events

The severity of each AE or SAE must be assessed by the Investigator or designee.

- Mild: The AE is transient or causes mild discomfort. There usually is no intervention/therapy required and the AE does not interfere with the participant's normal activities.
- Moderate: The AE causes some limitation in activity and some assistance may be needed. Treatment for the AE may be needed.
- Severe: The AE causes marked limitation in activity. The participant's usual daily activity is interrupted. The participant may require medical intervention/therapy; hospitalization is possible.

10.4 Outcome of Adverse Events and Serious Adverse Events

The outcome of each AE and SAE must be noted in the source documents and CRF.

- Not recovered / not resolved – the event has not improved
- Recovered / resolved – the participant has recuperated from the event
- Recovered / resolved with sequelae – the participant has recuperated from the event but retained pathological conditions resulting from the event
- Recovering / resolving – event is improving

- Unknown – either not known, not observed, not recorded, or participant refused to provide information

- Fatal – death as a result of the AE

10.5 Assessment of Relatedness to Study Product

The Investigator or designee will assess each AE and SAE for causality based on their best medical judgement, the observed symptoms associated with the event, and the available information on the investigational product.

- Not Related: No evidence of any causal relationship with the investigational device or trial intervention.

- Possibly Related: Some evidence to suggest a causal relationship between the AE and the investigational device or trial intervention, but other factors may have contributed to the event.

- Probably Related: Evidence suggests a causal relationship with the investigational device or trial intervention; the influence of other factors is unlikely.

- Definitely Related: Evidence suggests a causal relationship to the investigational device or trial intervention, i.e., follows a known or suspected pattern of response, or is otherwise logically related to the investigational device or trial intervention, and no alternative cause is present.

10.6 Reporting Serious Adverse Events

Prompt notification of the Sponsor regarding SAEs is essential so that regulatory responsibilities and legal obligations can be satisfied. The Investigator must report SAEs according to the following time frames.

- Initial notification of all SAEs based on the available information must be provided to the Sponsor or designee by phone within 24 hours of the investigational site learning of the event. Initial notification within 24 hours of death or life-threatening events is extremely important.

- Follow-up information, when available, must be sent to the Sponsor or designee within 48 hours of receipt of the information by the investigational site

The Investigator must comply with all applicable local regulatory requirements related to reporting SAEs to the IRB.

The Investigator should institute appropriate therapeutic and follow-up measures in accordance with good medical practice and should notify the monitor of such actions and record them in the participant's CRF.

10.6.1 Serious Adverse Event Information to Report

At a minimum, SAE reports must contain the participant ID number, the serious adverse event term, onset date, relationship to study product, and a brief description of the event. Causality should be included in the initial report.

11 POTENTIAL ANTICIPATED ADVERSE EVENTS

The following potential anticipated AEs are transient and may be associated with use of the VCool Intranasal Cooling System:

- Nosebleed
- Shivering
- Irritation of the nasal passages
- Skin irritation at the site of the cannula
- Nasal congestion

11.1 Immediate Post-Procedure Observations

There are observations that are expected events immediately post-procedure that would be expected with any procedure. Any event that does not require medical management will not be considered significant but will be documented and reported.

11.2 Unanticipated Adverse Device Effect

An Unanticipated Adverse Device Effect (UADE) is any serious adverse effect on health or safety or any life-threatening problem or death caused by, or associated with use of the VCool device, if that effect, problem, or death is not identified in nature, severity, or degree of incidence in this Investigational Plan (protocol) or any other unanticipated serious problem associated with the VCool device that relates to the rights, safety, or welfare of the participants participating in this study.

11.3 Reporting

All AEs, UADEs, and SAEs will be reported via the CRFs. All AEs meeting the above definitions of an UADE and/or SAE, whether device-related or not, must also be reported to the study Sponsor within 24 hours of discovery and to the approving IRB as soon as possible, based on IRB requirements but in no event later than 10 working days after the Investigator first learns of the AE or effect.

12 DATA ANALYSIS

12.1 Primary Analysis

The time and flow rate that is required to reduce and maintain each participant's core temperature to 35.5℃ from measured baseline temperature will be calculated. The total number of participants who reach the target temperature in 2 hours or less will be calculated.

12.2 Secondary Analysis

The incidence of any shivering will be analyzed using the BSAS. The incidence and nature of any adverse events or participant intolerance will be tabulated.

12.3 Populations for Analysis

The study population will consist of 10 healthy volunteers who have signed the ICF and who meet all the inclusion criteria and none of the exclusion criteria.

12.4 Imaging Data analysis

All MRI data will be spatially aligned to the structural images to ensure accurate anatomical localization of measurements. Spectroscopic data will be reconstructed to obtain resonance frequencies for water and metabolites (N-acetylaspartate, choline, creatine). Temperature will be estimated based on the chemical shift difference between the water peak and metabolite peaks, which is temperature-dependent. Phase images from the thermometry acquisition will be unwrapped and converted to temperature change maps using the proton resonance frequency (PRF) shift method. These maps will capture dynamic temperature changes over the cooling period. CBF maps will be generated using standard perfusion modeling (e.g., ASL kinetic modeling) to quantify cerebral blood flow. Temporal changes in CBF will be evaluated across the cooling period. Imaging-derived measures (temperature, Δtemperature, CBF) will be extracted from predefined anatomical regions of interest or whole-brain parcellations to characterize regional variability in cooling response.

13 RESPONSIBILITIES

The PI or designee must ensure and maintain adequate and accurate records of all observations and other data pertinent to the clinical study for each study participant. The PI or designee will make all appropriate safety assessments on an ongoing basis. The PI or designee will prepare and maintain adequate and accurate study source documents (medical records, AE, raw data collection forms, etc.) designed to record all observations and other pertinent data for each participant. The PI will allow Sponsor representatives, contract designees, authorized regulatory authority inspectors, and the IRB to have direct access to all documents pertaining to the study.

13.1 Data Quality Assurance

Standardized paper CRFs will be used at the investigational site. The PI is responsible for ensuring completion and timely submission of the forms to the study Sponsor or designee for data processing. All data are reviewed to identify inconsistent or missing data and AEs. All

hard copy forms will be secured to ensure confidentiality. The PI is required to maintain all source documents as required by the protocol, including any applicable supporting medical records and Informed Consents. The source documents may be reviewed to verify information submitted on the CRFs.

13.2 Protocol Deviations

All protocol deviations (i.e., instance[s] of failure to follow, intentionally or unintentionally, the requirements of this protocol) must be documented in the appropriate Protocol Deviation CRF with an explanation for the deviation.

The Sponsor or representative is responsible for assessing protocol deviations in an ongoing manner and determining their impact on the conduct of the study. After evaluation of each deviation, the Sponsor or representative will determine if corrective and preventive actions need to occur.

Minor Deviations: A minor deviation does not impact the participants' rights, safety, or well-being, or the completeness, accuracy, or reliability of the study data. Minor deviations shall be documented in the study database along with a full description of the event and outcome. The Sponsor will analyze these deviations and assess their significance.

Major Deviations: A major deviation is a deviation from the protocol that may impact the participants' rights, safety, or well-being, or the completeness, accuracy or reliability of the study data related to key endpoints, or a significant deviation from national or local regulations or IRB guidelines. Major deviations should be reported to the Sponsor within 48 hours of site awareness of the event and must be documented in the study database along with a full description of the event and outcome. The Sponsor will analyze these deviations and assess their significance.

Major protocol deviations should be reported to the IRBs and the regulatory authorities as per national or local regulations.

14 PARTICIPANTS LOST TO FOLLOW-UP

Participants will be followed up for 24 hours for any unresolved adverse events. A participant will be considered lost to follow-up if reasonable efforts made to contact the participant have failed. At least three separate attempts should be made to contact the participant. All attempts to contact the participant will be documented with the last attempt made via registered mail.

15 INSTITUTIONAL REVIEW BOARD

Requirements for institutional review as set forth in Title 21 of the Code of Federal Regulations (21 CFR) or other relevant local regulations for institutional review will be followed. The Investigational Plan (Protocol), Informed Consent and other required documents must be approved by the IRB before enrollment of participants in the study. The Sponsor or representative must confirm that the IRB is in compliance with the general standards for the composition, operation, and responsibility of an IRB as set forth in ICH Guidelines for GCP, Sections 3.1 to 3.4, and 21 CFR, Part 56. Any member of the IRB who is directly affiliated with

this study as an Investigator or as site personnel must abstain from the IRB vote on the approval of the protocol. The PI, in collaboration with the Sponsor or representative, will be responsible for reporting to the IRB all changes in research activity, including protocol amendments, safety reports, all unanticipated problems involving risks to participants, and study termination. The PI or delegate will also be responsible for submitting progress reports to the IRB at regular intervals appropriate to the degree of patient risk involved, but no less than once per year as required by the IRB. Copies of all IRB notifications and approvals will be forwarded to the Sponsor or representative.

16 INVESTIGATOR REPORTS

The PI will be responsible for the following reports:

- AEs: SAEs, SADEs, and UADEs shall be reported to the Sponsor by fax or email within 24 hours of the Investigator becoming aware of the event
- All other AEs will be reported to the Sponsor via the Adverse Event CRF.
- AEs must also be reported to the reviewing IRB in compliance with its reporting requirements. For serious adverse events (whether or not device-related), this is typically no later than 10 business days of the Investigator becoming aware of the event.
- An Investigator shall submit to the reviewing IRB a report of any UADE occurring during an investigation as soon as possible, but in no event later than 10 working days after the Investigator first learns of the effect
- Protocol Deviations: The Investigator is responsible for reporting all protocol deviations to the Sponsor and to the IRB in compliance with the IRB's reporting requirements. The Sponsor will provide a deviation log and CRF to document protocol deviations.
- Withdrawal of IRB Approval: The Investigator shall report to the Sponsor within 5 working days if, for any reason, the IRB withdraws approval to conduct the investigation. The report will include a complete description of the reason(s) for which approval was withdrawn.

Other Reports: Upon request of the Sponsor, the FDA, or the IRB, the Investigator shall provide accurate, complete, and current information.

17 TRAINING

The Sponsor or representative will train all site staff on the device, protocol, and study procedures in accordance with their responsibilities.

As necessary, training will occur after any revisions to the device, Instructions for Use (IFU), protocol, or study procedures.

18 STUDY DEVICE ACCOUNTABILITY

The Investigator or designated site staff must maintain device accountability records throughout the course of the study. The PI will not supply the investigational device to other Investigators not participating in the study and the investigational device will be used only on consented participants as directed by this protocol. Use of the VCool Intranasal Cooling System, other than as directed by this protocol, is not allowed.

Device accountability will indicate receipt, use, and final disposition of each device. The Investigator or designee will complete a device accountability CRF.

19 ETHICS/PROTECTION OF HUMAN PARTICIPANTS

19.1 Ethical Conduct of the Study

The study will be conducted in compliance with the ICH Guidelines on GCP, 21 CFR 812 and ISO14155 Standards.

19.2 Participant Confidentiality

All clinical study findings and documents will be regarded as confidential. Study documents (protocols and other material) will be stored appropriately to ensure their confidentiality. Investigator and members of their research team (including the IRB) must not disclose such information without prior written approval from the Sponsor, except to the extent necessary to obtain informed consent from participants who wish to participate in the trial or to comply with regulatory requirements. The anonymity of participating participants must be maintained. Participants will be specified on study documents by their unique number and initials, not by name. Documents that identify the participant (e.g., the signed informed consent document) must be maintained in confidence by the Investigator.

20 ADMINISTRATIVE CONSIDERATIONS

20.1 Protocol Approval and Amendment(s)

No modifications to the protocol may be made without the written consent of the Sponsor. To alter the protocol, amendments must be written, released by the Sponsor, and approved by the IRB prior to implementation (as appropriate). Administrative protocol changes or protocol clarifications may be made without the need for a formal amendment, but these changes will be documented in an integrated clinical study report.

20.2 Case Report Form Completion

A Case Report Form (CRF) must be completed for every participant that signs an ICF. Either the PI or sub-investigator must sign and date the completed CRF within a reasonable time period after completion of data collection. If a participant does not complete the study, the reason for withdrawal must be documented.

The investigator is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported. All data collection should be completed in a neat, legible manner to ensure accurate interpretation of data.

Data collection and accurate documentation are the responsibility of the study staff under the supervision of the investigator. Data recorded on paper must use black ink and any changes or corrections made must be done by drawing a single line through the original entry. The corrected entry must be initialed and dated by the individual making the corrections and provide a reason for the change. **DO NOT ERASE, OVERWRITE, OR USE CORRECTION FLUID OR TAPE ON THE ORIGINAL**.

20.3 Archiving Study Documents

The PI shall maintain the records for this study for a period of 2 years after the latter of the following 2 dates: the date on which the investigation is terminated or completed, or the date that the records are no longer required for purposes of supporting a premarket approval application, a premarket notification submission, or a request for De Novo classification.

The Investigator will notify the Sponsor in the event he/she relocates, or for any reason desires to dispose of the records.

All source documents must be retained. Source documents (paper or electronic) are the original records of clinical findings, observations, or other activities in a clinical study necessary for the reconstruction and evaluation of that trial. Examples of source documents include but are not limited to hospital and clinic medical records, signed informed consent forms, evaluation checklists, scan or laboratory results, etc.

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